catalysts

MDPI

SUPPLEMENTARY MATERIALS:

Green Chemo-Enzymatic Protocols for Synthesis of Enantiopure β-Blockers (*S*)-Esmolol and (*S*)-Penbutolol

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* Correspondence: elisabeth.e.jacobsen@ntnu.no

Spectroscopic and chromatographic analysis data

*Methyl 3-(4-(oxiran-2-ylmethoxy)phenyl)propanoate (**2**)*

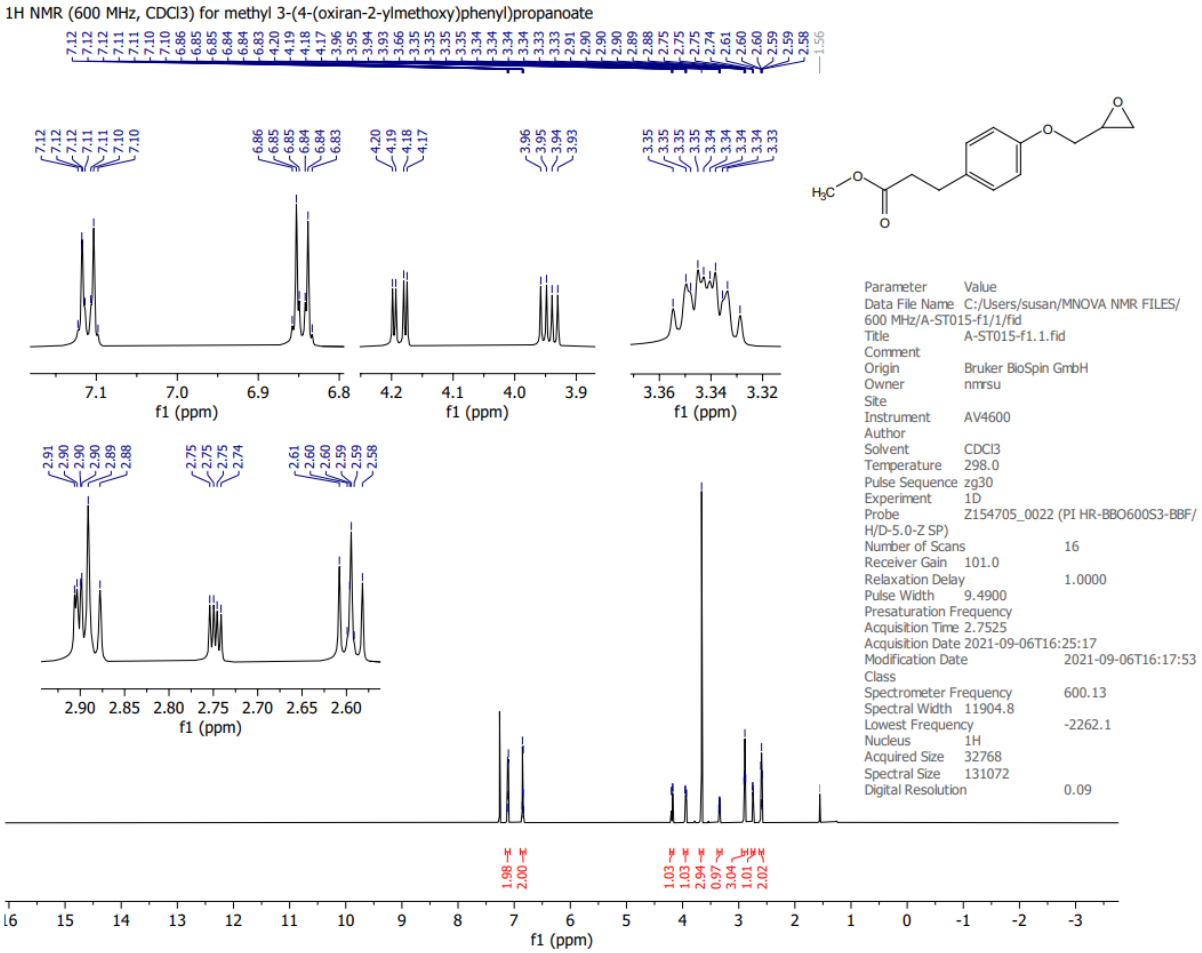


Figure S1. ^{1}H NMR spectrum (600 MHz, $CDCl_3$) of methyl 3-(4-(oxiran-2-ylmethoxy)phenyl)propanoate (**2**).

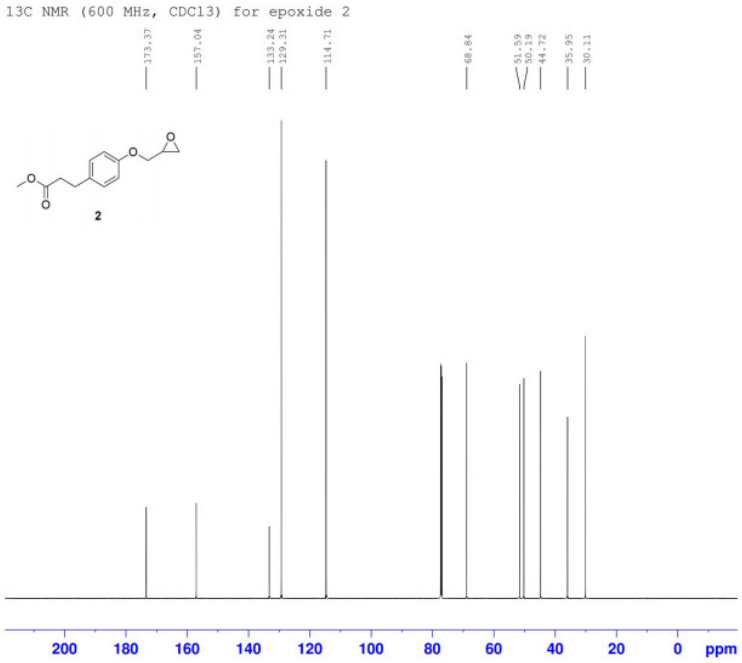


Figure S2. ^{13}C NMR spectrum (150 MHz, $CDCl_3$) of methyl 3-(4-(oxiran-2-ylmethoxy)phenyl)propanoate (**2**).

*Methyl 3-(4-(3-chloro-2-hydroxypropoxy)phenyl)propanoate (**3**)*

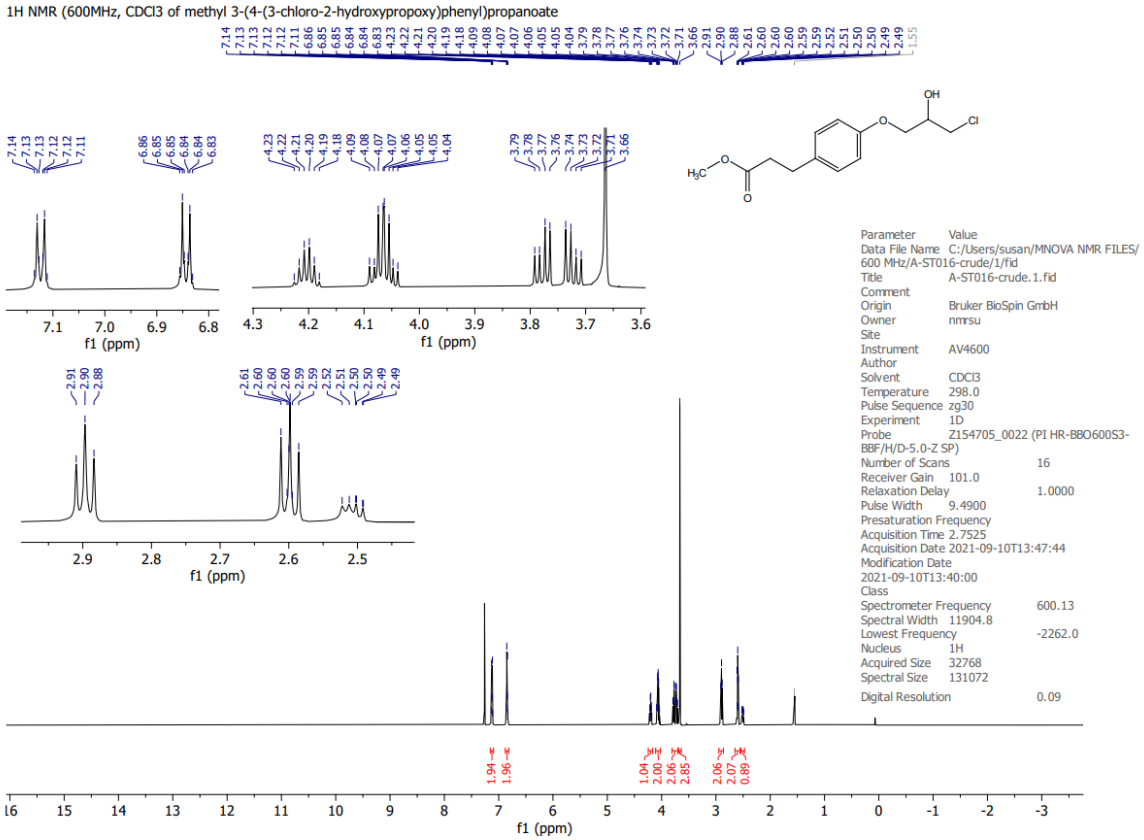


Figure S3. ^{1}H NMR spectrum (600 MHz, $CDCl_3$) of methyl 3-(4-(3-chloro-2-hydroxypropoxy)phenyl)propanoate (**3**).

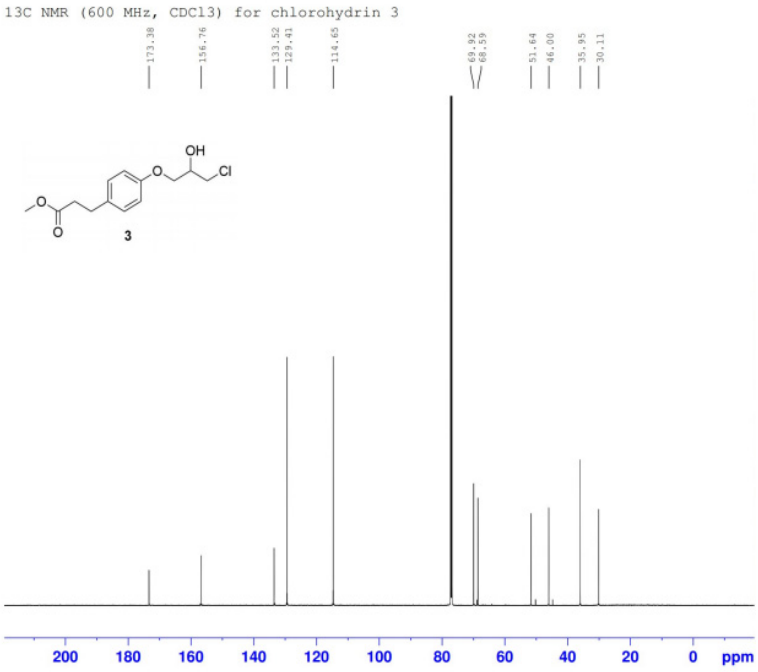


Figure S4. ^{13}C NMR spectrum (150 MHz, $CDCl_3$) of methyl 3-(4-(3-chloro-2-hydroxypropoxy)phenyl)propanoate (**3**).

*(S)-1-Chloro-3-(4-(3-methoxy-3-oxopropyl)phenoxy)propan-2-yl butanoate ((S)-**4**)*

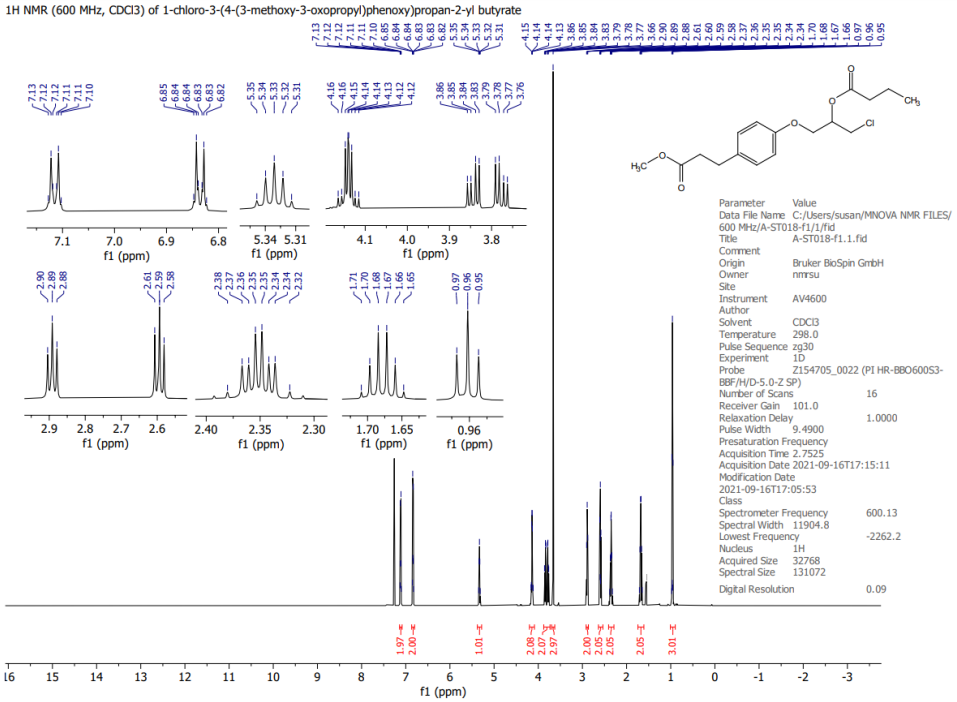


Figure S5. ^{1}H NMR spectrum (600 MHz, $CDCl_3$) of (*S*)-1-chloro-3-(4-(3-methoxy-3-oxopropyl)phenoxy)propan-2-yl butanoate ((*S*)-**4**).

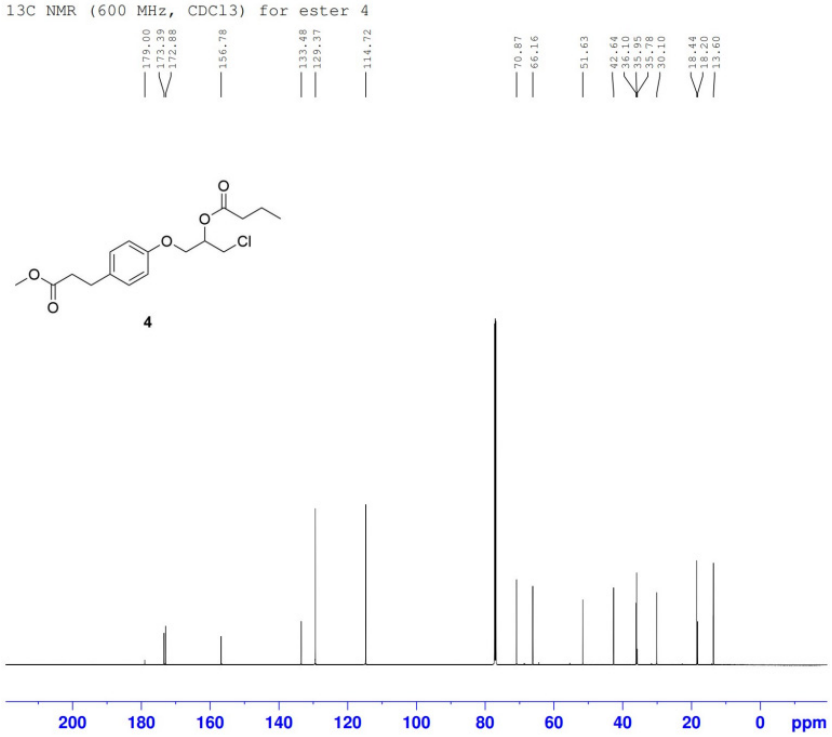


Figure S6. ^{13}C NMR spectrum (150 MHz, $CDCl_3$) of (*S*)-1-chloro-3-(4-(3-methoxy-3-oxopropyl)phenoxy)propan-2-yl butanoate ((*S*)-**4**).

*Esmolol (**5**)*

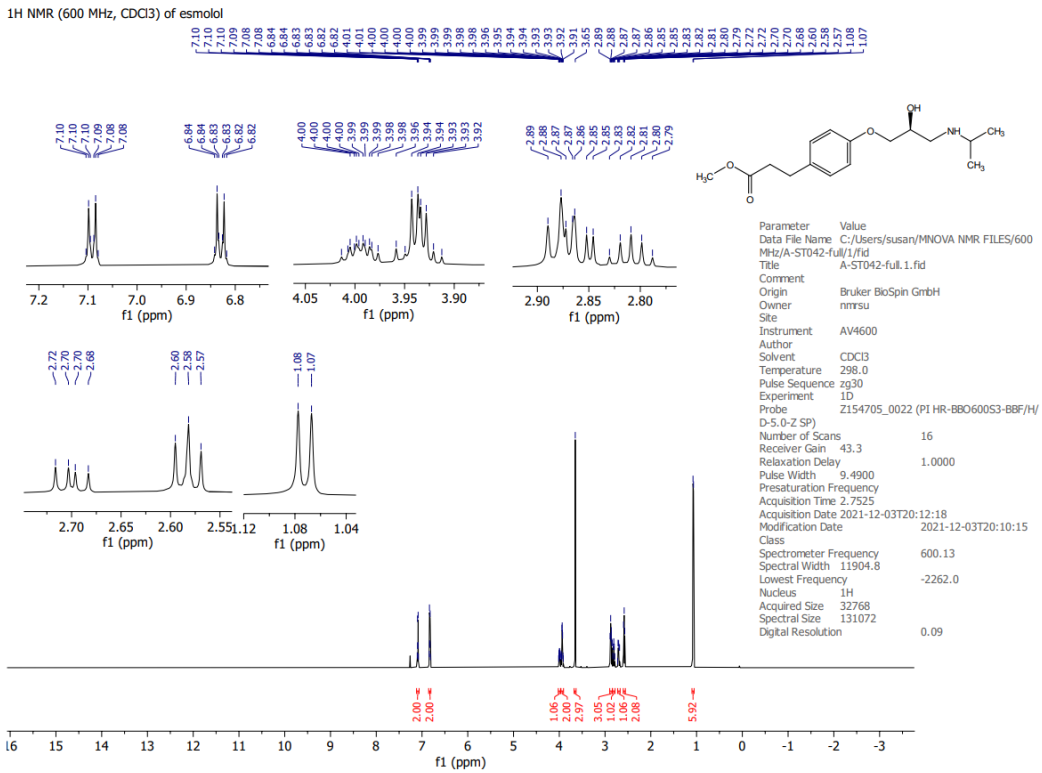


Figure S7. ^{1}H NMR spectrum (600 MHz, $CDCl_3$) of esmolol (**5**).

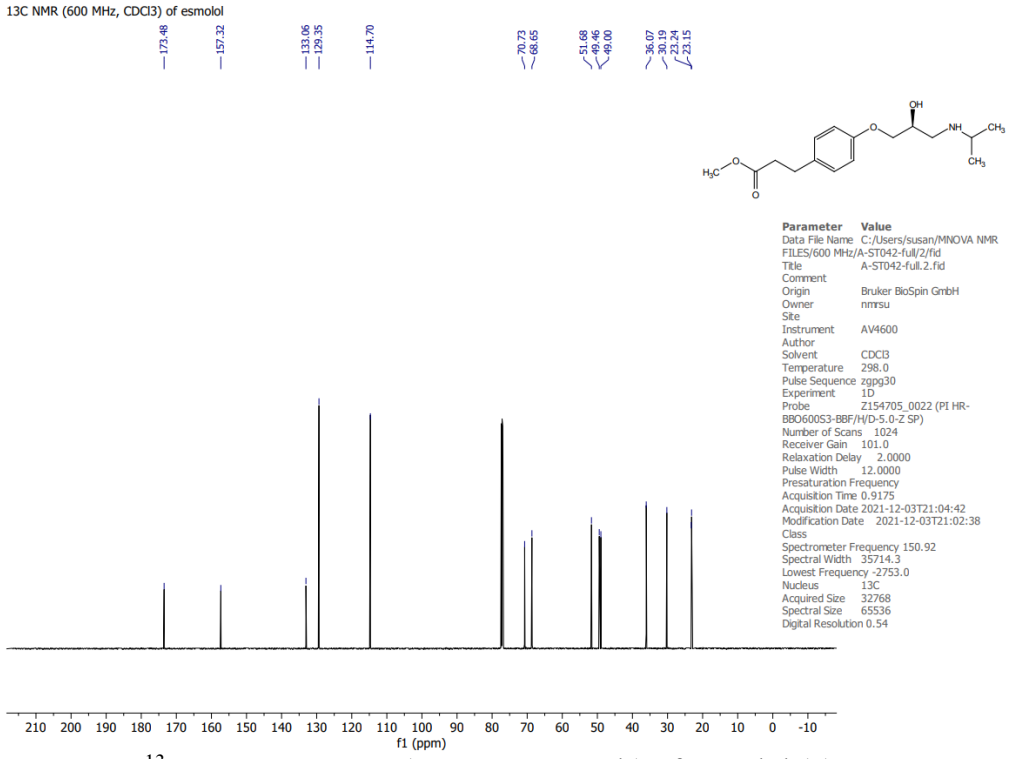


Figure S8. ^{13}C NMR spectrum (150 MHz, $CDCl_3$) of esmolol (**5**).

1-Chloro-3-(2-cyclopentylphenoxy)propan-2-ol (***8***)

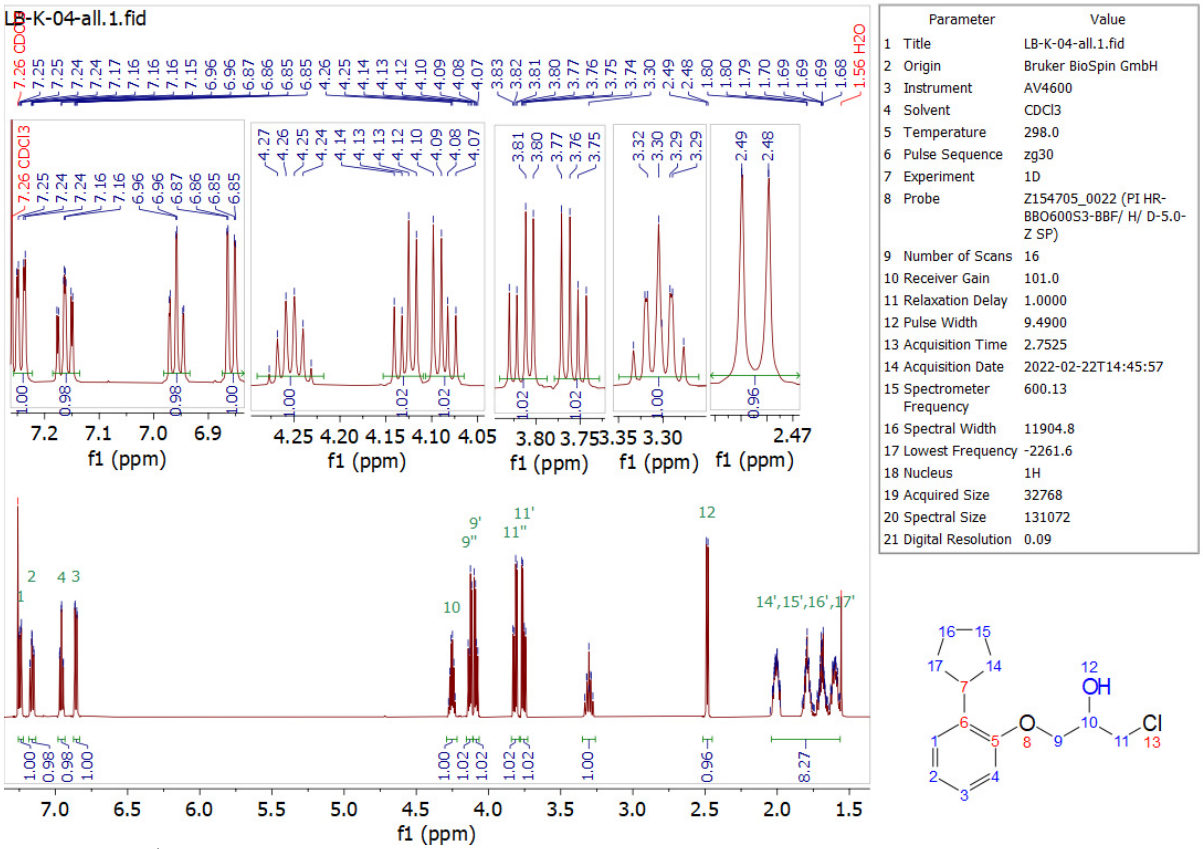


Figure S9. ^{1}H NMR spectrum (600 MHz, $CDCl_3$) of 1-chloro-3-(2-cyclopentylphenoxy)propan-2-ol (**8**).

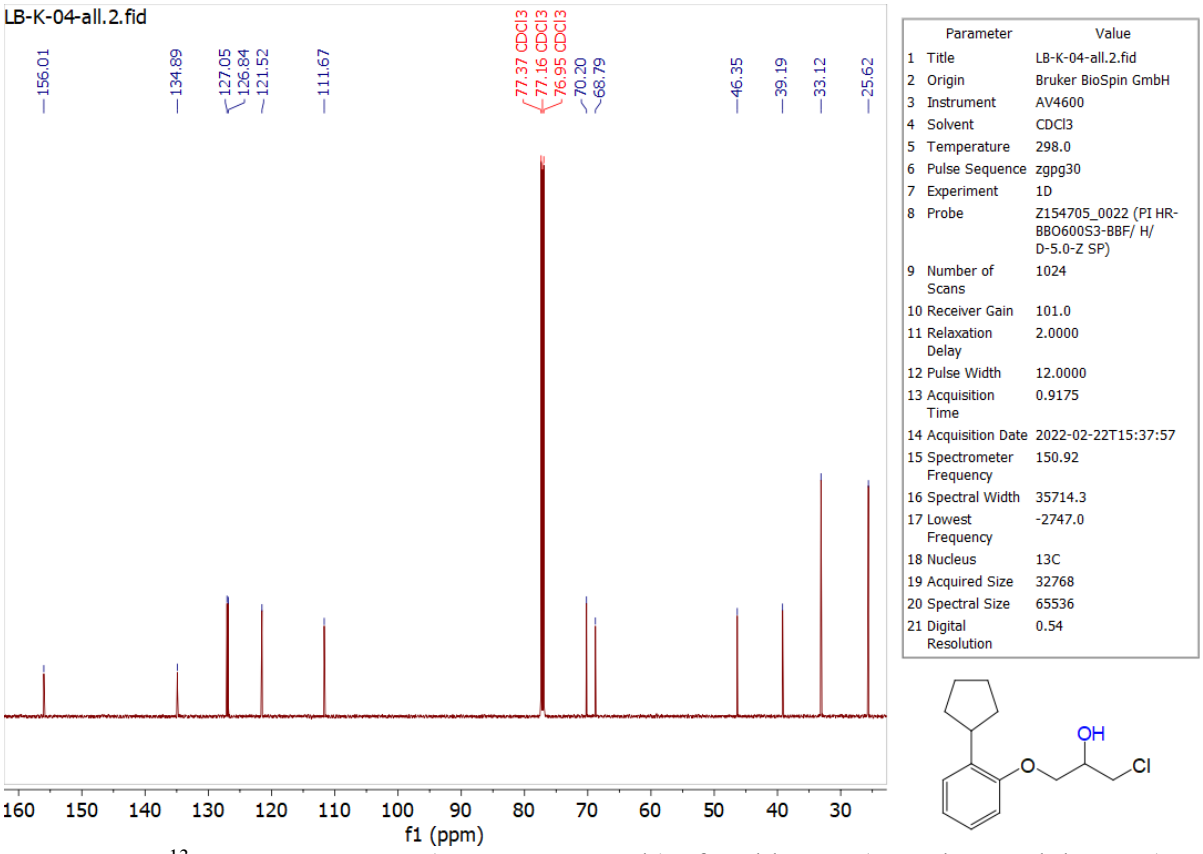


Figure S10. ^{13}C NMR spectrum (150 MHz, $CDCl_3$) of 1-chloro-3-(2-cyclopentylphenoxy)propan-2-ol (**8**)

*Penbutolol (**10**)*

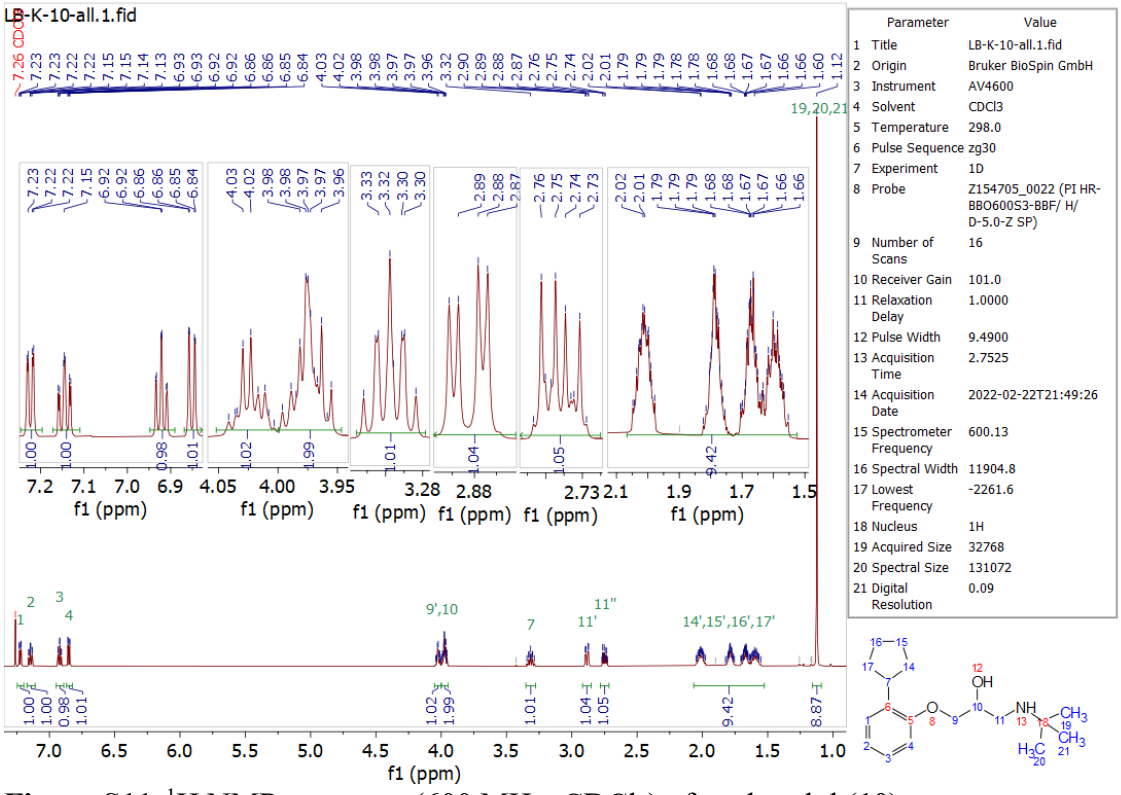
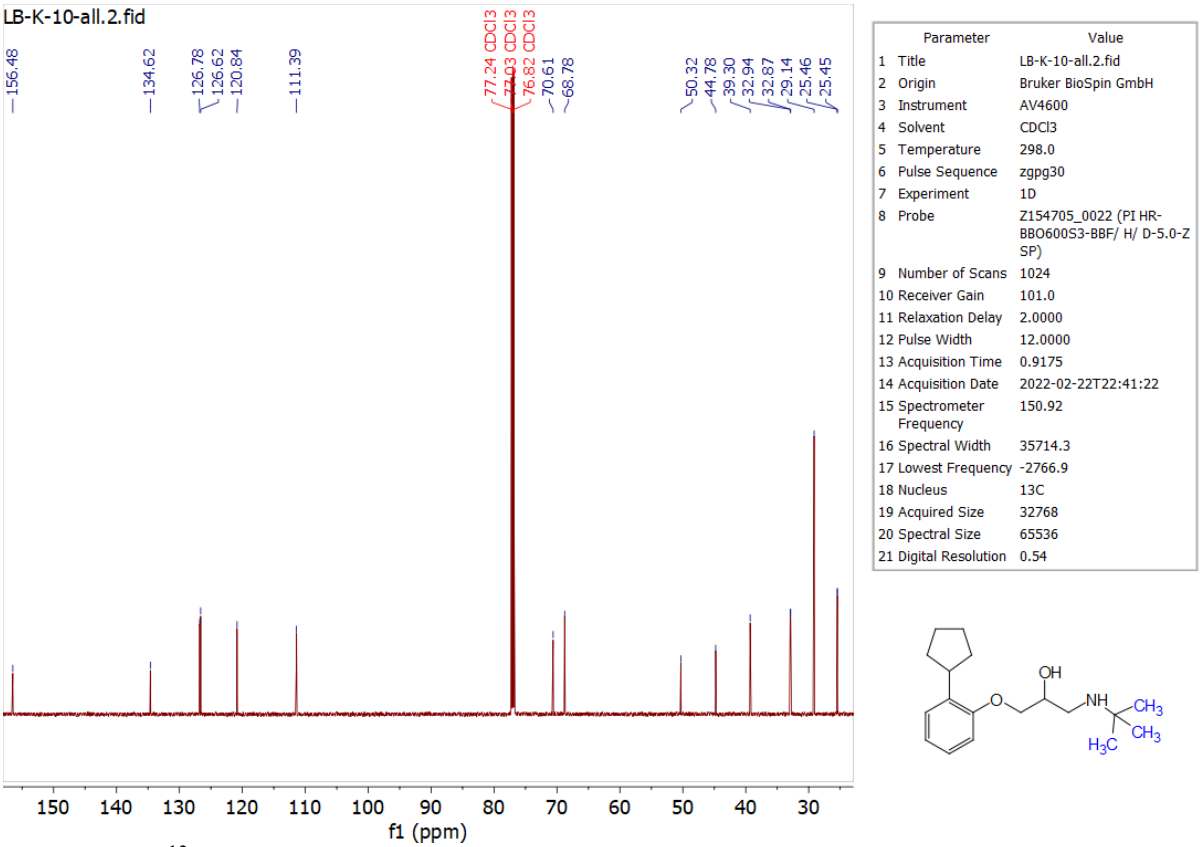


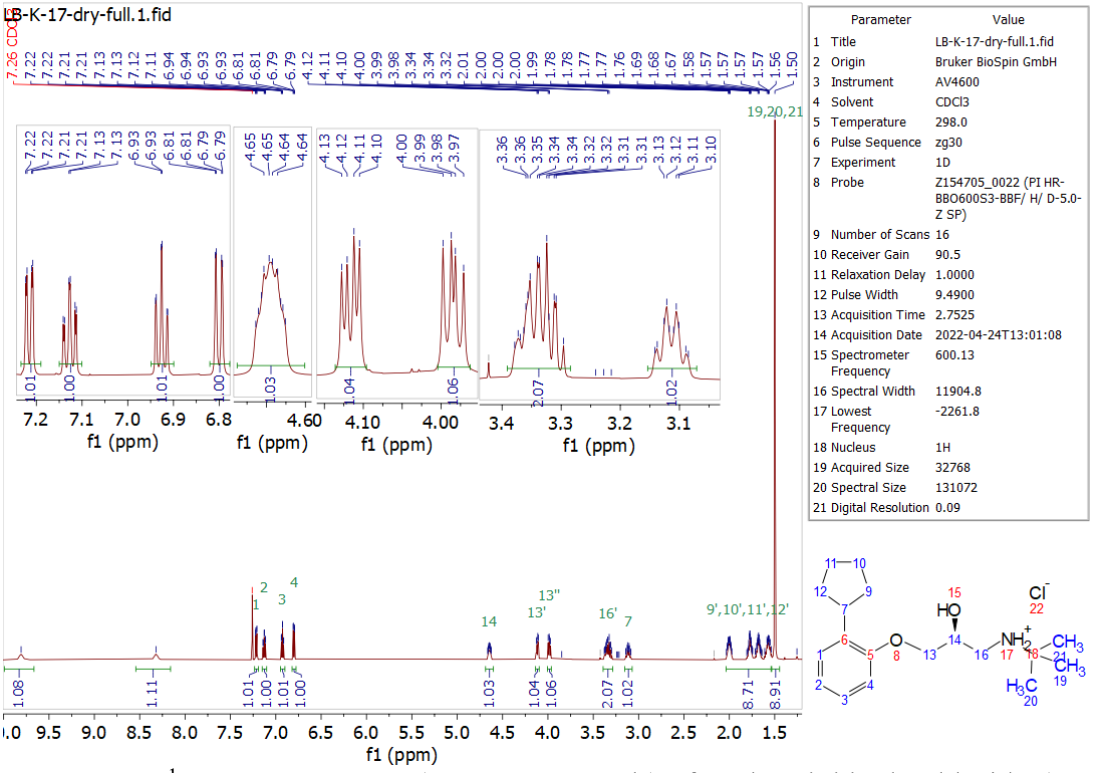
Figure S11. ^{1}H NMR spectrum (600 MHz, $CDCl_3$) of penbutolol (**10**).



Parameter	Value
1 Title	LB-K-10-all.2.fid
2 Origin	Bruker BioSpin GmbH
3 Instrument	AV4600
4 Solvent	CDCl3
5 Temperature	298.0
6 Pulse Sequence	zgpg30
7 Experiment	1D
8 Probe	Z154705_0022 (PI HR-BBO600S3-BBF/ H/ D-5.0-Z SP)
9 Number of Scans	1024
10 Receiver Gain	101.0
11 Relaxation Delay	2.0000
12 Pulse Width	12.0000
13 Acquisition Time	0.9175
14 Acquisition Date	2022-02-22T22:41:22
15 Spectrometer Frequency	150.92
16 Spectral Width	35714.3
17 Lowest Frequency	-2766.9
18 Nucleus	13C
19 Acquired Size	32768
20 Spectral Size	65536
21 Digital Resolution	0.54

Figure S12. ^{13}C NMR spectrum (150 MHz, $CDCl_3$) of penbutolol ***(10)***

*Penbutolol hydrochloride (***10**·HCl*)*



Parameter	Value
1 Title	LB-K-17-dry-full.1.fid
2 Origin	Bruker BioSpin GmbH
3 Instrument	AV4600
4 Solvent	CDCl3
5 Temperature	298.0
6 Pulse Sequence	zg30
7 Experiment	1D
8 Probe	Z154705_0022 (PI HR-BBO600S3-BBF/ H/ D-5.0-Z SP)
9 Number of Scans	16
10 Receiver Gain	90.5
11 Relaxation Delay	1.0000
12 Pulse Width	9.4900
13 Acquisition Time	2.7525
14 Acquisition Date	2022-04-24T13:01:08
15 Spectrometer Frequency	600.13
16 Spectral Width	11904.8
17 Lowest Frequency	-2261.8
18 Nucleus	1H
19 Acquired Size	32768
20 Spectral Size	131072
21 Digital Resolution	0.09

Figure S13. ^{1}H NMR spectrum (600 MHz, $CDCl_3$) of penbutolol hydrochloride ***(*10**·HCl).

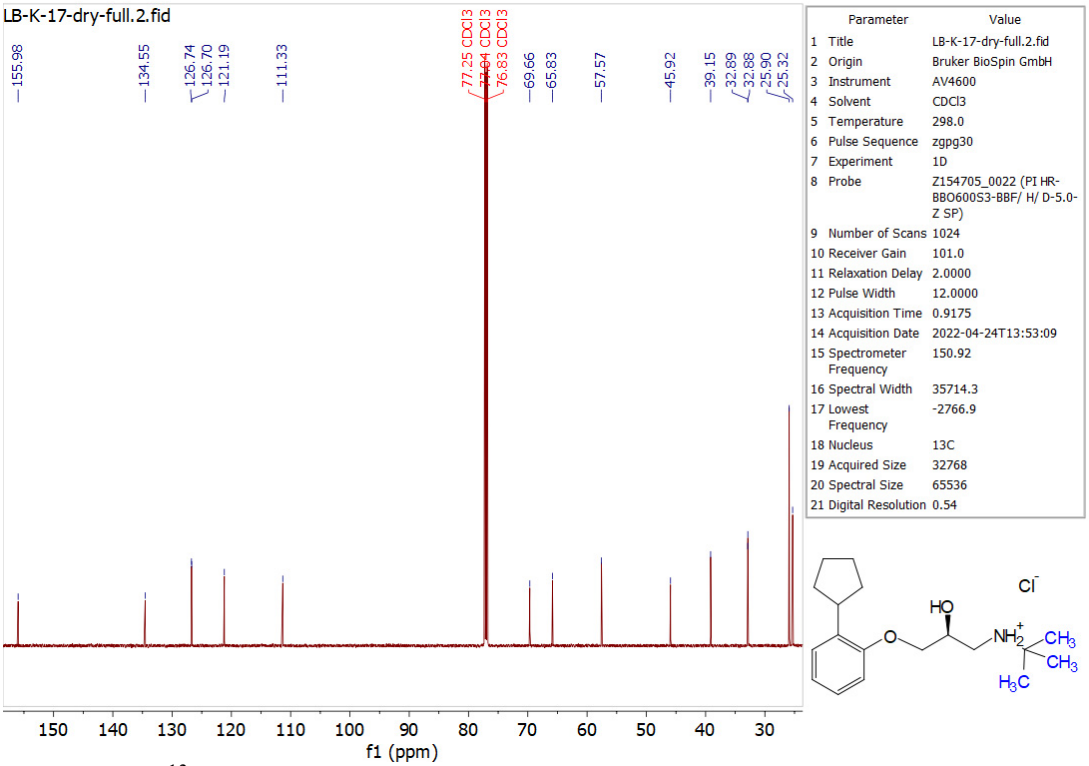


Figure S14. ^{13}C NMR spectrum (150 MHz, $CDCl_3$) of penbutolol hydrochloride (**10**·HCl).

HPLC DATA

*Methyl 3-(4-(3-chloro-2-hydroxypropoxy)phenyl)propanoate (**3**)*

```
Data File C:\CHEM32\1\DATA\SUSANNE HANSEN TROØYEN\20220410_A-ST016_NEW_RAC_CHLOROHYDR_C.D
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    ==================================================================
    Acq. Operator   : Susanne
    Acq. Instrument : Instrument 1                  Location : Vial 4
    Injection Date  : 10.04.2022 01:41:57
                                                  Inj Volume : 10 µl
    Acq. Method     : C:\CHEM32\1\METHODS\ELISABETH JACOBSEN\KIRAL_SHT_1.M
    Last changed    : 10.04.2022 01:36:59  by Susanne
    Analysis Method : C:\CHEM32\1\DATA\SUSANNE HANSEN TROØYEN\20220410_A-ST016_NEW_RAC_
                      CHLOROHYDR_C.D\DA.M (KIRAL_SHT_1.M)
    Last changed    : 10.04.2022 01:58:14  by Lucas
    Method Info     : Kiral separasjon med Chiralcel OD-H kolonne 250x4.6mm, 5um
                      Solventer: Heksan + ipa

    Sample Info     : 80% hexane 20% IPA sep of racemic chlorohydrin in n-hex
                      ane (esmolol)

    ==================================================================
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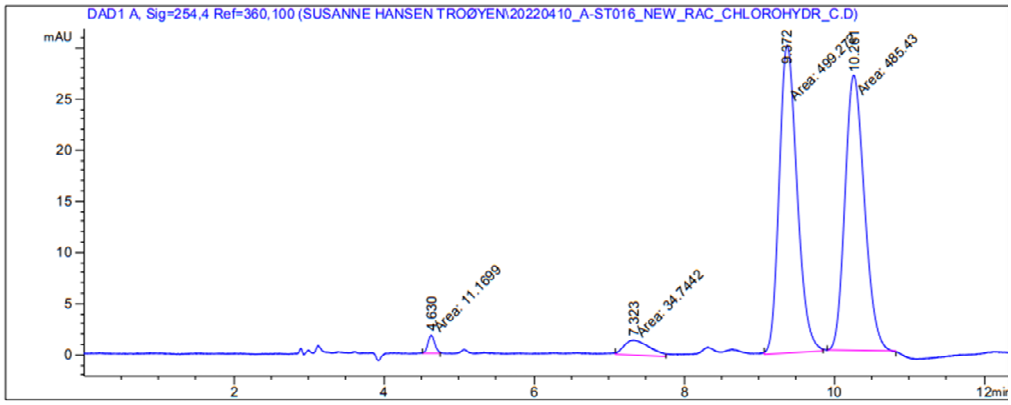


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    Dilution              :      1.0000
    Use Multiplier & Dilution Factor with ISTDs


    Signal 1: DAD1 A, Sig=254,4 Ref=360,100

    Peak RetTime Type  Width     Area      Height     Area
      #   [min]        [min]   [mAU*s]     [mAU]       %
    ----|-------|----|-------|----------|----------|--------|
       1   4.630 MM    0.1039   11.16994    1.79215   1.0838
       2   7.323 MM    0.3935   34.74419    1.47159   3.3712
       3   9.372 MM    0.2763  499.27225   30.11242  48.4440
       4  10.261 MM    0.3003  485.43021   26.93867  47.1010

    Totals :                  1030.61659   60.31484


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                            *** End of Report ***

Instrument 1 10.04.2022 01:58:57  Lucas                    Page   1 of 1
```

Figure S15. Chiral HPLC chromatogram of chlorohydrin **3**. The analysis was performed on a Chiralcel OD-H column with *n*-hexane and *i*-PrOH (80:20) as eluent and 1 mL/min flow, and with a detection wavelength of 254 nm. The retention times obtained are t_R((*S*)-**3**) = 9.4 min and t_R((*R*)-**3**) = 10.3 min. R_S((*S*)/(*R*)-**3**) = 1.86.

*(R)-Methyl 3-(4-(3-chloro-2-hydroxypropoxy)phenyl)propanoate ((R)-**3**)*

```
Data File C:\CHEM32\1\DATA\SUSANNE HANSEN TROØYEN\20220413_A-ST067_ALCOHOL_LOWC.D
Sample Name: A-ST067-F2-chloro_lowc
    ======================================================================
    Acq. Operator   : Susanne
    Acq. Instrument : Instrument 1                  Location : Vial 3
    Injection Date  : 13.04.2022 12:52:22
                                                  Inj Volume : 10 µl
    Acq. Method     : C:\CHEM32\1\METHODS\ELISABETH JACOBSEN\KIRAL_SHT_1.M
    Last changed    : 13.04.2022 12:33:23  by Susanne
    Analysis Method : C:\CHEM32\1\DATA\SUSANNE HANSEN TROØYEN\20220413_A-ST067_ALCOHOL_LOWC.D\DA.
                      M (KIRAL_SHT_1.M)
    Last changed    : 15.04.2022 05:11:51  by Lucas
    Method Info     : Kiral separasjon med Chiralcel OD-H kolonne 250x4.6mm, 5um
                      Solventer: Heksan + ipa

    Sample Info     : 80% hexane 20% IPA chlorohydrin sep for esmolol


    ======================================================================
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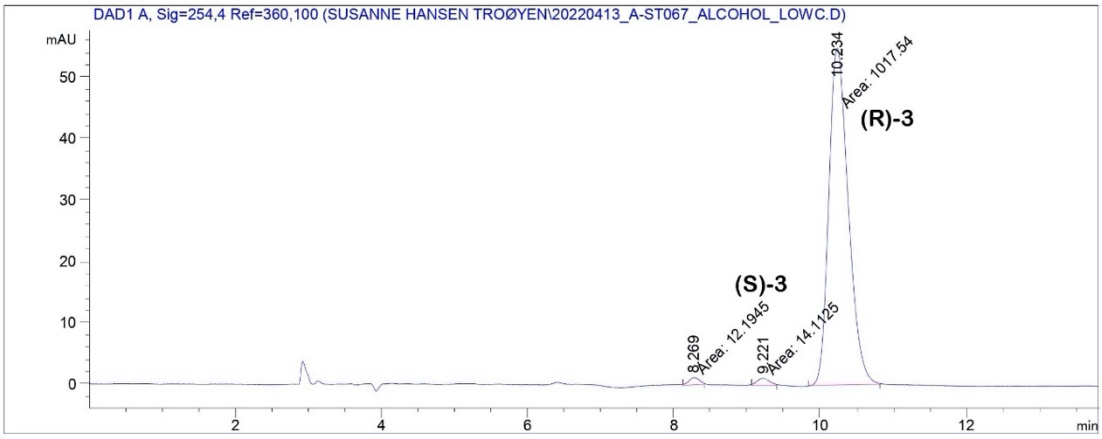


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    Use Multiplier & Dilution Factor with ISTDs


    Signal 1: DAD1 A, Sig=254,4 Ref=360,100

    Peak RetTime Type  Width     Area      Height     Area
      #   [min]        [min]   [mAU*s]     [mAU]       %
    ----|-------|----|-------|----------|----------|--------|
       1   8.269 MM    0.1678   12.19451    1.21100   1.1682
       2   9.221 MM    0.1956   14.11250    1.20225   1.3520
       3  10.234 MM    0.3090 1017.54163   54.88408  97.4798

    Totals :                  1043.84863   57.29733

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                              *** End of Report ***


Instrument 1 15.04.2022 05:13:23  Lucas                          Page   1 of 1
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Figure S16. Chiral HPLC chromatogram of (*R*)-chlorohydrin (*R*)-**3**. The analysis was performed on a Chiralcel OD-H column with *n*-hexane and *i*-PrOH (80:20) as eluent and 1 mL/min flow, and with a detection wavelength of 254 nm. The retention times obtained are t_R((*S*)-**3**) = 9.2 min and t_R((*R*)-**3**) = 10.2 min. R_S((*S*)/(*R*)-**3**) = 1.86.

(S)-1-chloro-3-(4-(3-methoxy-3-oxopropyl)phenoxy)propan-2-yl butanoate ((S)-4)

```
Data File C:\CHEM32\1\DATA\SUSANNE HANSEN TROØYEN\20220416_A-ST067_S-ESTER_PUR.D
Sample Name: A-ST067-ester
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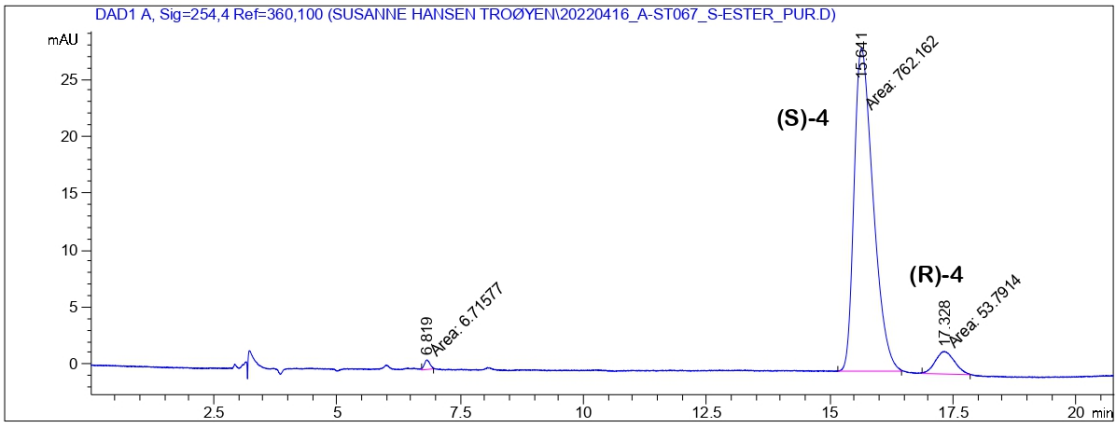
    Acq. Operator   : Susanne
    Acq. Instrument : Instrument 1                  Location : Vial 2
    Injection Date  : 16.04.2022 01:08:13
                                                   Inj Volume : 10 µl
    Acq. Method     : C:\CHEM32\1\METHODS\ELISABETH JACOBSEN\KIRAL_SHT_1.M
    Last changed    : 16.04.2022 01:07:32  by Susanne
    Analysis Method : C:\CHEM32\1\DATA\SUSANNE HANSEN TROØYEN\20220416_A-ST067_S-ESTER_PUR.D\DA.M
                      (KIRAL_SHT_1.M)
    Last changed    : 16.04.2022 01:30:38  by Susanne
    Method Info     : Kiral separasjon med Chiralcel OD-H kolonne 250x4.6mm, 5um
                      Solventer: Heksan + ipa

    Sample Info     : 97% hexane 3% IPA  ester esmolol sep, in IPA
```

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       1   6.819 MM    0.1314    6.71577 8.51701e-1   0.8163
       2  15.641 MM    0.4484  762.16235   28.32586  92.6450
       3  17.328 MM    0.4503   53.79138    1.99108   6.5386

    Totals :                  822.66950   31.16863

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                            *** End of Report ***

Instrument 1 16.04.2022 01:31:41  Lucas                          Page   1 of 1
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Figure S17. Chiral HPLC chromatogram of *S*-ester (*S*)-**4**. The analysis was performed on a Chiralcel OD-H column with *n*-hexane and *i*-PrOH (97:3) as eluent and 1 mL/min flow, and with a detection wavelength of 254 nm. The retention times obtained are t_R((*S*)-**4**) = 15.6 min and t_R((*R*)-**4**) = 17.3 min. R_S((*S*)/(*R*)-**4**) = 2.08.

*Esmolol (**5**)*

```
Data File C:\CHEM32\...ANNE HANSEN TROØYEN\20211205_ESMOLOL 2021-12-05 14-13-49\004-0401.D
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    Acq. Instrument : Instrument 1                   Location : Vial 4
    Injection Date  : 05.12.2021 03:42:58                 Inj :   1
                                                   Inj Volume : 10 µl
    Acq. Method     : C:\Chem32\1\DATA\SUSANNE HANSEN TROØYEN\20211205_ESMOLOL 2021-12-05 14-13-
                      49\KIRAL_3_DEA.M
    Last changed    : 05.12.2021 02:12:28  by Susanne
    Analysis Method : C:\CHEM32\1\DATA\SUSANNE HANSEN TROØYEN\20211205_ESMOLOL 2021-12-05 14-13-
                      49\004-0401.D\DA.M (KIRAL_3_DEA.M)
    Last changed    : 15.04.2022 05:17:31  by Lucas
                      (modified after loading)
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                      Solventer: Heksan + ipa

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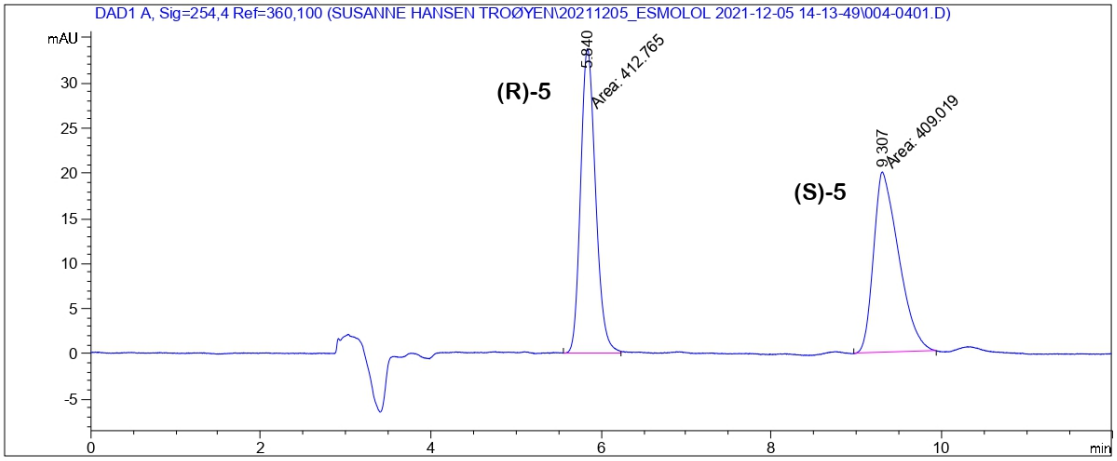


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       1   5.840 MM    0.2040  412.76514   33.72710  50.2280
       2   9.307 MM    0.3411  409.01855   19.98699  49.7720

    Totals :                    821.78369   53.71409


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Instrument 1 15.04.2022 05:17:34  Lucas                          Page   1 of 1
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Figure S18. Chiral HPLC chromatogram of racemic esmolol **5**. The analysis was performed on a Chiralcel OD-H column with n-hexane:*i*-PrOH:Et_2NH (80:19.6:0.4) as eluent and 1 mL/min flow, and with a detection wavelength of 254 nm. The retention times obtained are t_R((*R*)-**5**) = 5.8 min and t_R((*R*)-**5**) = 9.3 min. R_S((*S*)/(*R*)-**5**) = 11.5.

*(S)-Esmolol ((S)-**5**)*

```
Data File C:\CHEM32\1\DATA\SUSANNE HANSEN TROØYEN\20220415_A-ST069_S-ESMOLOL_PUR2.D
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    Acq. Operator   : Susanne
    Acq. Instrument : Instrument 1                  Location : Vial 1
    Injection Date  : 15.04.2022 04:53:28
                                                   Inj Volume : 10 µl
    Acq. Method     : C:\CHEM32\1\METHODS\ELISABETH JACOBSEN\KIRAL_3_DEA.M
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    Analysis Method : C:\CHEM32\1\DATA\SUSANNE HANSEN TROØYEN\20220415_A-ST069_S-ESMOLOL_PUR2.D\
                      DA.M (KIRAL_3_DEA.M)
    Last changed    : 15.04.2022 05:07:29  by Lucas
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                      Solventer: Heksan + ipa

    Sample Info     : 80% hexane 20% IPA w 2% DEA esmolol sep, in IPA
                      Purified by TLC, lower c



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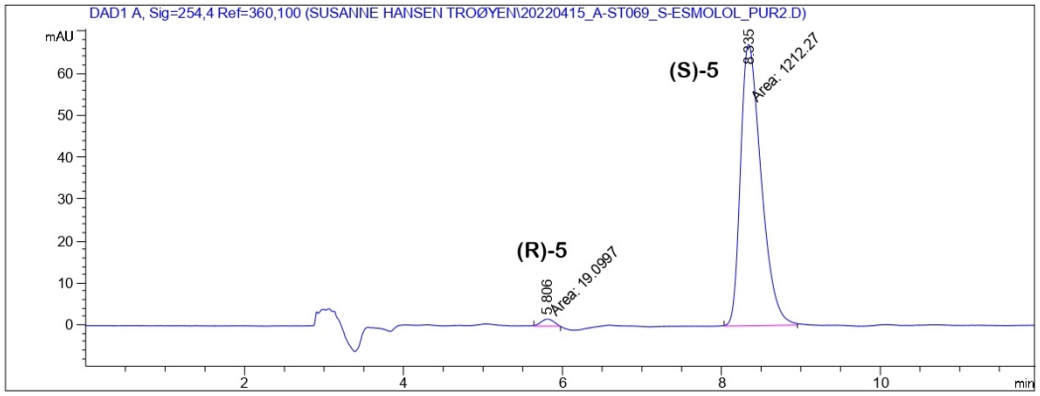


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       2   8.335 MM    0.3010 1212.27112   67.12397  98.4489

    Totals :                   1231.37085   68.87037

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                            *** End of Report ***

Instrument 1 15.04.2022 05:10:37  Lucas                          Page   1 of 1
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Figure S19. Chiral HPLC chromatogram of (*S*)-esmolol (*S*)-**5**. The analysis was performed on a Chiralcel OD-H column with n-hexane:*i*-PrOH:Et_2NH (80:19.6:0.4) as eluent and 1 mL/min flow, and with a detection wavelength of 254 nm. The retention times obtained are t_R((*R*)-**5**) = 5.8 min and t_R((*R*)-**5**) = 8.3 min. R_S((*S*)/(*R*)-**5**) = 11.5.

*3,3'-(((2-Hydroxypropane-1,3-diyl)bis(oxy))bis(4,1-phenylene))dipropanoate (**3d**)*

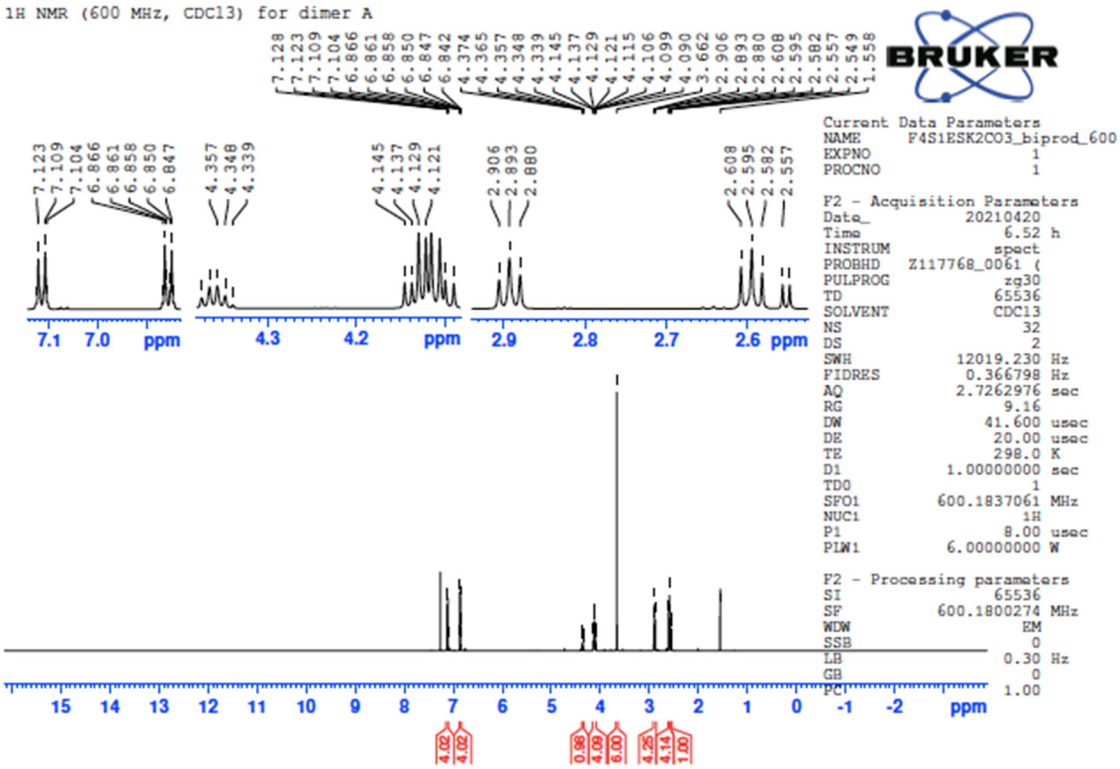

Figure S20. ^{1}H-NMR spectrum (600 MHz, $CDCl_3$) of dimer **3d**. δ = 1.56 ppm corresponds to water from the solvent.

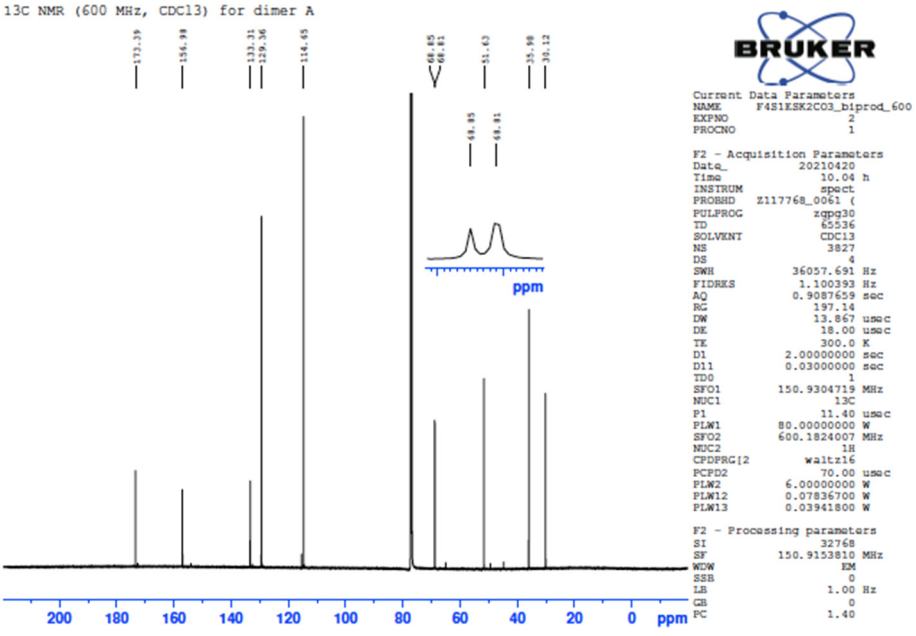

Figure S21. ^{13}C-NMR spectrum (600 MHz, $CDCl_3$) of dimer **3d**.

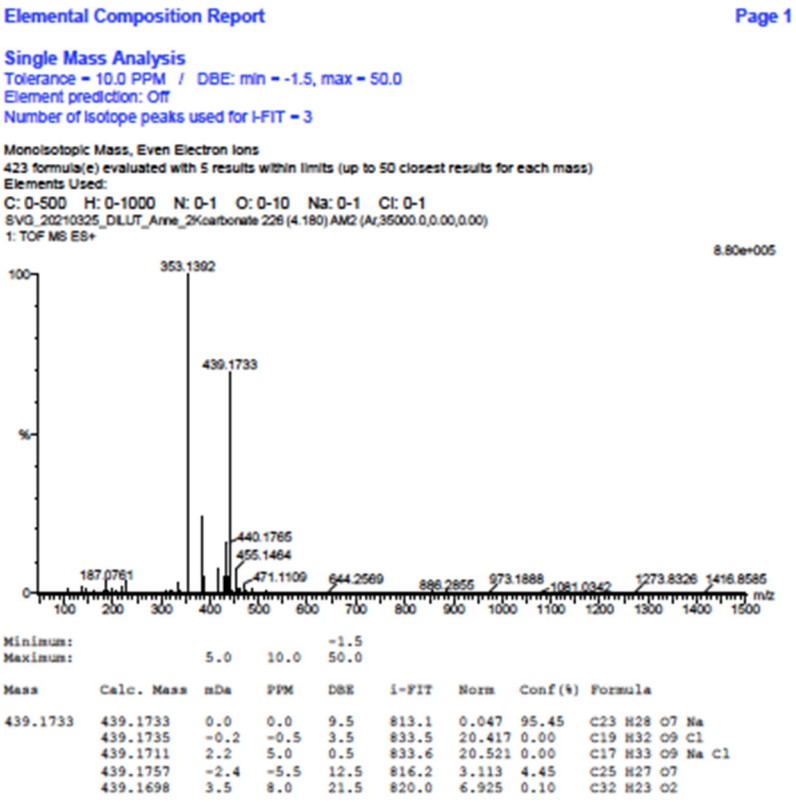

Figure S22. MS-spectrum of dimer **A** with *m/z* = 416.46

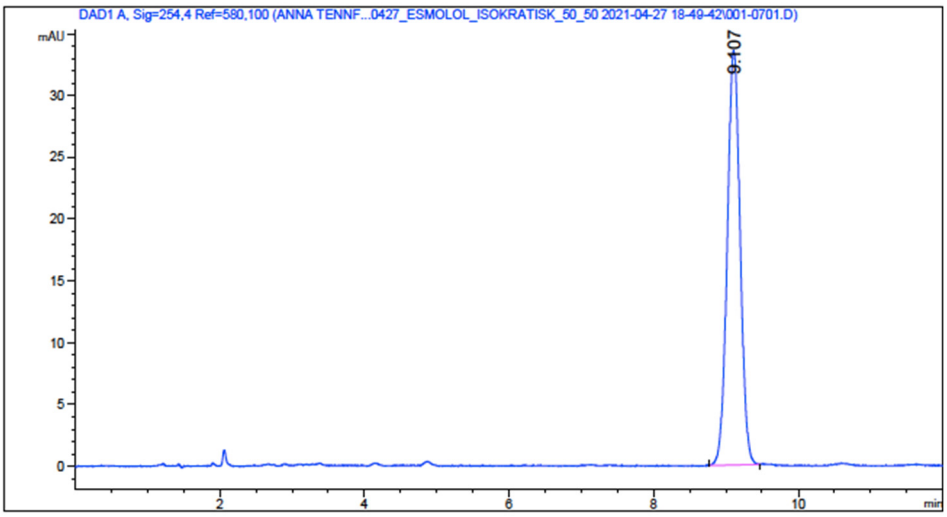

Figure S23. Achiral HPLC of isolated dimer **A** (t_R = 9.1 min) performed on an ACE Excel 5 C18 column with an isocratic mobile phase composition of water and acetonitrile (50:50) over 12 min, flow 1 mL/min.

*1-chloro-3-(2-cyclopentylphenoxy)propan-2-ol (**8**)*

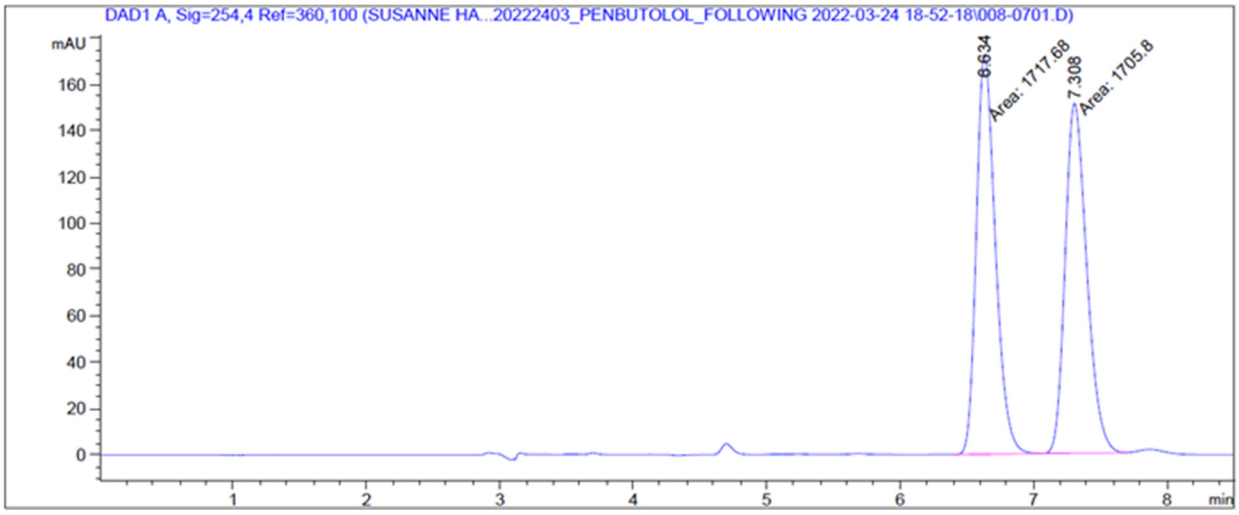

Figure S24. Chiral HPLC chromatogram of chlorohydrin **8**. The analysis was performed on a Chiralcel OD-H column with hexane and 2-propanol (90:10) as eluent and 1 mL/min flow, and with a detection wavelength of 280.8 nm. The retention times obtained are t_R((*S*)-**8**) = 6.63 min and t_R((*R*)-**8**) = 7.31 min. R_S((*S*)/(*R*)-**8**) = 2.41.

*(S)-1-chloro-3-(2-cyclopentylphenoxy)propan-2-ol ((S)-**8**)*

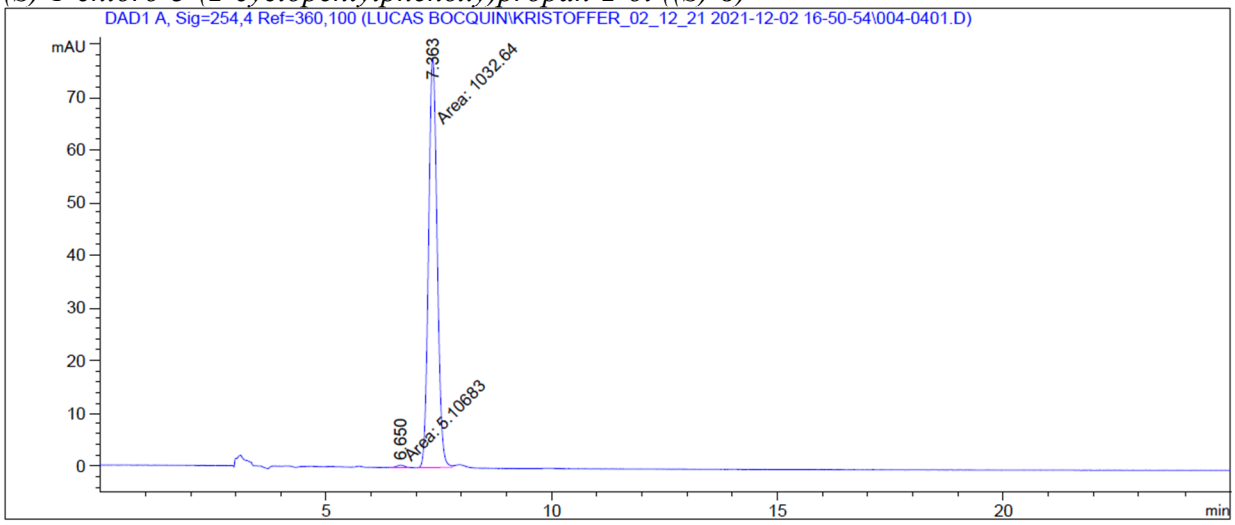

Figure S25. Chiral HPLC chromatogram of enantiopure chlorohydrin *(S)*-***8*** obtained by the CALB catalysed kinetic resolution of racemic chlorohydrin **8**. The analysis was performed on a Chiralcel OD-H column with hexane and 2-propanol (90:10) as eluent and 1 mL/min flow, and with a detection wavelength of 280.8 nm.

*Penbutolol (**10**)*

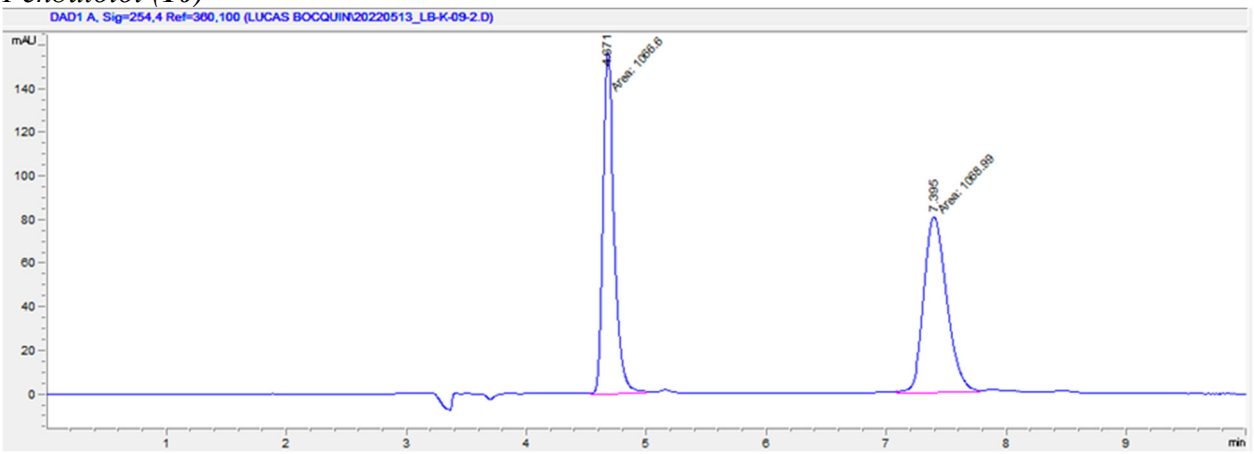

Figure S26. Chiral HPLC chromatogram of racemic of racemic penbutolol (**10**). The analysis was performed on a Chiralcel OD-H column with hexane (90%) and 2-propanol containing 2% diethanolamine (10%) as eluent and 1 mL/min flow, and with a detection wavelength of 254.4 nm. The retention times obtained are t_R((*R*)-**10**) = 4.671 min and t_R((*S*)-**10**) = 7.395 min. R_S((*S*)/(*R*)-**10**) = 10.5.

*(S)-Penbutolol ((S)-**10**)*

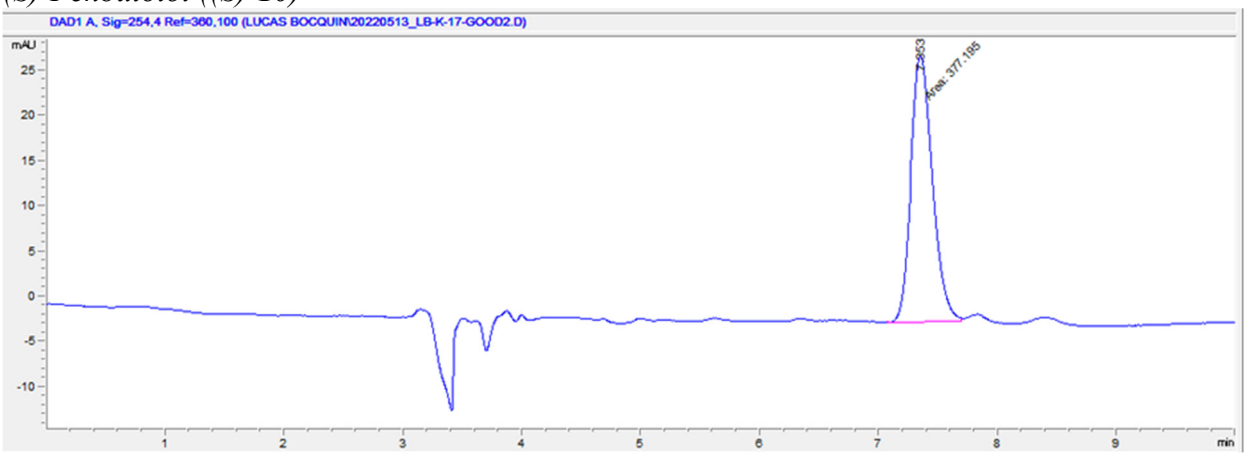

Figure S27. Chiral HPLC chromatogram of enantiopure (*S*)-penbutolol ((*S*)-**10**). The analysis was performed on a Chiralcel OD-H column with hexane (90%) and 2-propanol containing 2% diethanolamine (10%) as eluent and 1 mL/min flow, and with a detection wavelength of 254.4 nm.