Asymmetric Chemoenzymatic Synthesis of 1,3-Diols and 2,4-Disubstituted Aryloxetanes by Using Whole Cell Biocatalysts

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ELECTRONIC SUPPLEMENTARY INFORMATION

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Materials and methods

^{1}H NMR and ^{13}C NMR spectra were recorded on a Bruker 600 MHz or on a Varian Inova 400 MHz spectrometer and chemical shifts are reported in parts per million (δ). ^{19}F NMR spectra were recorded by using $CFCl_3$ as an internal standard. Absolute values of the coupling constants are reported. FT-IR spectra were recorded on a Perkin-Elmer 681 spectrometer. GC analyses were performed on a HP 6890 model, Series II by using a HP1 column (methyl siloxane; 30 m x 0.32 mm x 0.25 µm film thickness). Analytical thin-layer chromatography (TLC) was carried out on pre-coated 0.25 mm thick plates of Kieselgel 60 F_{254}; visualisation was accomplished by UV light (254 nm) or by spraying a solution of 5 % (w/v) ammonium molybdate and 0.2 % (w/v) cerium(III) sulfate in 100 mL 17.6 % (w/v) aq. sulfuric acid and heating to 473 K until blue spots appeared. Chromatography was conducted by using silica gel 60 with a particle size distribution 40-63 µm and 230-400 ASTM. GC-MS analyses were performed on HP 5995C model. MS-ESI analyses were performed on Agilent 1100 LC/MSD trap system VL. The high resolution mass spectrometry (HRMS) analyses were performed using a Bruker microTOF QII mass spectrometer equipped with an electrospray ion source (ESI) operating in positive ion mode. Optical rotation values were measured at 25 °C using a Perkin Elmer 341 polarimeter with a cell of 1 dm path length; the concentration (c) is expressed in g/100 mL. Enantiomeric ratios (er) were determined by HPLC analysis using Phenomenex LUX Cellulose-1 [Cellulose tris(3,5-dimethylphenylcarbamate)], LUX Cellulose 2 [Cellulose 2 tris(3-chloro-4-methylphenylcarbamate)] and LUX Cellulose-4 [Cellulose tris(4-chloro-3-methylphenylcarbamate)] columns (250 x 4.6 mm), or by GC-analyses performed on a Hewlett–Packard 6890 Series II chromatograph equipped with a Chirasil-DEX CB (250x0.25 µm) capillary column; column head pressure = 18 psi, He flow 2 mL/min, split ratio 100/1, T (oven) from 90 to 120 °C. All the chemicals and solvents were of commercial grade and further purified by distillation or crystallization prior to use.

Synthetic procedures. All the optically active aldols **2a**–**e** and diols **3a**–**e** obtained by bioreduction had analytical and spectroscopic data identical to those previously reported or to the commercially available compounds. The aldols **2a**–**e** and diols **3a**–**e** were prepared also as racemic mixtures, starting from the corresponding 1,3-diketones, (for HPLC references) by $NaBH_4$ reduction in EtOH in 89–95% yields, according to the reported procedures,[1] excepted when otherwise specified.

Microorganisms and cultures. *Saccharomyces cerevisiae* CBS 7336, *Kluyveromyces marxianus* CBS 6556,[1] *Yarrovia Lipolytica* Y-16,[2] *Trigonopsis variabilis* DSM 70714 were obtained from public type culture collections (CBS, DSM), and were cultivated under aerobic conditions in a medium containing 0.3% yeast extract, 0.3% malt extract, 0.5% peptone, 1% glucose. Agar-agar (2%) was added to the same medium for cells preservation on agar slants. *Lactobacillus reuteri* DSM 20016 was obtained from DSMZ culture collection (Germany).[3] Cells were maintained at –80°C in culture broth supplemented with 25% (w/v) glycerol. Pre-cultures and cultures were carried out in classical MRS medium[4] (Oxoid) containing: 20g/L glucose, 10 g/L peptone, 8 g/L meat extract, 4 g/L yeast extract, 1 g/L Tween 80, 2 g/L di-potassium hydrogen phosphate, 5 g/L sodium acetate·$3H_2O$, 2 g/L tri-ammonium citrate, 0.2 g/L of magnesium sulfate·$7H_2O$, 0.05 g/L manganese sulfate·$2H_2O$. Cells were incubated at 37 °C for 24 h, statically. Cell density was monitored using optical density at 620 nm (OD620) with a spectrophotometer Genesys TM 20 (Thermo Fisher Scientific Inc., Waltham, Massachusetts, USA).

Blank experiments. A 1 L flask containing 400 mL of the culture medium was stirred at 30 °C on an orbital shaker at 200 rpm and diketone (**1a**–**e**) (50 mg) was added. The reaction was monitored by TLC and stopped after 24 or 96 h. The content of the flask was extracted with Et_2O and analyzed by GC-MS or NMR analyses.

Bioreduction of 1,3-aryldiketones 1a–e by yeast growing cells (GC). General procedure. Cells preserved on agar slants at 4 °C were used to inoculate 250 mL flasks containing 100 mL of the culture medium. The flasks were incubated aerobically at 30 °C on an orbital shaker and stirred at 250 rpm. Flasks (250 mL) containing 100 mL of the culture medium were then inoculated with 5 mL of the 24-h-old suspension and incubated in the same conditions for 24 h. Flasks (1 L) containing 400 mL of the culture medium were then inoculated with 5 mL of the latter suspension and incubated for 24 h. The optical density was checked at 620 nm for all cultures before aryldiketone (**1a**–**e**) (100 mg), dissolved in 1 mL of EtOH, was added. The progress of the reactions was monitored by TLC and/or GC and stopped at the times indicated in Table 1. The content of the flask was then centrifuged and the supernatant extracted with EtOAc. All the reactions were repeated at least twice without any noticeable bias in the results. With the exception of diketone **1a**, all the other substrates were successfully reduced in all the conditions used (data reported in Table 1 show that each yeast behaves differently for each substrate). The residue was purified by silica gel column chromatography using hexane and EtOAc (90:10 or 80:20) as eluents to yield the desired aldol (**2a**–**e**), as reported in Table 1.

Bioreduction of 1,3-aryldiketones 1a-e by yeast resting cells (RC). General procedure. Culture conditions were the same as in the case of growing cells. Once cell growth reached the stationary phase, cells were harvested by centrifugation (20 min and 4000 rpm) at 4 °C. The cells were then recovered and washed three times (20 mL each time) with 0.1 M KH_2PO_4 buffer pH = 7. The cells were collected by centrifugation as previously described. The cell wet mass, free from culture medium, was re-suspended in the same buffer solution enriched with 1% glucose to reach a concentration of 0.5 g/L. Then, the substrate of the reaction (2 mM final concentration) was added to the reaction medium. The flask was shaken (250 rpm) in an orbital shaker and kept at 30 °C. The reaction progress was monitored by TLC analysis and stopped at times indicated in Table 1. The content of the flask was then centrifuged and the supernatant extracted with EtOAc. All the reactions were repeated at least twice without any noticeable change in the results.

Synthesis of racemic aldol 2b with $NaBH_4$. Typical procedure. To a solution of 4,4,4-trifluoro-1-phenylbutane-1,3-dione (**1b**) (108 mg, 0.5 mmol) in EtOH (1 mL), stirred at 0 °C, $NaBH_4$ (38 mg, 1 mmol) was added. After 16 h, water was added and the aqueous solution extracted with EtOAc. The residue was purified by silica gel column chromatography using hexane and EtOAc (80:20) as eluents to yield 6% of 4,4,4 trifluoro-1-phenylbutane-1,3 diol (**3b**), together with 87% of (*rac*)-4,4,4-trifluoro–3-hydroxy-1-phenylbutan-1-one (**2b**).[5,6]

Synthesis of (*rac*)-4,4,4-trifluoro-1-(furan-2-yl)-3-hydroxybutane-1-one (**2c**). To a solution of 4,4,4-trifluoro-1-(furan-2-yl)butane-1,3-dione (**1c**) (0.184 g, 4.6 mmol) in dry hexane (10 mL) kept at –78 °C, a solution of DIBAL-H (4.16 ml, 4.16 mmol, 1M in THF) was added dropwise. After stirring the yellow reaction mixture for 2 h, water and a sat. aq. solution of $NaHCO_3$ (5 mL) were sequentially added, and the solution extracted with EtOAc (3 x 15 mL). The residue was purified by silica gel column chromatography (hexane: EtOAc = 7:3), and (*rac*)-4,4,4-trifluoro-1-(furan-2-yl)-3-hydroxybutane-1-one (**2c**)[5,6] was isolated in 90% yield.

Synthesis of (*rac*)-3-hydroxy-1-phenylbutan-1-one (2d). To a solution of NaOH (0.184 g, 4.6 mmol) in water (1 mL) and EtOH (2 mL) kept at 0 °C, acetophenone (0.5 g, 4.16 mmol) and acetaldehyde (0.3 mL, 5.3 mmol) were sequentially added. After stirring for 15 min, NH_4Cl (5 mL) was added, and the solution extracted with EtOAc (3 x 15 mL). The residue was purified by silica gel column chromatography using hexane and EtOAc (7:3) as eluents to yield 66% of (*rac*)-3-hydroxy-1-phenylbutan-1-one (**2d**).[7]

Synthesis of (*rac*)-4,4,4-trifluoro-1-(furan-2-yl)butane-1,3-diol (3c).[5] To a solution of (*rac*)-4,4,4-trifluoro-1-(furan-2-yl)-3-hydroxybutane-1-one **(2c)** (0. 296 g, 1.42 mmol) in dry hexane (6 mL) kept at 0 °C, a solution of DIBAL-H (2.8 mL, 2.8 mmol, 1M in THF) was added dropwise. After stirring the yellow reaction mixture for 2 h, HCl 1 M was added, and the solution extracted with EtOAc (3 x 15 mL). The residue was purified by silica gel column chromatography (hexane: EtOAc = 8:2), affording the expected diol **3c** in 91% yield (0.260 g).

Spectroscopic Data

3-Hydroxy-1,3-diphenylpropan-1-one (2a).[8] ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.97–7.94 (m, 2 H), 7.61–7.56 (m, 1 H), 7.49–7.43 (m, 4 H), 7.40–7.35 (m, 2 H), 7.33–7.28 (m, 1 H), 5.38–5.33 (m, 1 H), 3.57 (d, *J* = 2.86 Hz, 1 H, exchanges with D_2O), 3.38 (d, *J* = 6.03 Hz, 1 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 211.4, 133.6, 128.7, 128.6, 128.1, 127.7, 125.7, 70.1, 47.4; FT IR (neat): 3386, 3014, 2852, 1682, 1600, 1582, 749, 698 cm^{-1}. ESI-MS: 249 $[M+Na]^+$; m^2 MS/MS: 129 ($C_6H_5COCH_2Na^+$). **(*S*)-2a**: 50% yield (52 mg) from bioreduction of **1a** with *Saccharomyces cerevisiae* CBS 7336 (Table 1, entry 5), purified by silica gel column chromatography using hexane and EtOAc (7:3) as eluent. Er (*S*):(*R*) = 79:21 determined by HPLC, LUX Cellulose-1 column (hexane:2-propanol 90:10), 0.5 mL/min, t_R [major (*S*)-enantiomer] = 26.1 min, t_R [minor (*R*)-enantiomer] = 28.4 min. $[\alpha]_D^{20}$= –13.6° (c 0.83, $CHCl_3$).

4,4,4-Trifluoro–3-hydroxy-1-phenylbutan-1-one (2b).[5,6] ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.01–7.96 (m, 2 H), 7.66–7.61 (m, 1 H), 7.55–7.48 (m, 2 H), 4.75–4.65 (m, 1 H), 3.64–3.56 (bs, 1 H, exchanges with D_2O), 3.39 (dd, *J* =17.8, 9.2 Hz, 1 H,), 3.31 (dd, *J* = 17.8, 2.8 Hz, 1 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 197.5, 136.0, 134.1, 128.9, 128.2, 124.7 (q, $^1J_{C\text{-}F}$ = 280.0 Hz), 123.3, 67.0 (q, $^2J_{C\text{-}F}$ = 32.0 Hz), 38.2; ^{19}F NMR (376 MHz, $CDCl_3$) δ –81.0 (d, *J* = 6.6 Hz); FT IR (neat): 3386, 2917, 2849, 1685, 1596, 1581, 1452, 1385, 1349, 1279, 1228, 1154, 1125, 1105, 883, 755, 686 cm^{-1}. GC MS (70 eV) *m/z* (%) 218 (M^+, 4), 198 (9), 105 (100), 77 (46), 69 (6), 51 (13). **(*S*)-2b**: 80% yield (81 mg) by bioreduction of **1b** with *Lactobacillus reuteri* (Table 1, entry 14), purified by silica gel column chromatography using hexane and EtOAc (8:2) as eluent. Er (*S*):(*R*) = 92:8, determined by HPLC, LUX Cellulose-1 column (hexane:2-propanol 90:10), 1 mL/min, t_R [major (*S*)-enantiomer) = 7.2 min, t_R [minor (*R*)-enantiomer] = 8.0 min. $[\alpha]_D^{20}$ = – 1 (*c* = 1, MeOH).

4,4,4-Trifluoro-1-(furan-2-yl)-3-hydroxybutane-1-one (2c).[5,6] ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.62–7.60 (m, 1 H), 7.29–7.27 (m, 1 H), 6.57–6.54 (m, 1 H), 4.70–4.64 (m, 1 H), 4.1–3.90 (bs, 1 H, exchanges with D_2O), 3.32–3.19 (m, 1 H), 3.14–3.08 (m, 1 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 185.8, 151.9, 147.7, 124.7 (q, $^1J_{C\text{-}F}$ = 280.0 Hz), 66.5 (q, $^1J_{C\text{-}F}$ = 32.0 Hz), 38.1; ^{19}F NMR (376 MHz, $CDCl_3$) δ –79.6 (d, $^1J_{C\text{-}F}$ = 32.0 Hz); GC MS (70 eV) *m/z* (%): 208 (M^+, 9), 188 (16), 110 (5), 95 (100), 68 (3), 43 (3). **(*S*)-2c**: 78% yield (79 mg) by bioreduction of **1c** with *Kluyveromyces marxianus* CBS 6556 (Table 1, entry 16), purified by silica gel column chromatography using hexane and EtOAc (7:3) as eluent. Er (*S*):(*R*) = 69:31, determined by HPLC, LUX Cellulose-1 column (hexane:2-propanol 90:10), 0.5 mL/min, t_R [major (*S*)-enantiomer] = 15.9 min, t_R [minor (*R*)-enantiomer] = 20.3 min, $[\alpha]_D^{20}$ = –12.6° (c = 1.0, $CHCl_3$).

3-Hydroxy-1-phenylbutan-1-one (2d).[7] ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.96–7.93 (m, 2 H), 7.60–7.56 (m, 1 H), 7.50–7.45 (m, 2 H), 4.45–4.37 (m, 1 H), 3.60–3.20 (bs, 1 H, exchanges with D_2O), 3.16 (dd, *J* = 17.6, 2.7 Hz, 1 H), 3.05 (dd, *J* = 17.6, 8.8 Hz, 1 H), 1.30 (d, *J* = 6.6 Hz, 3 H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 200.8, 136.7, 133.5, 128.6, 128.0, 64.0, 46.5, 22.4; FT-IR (neat): 3439, 3063, 3023, 2923, 2854, 1739, 1682, 1598, 1581, 1449, 1416, 1376, 1211, 1104, 1002, 937, 800, 754, 690 cm^{-1}; GC MS (70 eV) *m/z* (%): 164 (M^+, 3), 146 (14), 120 (15), 106 (12), 105 (100), 78 (11), 77 (47), 51 (13). **(*R*)-2d**: 95% yield (96 mg) by bioreduction of **1d** with *Lactobacillus reuteri* (Table 1, entry 20), purified by silica gel column chromatography using hexane and EtOAc (7:3) as eluent. Er (*S*):(*R*) = 2:98, determined by HPLC, LUX Cellulose-1 column (hexane:2-propanol 90:10), 1 mL/min, t_R [major (*R*)-enantiomer] = 8.1 min, t_R [minor (*S*)-enantiomer] = 8.9 min. $[\alpha]_D^{20}$ = – 60 (*c* = 1, $CHCl_3$).

4,4,4-Trifluoro-3-hydroxy-1-(naphthalen-2-yl)butan-1-one (2e).[5] ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.48 (s, 1 H), 8.04–7.98 (m, 2 H), 7.94–7.89 (m, 2 H), 7.67–7.57 (m, 2 H), 4.78–4.73 (m, 1 H), 3.80–3.40 (bs, 1 H, exchanges with D_2O), 3.57–3.42 (m, 2 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 200.8, 136.7, 133.5, 128.6, 128.0, 64.0, 46.5, 22.4; FT-IR (neat): 3439, 3063, 3023, 2923, 2854, 1739, 1682, 1598, 1581, 1449, 1416, 1376, 1211, 1104, 1002, 937, 800, 754, 690 cm^{-1}; GC MS (70 eV) *m/z* (%): 164 (M^+, 3), 146 (14), 120 (15), 106 (12), 105 (100), 78 (11), 77 (47), 51 (13). **(*R*)-2e**: 40% yield (41 mg) by bioreduction of **1e** with *Baker's yeast* (Table 1, entry 23), purified by silica gel column chromatography using hexane and EtOAc (8:2) as eluent. Er (*S*):(*R*) = 10:90, determined by HPLC, LUX Cellulose-1 column (hexane:2-propanol 90:10), 0.8 mL/min, t_R [minor (*S*)-enantiomer] = 15.8 min, t_R [major (*R*)-enantiomer] = 26.4 min, $[\alpha]_D^{20}$ = +12.9 (*c* = 1, $CHCl_3$).

***anti*-1,3-Diphenylpropan-1,3-diol (3a).**[9] ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.39–7.25 (m, 10 H), 4.96 (dt, *J* = 6.1, 2.8 Hz, 2 H), 2.98–2.90 (bs, 2 H, exchange with D_2O), 2.16 (t, *J* = 6.1 Hz, 2 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 144.1, 128.4, 127.7, 125.6, 72.7, 46.4; FT-IR (neat): 3361, 3080, 3062, 3030, 2920, 2854, 1603, 1494, 1454, 1403, 1323, 1202, 1089, 1060, 1028, 914, 854, 754, 699 cm^{-1}; GC MS (70 eV) *m/z* (%): 226 (M^+, 2), 210 (46), 107 (22), 106 (13), 105 (57), 104 (100), 103 (12), 79 (31), 77 (47), 51 (12). ***anti*-(1*S*,3*S*)-3a:** 76% yield (77 mg) by bioreduction of **1a** with *Kluyveromyces marxianus* CBS 6556 (Table 1, entry 6), purified by silica gel column chromatography using hexane and EtOAc (6:4) as eluent. Er (1*S*,3*S*):(1*R*,3*R*) = 54:46 [HPLC, LUX Cellulose-1 column (hexane:2-propanol 90:10), 0.8 mL/min, t_R [major (*S,S*)-enantiomer] = 7.2 min, t_R [minor (*R,R*)-enantiomer] = 7.8 min, $[\alpha]_D^{20}$ = –15.4 (c =1.0, $CHCl_3$).

***anti*-4,4,4-Trifluoro-1-phenylbutane-1,3 diol (3b).**[5,6] ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.65–7.25 (m, 5 H), 5.12 (dd, *J* = 8.9, 3.3 Hz, 1 H), 4.32–4.18 (m, 1 H), 3.40–3.10 (bs, 1 H, exchanges with D_2O), 2.30–1.95 (bs, 1 H, exchanges with D_2O), 2.18–1.99 (m, 2 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 143.1 (major stereoisomer), 128.9, 128.7, 128.5, 125.2 (q, $^{1}J_{C\text{-}F}$ = 281.0 Hz), 70.7, 67.9 (q, $^{2}J_{C\text{-}F}$ = 31.0 Hz), 37.5; ^{19}F NMR (376 MHz; $CDCl_3$) δ –79.4 (d, *J* = 6.9 Hz); FT-IR (neat): 3391, 3036, 2920, 2852, 1455, 1131, 918, 876, 752,701 cm^{-1}; GC MS (70 eV) *m/z* (%) 220 (M^+, 7), 201 (1), 133 (4), 108 (8), 107 (100), 105 (14), 91 (4), 79 (51), 77 (30), 65 (2), 51 (9), 43 (2). ***anti*-(1*R*,3*R*)-3b:** 89% yield (73% overall yield, 74 mg starting from 100 mg of **1b**), Table 2, entry 1, purified by silica gel column chromatography using hexane and EtOAc (8:2) as eluent. Er (1*R*,3*R*):(1*S*,3*S*) = 90:10 determined by HPLC, (*R*,*R*) Whelk 02 column (hexane:2-propanol 95:5), 0.5 mL/min, t_R [major (*R*,*R*)-enantiomer] = 16.7 min, t_R [minor (*S*,*S*)-enantiomer] = 18.3 min, $[\alpha]_D^{20}$= +51.4° (*c* 1, $CHCl_3$).

***syn*-4,4,4-Trifluoro-1-phenylbutane-1,3 diol (3b)**.[5,6] ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.43–7.29 (m, 5 H), 5.00 (dd, *J* = 9.8, 3.7 Hz, 1 H), 4.32–4.18 (m, 2 H), 3.35–3.06 (bs, 1 H, exchanges with D_2O), 2.40–1.95 (bs, 1 H, exchanges with D_2O), 2.18–1.99 (m, 2 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 142.9, 128.8, 128.5, 128.1, 125.2, (q, $^{1}J_{C\text{-}F}$ = 281.0 Hz), 74.1, 70.6 (q, $^{2}J_{C\text{-}F}$ = 31.0 Hz), 37.7; ^{19}F NMR (376 MHz, $CDCl_3$) δ –80.4 (d, *J* = 6.5 Hz); FT-IR (neat): 3069, 2964, 2920, 1640, 1495, 1277, 1168, 1057, 916, 876, 762, 701 cm^{-1}; GC MS (70 eV) *m/z* (%) 220 (M^+, 6), 133 (3), 107 (100), 105 (14), 91 (5), 79 (51), 77 (32), 65 (2), 51 (9), 43 (2). ***syn*-(1*S*,3*R*)-3b:** 8% yield (7% overall yield, 8 mg starting from 100 mg of **1b**), Table 2, entry 1, purified by silica gel column chromatography using hexane and EtOAc (8:2) as eluent. Er (1*S*,3*R*):(1*R*,3*S*) = 76:24, t_R [major (*S*,*R*)-enantiomer] = 19.8 min, t_R [minor (*R*,*S*)-enantiomer] = 27.9 min, $[\alpha]_D^{20}$= +13.4 (*c* 1, MeOH).

4,4,4-Trifluoro-1-(furan-2-yl)butane-1,3-diol (3c).[5] 78% yield (61% overall yield, 62 mg starting from 100 mg of **1c**), Table 2, entry 2, inseparable mixture of diastereoisomers, *anti:syn*= 75:25, purified by silica gel column chromatography using hexane and EtOAc (8:2) as eluent. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.41–7.40 (m, 1 H major stereoisomer + 1 H, minor stereoisomer), 6.37–6.35 (m, 1 H major stereoisomer + 1 H, minor stereoisomer), 6.32 (d, *J* = 3.1 Hz, 1 H major stereoisomer), 6.30 (d, *J* = 3.1 Hz, 1 H, minor stereoisomer), 5.12–5.09 (m, 1 H, minor stereoisomer), 5.06–5.03 (m, 1 H, major stereoisomer), 4.37–4.33 (m, 1 H, minor stereoisomer), 4.23–4.19 (m, 1 H, major stereoisomer), 3.50 (d, *J* = 3.0 Hz, 1 H, exchanges with D_2O, major stereoisomer), 2.93 (d, *J* = 5.0 Hz 1 H, exchanges with D_2O, minor stereoisomer), 2.49 (d, *J* = 3.3 Hz, 1 H, exchanges with D_2O, major stereoisomer), 2.28–2.20 (m, 2 H, major stereoisomer + 1 H, which exchanges with D_2O, minor stereoisomer), 2.15–2.09 (m, 2 H, minor stereoisomer); ^{13}C NMR (100 MHz, $CDCl_3$,) δ 155.2 (minor stereoisomer), 154.6 (major stereoisomer), 142.6 (major stereoisomer), 142.4 (minor stereoisomer), 124.5 (q, $^{1}J_{C\text{-}F}$ = 281.0 Hz, major and minor stereoisomer), 110.4 (major stereoisomer), 110.3 (minor stereoisomer), 106.7 (major stereoisomer), 106.4 (minor stereoisomer), 70.1 (q, $^{2}J_{C\text{-}F}$ = 32.0 Hz, major stereoisomer), 67.7 (q, $^{2}J_{C\text{-}F}$ = 31.0 Hz, minor stereoisomer), 66.8 (major stereoisomer), 64.2 (minor stereoisomer), 34.4 (major stereoisomer), 34.2 (minor stereoisomer); ^{19}F NMR (376 MHz, $CDCl_3$) δ –79.7 (d, *J* = 6.7 Hz, minor stereoisomer), –80.4 (d, *J* = 6.5 Hz, major stereoisomer); GC MS (70 eV) *m/z* (%) major stereoisomer: 210 (M^+, 14), 192 (2), 123 (6), 97 (100), 95 (15), 69 (11), 65 (4), 41 (11). HRMS (ESI-TOF) *m/z*: $[M - H]^-$ Calcd for $C_8H_8F_3O_3$ 209.0431; Found 209.0417. ***anti*-(1*R*,3*R*)-3c**: er (1*R*,3*R*):(1*S*,3*S*) = 71:29 determined by HPLC, LUX Cellulose-1 column (hexane:2-propanol 90:10), 0.8 mL/min, t_R [minor (*S*,*S*)-enantiomer] = 14.8 min, t_R [major (*R*,*R*)-enantiomer] = 16.1 min. ***syn*-(1*S*,3*R*)-3c**: er (1*S*,3*R*):(1*R*,3*S*) = 67:33, determined by HPLC, LUX Cellulose-1 column (hexane:2-propanol 90:10), 0.8 mL/min, t_R [major (*S*,*R*)-enantiomer] = 11.6 min, t_R [minor (*R*,*S*)-enantiomer] = 13.0 min.

***anti*-1-Phenylbutane-1,3-diol (3d)**.[7] ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.37–7.34 (m, 4 H), 7.29–7.27 (m, 1 H), 5.05 (dd, 1 H, *J* = 7.9, 3.5 Hz), 4.10–4.05 (m, 1 H), 1.95-1.83 (m, 2 H, CH_2), 1.85 (ddd, 1 H, *J* = 8.4, 3.5, 14.5 Hz), 1.25 (d, 3 H, *J* = 6.3 Hz); ^{13}C NMR (150 MHz, $CDCl_3$) δ 144.4, 128.3, 127.2, 125.6, 71.5, 65.2, 46.2, 23.4; FT-IR (neat): 3350, 3087, 3063, 3030, 2966, 2924, 2853, 1603, 1585, 1494, 1454, 1376, 1199, 1130, 1087, 974, 913, 866, 854, 811, 756, 700 cm^{-1}; GC-MS (70 eV) *m/z* (%) 166 (M^+, 6), 148 (37), 147 (13), 133 (16), 108 (210), 107 (100), 106 (16), 105 (74), 104 (23), 103 (15), 79 (65), 78 (20), 77 (48), 65 (3), 51 (12). ***anti*-(1*R*,3*S*)-3d:** 78% yield, Table 2, entry 3 (74% overall yield, 75 mg starting from 100 mg of **1d**), purified by silica gel column chromatography using hexane and EtOAc (7:3) as eluent. Er (1*R*,3*S*):(1*S*,3*R*) = 93:7 determined by HPLC, LUX Cellulose-4 column (hexane:2-propanol 92:8), 0.5 mL/min, t_R [minor (*R*,*S*)-enantiomer] = 33.7, t_R [major (*S*,*R*)-enantiomer] = 34.9 min, $[\alpha]_D^{20}$= +24.5° (c 1, CH_3OH).

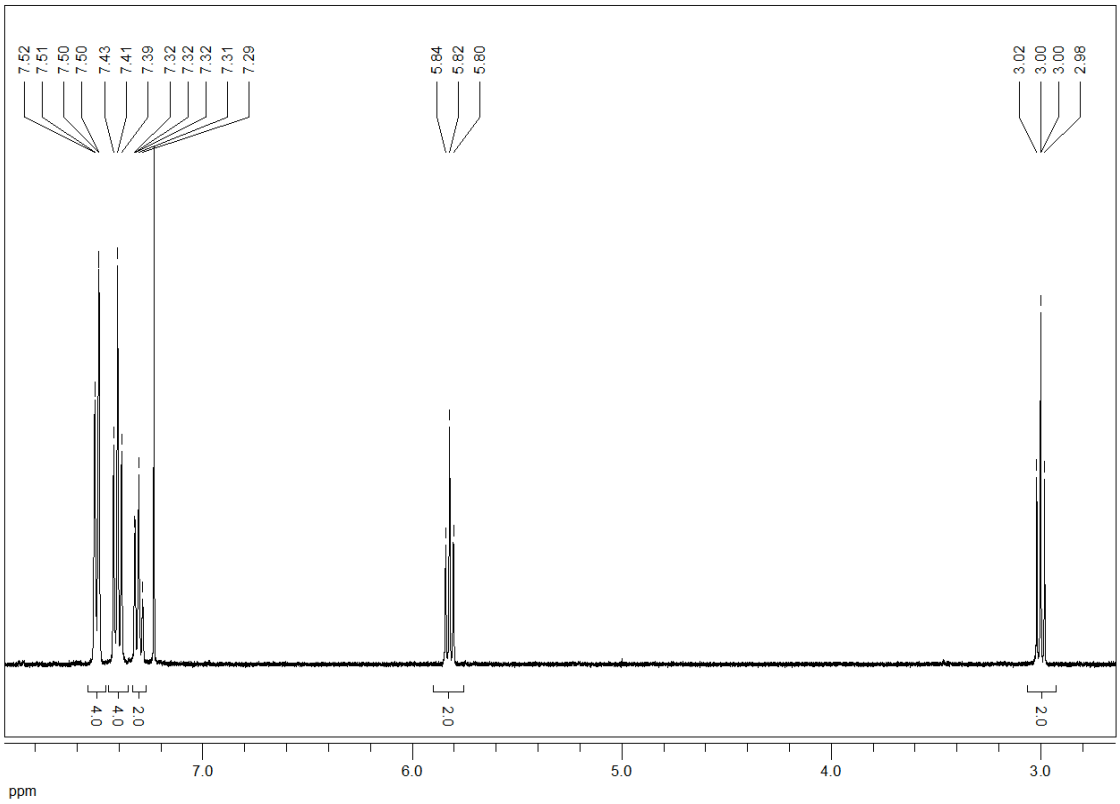
***syn*-1-Phenylbutane-1,3-diol (3d).** ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.38–7.34 (m, 4 H), 7.29–7.27 (m, 1 H), 4.95 (dd, 1 H, *J* = 10.1, 2.9 Hz), 4.18–4.13 (m, 1 H), 2.70–1.90 (bs, 2 H, exchange with D_2O), 1.87 (dt, 1 H, *J* = 10.1, 14.6 Hz), 1.77 (m , 1 H), 1.24 (d, 3 H, *J* = 6.2 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) δ 144.4, 128.5, 127.7, 125.6, 75.4, 68.9, 47.1, 24.1; GC MS (70 eV) *m/z* (%): 166 (M^+, 6), 148 (34), 147 (12), 133 (12), 108 (13), 107 (100), 106 (16), 105 (68), 104 (25), 103 (13), 79 (61), 78 (19), 77 (45), 65 (3), 51 (11). ***syn*-(*1R,3R*)-3d:** 95% yield, Table 2, entry 4 (85% overall yield, 86 mg starting from 100 mg **1d**) purified by silica gel column chromatography using hexane and EtOAc (6:4) as eluent. Er (1*S,*3*S*):(1*R,*3*R*) = 1:99 determined by HPLC, LUX Cellulose-4 column (hexane:2-propanol 92:8), 0.5 mL/min, t_R [minor (*R*,*S*)-enantiomer] = 41.9, t_R [major (*R*,*R*)-enantiomer] = 43.3 min.

***anti*-4,4,4-Trifluoro-1-(naphthalen-2-yl)butane-1,3-diol (3e).**[5] ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.90–7.77 (m, 4 H), 7.53–7.51 (m, 3 H), 5.21–5.19 (m, 1 H), 4.55–4.51 (bs, 1 H, exchanges with D_2O), 4.36–4.23 (m, 1 H), 3.24–3.22 (bs, 1 H, exchanges with D_2O), 2.15–2.01 (m, 2 H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 141.5, 133.2, 132.7, 128.1, 127.8, 127.5, 126.1, 125.7, 125.5 (q, $^{1}J_{C\text{-}F}$ = 281.0 Hz), 124.1, 123.8, 69.6, 67.2 (q, $^{2}J_{C\text{-}F}$ = 31.0 Hz), 38.1, 31.6; ^{19}F NMR (376 MHz, $CDCl_3$) δ –72.2 (d, *J* = 7.1 Hz); FT-IR (neat): 3310, 3060, 2960 2918, 2846, 1626, 1601, 1405, 1273, 1172, 1135, 1105, 1051, 962, 901, 864, 820, 749 cm^{-1}. HRMS (ESI-TOF) m/z: $[M - H]^-$ Calcd for $C_{14}H_{12}F_3O_2$ 269.0789; Found 269.0795. ***anti*-(1*R,*3*R*)-3e:** 76% yield, Table 2, entry 7 (30% overall yield, 32 mg starting from 100 mg of **1e**), purified by silica gel column chromatography using hexane and EtOAc (9:1) as eluent. Er (1*R*,3*R*):(1*S*,3*S*) = 80:20 determined by HPLC, (*R,R*) Whelk 02 column (hexane:2-propanol 95:5), 0.5 mL/min, t_R [major (*R*,*R*)-enantiomer] = 29.6 min, t_R [minor (*S*,*S*)-enantiomer] = 39.4 min, $[\alpha]_D^{20}$ = +19.8° (*c* 1, $CHCl_3$).

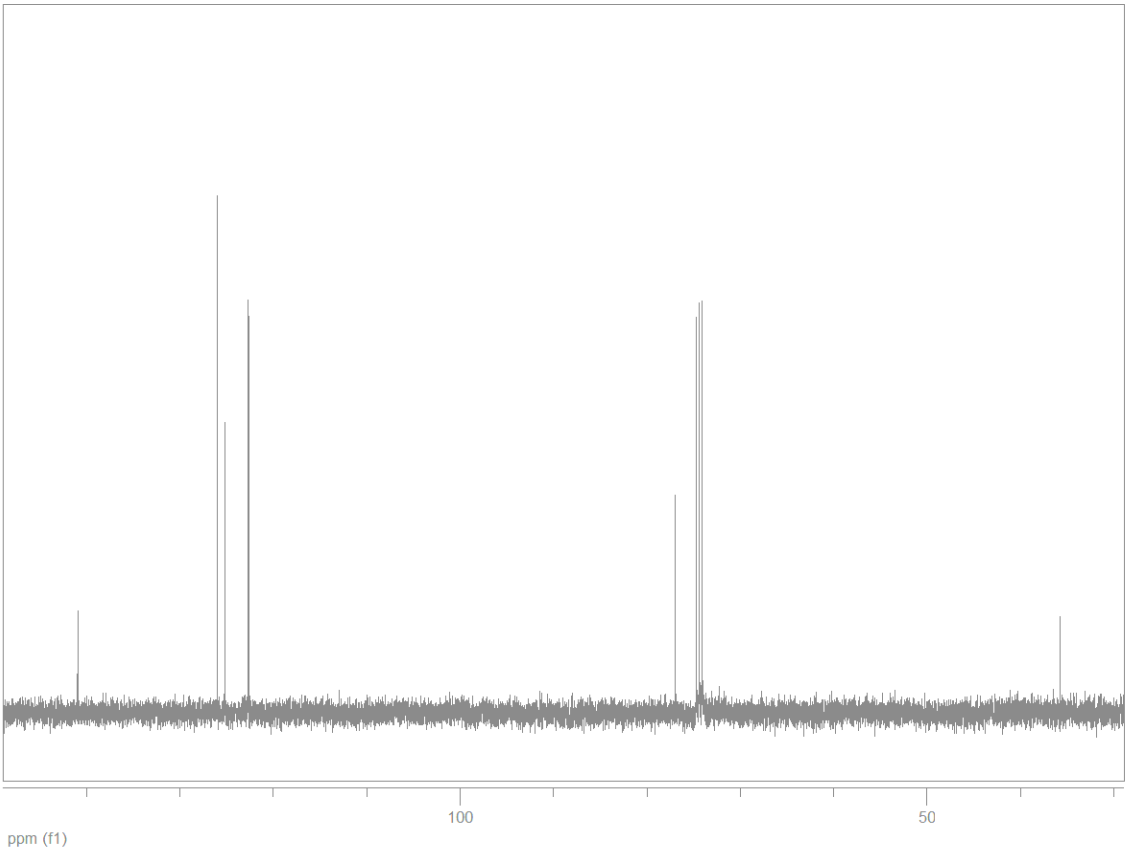
***syn*-4,4,4-Trifluoro-1-(naphthalen-2-yl)butane-1,3-diol (3e).**[5] ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.87–7.82 (m, 4 H), 7.51–7.48 (m, 3 H), 5.18 (dd, *J* = 9.6, 3.6 Hz, 1 H), 4.29–4.21 (m, 1 H), 3.89–3.80 (bs, 1 H, exchanges with D_2O), 2.45–2.38 (bs, 1 H, exchanges with D_2O), 2.25–2.10 (m, 2 H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 140.2, 133.3, 133.2, 128.9, 128.0, 127.7, 126.5, 126.3, 124.5 (q, $^{1}J_{C\text{-}F}$ = 283.0 Hz), 124.1, 123.8, 74.2, 70.6 (q, $^{2}J_{C\text{-}F}$= 32.0 Hz), 64.2, 37.7; ^{19}F NMR (376 MHz, $CDCl_3$) δ –73.1 (d, *J* = 6.6 Hz). HRMS (ESI-TOF) *m/z*: $[M - H]^-$ Calcd for $C_{14}H_{12}F_3O_2$ 269.0795; Found 269.0793. ***syn*-(1*S,*3*R*)-3e:** 88% yield, Table 2, entry 8 (34% overall yield, 37 mg starting from 100 mg of **1e**), purified by silica gel column chromatography using hexane and EtOAc (9:1) as eluent. Er (1*S*,3*R*):(1*R*,3*S*) = 86:14 [HPLC, (*R,R*) Whelk 02 column (hexane:2-propanol 95:5), 0.5 mL/min, t_R [major (*S*,*R*)-enantiomer] = 46.9 min, t_R [minor (*R*,*S*)-enantiomer] = 25.2 min; $[\alpha]_D^{20}$ = –3.7 (*c* 1, $CHCl_3$).

Spectra of *trans*-2,4-diphenyloxetane (4a)

^{1}H NMR, 600 MHz, $CDCl_3$

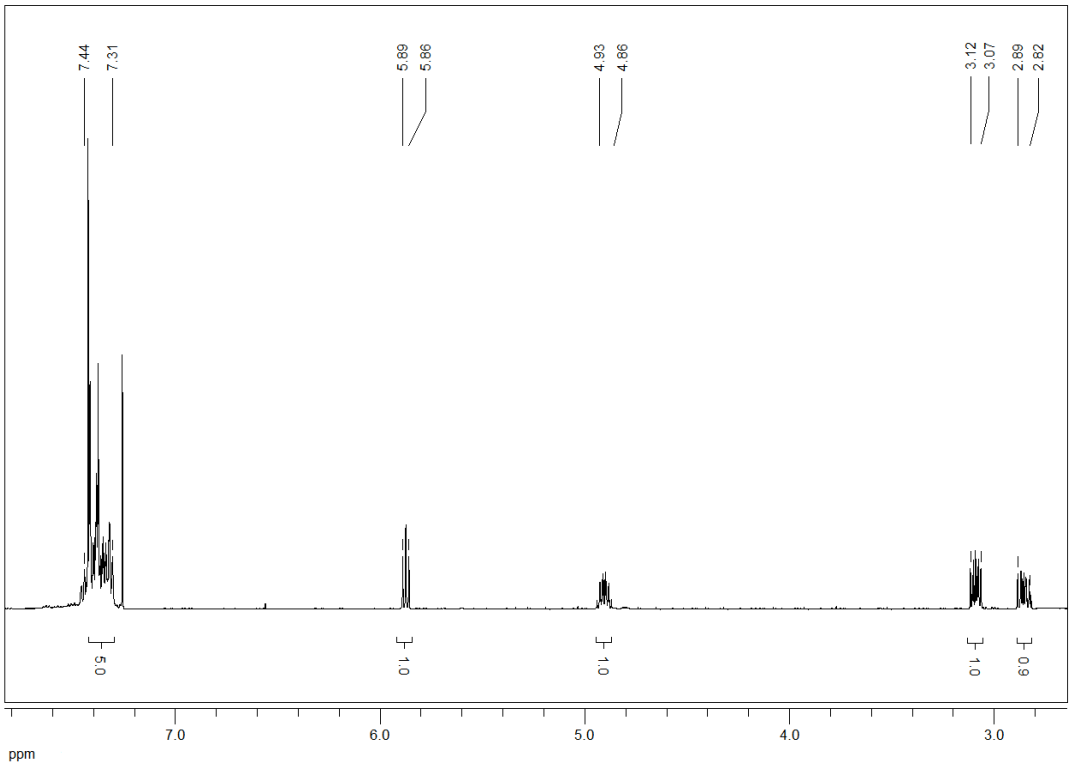


^{13}C NMR, 100 MHZ, $CDCl_3$

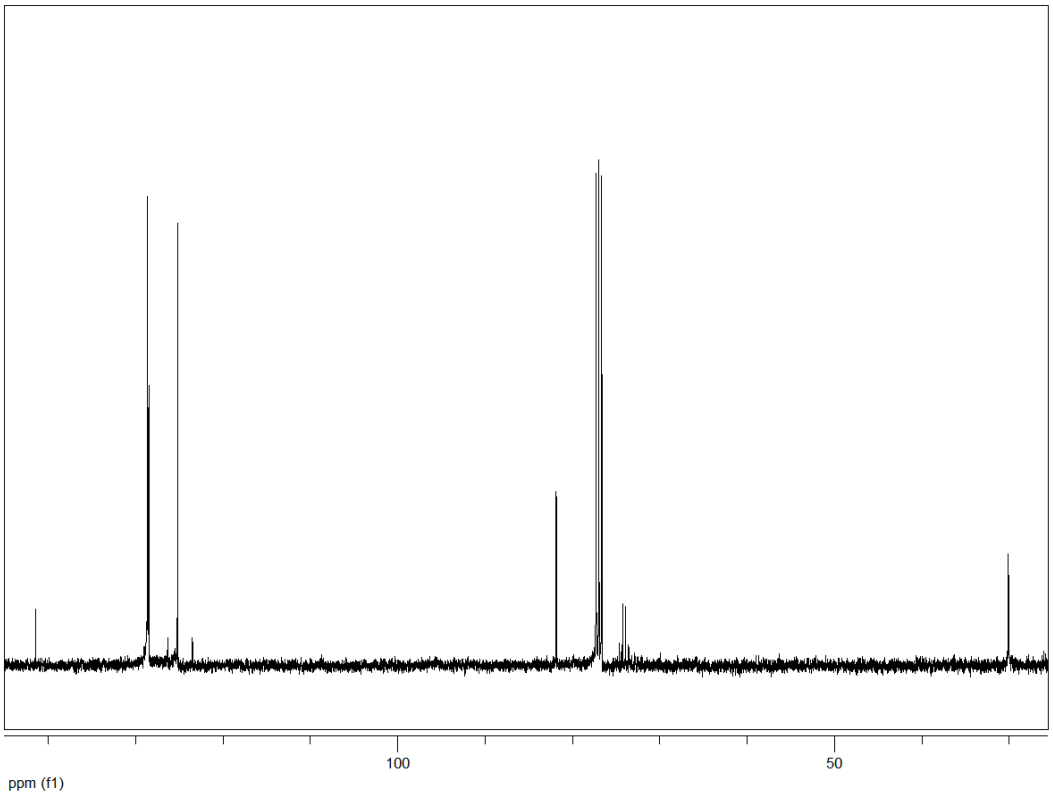


Spectra of *trans*-2-phenyl-4-(trifluoromethyl)oxetane (4b)

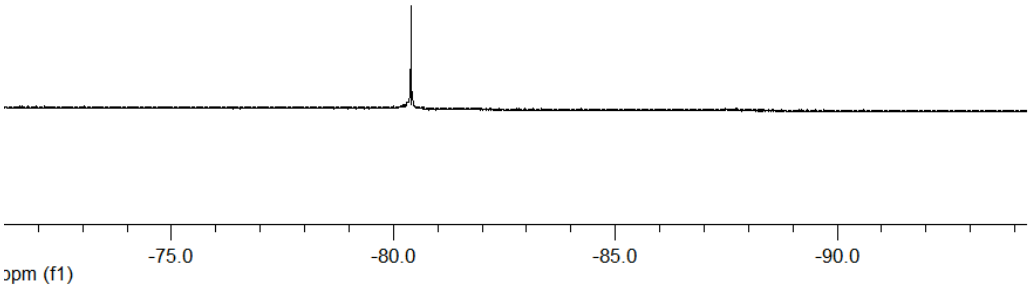
^{1}H NMR, 400 MHz, $CDCl_3$



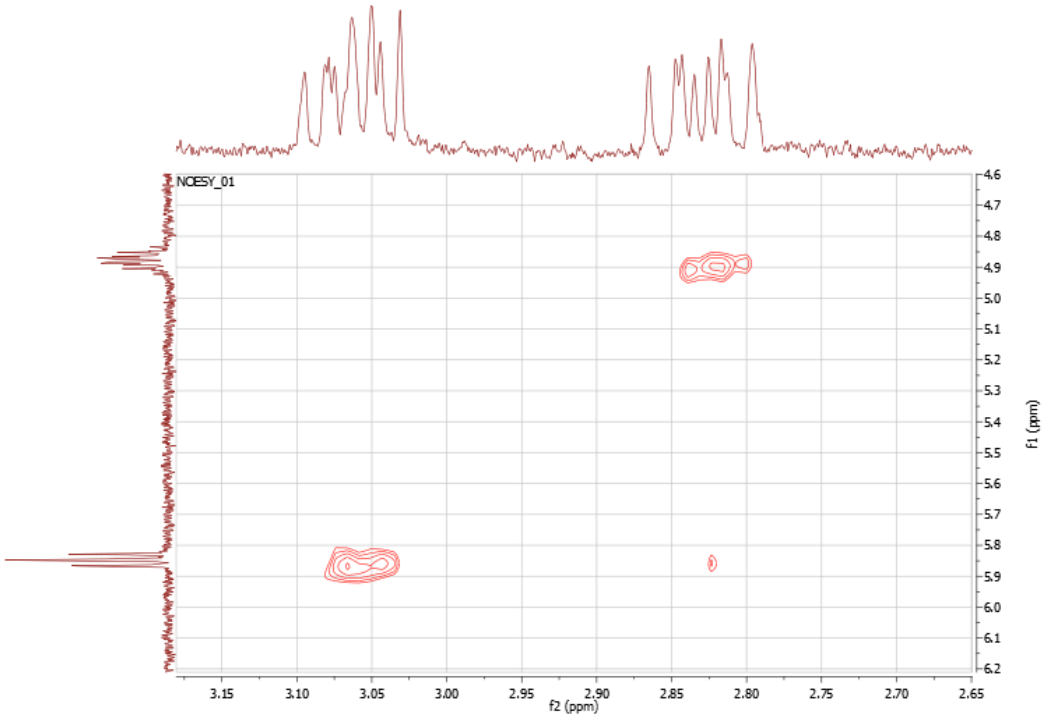
^{13}C NMR, 100 MHZ, $CDCl_3$



^{19}F NMR, 376 MHz, $CDCl_3$

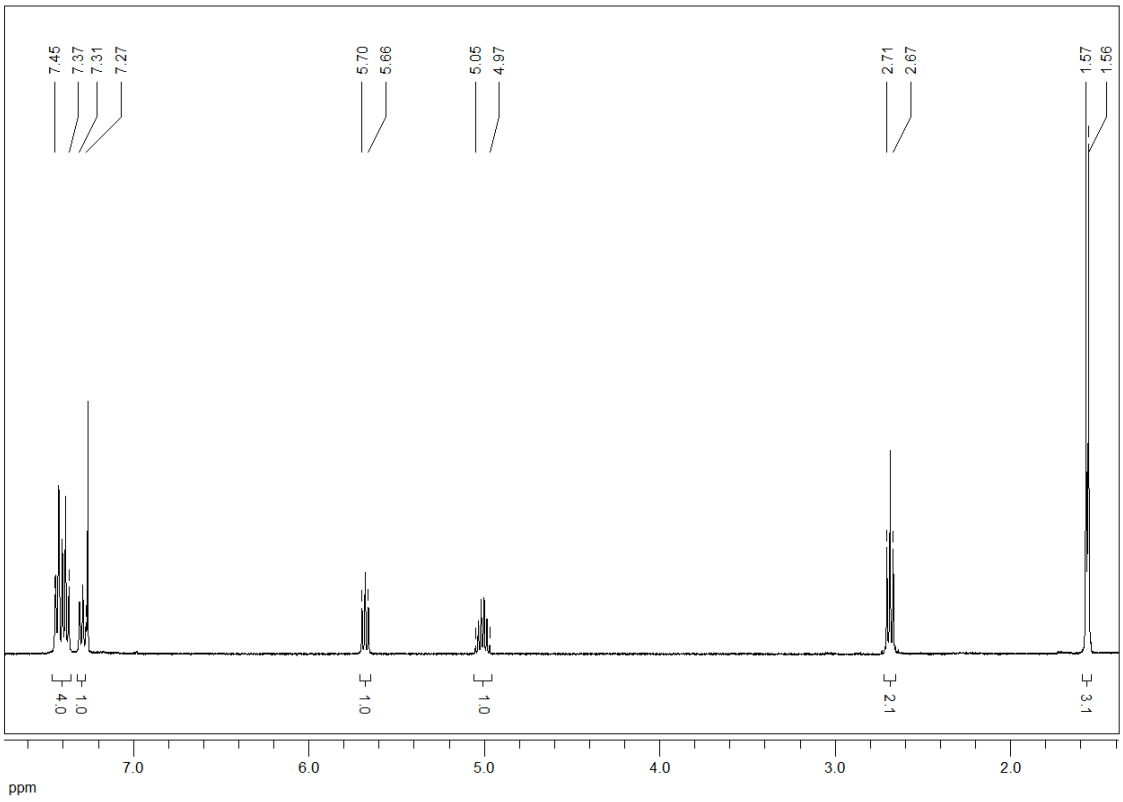


2D-NOESY Proton NMR Spectrum, $CDCl_3$, ***trans*-2-phenyl-4-(trifluoromethyl)oxetane (4b)**

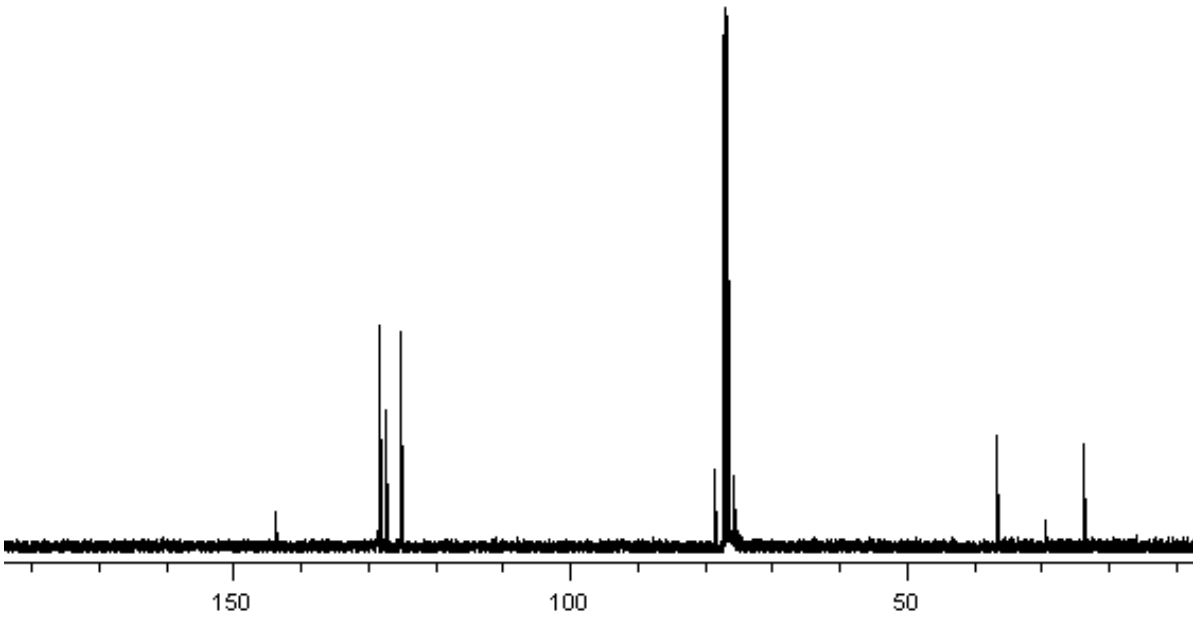


Spectra of *trans*-2-phenyl-4-methyloxetane (4d)

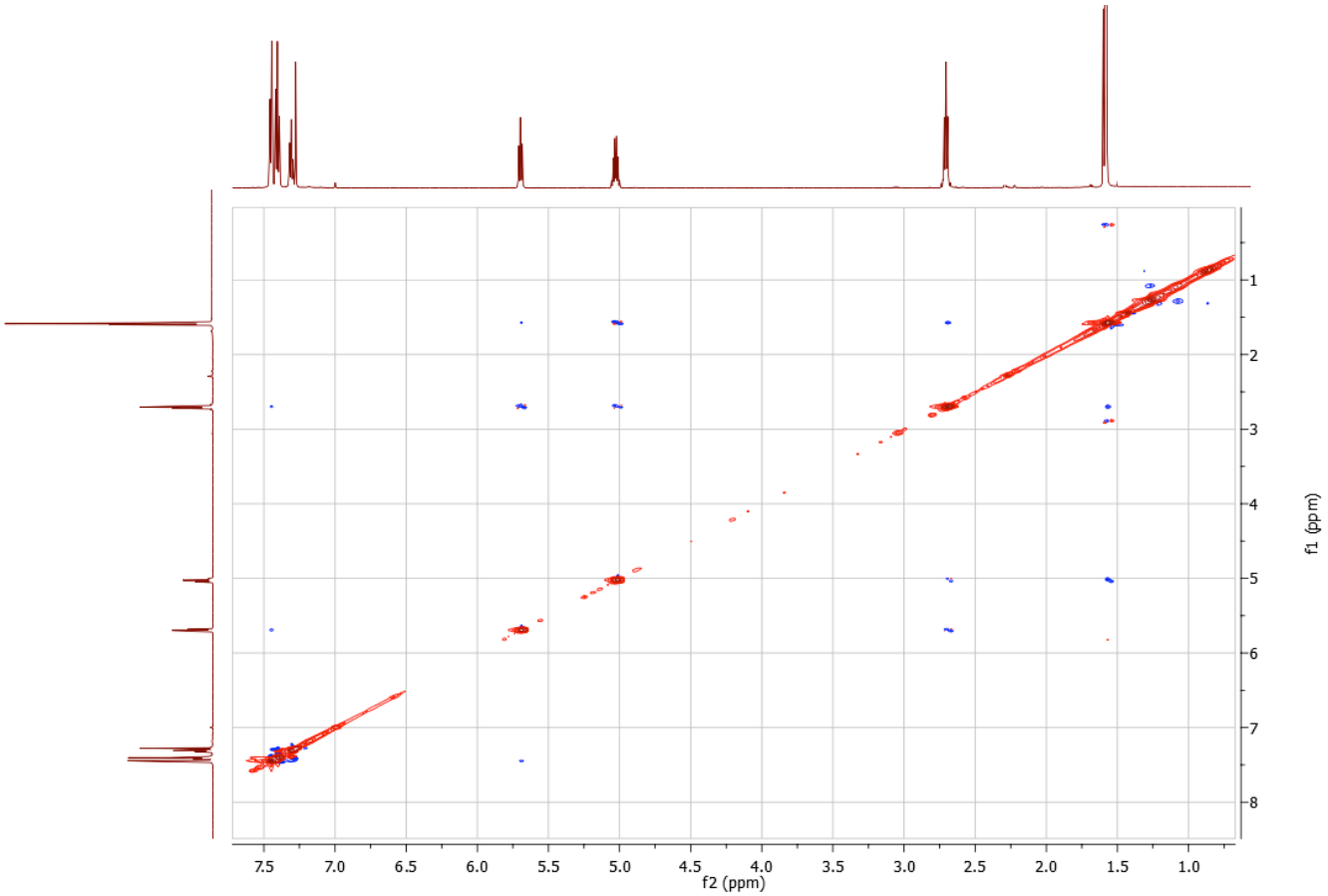
^{1}H NMR, 600 MHz, $CDCl_3$



^{13}C NMR, 125 MHZ, $CDCl_3$

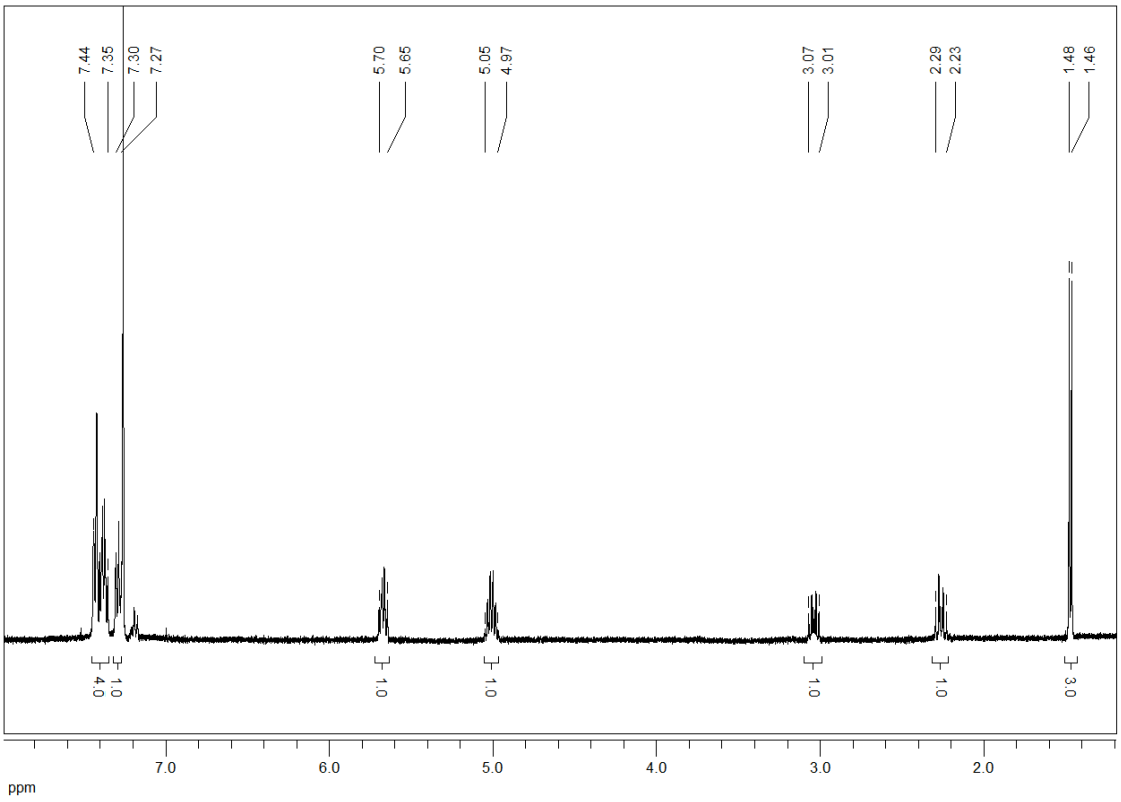


2D-NOESY Proton NMR Spectrum, $CDCl_3$, ***trans*-2-phenyl-4-methyloxetane (4d)**

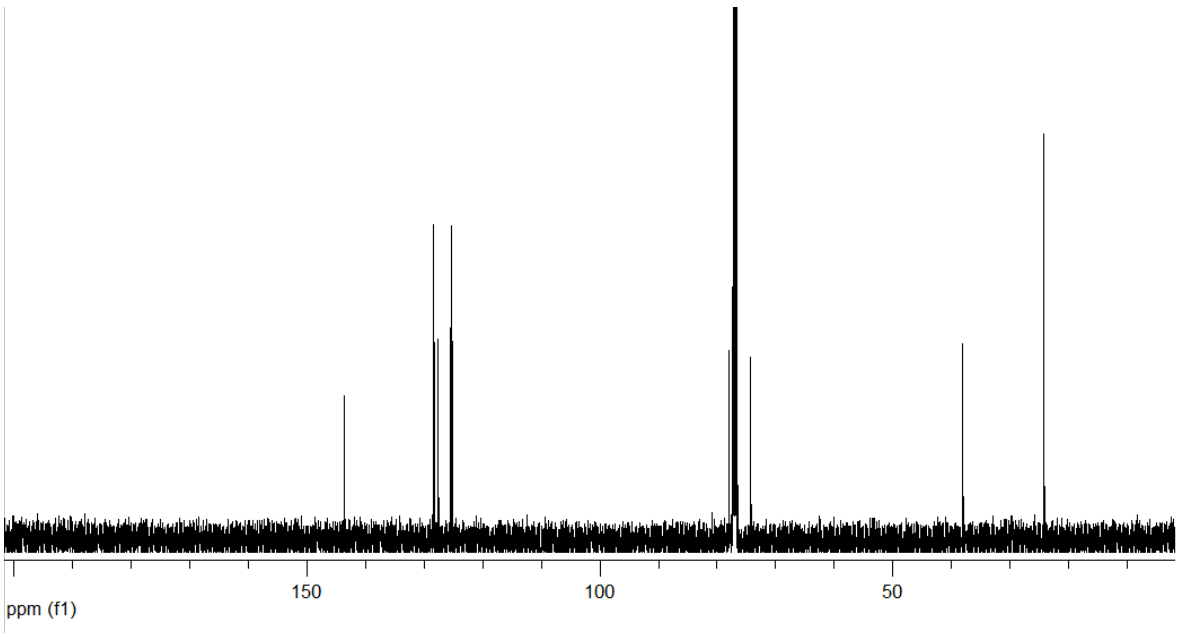


Spectra of *cis*-2-phenyl-4-methyloxetane (4d)

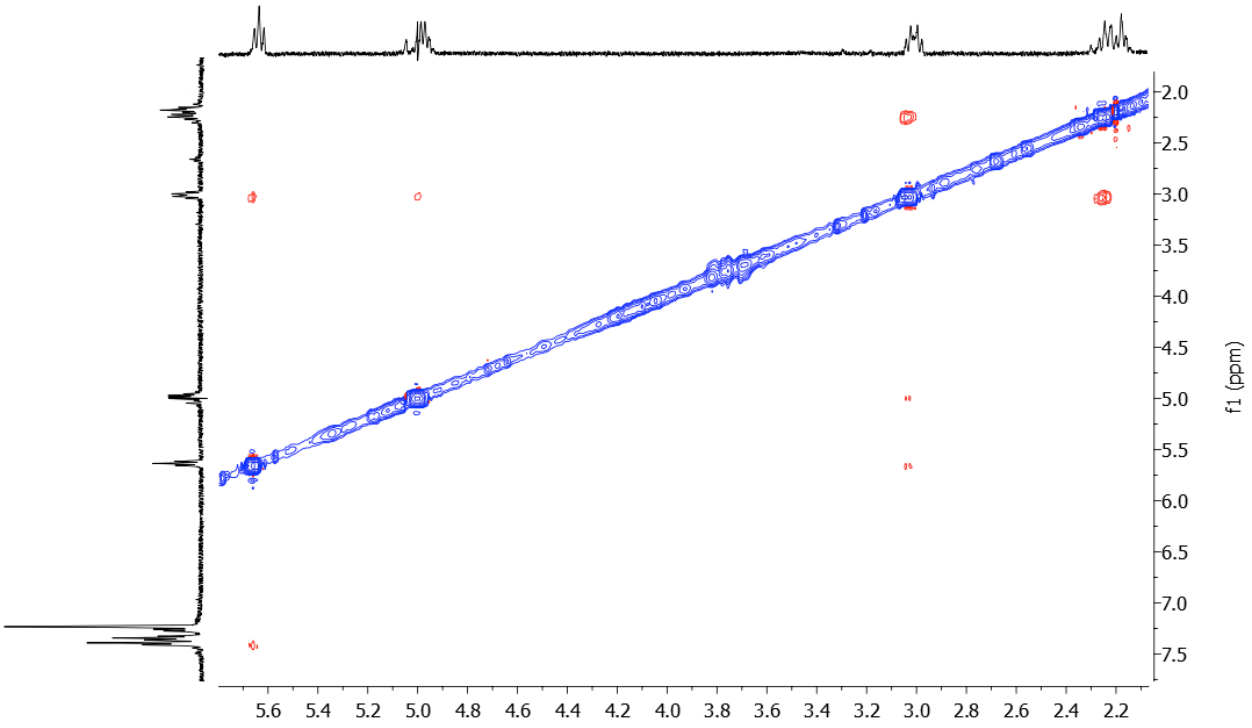
^{1}H NMR, 600 MHz, $CDCl_3$



^{13}C NMR, 125 MHZ, $CDCl_3$

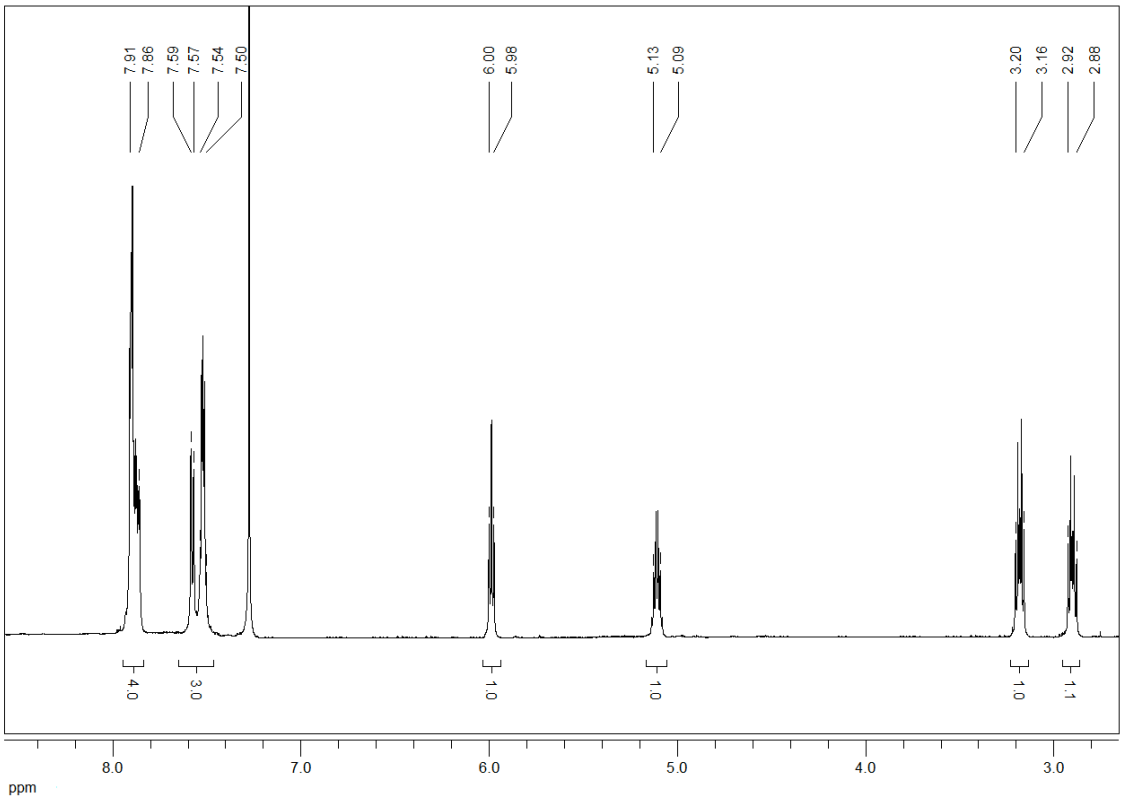


2D-NOESY Proton NMR Spectrum,$CDCl_3$, ***cis*-2-phenyl-4-methyloxetane (4d)**

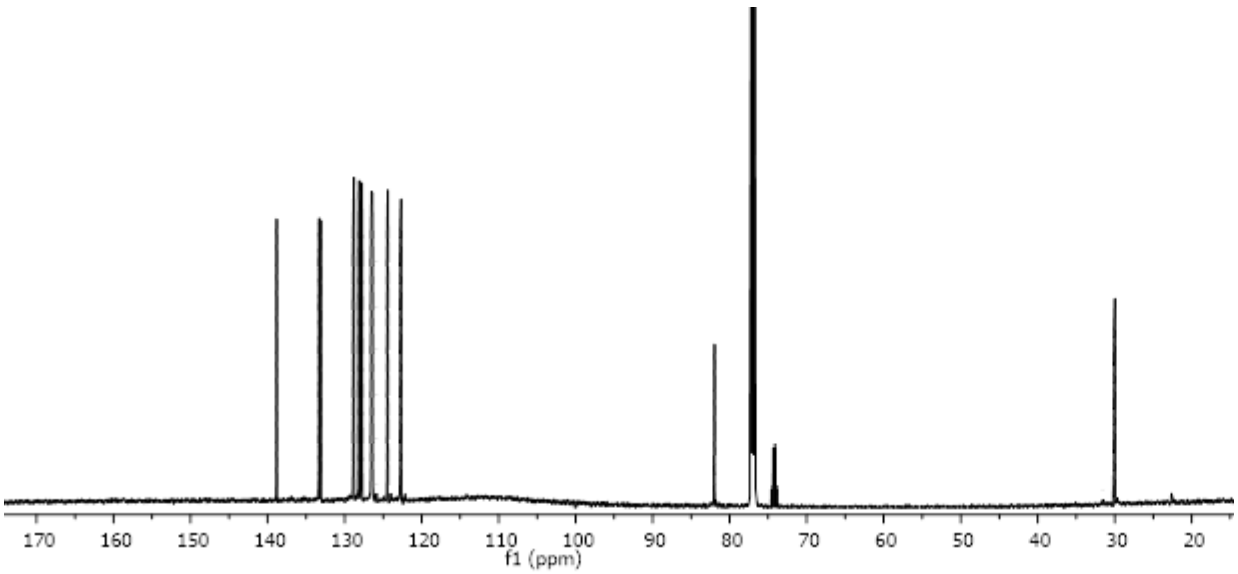


Spectra of *cis* 2-(naphthalen-2-yl)-4-(trifluoromethyl)oxetane (4e)

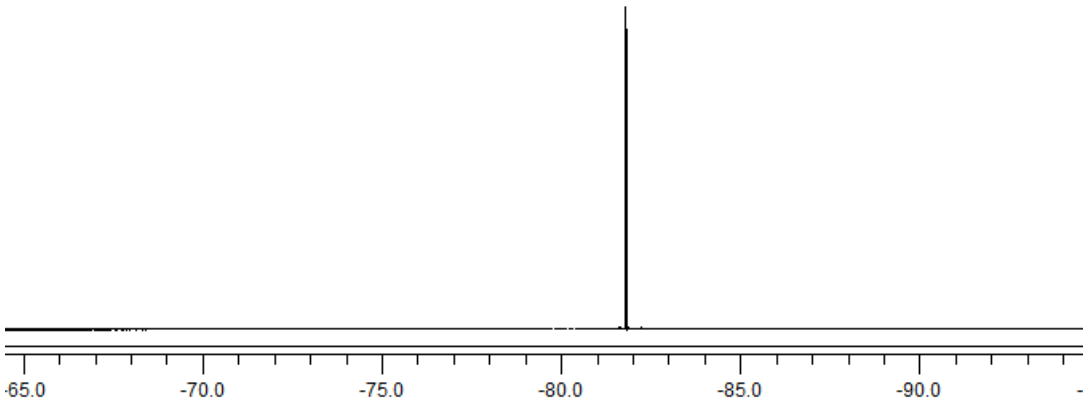
^{1}H NMR, 600 MHz, $CDCl_3$



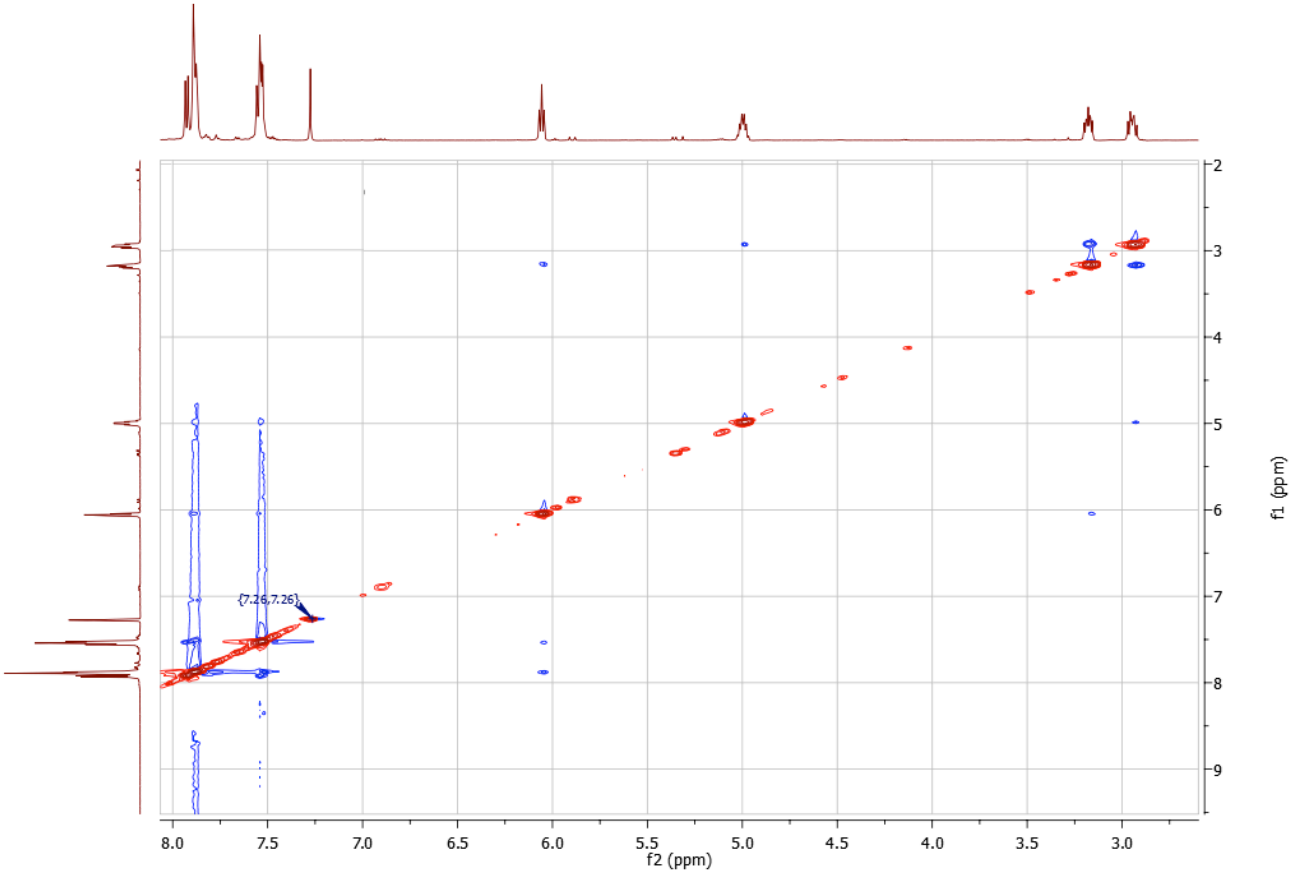
^{13}C NMR, 125 MHZ, $CDCl_3$



^{19}F NMR, 376 MHz, $CDCl_3$



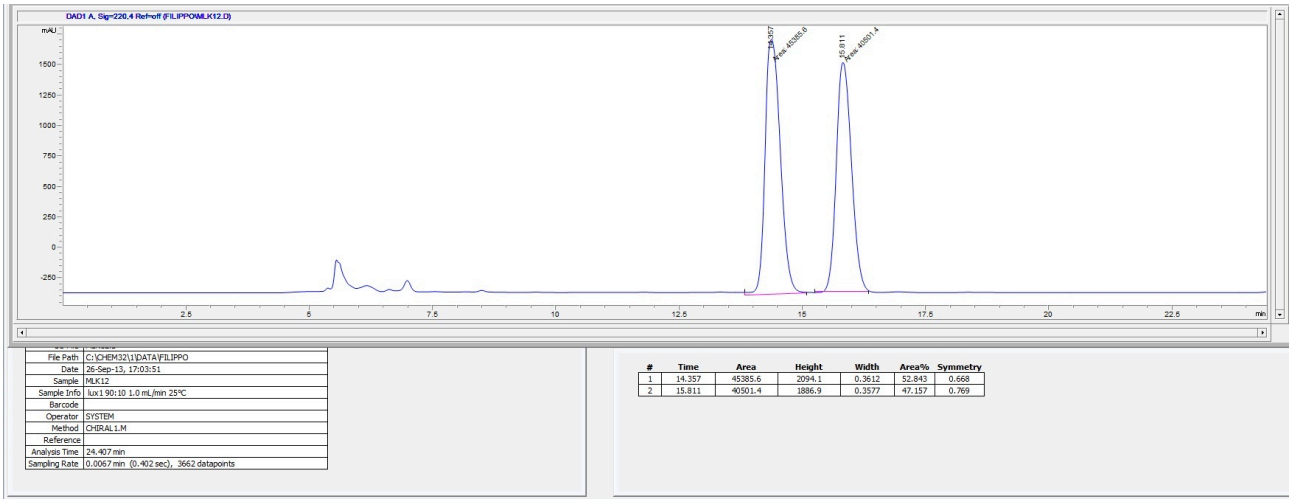
2D-NOESY Proton NMR Spectrum, $CDCl_3$, ***cis*** **2-(naphthalen-2-yl)-4-(trifluoromethyl)oxetane (4e)**



Chromatograms for er determination

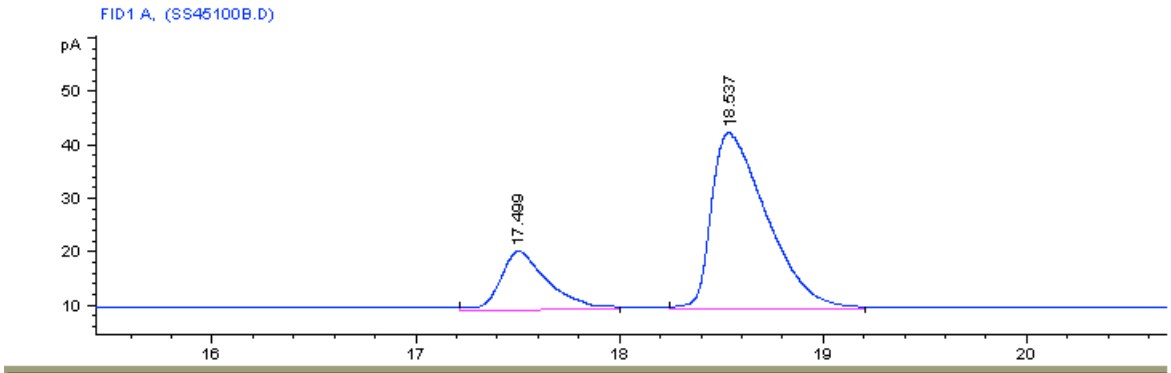
trans-2,4-Diphenyloxetane (4a)

HPLC, Lux Cellulose-1 column, hexane:2-propanol = 90:10, 0.8 mL/min, t_R [major (*S*,*S*)-enantiomer] = 14.4 min, t_R [minor (*R*,*R*)-enantiomer] = 15.8 min



(2*R*,4*R*)-2-Phenyl-4-(trifluoromethyl)oxetane (4b)

GC-Chirasil-DEX CB capillary column, (He flow 1 mL/min, 100 °C), t_R [major (*R*,*R*)-enantiomer] = 18.5 min, t_R [minor (*S*,*S*)-enantiomer] = 17.5 min



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Date	01/01/88 2.36.35
Sample	SS45

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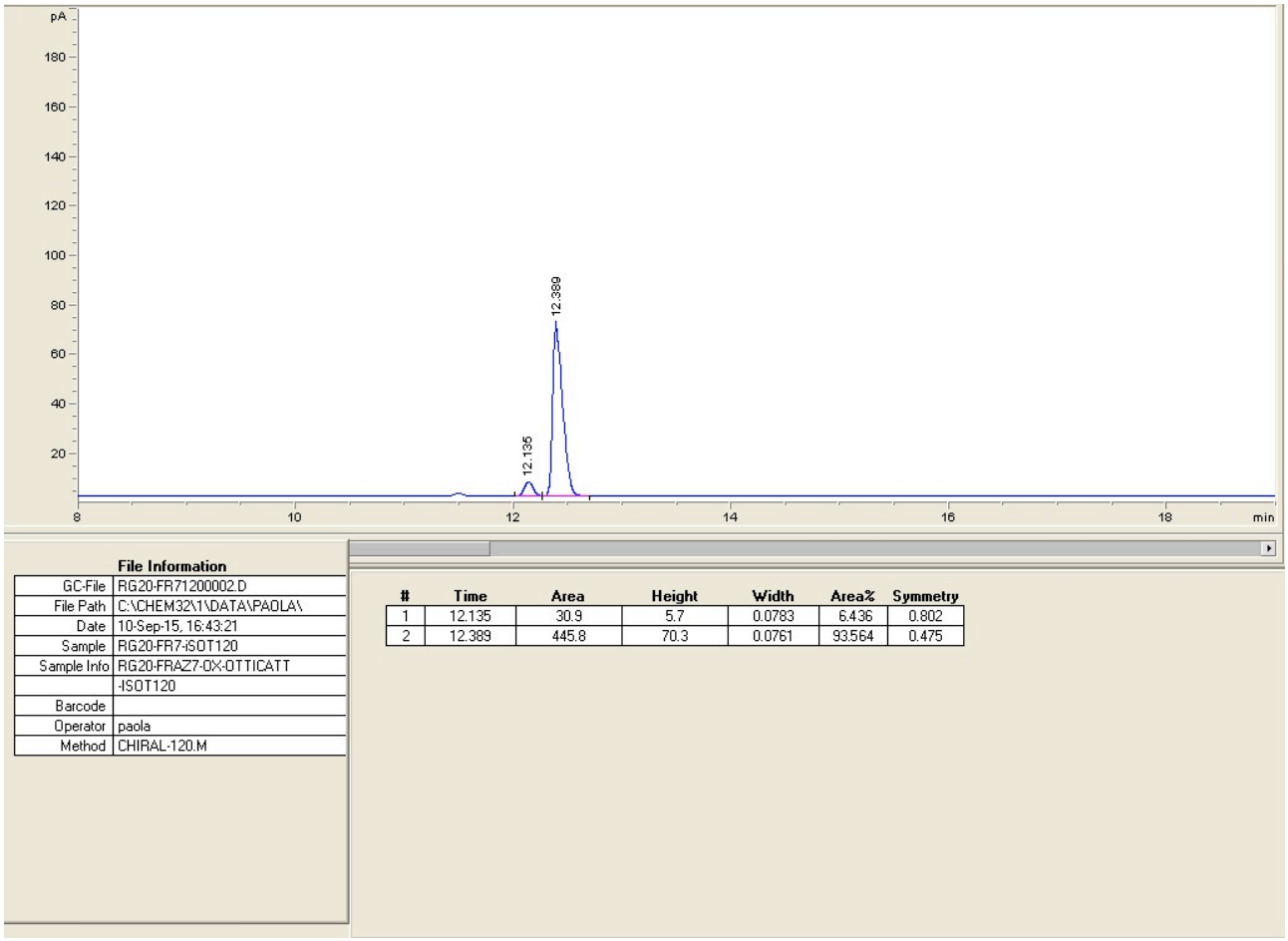
2	18.537	624.9	33.1	0.3145	

(2*R*,4*S*)-2-Phenyl-4-methyloxetane (4d).

GC-Chirasil-DEX CB capillary column, (He flow 2 mL/min, 100 °C), t_R [minor (*S*,*R*)-enantiomer] = 12.1 min, t_R [major (*R*,*S*)-enantiomer] = 12.4 min)



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Barcode	
Operator	paola
Method	CHIRAL-120.M

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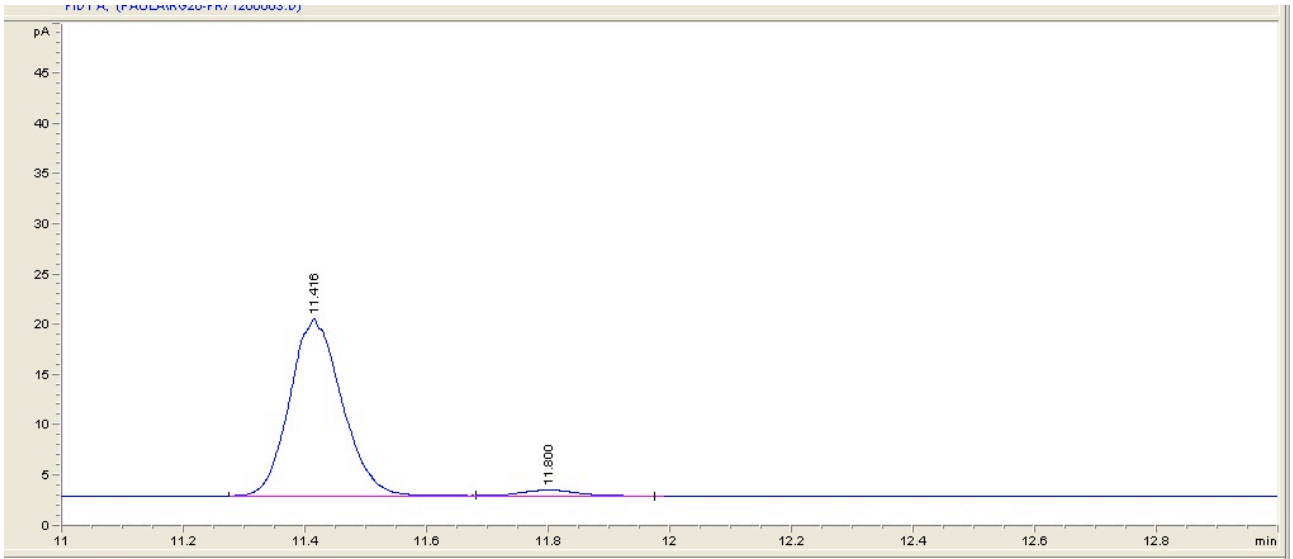
1	12.135	30.9	5.7	0.0783	6.436	0.802
2	12.389	445.8	70.3	0.0761	93.564	0.475

(2*S*,4*S*)-2-Phenyl-4-methyloxetane (4d).

GC-Chirasil-DEX CB capillary column, (He flow 2 mL/min, 100 °C), t_R [major (*S*,*S*)-enantiomer] = 11.4 min, t_R [minor (*R*,*R*)-enantiomer] = 11.8 min



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	-ISOT120
Barcode	
Operator	paola
Method	CHIRAL-120.M

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2	11.8	3.6	5.8E-1	0.0811	3.273

(2*S*,4*R*)-2-(Naphthalen-2-yl)-4-(trifluoromethyl)oxetane (4e).

HPLC, Lux Cellulose-1 column, hexane:2-propanol = 90:10, 0.8 mL/min), t_R [major (*S*,*R*)-enantiomer] = 7.6 min, t_R [minor (*R*,*S*)-enantiomer] = 7.0 min

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ata File C:\HPCHEM\1\DATA\CAPRIATI\SS47OA.D                    Sample Name: SS47OA-lux1-0.8

   SS47OA-LUX-1;
    esano/i-propanolo 90:10 0.8 mL/min

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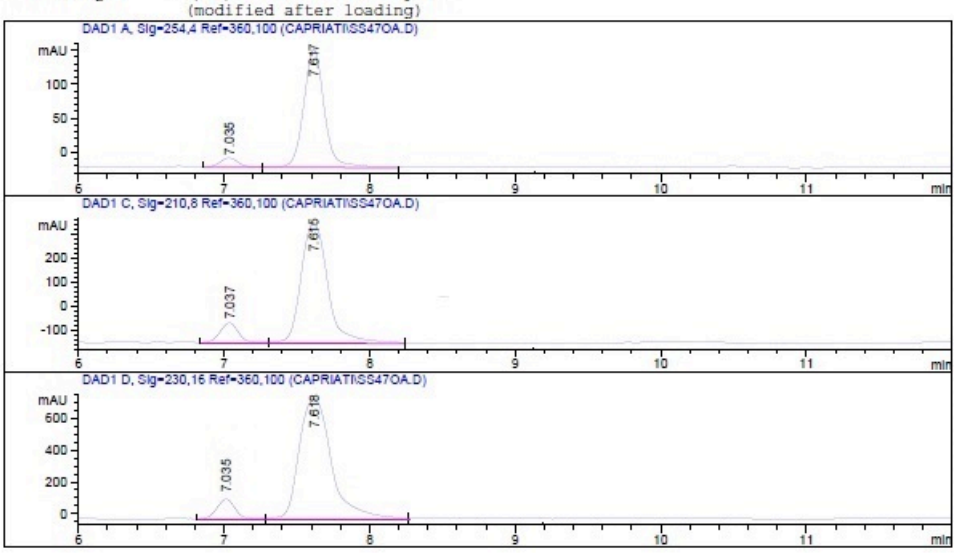


```
   ===================================================================
                              Area Percent Report
   ===================================================================

   Sorted By             :      Signal
   Multiplier            :      1.0000
   Dilution              :      1.0000


   Signal 2: DAD1 C, Sig=210,8 Ref=360,100

   Peak RetTime Type  Width     Area      Area
     #   [min]       [min]    [mAU*s]      %
   ----|-------|----|-------|----------|--------|
      1   0.075 BV    0.1089   16.08431   0.1985
      2   7.037 VV    0.1428  716.52555  10.0597
      3   7.615 VV    0.2035 6448.41553  89.5820
```

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