

Supplement

Kinetic Resolution of Cyclic β -Amino Esters with Lipase B from *Candida antarctica* on Magnetic Nanoparticles

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1 General

The materials and methods used are described in detail in Section 4 (Experimental Section) of the main article.

2 Characterization of compounds

2.1 Gas chromatography

Details of the gas chromatography methods are provided in Section 4.2 of the main article, and in Tables S1 and S2. Methyl *tert*-butyl ether (MTBE, 500 μ L) was added to samples (5 μ L) taken from the CaLB-MNP-catalyzed acylation reactions (see Section 4.8 of the main article), followed by the addition of acetic anhydride (10 μ L) to the mixture at room temperature. The diluted sample was then shaken in

a shaker at 350 rpm for 30 min (to convert the unreacted fraction of amines **1a–c** into *N*-acetamides **1*a–c**), and the reaction was quenched with a drop of distilled water. After drying over Na₂SO₄, the enantiomeric composition of the samples was analyzed by GC using a chiral column (see representative GC results in Figs. S1–S13).

2.2 NMR

Details on the NMR measurements can be found in Section 4.2 of the main article. The experimental ¹H-NMR and ¹³C-NMR spectra of the products are shown in Figs. S20–S36.

Table S1 GC methods, and retention times of acetamides [(*S*)- and (*R*)-**1*a–c**] and amides [(*S*)- and (*R*)-**2a–c**] in quantitative GC analysis on Hydrodex β -6TBDM column

Substrate + acylating agent	Temperature program	Retention times ^a [min]				
		(<i>S</i>)- 1*a–c	(<i>R</i>)- 1*a–c	(<i>S</i>)- 2a–cE / <i>-C</i> / <i>-M</i>	(<i>R</i>)- 2a–cE / <i>-C</i> / <i>-M</i>	($\pm$)- 2a–cE / <i>-C</i> / <i>-M</i>
($\pm$)- 1a + M	140–190 °C, 3 °C min ⁻¹ ; 190 °C, 7 min	7.8	7.9	-	-	19.8
($\pm$)- 1b + M	140–190 °C, 3 °C min ⁻¹ ; 190 °C, 7 min	10.9	11.1	..**	..**	..**
($\pm$)- 1c + M	140–160 °C, 0.9 °C min ⁻¹ ; 160 °C, 25 min	20.8	20.5	..**	..**	..**
($\pm$)- 1a + C	140–190 °C, 3 °C min ⁻¹ ; 190 °C, 7 min	7.8	7.9	-	-	18.3
($\pm$)- 1b + C	140–190 °C, 3 °C min ⁻¹ ; 190 °C, 7 min	10.9	11.1	..**	..**	..**
($\pm$)- 1a + E	145 °C, 25 min	10.2	10.5	23.1	23.5	-
($\pm$)- 1b + E	140–170 °C, 3 °C min ⁻¹ ; 170–180 °C, 1 °C min ⁻¹	10.9	11.0	-	-	16.9
($\pm$)- 1c + E	140–160 °C, 0.9 °C min ⁻¹ ; 160 °C, 15 min	20.5	20.2	-	-	32.4

^a The acetamides **1*a–c** were prepared by derivatization of the residual amines **1a–c** in the samples from enzymatic reactions with acetic anhydride as described in Section 2.1 of this Supplement. ** No detectable product formation was observed under the applied reaction conditions.

Table S2 GC methods, and retention times of acetamides [(*S*)- and (*R*)-**1**b*-c**] and amides [(*S*)- and (*R*)-**2*b*-cE**] in quantitative GC analysis on Hydrodex β -6TBDAC column

Substrate + acylating agent	Temperature program	Retention times ^a [min]				
		(<i>S</i>)- 1*<i>a</i>-c	(<i>R</i>)- 1*<i>a</i>-c	(<i>S</i>)- 2<i>b</i>-cE	(<i>R</i>)- 2<i>b</i>-cE	($\pm$)- 2<i>a</i>-cE
($\pm$)- 1<i>b</i> + E	170 °C, 30 min	9.9	10.2	-	-	19.0
($\pm$)- 1<i>c</i> + E	150–180 °C, 1.5 °C min ⁻¹ ; 180 °C, 10 min	16.8	16.5	-	-	23.7

^a The acetamides **1**b*-c** were prepared by derivatization of the residual amines **1*b*-c** in the samples from enzymatic reactions with acetic anhydride as described in Section 2.1 of this Supplement.

2.3 Representative GC data for biocatalysed kinetic resolution of various cyclic β -amino acids ($\pm$)-**1*a***

Details on the GC measurements can be found in Section 2.1 of the Supplement. Representative chromatograms for KRs of ($\pm$)-**1*a*-c** with ester **E** are shown in Figs. S1–S20.

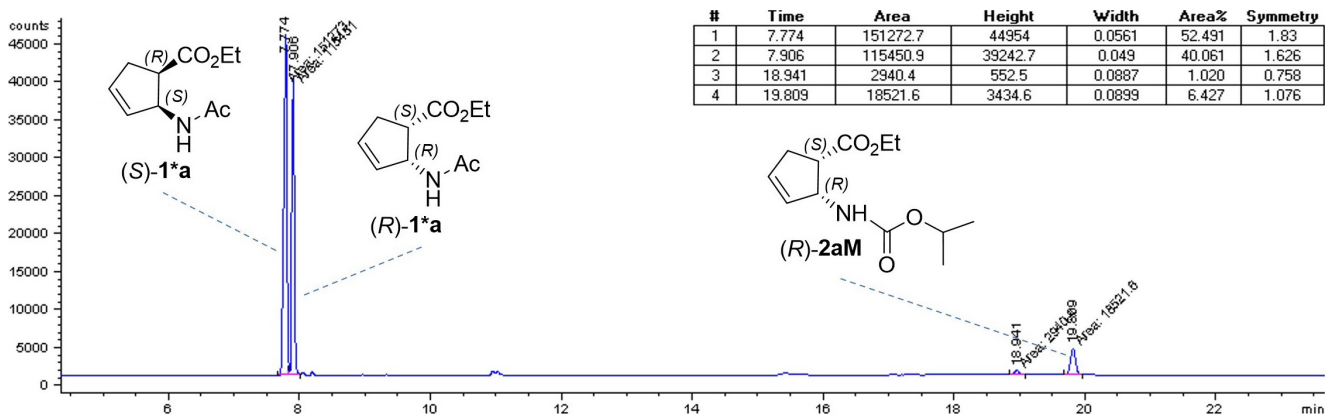


Fig. S1 Chromatogramm of CaLB-MNP catalyzed *N*-acylation of ($\pm$)-ethyl *cis*-2-aminocyclopent-3-enecarboxylate ($\pm$)-**1*a*** with diisopropyl malonate **M** after 24 h on Hydrodex β -6TBDM column

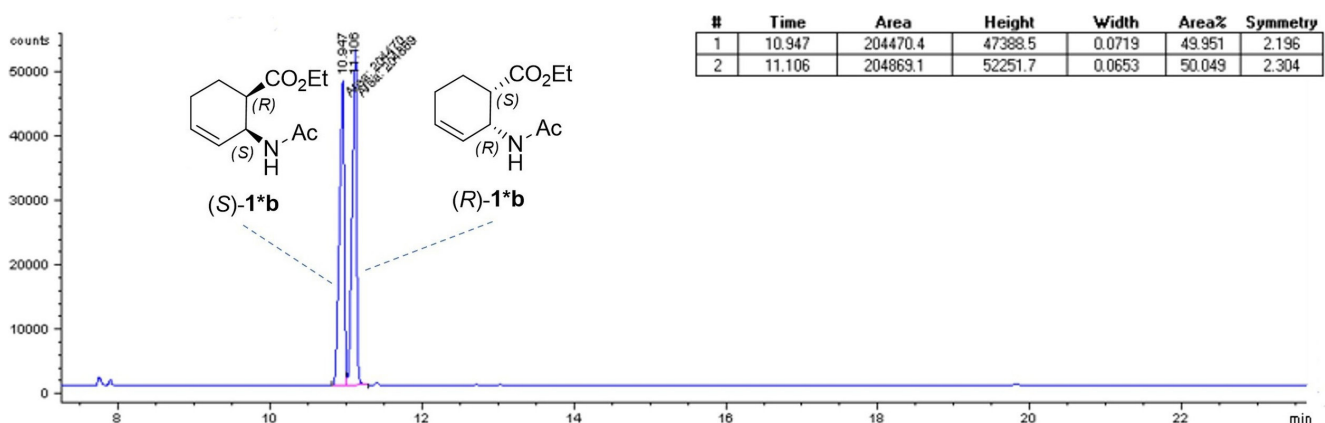


Fig. S2 Chromatogramm of CaLB-MNP catalyzed *N*-acylation of ($\pm$)-ethyl *cis*-2-aminocyclohex-3-enecarboxylate ($\pm$)-**1*b*** with diisopropyl malonate **M** after 24 h on Hydrodex β -6TBDM column

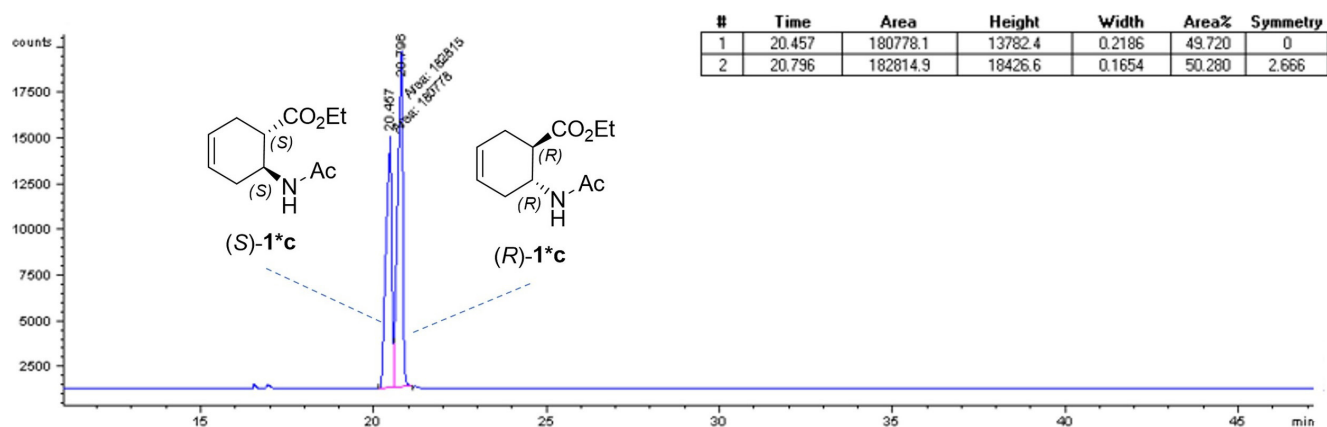


Fig. S3 Chromatogram of CaLB-MNP catalyzed *N*-acylation of (±)-ethyl *trans*-6-aminocyclohex-3-enecarboxylate (±)-**1c** with diisopropyl malonate **M** after 24 h on Hydrodex β -6TBDM column

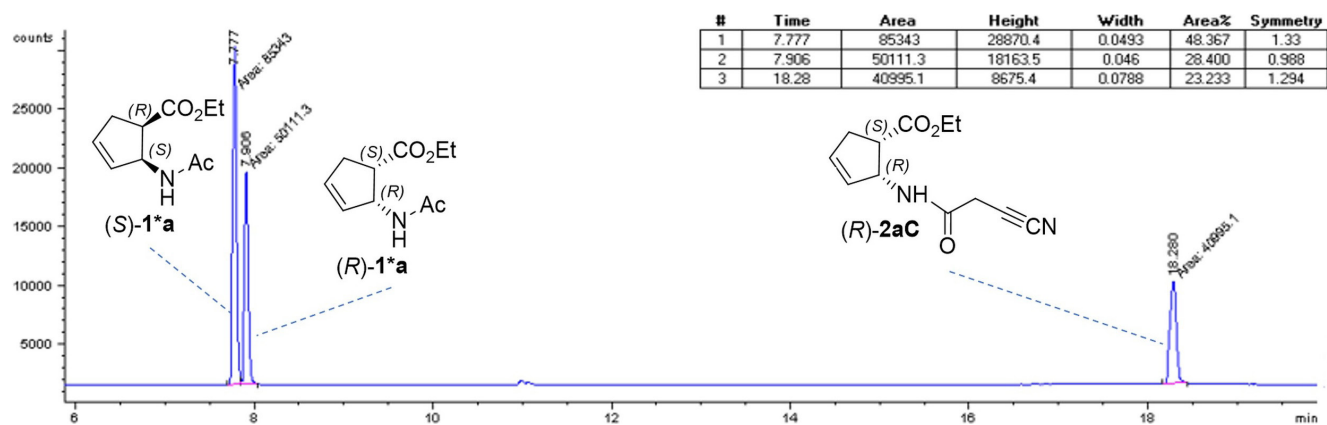


Fig. S4 Chromatogram of CaLB-MNP catalyzed *N*-acylation of (±)-ethyl *cis*-2-aminocyclopent-3-enecarboxylate (±)-**1a** with ethyl 2-cyanoacetate **2c** after 24 h on Hydrodex β -6TBDM column

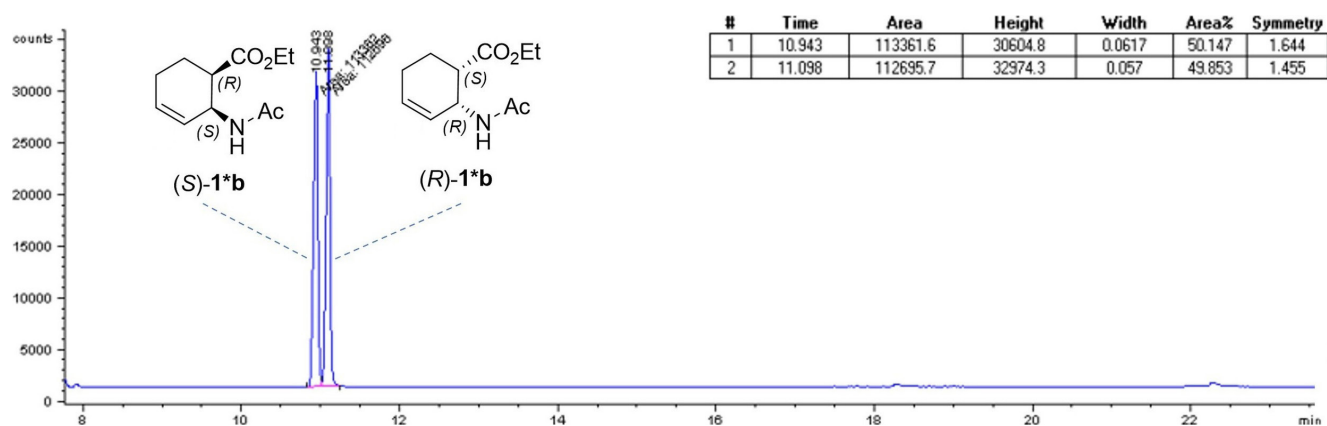


Fig. S5 Chromatogram of CaLB-MNP catalyzed *N*-acylation of (±)-ethyl *cis*-2-aminocyclohex-3-enecarboxylate (±)-**1b** with ethyl 2-cyanoacetate **C** after 24 h on Hydrodex β -6TBDM column

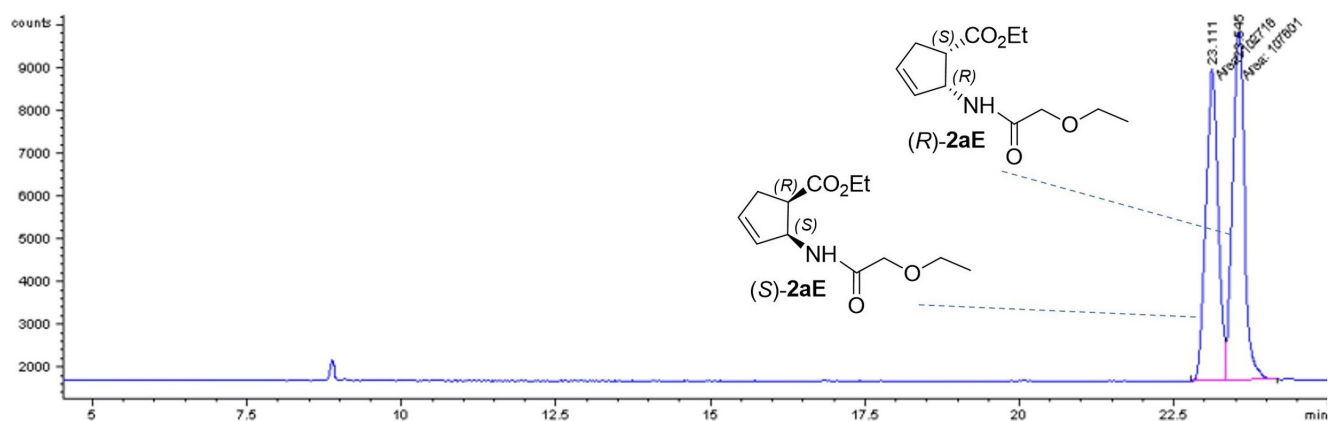


Fig. S6 Chromatogram of (±)-ethyl *cis*-2-(2-ethoxyacetamido)cyclopent-3-ene-1-carboxylate (±)-2aE on Hydrodex β-6TBDM column

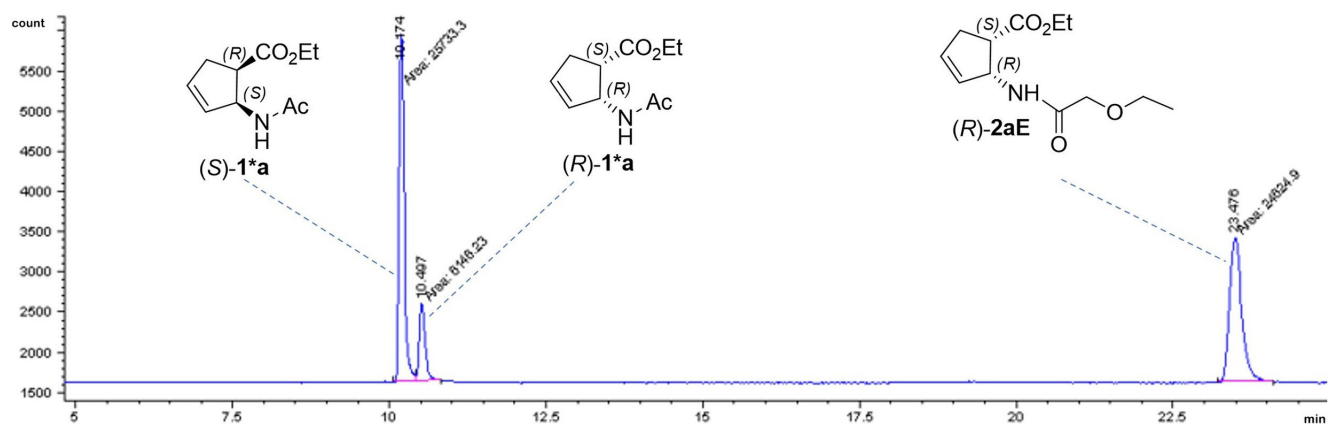


Fig. S7 Chromatogram of CaLB-MNP *N*-acylation of (±)-ethyl *cis*-2-aminocyclohex-3-enecarboxylate (±)-1a with isopropyl 2-ethoxyacetate E after 4 h on Hydrodex β-6TBDM column

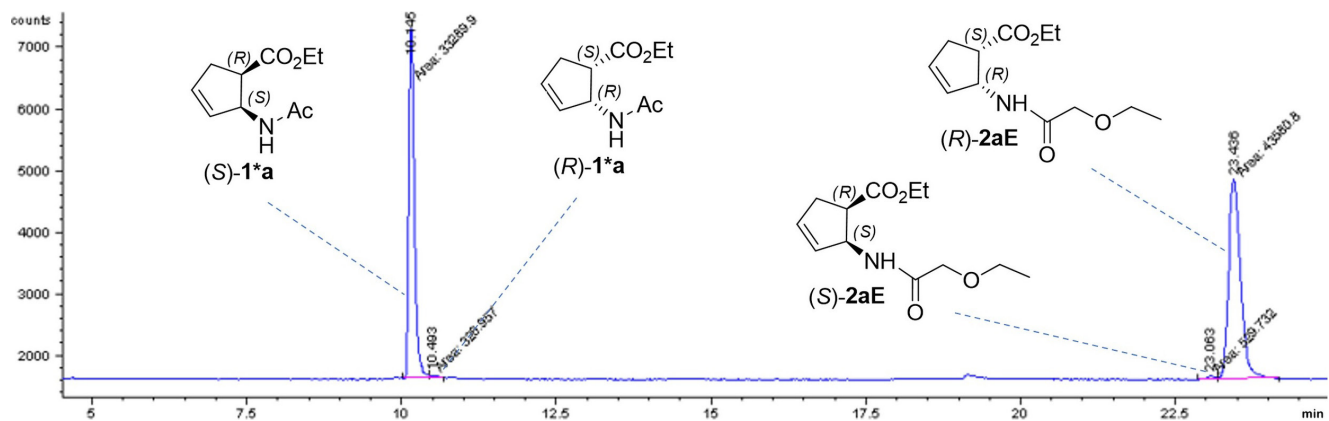


Fig. S8 Chromatogram of CaLB-MNP *N*-acylation of (±)-ethyl *cis*-2-aminocyclohex-3-enecarboxylate (±)-1a with isopropyl 2-ethoxyacetate E after 24 h on Hydrodex β-6TBDM column

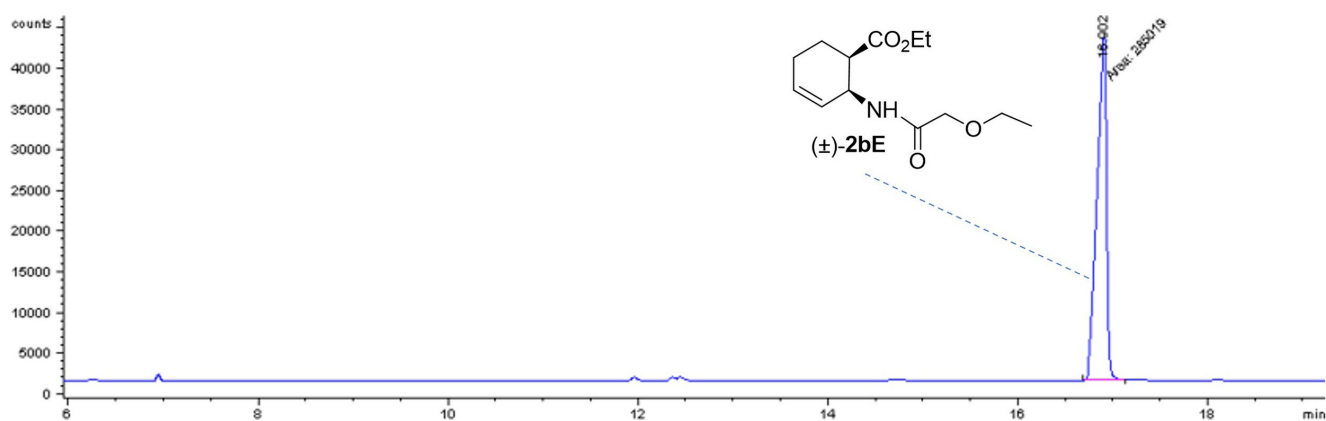


Fig. S9 Chromatogramm of (±)-ethyl *cis*-2-(2-ethoxyacetamido)cyclohex-3-ene-1-carboxylate (±)-**2bE** on Hydrodex β -6TBDM column

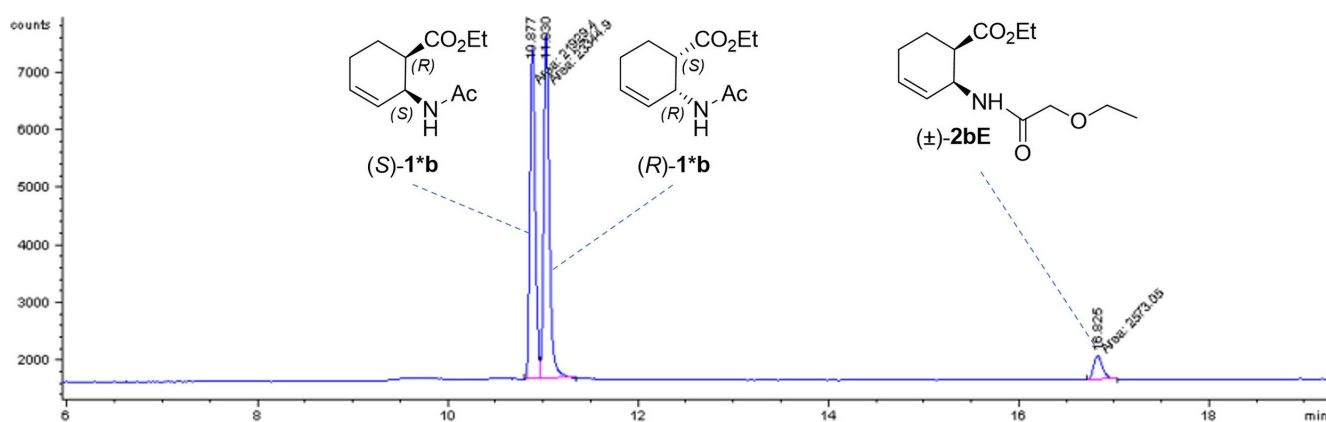


Fig. S10 Chromatogramm of CaLB-MNP *N*-acylation of (±)-ethyl *cis*-2-aminocyclohex-3-enecarboxylate (±)-**1b** with isopropyl 2-ethoxyacetate **E** after 4 h on Hydrodex β -6TBDM column

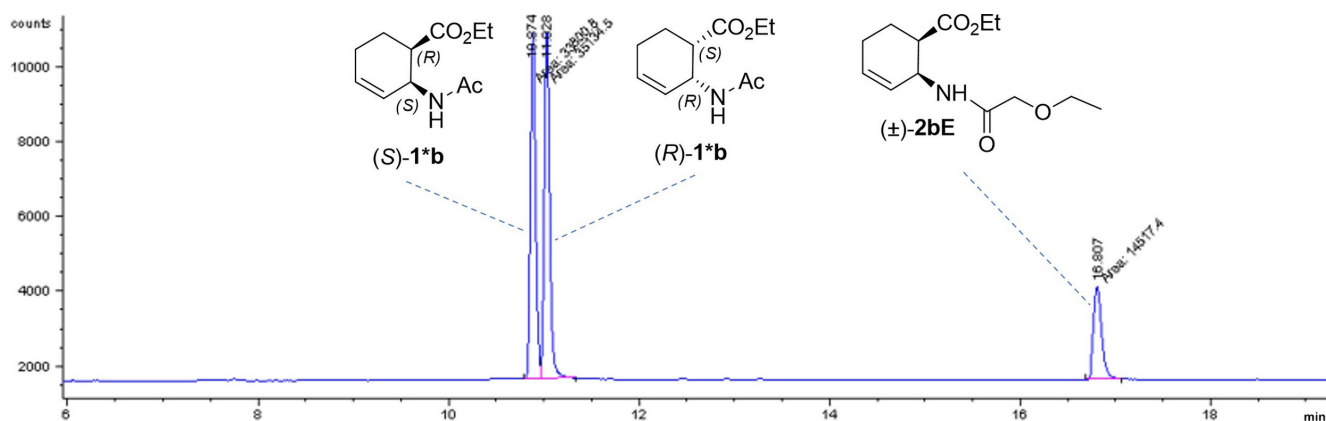


Fig. S11 Chromatogramm of CaLB-MNP *N*-acylation of (±)-ethyl *cis*-2-aminocyclohex-3-enecarboxylate (±)-**1b** with isopropyl 2-ethoxyacetate **E** after 24 h on Hydrodex β -6TBDM column

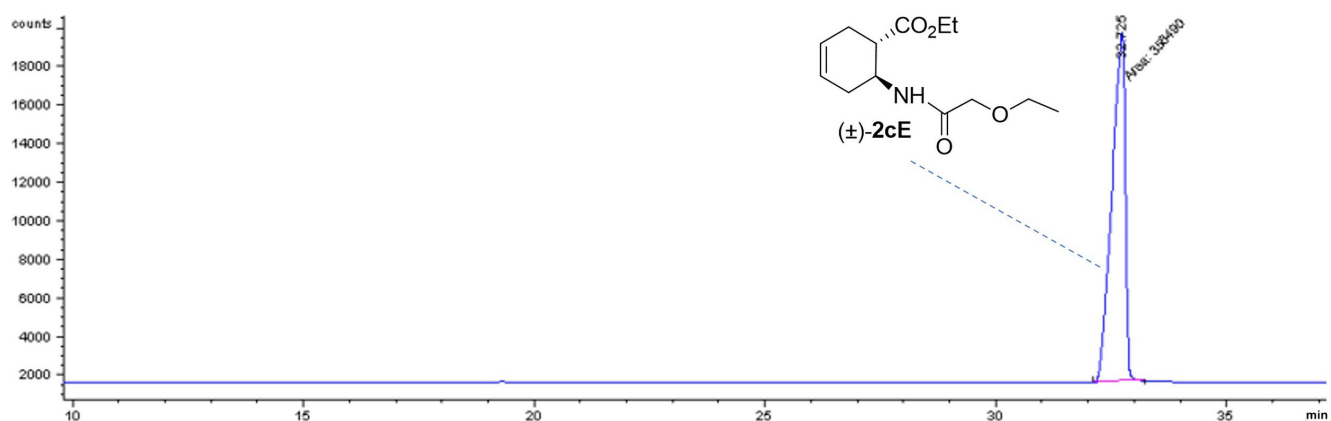


Fig. S12 Chromatogramm of (±)-ethyl *trans*-6-(2-ethoxyacetamido)cyclohex-3-ene-1-carboxylate (±)-2cE on Hydrodex β-6TBDM column

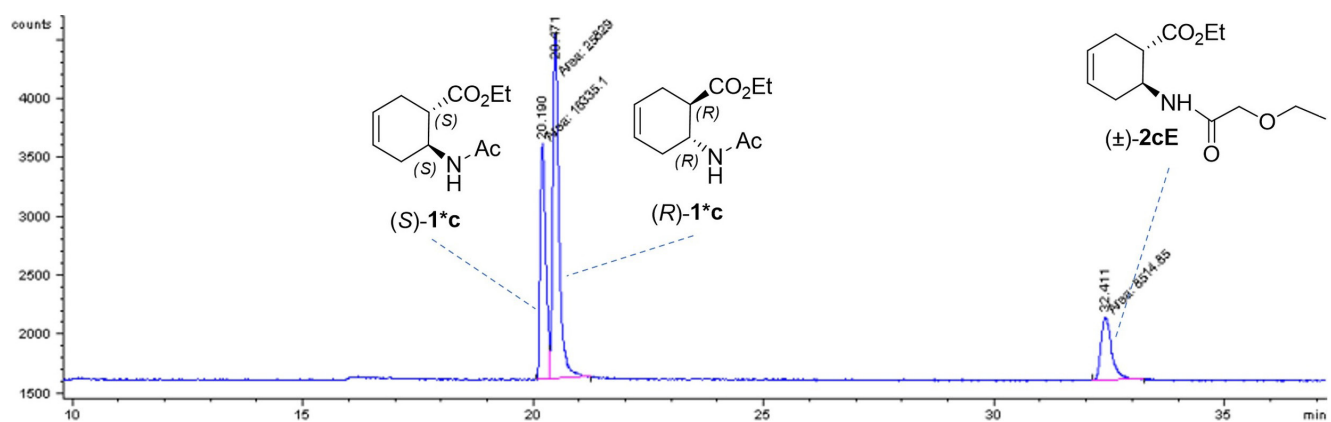


Fig. S13 Chromatogramm of CaLB-MNP *N*-acylation of (±)-ethyl *trans*-6-aminocyclohex-3-enecarboxylate (±)-1c with isopropyl 2-ethoxyacetate E after 4 h on Hydrodex β-6TBDM column

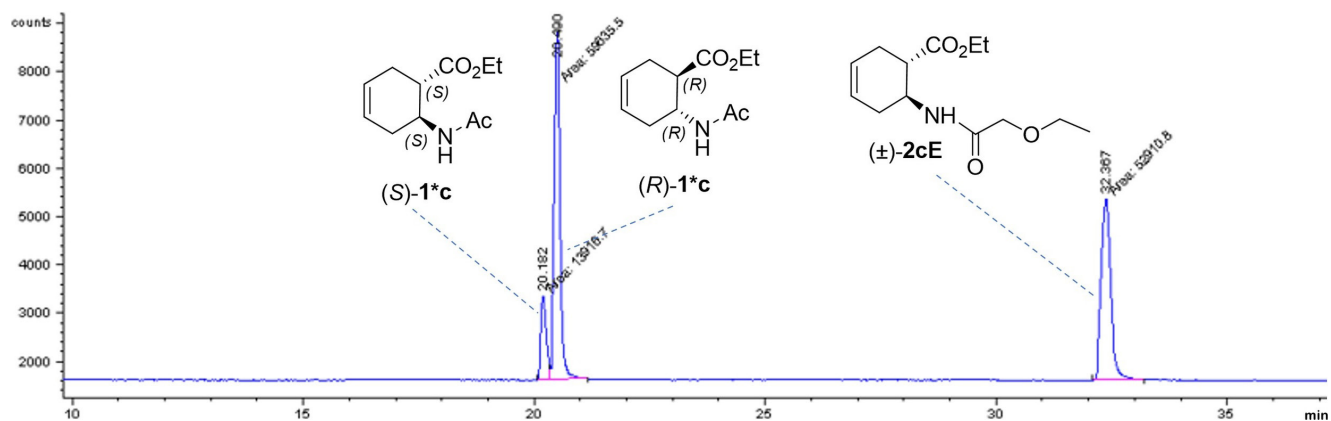


Fig. S14 Chromatogramm of CaLB-MNP *N*-acylation of (±)-ethyl *trans*-6-aminocyclohex-3-enecarboxylate (±)-1c with isopropyl 2-ethoxyacetate E after 24 h on Hydrodex β-6TBDM column

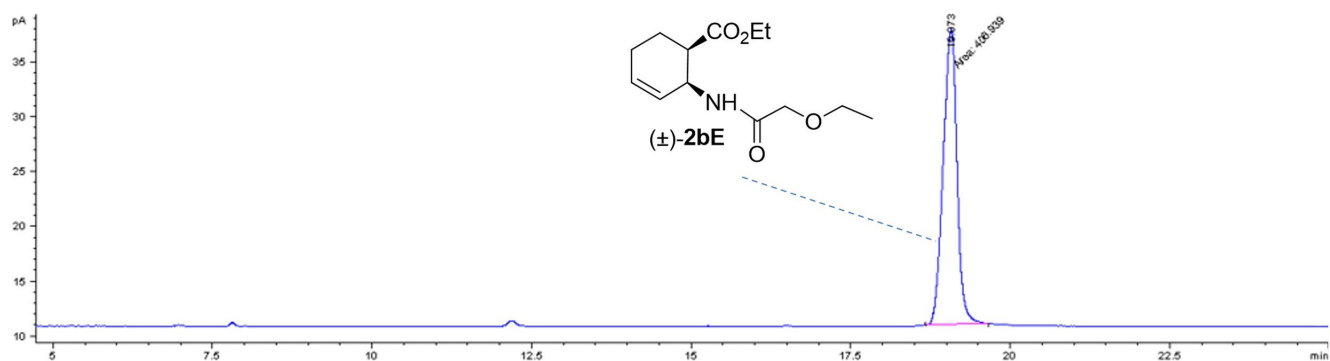


Fig. S15 Chromatogram of (±)-ethyl *cis*-2-(2-ethoxyacetamido)cyclohex-3-ene-1-carboxylate (±)-2bE on Hydrodex β -6TBDAC column

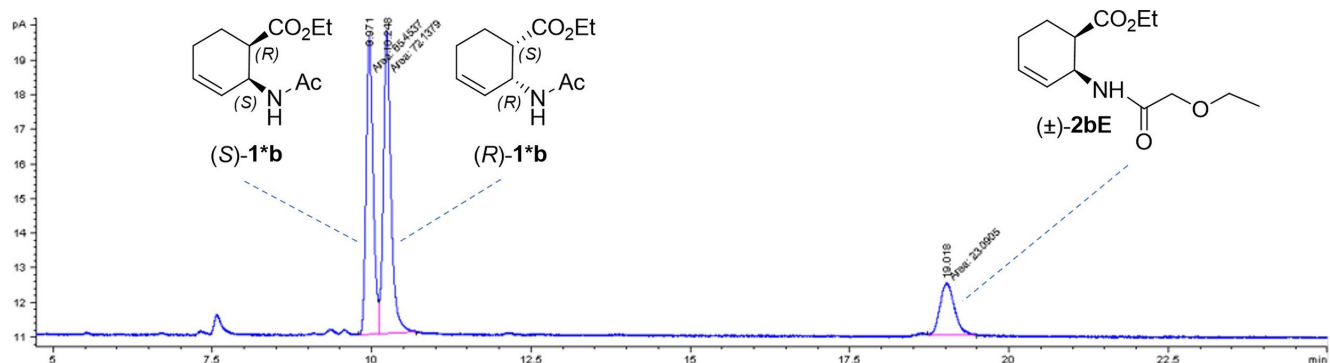


Fig. S16 Chromatogram of CaLB-MNP *N*-acylation of (±)-ethyl *cis*-2-aminocyclohex-3-enecarboxylate (±)-1b with isopropyl 2-ethoxyacetate E after 24 h on Hydrodex β -6TBDAC column

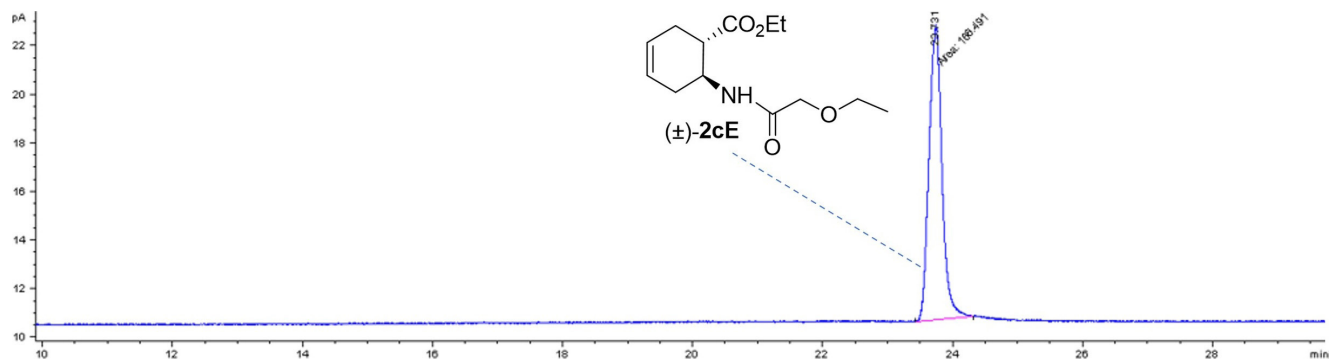


Fig. S17 Chromatogram of (±)-ethyl *trans*-6-(2-ethoxyacetamido)cyclohex-3-ene-1-carboxylate (±)-2cE on Hydrodex β -6TBDAC column

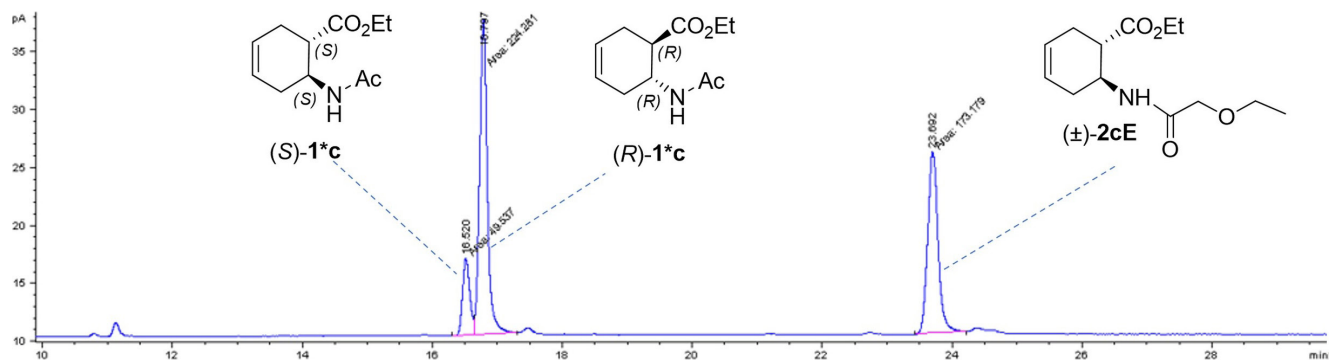


Fig. S18 Chromatogram of CaLB-MNP *N*-acylation of (±)-ethyl *trans*-6-aminocyclohex-3-enecarboxylate (±)-1c with isopropyl 2-ethoxyacetate E after 24 h on Hydrodex β -6TBDAC column

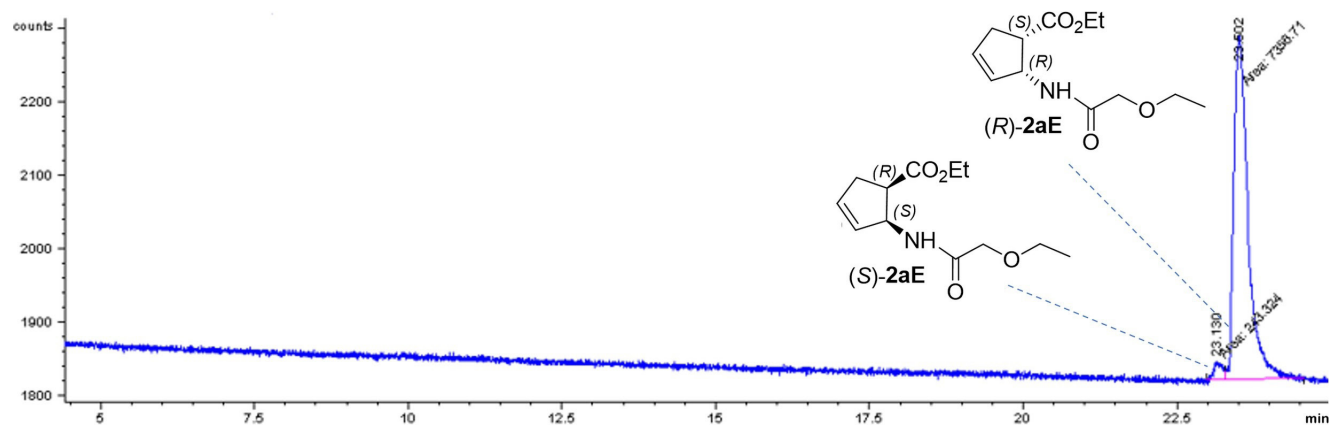


Fig. S19 Chromatogramm of (*R*)-ethyl *cis*-2-(2-ethoxyacetamido)cyclopent-3-ene-1-carboxylate (*R*)-2aE on Hydrodex β -6TBDM column

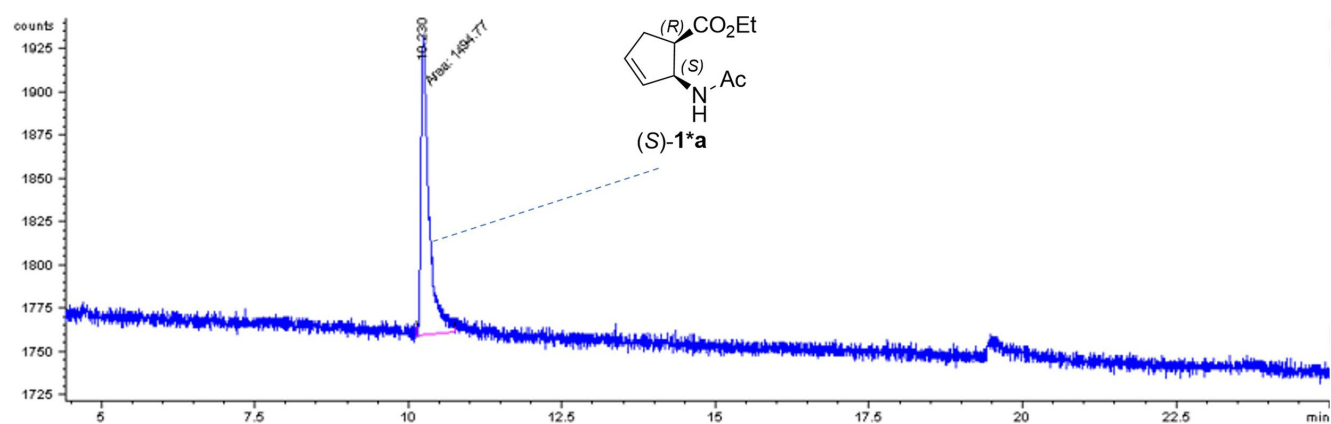


Fig. S20 Chromatogramm of (*S*)-ethyl *cis*-2-aminocyclopent-3-enecarboxylate (*S*)-1a on Hydrodex β -6TBDM column

2.4 NMR spectra of cyclopentyl β -amino ester derivatives

NMR spectra of the isolated compounds are shown in Figs. S21–S36.

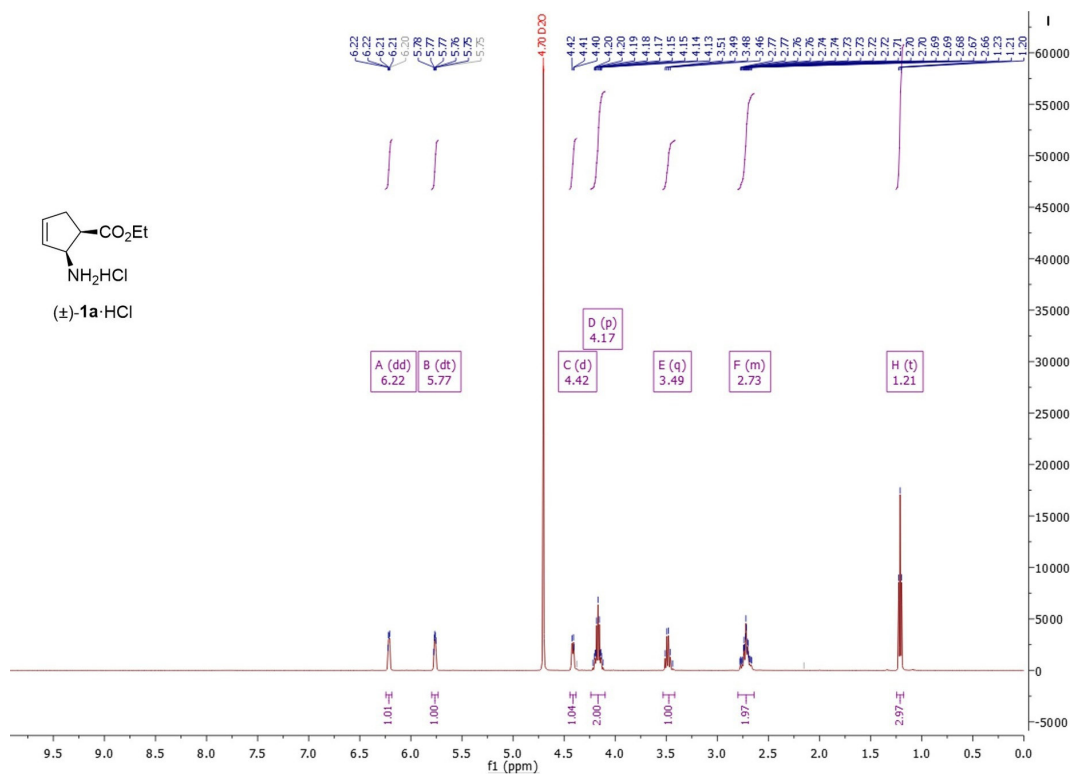


Fig. S21 $^1\text{H-NMR}$ spectrum of $(\pm)$ -ethyl *cis*-2-aminocyclopent-3-enecarboxylate·HCl ($\pm$)-1a·HCl

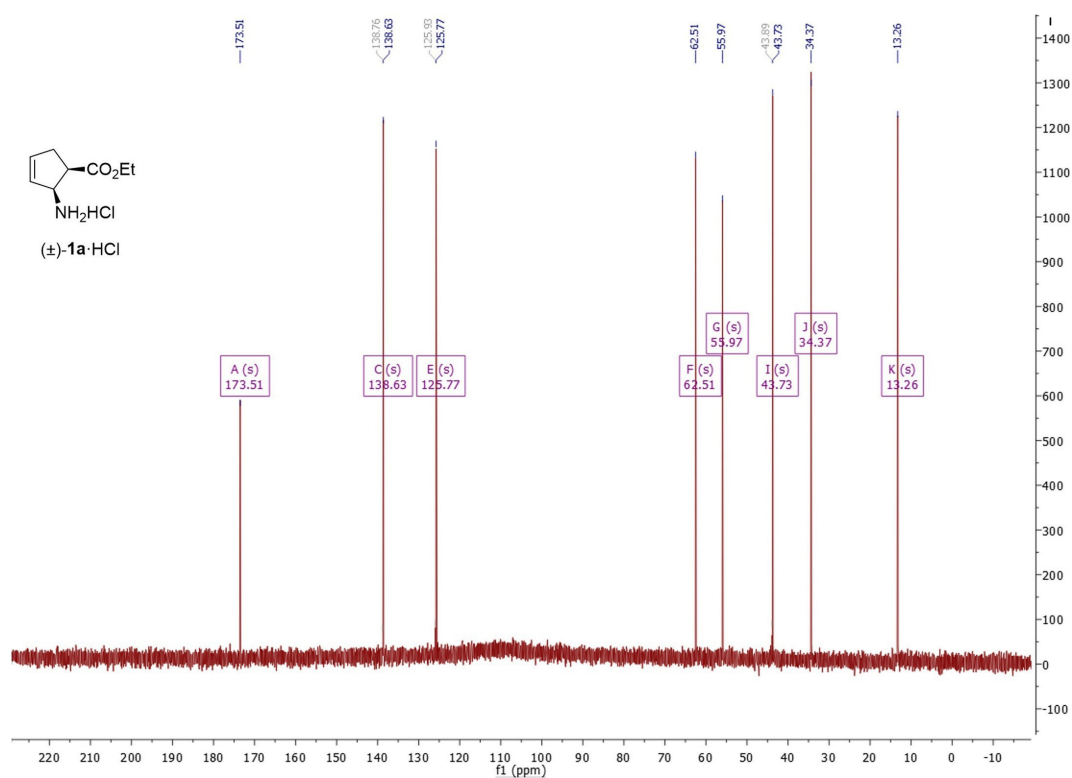


Fig. S22 $^{13}\text{C-NMR}$ spectrum of $(\pm)$ -ethyl *cis*-2-aminocyclopent-3-enecarboxylate·HCl ($\pm$)-1a·HCl

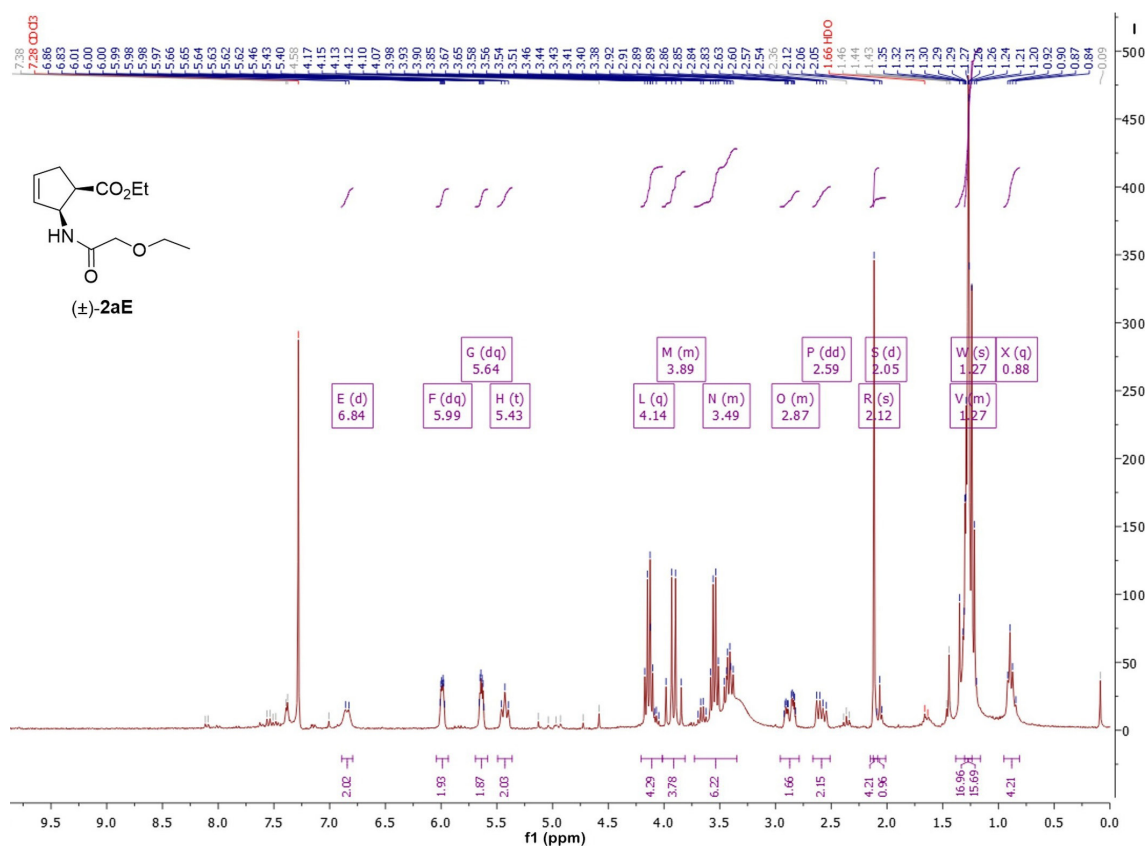


Fig. S23 ¹H-NMR spectrum of (±)-ethyl *cis*-2-(2-ethoxyacetamido)cyclopent-3-ene-1-carboxylate (±)-2aE

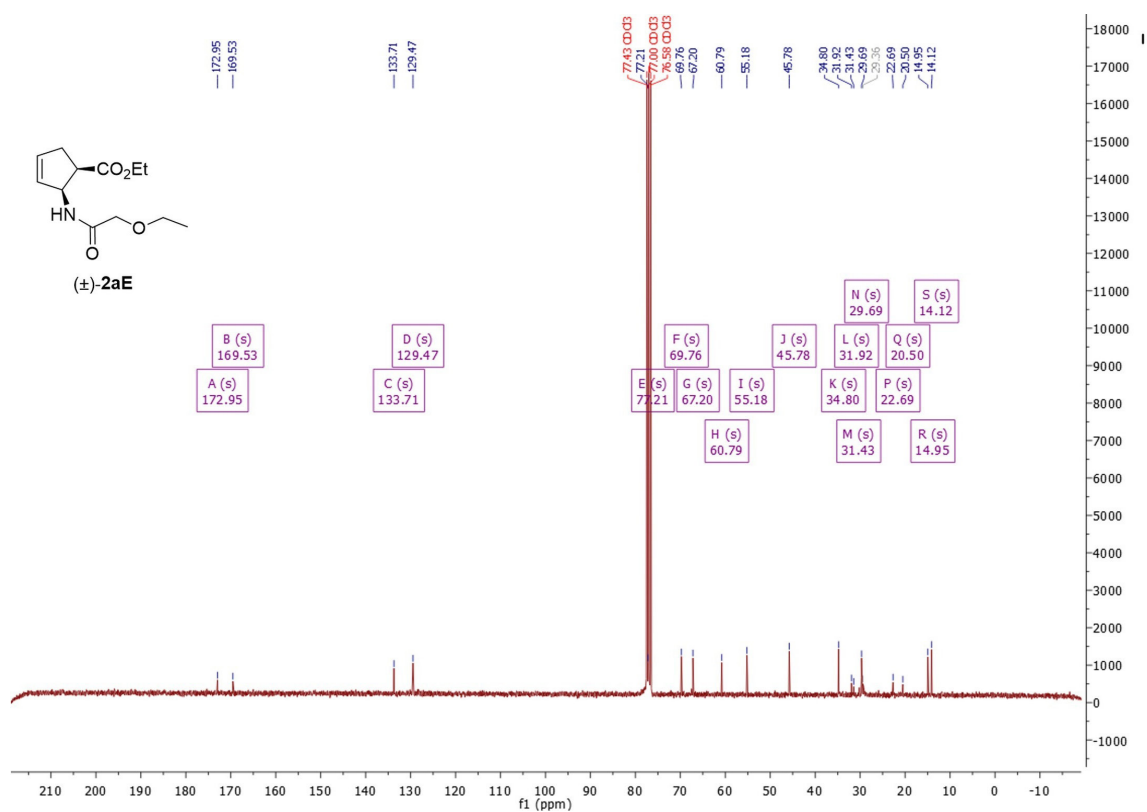


Fig. S24 ¹³C-NMR spectrum of (±)-ethyl *cis*-2-(2-ethoxyacetamido)cyclopent-3-ene-1-carboxylate (±)-2aE

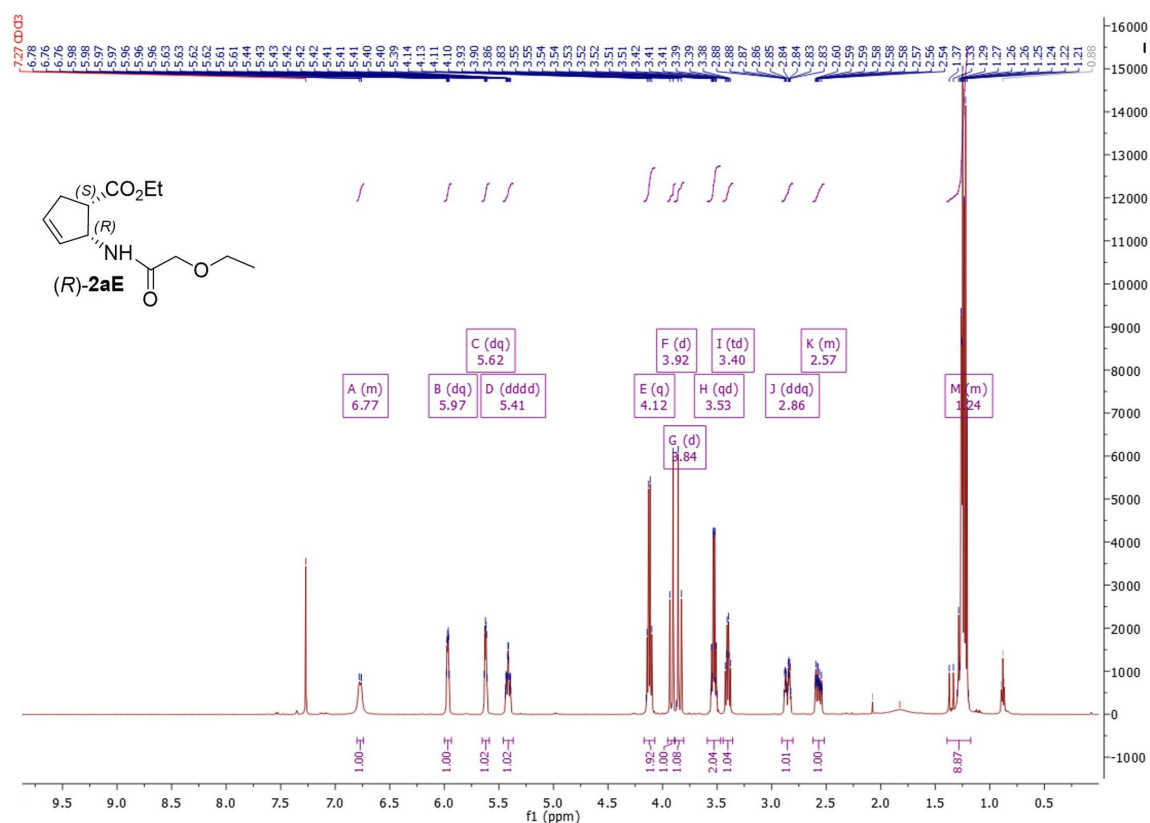


Fig. S25 ¹H-NMR spectrum of (R)-ethyl *cis*-2-(2-ethoxyacetamido)cyclopent-3-ene-1-carboxylate (R)-2aE

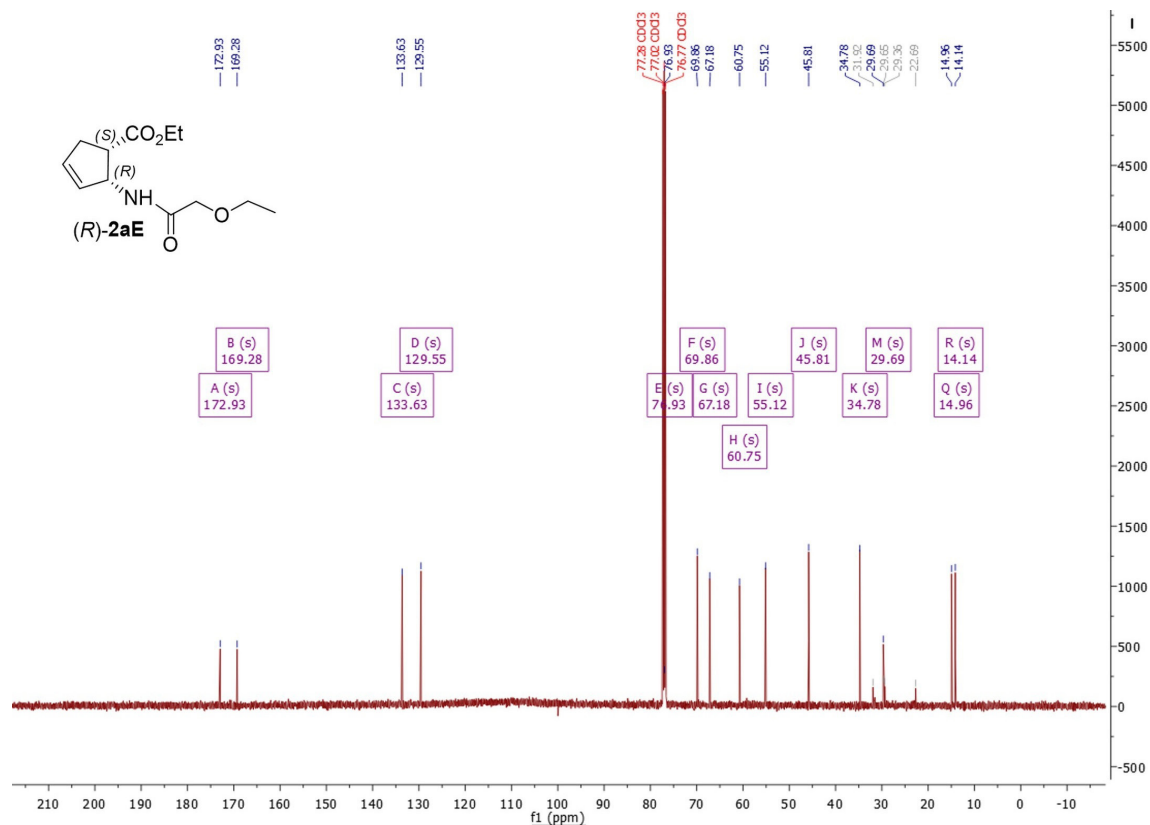


Fig. S26 ¹³C-NMR spectrum of (R)-ethyl *cis*-2-(2-ethoxyacetamido)cyclopent-3-ene-1-carboxylate (R)-2a

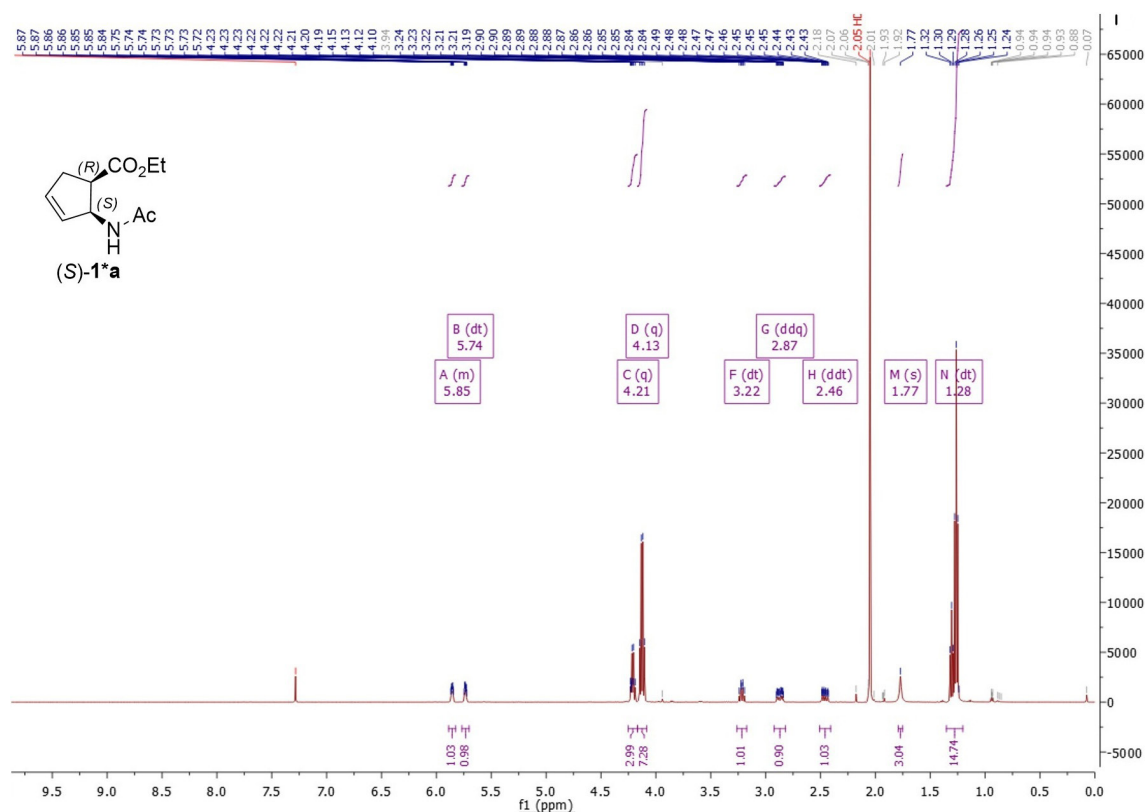


Fig. S27 ¹H-NMR spectrum of (S)-ethyl *cis*-2-aminocyclopent-3-enecarboxylate (S)-1a

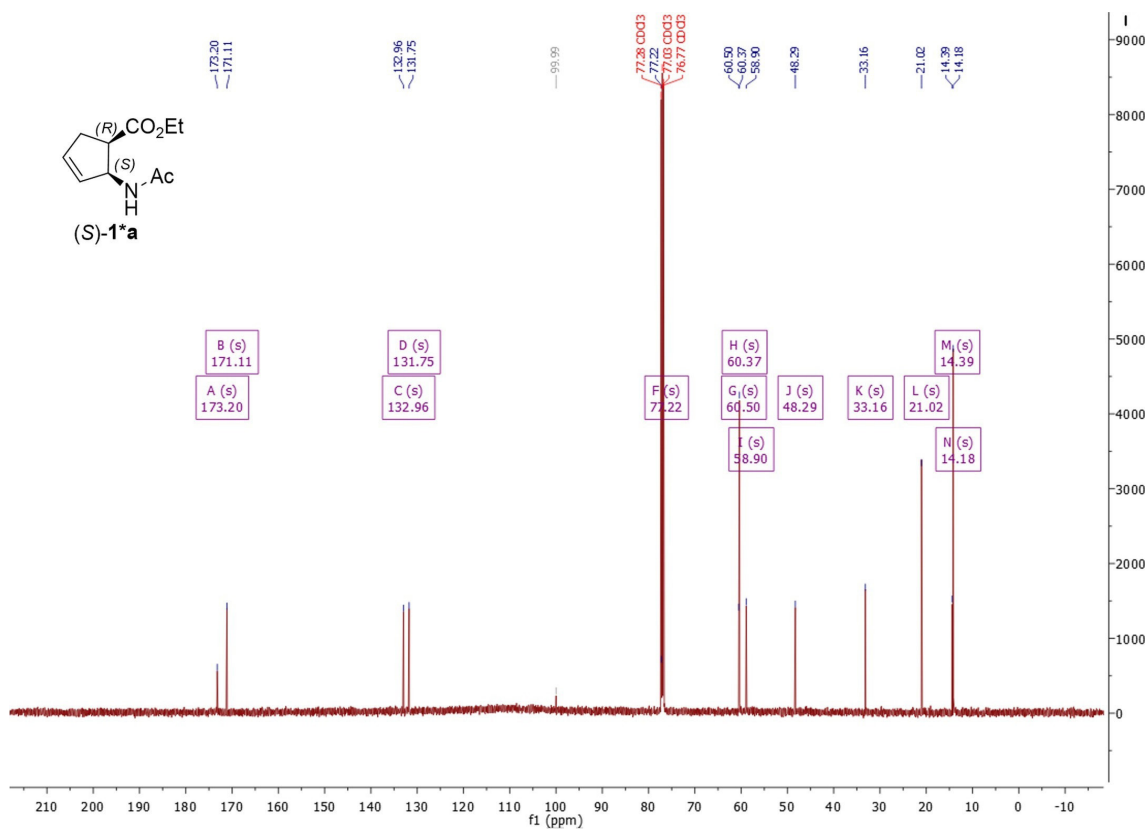


Fig. S28 ¹³C-NMR spectrum of (S)-ethyl *cis*-2-aminocyclopent-3-enecarboxylate (S)-1a

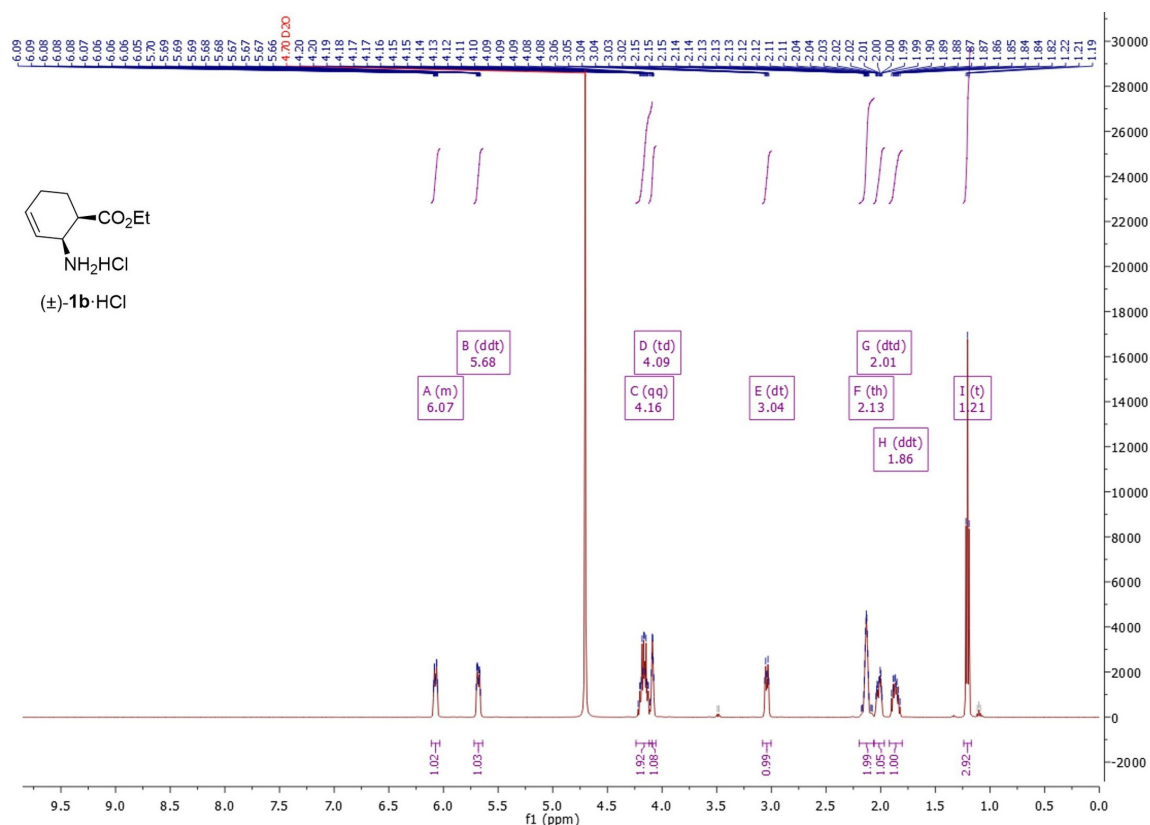


Fig. S29 $^1\text{H-NMR}$ spectrum of $(\pm)$ -ethyl *cis*-2-aminocyclohex-3-enecarboxylate·HCl ($\pm$)-**1b**·HCl

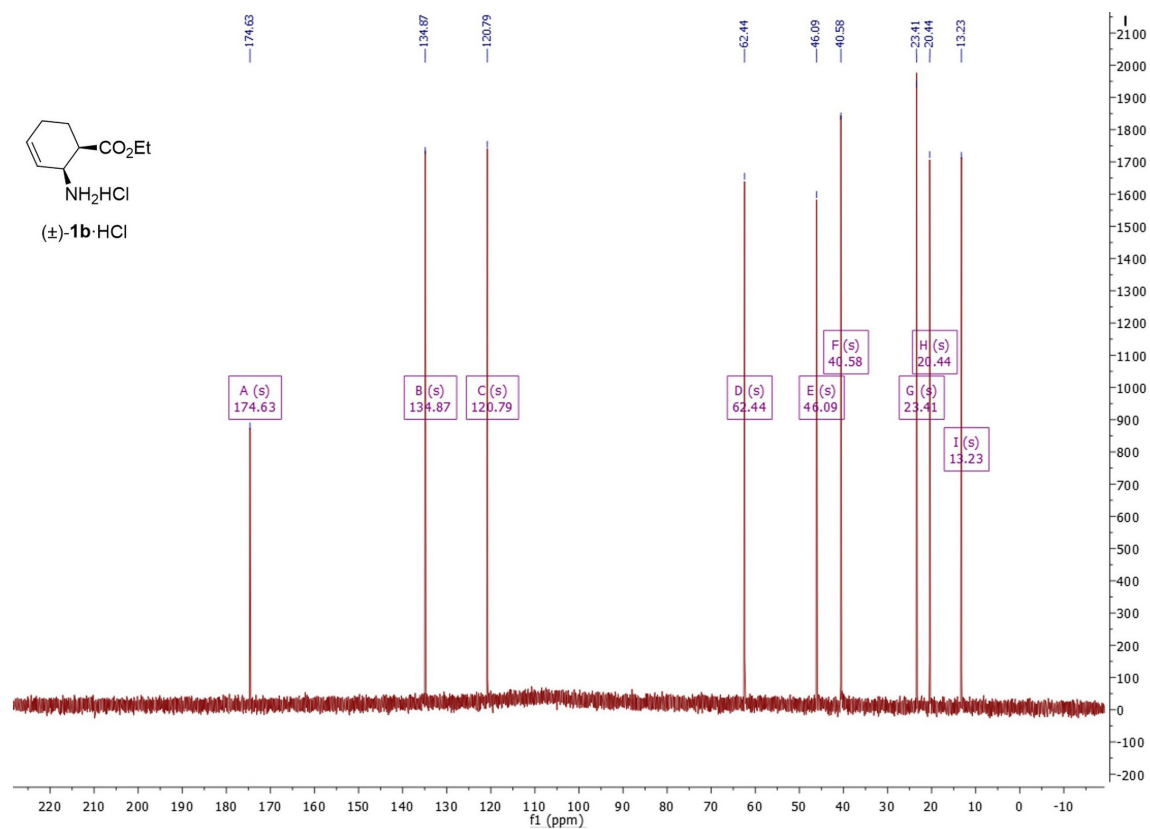


Fig. S30 $^{13}\text{C-NMR}$ spectrum of $(\pm)$ -ethyl *cis*-2-aminocyclohex-3-enecarboxylate·HCl ($\pm$)-**1b**·HCl

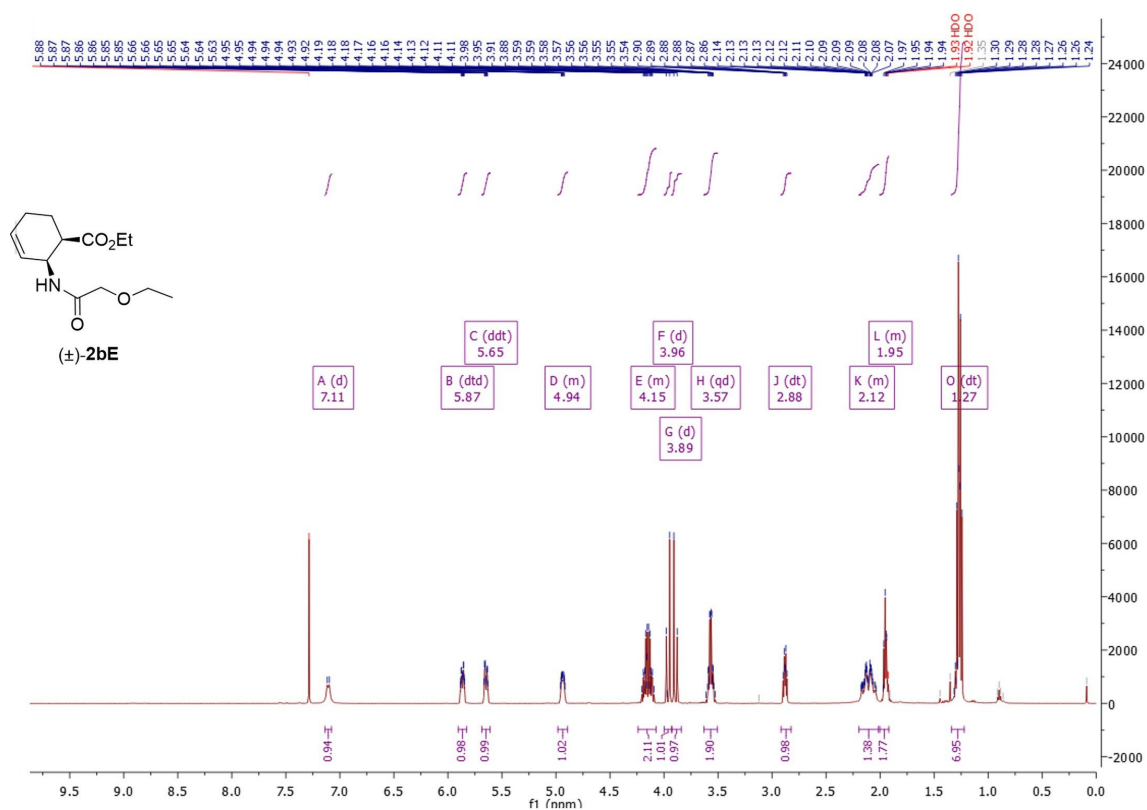


Fig. S31 ^1H -NMR spectrum of (±)-ethyl *cis*-2-(2-ethoxyacetamido)cyclohex-3-ene-1-carboxylate (±)-2bE

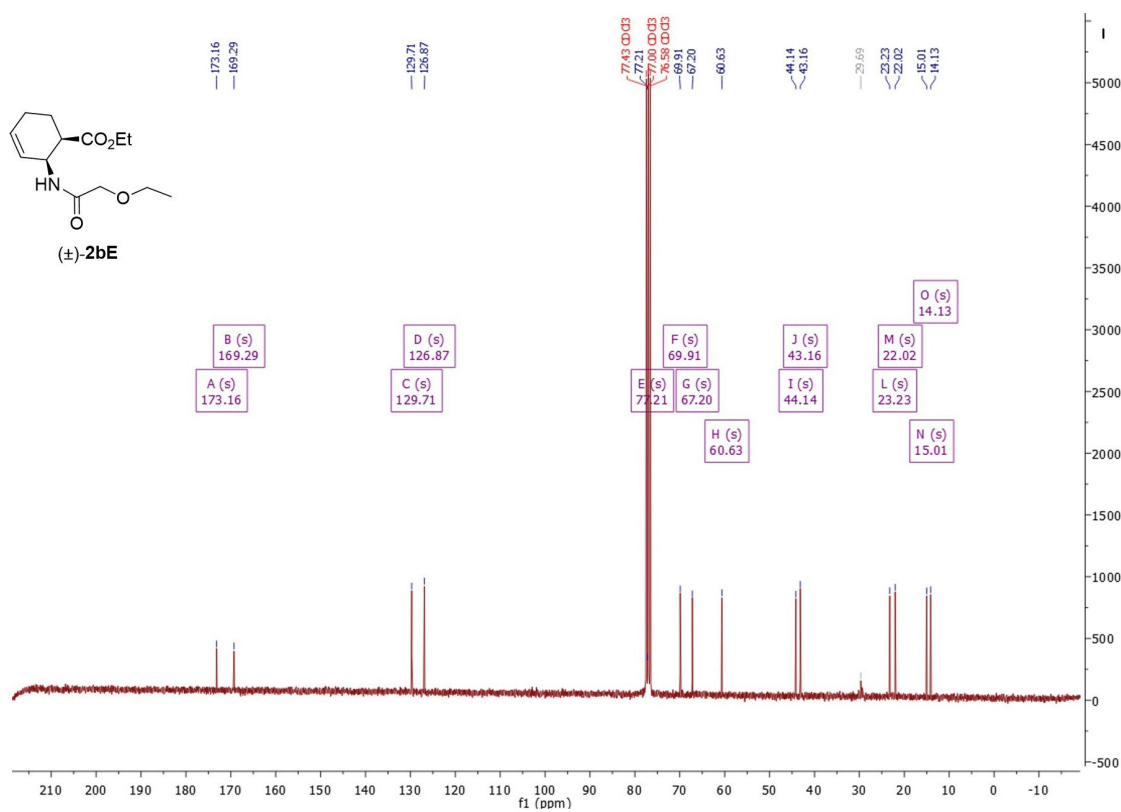


Fig. S32 ^{13}C -NMR spectrum of (±)-ethyl *cis*-2-(2-ethoxyacetamido)cyclohex-3-ene-1-carboxylate (±)-2bE

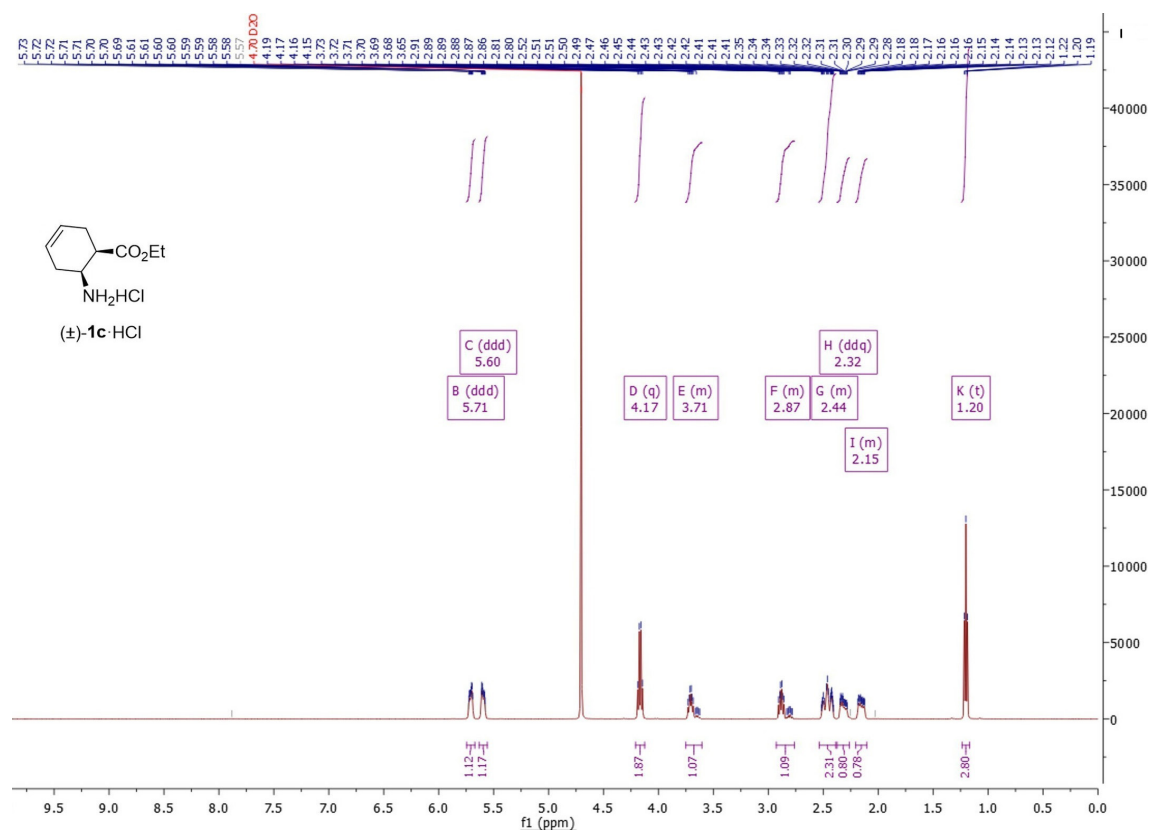


Fig. S33 ^1H -NMR spectrum of (±)-ethyl *trans*-6-aminocyclohex-3-enecarboxylate·HCl (±)-1c·HCl

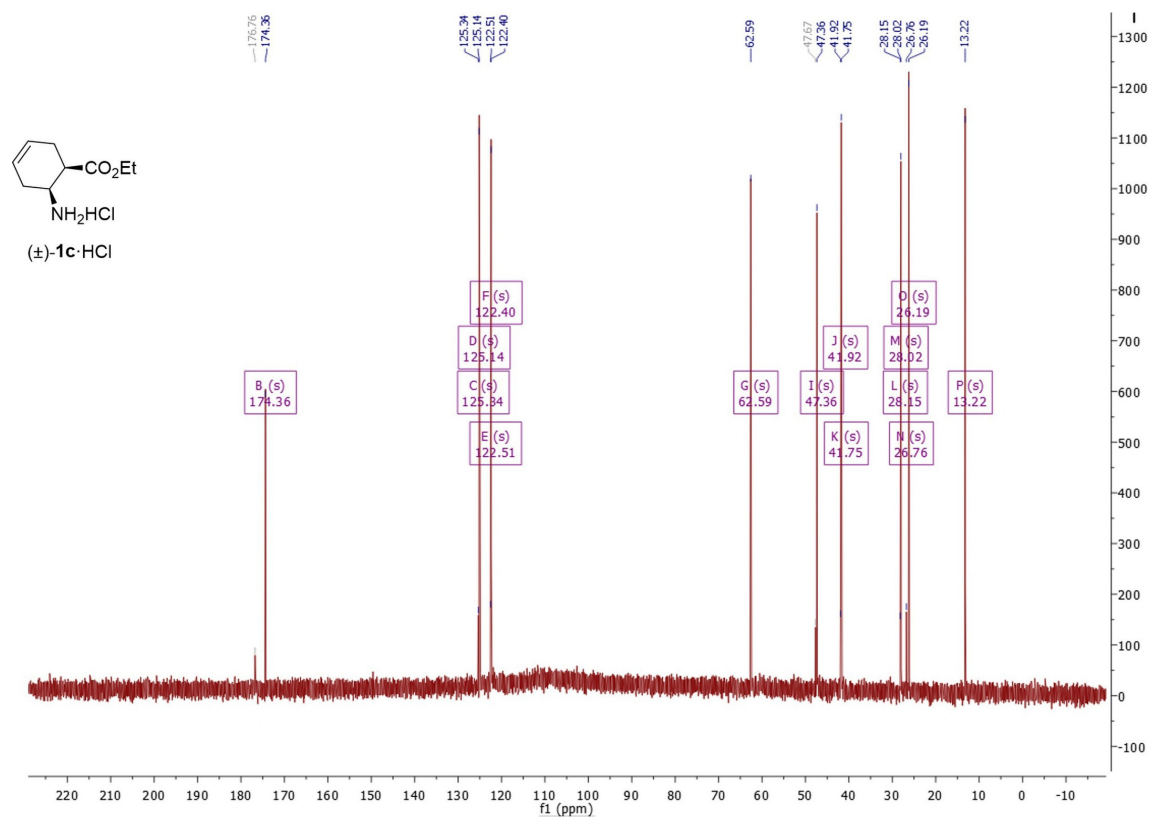


Fig. S34 ^{13}C -NMR spectrum of (±)-ethyl *trans*-6-aminocyclohex-3-enecarboxylate·HCl (±)-1c·HCl

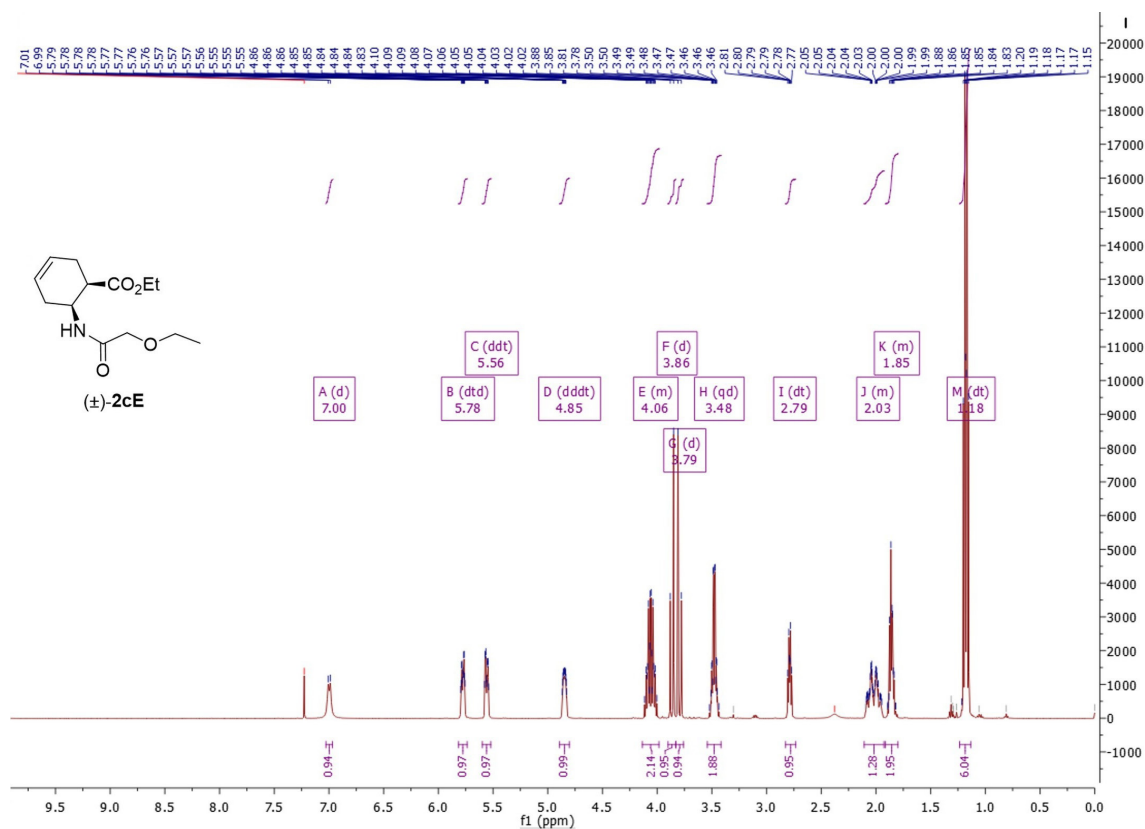


Fig. S35 ¹H-NMR spectrum of (±)-ethyl *trans*-6-(2-ethoxyacetamido)cyclohex-3-ene-1-carboxylate (±)-2cE

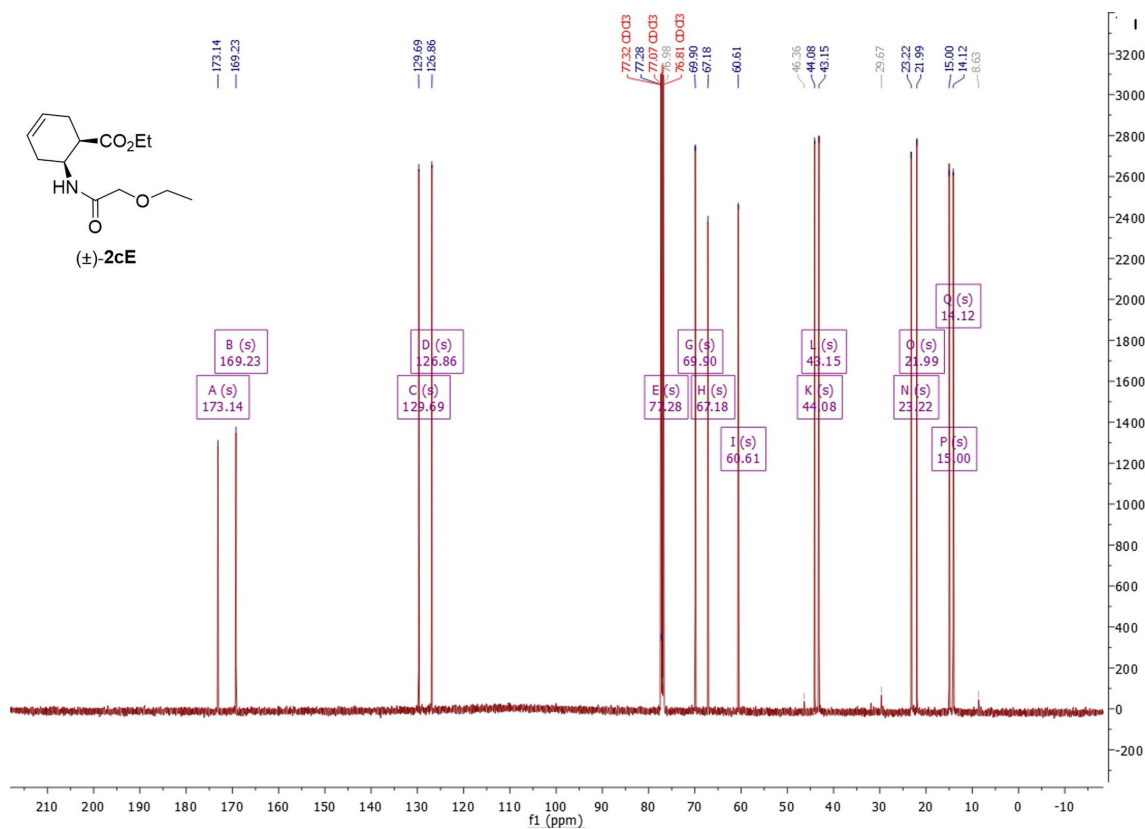


Fig. S36 ¹³C-NMR spectrum of (±)-ethyl *trans*-6-(2-ethoxyacetamido)cyclohex-3-ene-1-carboxylate (±)-2cE