

## Electronic Supplementary Information

### Broadening the Chemical Scope of Laccases: Selective Deprotection of *N*-Benzyl Groups

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## I. General considerations

Amines **1b-6b**, **L-8b**, **11b**, and **12b** were purchased from Aldrich and used as received. Protected *N*-benzylamine **11a** was purchased from Aldrich and used as received and ( $\pm$ )-*trans*-**10a** was obtained from *EntreChem* SL. Laccase from *Trametes versicolor* was purchased from Sigma-Aldrich. All other reagents were obtained from commercial sources and used as received unless otherwise indicated.

Oxidation reactions using the laccase from *Trametes versicolor*/TEMPO catalytic system were performed in an Erlenmeyer flask, open to air and with orbital stirring (250 rpm).

NMR spectra were recorded on Bruker DPX 300 MHz and AV300 MHz spectrometers. All chemical shifts ( $\delta$ ) are given in parts per million and referenced to the residual solvent signal as internal. All coupling constants ( $J$ ) are reported in Hz with the following abbreviations: *s* = singlet, *d* = doublet, *dd* = double doublet, *ddd* = double doublet of doublets, *t* = triplet, *dt* = double triplet, *q* = quatriplet, *m* = multiplet, *br* = broad. High resolution mass spectra (HRMS) were obtained using a Bruker MicroQtof spectrometer by positive electrospray ionization (ESI<sup>+</sup>). Gas chromatography (GC) analyses were performed on a Agilent 7820 A GC chromatograph equipped with a FID detector. IR spectra were recorded as thin films on NaCl plates on a Perkin-Elmer Spectrum 100 FT-IR and are reported in frequency of absorption (cm<sup>-1</sup>). Thin-layer chromatography (TLC) was conducted with Merck Silica Gel 60 F<sub>254</sub> precoated plates and visualized with UV and potassium permanganate stain. Column chromatography was performed using Merck Silica Gel 60 (230-400 mesh).

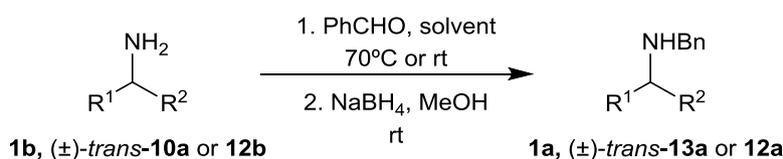
In addition to those specified above, the following abbreviations, designations and formulas are used throughout the Supporting Information; CH<sub>2</sub>Cl<sub>2</sub>= dichloromethane; Na<sub>2</sub>SO<sub>4</sub>= sodium sulfate; MeCN= acetonitrile; EtOAc= ethyl acetate; NaHCO<sub>3</sub>= sodium hydrogen carbonate; Na<sub>2</sub>CO<sub>3</sub>=sodium carbonate; TEMPO: 2,2,6,6-tetramethyl-1-piperidinyloxy radical; Bn= Benzyl group.



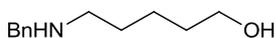
final amine as colorless oil in 69% yield.  $^1\text{H}$  NMR (300 MHz,  $\text{CDCl}_3$ )  $\delta$  7.35-7.19 (*m*, 5H), 3.20 (*m*, 1H), 2.74 (*dd*, 1H,  $J = 13.2, 5.4$  Hz), 2.55 (*dd*, 1H,  $J = 13.2, 8.0$  Hz), 1.52 (*br s*, 2H), 1.15 (*d*, 3H,  $J = 6.3$  Hz).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  140.1, 129.6 (2C), 128.8 (2C), 126.5, 48.9, 47.1, 24.0. MS (APCI<sup>+</sup>,  $m/z$ ) 136.1 [(M+H)<sup>+</sup>, 100%]. Spectral data are consistent with the literature values.<sup>1</sup>

## II.2. General procedure for the preparation of *N*-benzylamines **1a**, **12a**, and ( $\pm$ )-*trans*-**13a**

The corresponding protected amines **1a**, **12a** and ( $\pm$ )-*trans*-**13a** were synthesized in two steps using a condensation reaction with benzaldehyde followed by a reduction reaction with sodium borohydride as showed in Scheme S1.



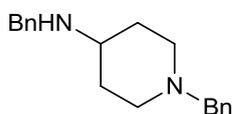
**Scheme S1** General reaction to obtain *N*-benzylamines **1a**, **12a** and ( $\pm$ )-*trans*-**13a**.



**1a**

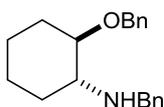
**5-(Benzylamino)pentan-1-ol (1a).** A solution of benzaldehyde (6 mmol, 610.7  $\mu\text{L}$ ) and **1b** (5 mmol, 515.8 mg) in MeOH (10 mL) was heated under reflux overnight. Then, the reaction mixture was cooled to ambient temperature, and  $\text{NaBH}_4$  (9 mmol, 340 mg,) was added portionwise to the stirred solution over a period of 0.5 h. The reaction was maintained under room temperature conditions for further 24 h, and after that time HCl 6 M was added to the reaction mixture to neutralize the excess of  $\text{NaBH}_4$ . The mixture was filtered and MeOH was removed under reduced pressure. The residue was extracted with ethyl acetate (3 x 10 mL) and the extract was concentrated. The residue was purified by flash column chromatography on silica gel (0-20% MeOH/EtOAc) to afford the *N*-protected amino alcohol **1a** as a yellow oil in 46% yield.  $^1\text{H}$  NMR (300 MHz,  $\text{CDCl}_3$ )  $\delta$  7.40-7.25 (*m*, 5H), 3.79 (*s*, 2H), 3.63 (*dt*, 2H,  $J = 6.4, 2.2$  Hz), 2.66 (*dt*, 2H,  $J = 6.7, 1.4$  Hz), 2.05-1.91 (*br s*, 2H), 1.58 (*m*, 4H), 1.45 (*m*, 2H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  140.5, 128.8 (2C), 128.6 (2C), 127.4, 62.9, 54.4, 49.6, 32.8, 30.0, 23.8. MS

(APCI<sup>+</sup>, m/z) 194.1 [(M+H)<sup>+</sup>, 100%]. Spectral data are consistent with the literature values.<sup>2</sup>



**12a**

***N*-Benzylpiperidin-4-benzylamine (12a)**. The title compound was prepared from **12b** in two steps. The starting amine **12b** (2.45 mmol, 500  $\mu$ L,) was added to a Schlenk tube containing dry MgSO<sub>4</sub> under nitrogen atmosphere, then dichloromethane (9.8 mL) and benzaldehyde (2.69 mmol, 275  $\mu$ L) were added. The resulting mixture was stirred at room temperature during 12 h. After that time the solvent was removed and the crude residue redissolved in dry MeOH (11 mL) under nitrogen atmosphere. Then, sodium borohydride (7.08 mmol, 268 mg) was added. The mixture was stirred during 16 h at room temperature. After that time, HCl 3 M was added to the reaction mixture to neutralize the excess of NaBH<sub>4</sub>. The mixture was filtered and MeOH was removed under reduced pressure. The residue was basified with NaOH 3N and then extracted with CH<sub>2</sub>Cl<sub>2</sub> (3 x 10 mL). The combined organic layers were dried over Na<sub>2</sub>SO<sub>4</sub>, filtered and concentrated under vacuum. This crude was purified by flash column chromatography on silica gel (30% MeOH/EtOAc) to afford the *N,N'*-diprotected compound **12a** as a yellow oil in 90% yield. <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>)  $\delta$  7.35-7.27 (*m*, 10H), 3.84 (*s*, 2H), 3.53 (*s*, 2H), 2.88 (*d*, 2H, *J*= 11.9 Hz), 2.54 (*m*, 1H), 2.05 (*td*, 2H, *J*= 11.6, 2.5 Hz), 1.91 (*d*, 2H, *J*= 12.5 Hz), 1.70 (*br s*, 1H), 1.49 (*m*, 2H). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>)  $\delta$  141.1 138.9, 129.5 (2C), 128.8 (2C), 128.6 (2C), 128.5 (2C), 127.3 (2C), 127.2, 63.5, 54.6, 52.8, 51.2, 33.1. MS (APCI<sup>+</sup>, m/z) 281.2 [(M+H)<sup>+</sup>, 100%]. IR (neat):  $\nu$  2934, 2799, 2758, 1493, 1452, 1366 and 1112 cm<sup>-1</sup> MS (APCI<sup>+</sup>, m/z) 280.2 [(M+H)<sup>+</sup>, 100%]. HRMS (ESI<sup>+</sup>) calcd for C<sub>19</sub>H<sub>25</sub>N<sub>2</sub> [(M+Na)<sup>+</sup>] 281.2012 found, 281.2011.



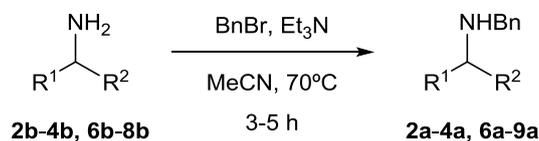
**(±)-*trans*-13a**

**(±)-*trans*-*N*-Benzyl-2-(benzyloxy)cyclohexan-1-amine [(±)-*trans*-13a]**. The title compound was prepared from (±)-*trans*-**10a** in two steps. The starting *O*-benzylated

amine ( $\pm$ )-*trans*-**10a** (2.44 mmol, 500 mg,) was added to a Schlenk tube containing dry  $\text{MgSO}_4$  under nitrogen atmosphere. Then, dichloromethane (10 mL) and benzaldehyde (2.44 mmol, 248  $\mu\text{L}$ ) were added. The resulting mixture was stirred at room temperature during 6 h. After that time the solvent was removed and the crude residue was redissolved in dry MeOH (7.5 mL) under nitrogen atmosphere. Then, sodium borohydride (4.8 mmol, 181 mg) was added. The mixture was stirred during 12 h at room temperature. After that time, HCl 3 M was added to the reaction mixture to neutralize the excess of  $\text{NaBH}_4$ . The mixture was filtered and MeOH was removed under reduced pressure. The residue was basified with a saturated solution of  $\text{Na}_2\text{CO}_3$  and then extracted with  $\text{CH}_2\text{Cl}_2$  (3 x 10 mL). The combined organic layers were dried over  $\text{Na}_2\text{SO}_4$ , filtered and concentrated under vacuum. The purity of this compound was excellent, so further purification was not necessary, obtaining ( $\pm$ )-*trans*-**13a** as a yellow oil (87% yield).  $^1\text{H}$  NMR (300 MHz,  $\text{CDCl}_3$ )  $\delta$  7.35-7.32 (*m*, 10H), 4.68 (*d*, 1H,  $J$ = 11.5 Hz), 4.47 (*d*, 1H,  $J$ = 11.5 Hz), 3.89 (*d*, 1H,  $J$ = 13.0 Hz), 3.71 (*d*, 1H,  $J$ = 13.0 Hz), 3.31 (*m*, 1H), 2.61 (*m*, 1H), 2.25-2.00 (*m*, 3H), 1.75 (*m*, 2H), 1.30-1.10 (*m*, 4H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  140.8, 138.8, 128.4 (4C), 128.0 (2C), 127.7 (2C), 127.6, 126.7, 82.0, 70.7, 61.2, 51.1, 30.3, 30.0, 24.4, 24.2. IR (neat):  $\nu$  2929, 2853, 2359, 1495, 1542 and 1093  $\text{cm}^{-1}$ . MS (APCI $^+$ ,  $m/z$ ) 296.2 [( $\text{M}+\text{H}$ ) $^+$ , 100%]. HRMS (ESI $^+$ ) calcd for  $\text{C}_{20}\text{H}_{25}\text{NO}$  [( $\text{M}+\text{Na}$ ) $^+$ ] 296.2009 found, 296.2009.

### II.3. General procedure for the preparation of *N*-benzylamines **2a-4a**, **6a-9a**

The corresponding *N*-protected amines were synthesized using nucleophilic substitution reactions with benzyl bromide as showed below (Scheme S2).

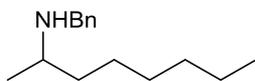


**Scheme S2** General alkylation reaction protocol.

To a solution of the corresponding deprotected amine (3 mmol) and triethylamine (3 mmol, 417  $\mu\text{L}$ ) in dry acetonitrile (2.5 mL), was added benzyl bromide (3.3 mmol, 392  $\mu\text{L}$ ). The reaction mixture was stirred at 70 $^\circ\text{C}$  in a sealed tube in a sand bath for 3-5 h until disappearance of the starting material and then cooled to room temperature.

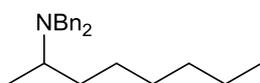
Acetonitrile was removed under reduced pressure and the crude material was purified by *flash* chromatography leading to the corresponding benzylated amine.

Characterization data are given below:



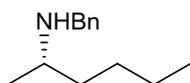
(±)-**2a**

(±)-***N*-Benzyl-octan-2-amine [(±)-2a]**. The title compound was obtained from (±)-**2b** according to the general procedure after 3 h and was isolated after column chromatography on silica gel (50% EtOAc/hexane) as a yellow oil (79% yield). <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 7.39-7.25 (*m*, 5H), 3.88-3.74 (apparent *q*, 2H, *J*= 13.1 Hz), 2.70 (*m*, 1H), 1.53 (*m*, 2H), 1.39-1.25 (*m*, 8H), 1.12 (*d*, 3H, *J*= 6.2 Hz), 0.91 (*t*, 3H, *J*= 6.2 Hz). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>) δ 140.8, 128.4 (2C), 128.1 (2C), 126.8, 52.5, 51.4, 37.1, 31.9, 29.5, 26.0, 22.7, 20.3, 14.1. MS (APCI<sup>+</sup>, *m/z*) 220.2 [(M+H)<sup>+</sup>, 100%]. Spectral properties are consistent with the literature values.<sup>3</sup>



(±)-**9a**

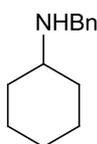
(±)-***N,N*-dibenzyl-octan-2-amine [(±)-9a]**. The title compound was obtained as byproduct in the previous reaction from (±)-**2b** and separated from the monobenzylated compound by silica gel column chromatography (50% EtOAc/hexane) to afford a yellow oil (12% yield). <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 7.45-7.20 (*m*, 10H), 3.72 (*d*, 2H, *J*= 13.9 Hz), 3.44 (*d*, 2H, *J*=13.8 Hz), 2.72 (*m*, 1H), 1.71-1.64 (*m*, 2H), 1.34-1.16 (*m*, 8H), 1.03 (*d*, 3H, *J*= 6.5 Hz), 0.90 (*t*, 2H, *J*= 6.8 Hz). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>) δ 140.9 (2C), 128.7 (4C), 128.1 (4C), 126.5 (2C), 53.3, 52.3, 33.8, 31.9, 29.4, 26.8, 22.7, 14.2, 13.5. MS (APCI<sup>+</sup>, *m/z*) 310.2 [(M+H)<sup>+</sup>, 100%].



(*S*)-**3a**

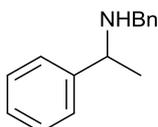
(*S*)-***N*-Benzylhexan-2-amine [(*S*)-3a]**. The title compound was prepared using the general procedure after 4.5 h and was isolated after column chromatography on silica

gel (70% EtOAc/hexane) as a colorless oil (83% yield).  $^1\text{H}$  NMR (300 MHz,  $\text{CDCl}_3$ )  $\delta$  7.35-7.25 (*m*, 5H), 3.80 (*q*, 2H,  $J= 13.0$  Hz), 2.69 (*m*, 1H), 1.53-1.28 (*m*, 7H), 1.10 (*d*, 3H,  $J= 6.0$  Hz), 0.92 (*t*, 3H,  $J= 6.8$  Hz).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ ) 130.4, 130.2, 129.3, 129.1, 52.5, 47.4, 32.4, 27.6, 22.2, 16.1, 13.8. MS (APCI $^+$ ,  $m/z$ ) 192.2 [(M+H) $^+$ , 100%]. Spectral data were consistent with the literature values.<sup>4</sup>



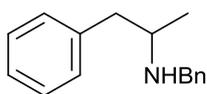
**4a**

***N*-benzylcyclohexanamine (4a).** The title compound was prepared using the general procedure after 4 h and was isolated after column chromatography on silica gel (70% EtOAc/hexane) as a colorless oil (86% yield).  $^1\text{H}$  NMR (300 MHz,  $\text{CDCl}_3$ )  $\delta$  7.49 (*d*, 2H,  $J= 6.6$  Hz), 7.40-7.29 (*m*, 3H), 5.50 (*br s*, 1H), 3.93 (*s*, 2H), 2.65 (*m*, 1H), 2.03 (*m*, 2H), 1.79 (*m*, 2H), 1.64 (*m*, 1H), 1.44-1.15 (*m*, 5H).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  136.2, 129.2 (2C), 128.7 (2C), 127.9, 55.9, 49.4, 31.5 (2C), 25.6, 24.8 (2C). MS (APCI $^+$ ,  $m/z$ ) 190.1 [(M+H) $^+$ , 100%]. Spectral data are consistent with the literature values.<sup>5</sup>



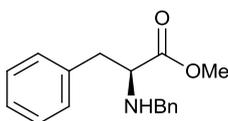
**(±)-6a**

**(±)-*N*-benzyl-1-phenylethanamine [(±)-6a].** The title compound was prepared using the general procedure after 5 h and was isolated after column chromatography (60% EtOAc/hexane) as a colorless oil (72% yield).  $^1\text{H}$  NMR (300 MHz,  $\text{CDCl}_3$ )  $\delta$  7.48-7.34 (*m*, 10H), 3.92 (*q*, 1H,  $J= 5.0$  Hz), 3.72 (apparent *q*, 2H,  $J= 9.9$  Hz), 1.74 (*br s*, 1H), 1.48 (*d*, 3H,  $J= 5.1$  Hz).  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ )  $\delta$  145.7, 140.8, 128.6 (2C), 128.5 (2C), 128.2 (2C), 127.0, 126.9, 126.8 (2C), 57.6, 51.8, 24.6. MS (APCI $^+$ ,  $m/z$ ) 212.1 [(M+H) $^+$ , 100%]. Spectral data are consistent with the literature values.<sup>6</sup>



(±)-7a

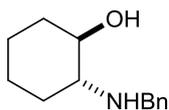
(±)-**N-benzyl-1-phenylpropan-2-amine** [(±)-7a]. The title compound was prepared using the corresponding deprotected amine (±)-7b, using the general procedure after 3 h and was isolated after column chromatography (100% EtOAc) as a colorless oil (81% yield). <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 7.34-7.17 (*m*, 10H), 3.88 (*q*, 2H, *J*= 13.3 Hz), 2.98 (*m*, 1H), 2.81 (*dd*, 1H, *J*= 13.2, 6.9 Hz), 2.67 (*dd*, 1H, *J*= 13.3, 6.5 Hz), 1.65 (*br s*, 1H), 1.13 (*d*, 3H, *J* = 6.2 Hz). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>) δ 140.9, 139.8, 129.7, 128.8, 128.4, 127.2, 126.6, 54.1, 51.7, 44.0, 20.6. MS (APCI<sup>+</sup>, *m/z*) 226.1 [(M+H)<sup>+</sup>, 100%]. Spectral data are consistent with the literature values.<sup>7</sup>



L-8a

**Methyl L-2-benzylamino-3-phenylpropionate** (L-8a). The title compound was prepared using the general procedure after 5 h and was isolated after column chromatography on silica gel (10% EtOAc/hexane) as a yellow oil (74% yield). <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 7.34-7.21 (*m*, 10H), 3.88 (*d*, 1H, *J*= 13.1 Hz), 3.69 (*m*, 4H), 3.60 (apparent *t*, 1H, *J*= 7.3 Hz), 3.02 (*d*, 2H, *J*= 7.3 Hz), 1.93 (*br s*, 1H). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>) δ 175.1, 139.6, 137.3, 129.3 (2C), 128.4 (2C), 128.3 (2C), 128.2 (2C), 127.1, 126.7, 62.1, 52.0, 51.7, 39.8. MS (APCI<sup>+</sup>, *m/z*) 270.1 [(M+H)<sup>+</sup>, 100%]. Spectral data are consistent with the literature values.<sup>8</sup>

#### II.4. General procedure for the preparation of N-benzylamine (±)-trans-5a



(±)-trans-5a

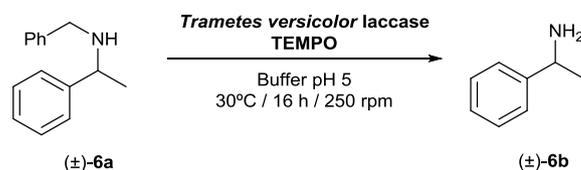
(±)-**trans-2-(Benzylamino)cyclohexanol** [(±)-trans-5a]. The title compound was prepared using a previously described procedure.<sup>9</sup> Cyclohexene oxide (5 mmol) and water (2 mL) were placed in a test tube equipped with a magnetic stirring bar. Then,

benzylamine (6 mmol) was added in one portion, and the reaction mixture was vigorously stirred at room temperature for 16 h. The reaction was monitored by TLC. Then water (2 mL) was added, and the organic layer was separated and dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>. The solvent was removed and the residue was purified by column chromatography on silica gel (5% MeOH/ EtOAc), obtaining (±)-*trans*-**5a** as a yellow solid (64% yield). <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 7.37-7.26 (*m*, 5H), 3.98 (*d*, 1H, *J*= 12.9 Hz), 3.72 (*d*, 1H, *J*= 12.9 Hz), 3.23 (*ddd*, 1H, *J* = 10.3, 9.2, 4.6 Hz), 2.32 (*ddd*, 1H, *J*= 11.1, 9.3, 3.9 Hz), 2.18 (*m*, 1H), 2.05 (*m*, 1H), 1.75 (*m*, 2H), 1.27 (*m*, 3H), 1.02 (*m*, 1H). <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>) δ 140.9, 128.8 (2C), 128.5 (2C), 127.4, 74.2, 63.5, 51.2, 33.7, 30.9, 25.6, 24.7. MS (APCI<sup>+</sup>, *m/z*) 206.1 [(M+H)<sup>+</sup>, 100%]. Spectral data are consistent with the literature values.<sup>10</sup>

### III. Laccase/TEMPO reaction optimizations

#### III.1. Effect of the buffer

**Table S1** Debenzylation of (±)-**6a** using the laccase/TEMPO system in different buffers at pH 5.<sup>a</sup>

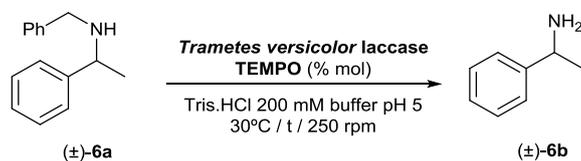


Entry	Buffer	(%) <sup>b</sup>
1	Tris.HCl 200 mM	98
2	Citrate 50 mM	99 <sup>c</sup>
3	Acetate 50 mM	12

<sup>a</sup> Reaction conditions: (±)-**6a** (0.08 mmol), laccase from *T. versicolor* (7 U) and TEMPO (33% mol) in an oxygen-saturated buffer pH 5 at 30°C for 16 h at 250 rpm. <sup>b</sup> Conversion values measured by GC. <sup>c</sup> 35% of acetophenone was found as by-product.

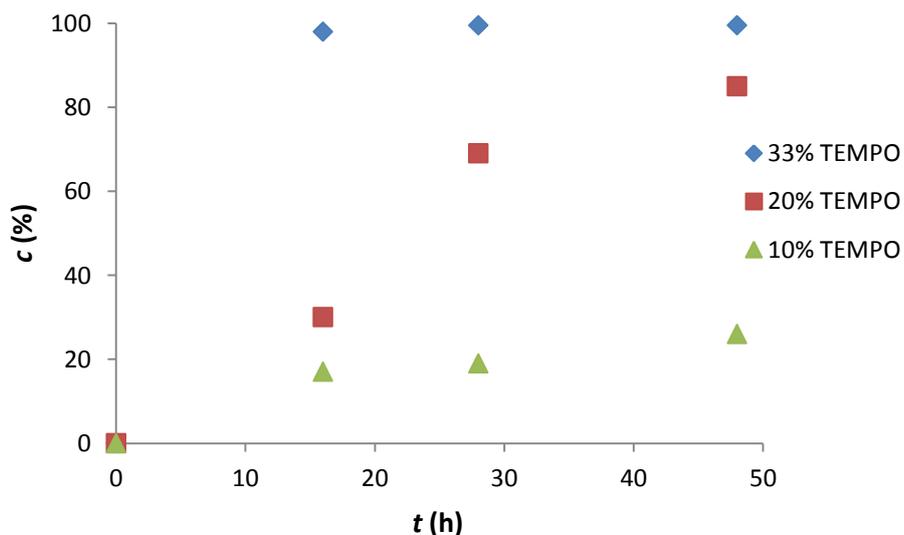
### III.2. Effect of the TEMPO concentration

**Table S2** Effect of the TEMPO concentration in the debenzylation of ( $\pm$ )-**6a** using the laccase/TEMPO system.<sup>a</sup>



Entry	TEMPO (%) <sup>b</sup>	t (h)	(%) <sup>c</sup>
1	33	16	98
2	33	28	>99
3	33	48	>99
2	20	16	30
3	20	28	69
6	20	48	85
7	10	16	17
8	10	28	19
9	10	48	26

<sup>a</sup> Reaction conditions: ( $\pm$ )-**6a** (0.08 mmol), *T. versicolor* laccase (7 U) and TEMPO (% mol) in an oxygen saturated Tris.HCl 200 mM buffer pH 5 at 30°C at 250 rpm. <sup>b</sup> % mol regarding the substrate. <sup>c</sup> Conversion values measured by GC.



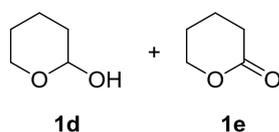
**Fig. S2** Effect of the TEMPO concentration in the debenylation of (±)-**6a** using the laccase/TEMPO system. Reaction conditions: (±)-**6a** (0.08 mmol), *T. versicolor* laccase (7 U) and TEMPO (% mol) in an oxygen saturated Tris.HCl 200 mM buffer pH 5 at 30°C at 250 rpm.

#### **IV. General procedure for the *N*-benzyl deprotection reactions using laccase from *Trametes versicolor*/TEMPO**

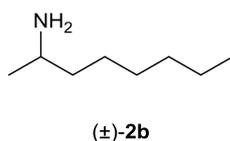
In an open-to-air Erlenmeyer flask (10 mL), a solution of the corresponding *N*-benzyl amines, amino alcohols or amino esters **1a-13a** (0.08 mmol, 47 mM) in previously O<sub>2</sub>-saturated Tris.HCl buffer 200 mM pH 5 (1.7 mL), was treated with TEMPO (33% mol, 4.17 mg), and this mixture was stirred until complete solubilization of the reactants. Then, the laccase was added (5.17 mg, 41 U/mL) and the reaction was stirred overnight in an orbital shaker at 250 rpm at 30°C. The reaction was monitored by GC until no starting material remained. The reaction media was basified with NaOH 3N or a saturated solution of NaHCO<sub>3</sub> for amino alcohol **1a** and amino ester L-**8a** (to avoid its hydrolysis), and then extracted with CH<sub>2</sub>Cl<sub>2</sub> or EtOAc for L-**8a** (3 x 5 mL). The combined organic layers were washed with brine and dried over Na<sub>2</sub>SO<sub>4</sub>. For volatile compounds, careful evaporation of the organic solvent was carried out in the rotary evaporator at 10°C.

## V. Procedure for large-scale deprotection reactions

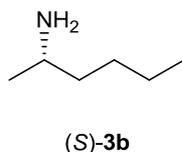
In an open-to-air Erlenmeyer flask (100 mL), protected amine ( $\pm$ )-**6a**, ( $\pm$ )-**7a**, or ( $\pm$ )-*trans*-**13a** (100 mg) in previously O<sub>2</sub>-saturated Tris.HCl 200 mM buffer pH 5 (11.1 mL), was treated with TEMPO (20% mol) and the mixture was stirred until complete solubilization of the reactants. The laccase was added (41 U/mL) and the reaction was stirred 16 h under orbital shaking (250 rpm) at 30°C. Then, the reaction was basified until pH 10 with NaOH 3N and extracted with CH<sub>2</sub>Cl<sub>2</sub> (3 x 10 mL). The combined organic layers were washed and dried over Na<sub>2</sub>SO<sub>4</sub>. Amines ( $\pm$ )-**6b**, ( $\pm$ )-**7b**, or ( $\pm$ )-*trans*-**10a**, were obtained as pure compounds based on GC and NMR analysis (>90% yield).



**Hemiacetal **1d** and  $\delta$ -valerolactone (**1e**).** This mixture of compounds was obtained after extraction by addition of a saturated solution of NaHCO<sub>3</sub> until pH 7 in >99% conversion in a proportion 1.5:1 (**1d**:**1e**). Spectral data are consistent with the literature values.<sup>11</sup>

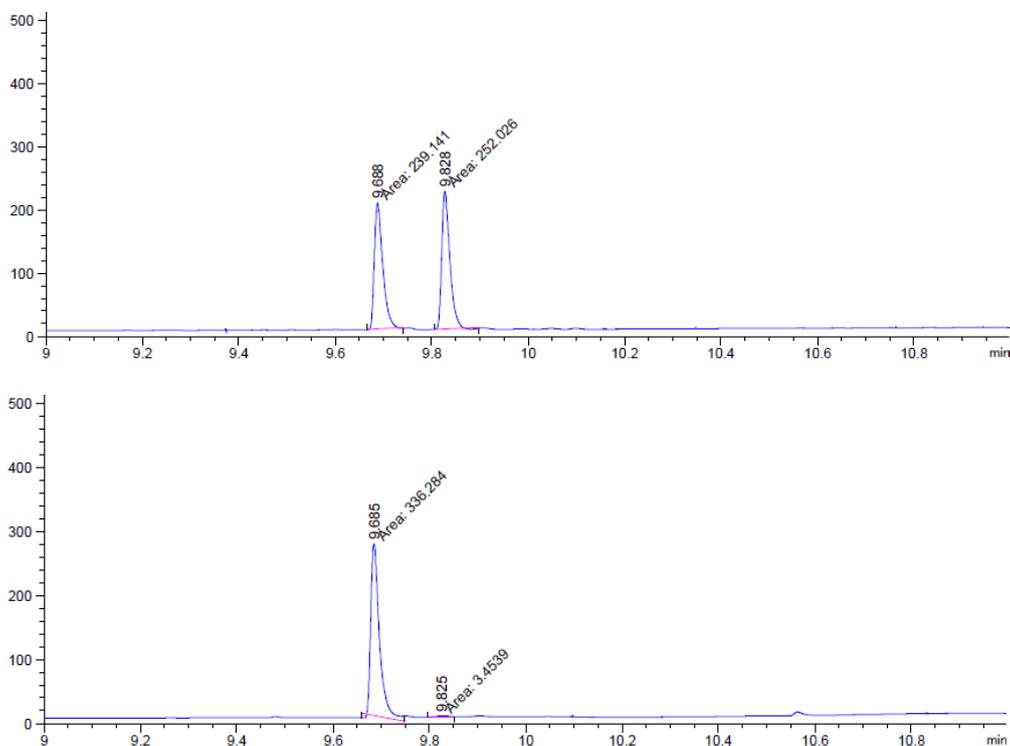


**( $\pm$ )-Octan-2-amine [( $\pm$ )-**2b**].** The title compound was obtained after basic extraction using an aqueous solution of NaOH 3 N until pH 10 in 97% conversion. <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>)  $\delta$  2.82 (*m*, 1H), 1.58 (*br s*, 2H), 1.25 (*m*, 10H), 1.00 (*d*, 3H, *J*= 6.5 Hz), 0.84 (*t*, 3H, *J*= 6.3 Hz). Spectral data are consistent with the sample obtained from the commercial source.

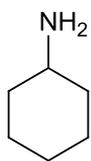


**(*S*)-Hexan-2-amine [(*S*)-**3b**].** The title compound was obtained after basic extraction using an aqueous solution of NaOH 3 N until pH 10 in 97% conversion. <sup>1</sup>H NMR (300

MHz,  $\text{CDCl}_3$ )  $\delta$  2.86 (*m*, 1H), 1.32-1.22 (*m*, 6H), 1.00 (*d*, 3H,  $J= 6.2$  Hz), 0.90 (*t*, 3H,  $J= 6.8$  Hz). For determining the *ee* from (*S*)-**3b**, it was acetylated using a standard procedure and compared to the acetylated racemic compound (Fig. S3). Spectral data are consistent with the sample obtained from the commercial source.

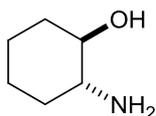


**Fig. S3** Analytical separation for racemic (up) and enantiopure (below) *N*-(hexan-2-yl)acetamide. CP-Chirasil-DEX CB 25 m x 0.25 mm. GC method: 70°C /5 min /20°C/min /180°C /3 min. Retention times:  $t_R$  (*S*)= 9.68 min,  $t_R$  (*R*)= 9.82 min.



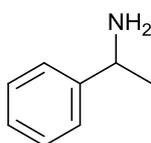
**4b**

**Cyclohexanamine (4b).** The title compound was obtained after basic extraction using an aqueous solution of NaOH 3 N until pH 10 in 99% conversion.  $^1\text{H}$  NMR (300 MHz,  $\text{CDCl}_3$ )  $\delta$  2.61 (*m*, 1H), 1.84-1.56 (*m*, 4H), 1.33-0.96 (*m*, 6H). Spectral data are consistent with the sample obtained from the commercial source.



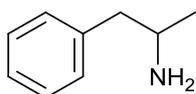
(±)-*trans*-**5b**

(±)-*trans*-2-Aminocyclohexanol [(±)-**5b**]. The title compound was after basic extraction using an aqueous solution of NaOH 3 N until pH 10 in 99% conversion. <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 3.13 (*m*, 1H), 2.42 (*m*, 1H), 2.02-1.85 (*m*, 2H), 1.76-1.67 (*m*, 2H), 1.34-1.23 (*m*, 3H), 1.19-1.05 (*m*, 1H). Spectral data are consistent with the sample obtained from the commercial source.



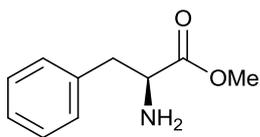
(±)-**6b**

(±)-1-Phenylethylamine [(±)-**6b**]. The title compound was obtained after basic extraction using an aqueous solution of NaOH 3 N until pH 10 in 97% conversion and 80% isolated yield in a large scale reaction using 100 mg of substrate. <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 7.35 (*m*, 4H), 7.25 (*m*, 1H), 4.13 (*q*, 1H, *J*= 6.7 Hz), 1.58 (*br s*, 2H), 1.41 (*d*, 3H, *J*= 6.6 Hz). Spectral data are consistent with the sample obtained from the commercial source.



(±)-**7b**

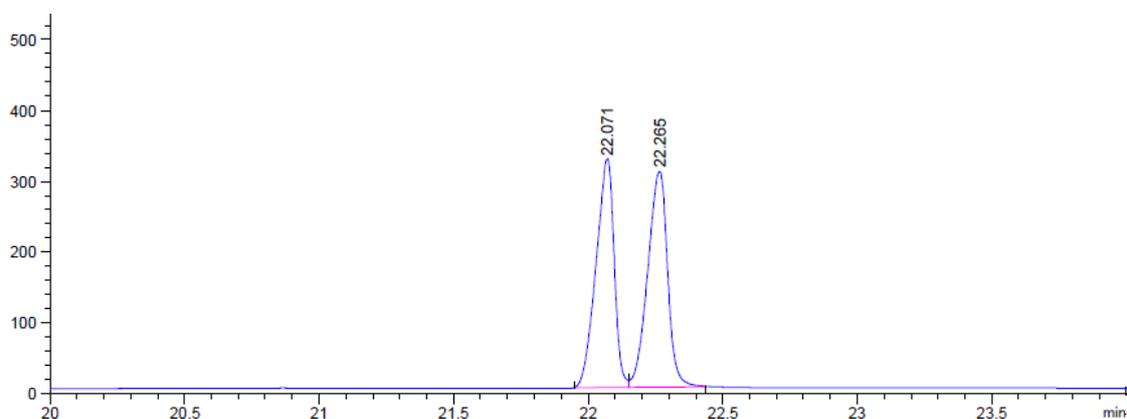
(±)-1-Phenylpropan-2-amine [(±)-**7b**]. The title compound was obtained after basic extraction using an aqueous solution of NaOH 3 N until pH 10 in >99% conversion and 90% isolated yield in a large scale reaction using 100 mg of substrate. Spectral data were previously shown (pages S3-S4).



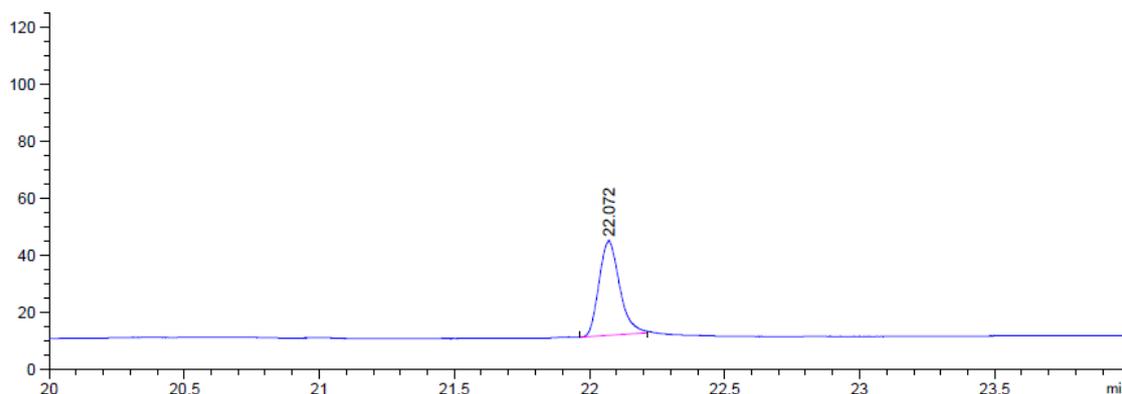
**L-8b**

**L-Phenylalanine methyl ester (L-8b).** The title compound was obtained after basic extraction using a saturated solution of NaHCO<sub>3</sub> until pH 7 and EtOAc in 99% conversion after 8 h in enantiopure form (Fig. S4). Longer reaction times afforded hydrolysis of methyl ester. <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 7.35-7.19 (*m*, 5H), 3.75 (*m*, 4H), 3.11 (*dd*, 1H, *J*= 13.5, 5.1 Hz), 2.88 (*dd*, 1H, *J*= 13.5, 7.9 Hz), 1.56 (*br s*, 2H). Spectral data are consistent with the sample obtained from the commercial source.

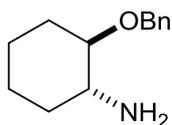
GC chromatogram for the racemic mixture of (±)-**8b**.



GC chromatogram for enantioenriched L-**8b**, 99% *ee*.

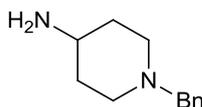


**Fig. S4** Analytical separation for **8b**. RtbDEXse (30 m x 0.25 mm). GC method: 110°C/ 3 min/ 3°C/min/ 200°C/ 3 min. Retention times:  $t_R$  L-**8b**= 22.0 min,  $t_R$  D-**8b**= 22.2 min.



(±)-*trans*-**10a**

(±)-*trans*-2-(Benzyloxy)cyclohexanamine [(±)-**10a**]. The title compound was obtained after basic extraction using an aqueous solution of NaOH 3 N until pH 10 in 99% conversion and 95% isolated yield in a large scale reaction using 100 mg of substrate (±)-*trans*-**13a**. <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 7.38-7.28 (*m*, 5H), 4.70 (*d*, 1H, *J*= 11.5 Hz), 4.48 (*d*, 1H, *J*= 11.5 Hz), 3.04 (*dt*, 1H, *J*= 9.6, 4.4 Hz), 2.70 (*ddd*, 1H, *J*= 10.7, 8.9, 4.2 Hz), 2.17 (*m*, 1H), 1.89 (*m*, 1H), 1.78 (*m*, 1H), 1.69 (*m*, 4H), 1.23 (*m*, 2H). Spectral data are consistent with the sample obtained from the commercial source.



**12b**

**4-Amino-1-benzylpiperidine (12b)**. The title compound was obtained after basic extraction using an aqueous solution of NaOH 3 N until pH 10 in 99% conversion. <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 7.33-7.25 (*m*, 5H), 3.50 (*s*, 1H), 2.84 (*d*, 2H, *J*= 12.1 Hz), 2.65 (*m*, 1H), 2.03 (*td*, 2H, *J*= 11.6, 2.7 Hz), 1.79 (*d*, 2H, *J*= 12.8 Hz), 1.47-1.34 (*m*, 4H). Spectral data are consistent with the sample obtained from the commercial source.

## VI. Control experiments

**Without laccase.** A solution of the protected amine (±)-**2a** (47 mM) in Tris.HCl buffer 200 mM pH 5 (1.7 mL) was treated with TEMPO (33% mol). The reaction was stirred 16 h in an orbital shaker (250 rpm) in an open-to-air Erlenmeyer flask at 30°C in the absence of the laccase and monitored by GC. Only the starting material was detected in all cases.

**Without TEMPO.** A solution of the corresponding protected amine (±)-**2a** (47 mM) in Tris.HCl buffer 200 mM pH 5 (1.7 mL) was treated with laccase (70 U). The reaction was stirred 16 h in an orbital shaker (250 rpm) in an open-to-air Erlenmeyer flask at 30°C in the absence of TEMPO and monitored by GC. Only the starting material was detected.

**Without TEMPO and laccase.** A solution of the corresponding protected amine ( $\pm$ )-**2a** (47 mM) in Tris.HCl buffer 200 mM pH 5 (1.7 mL) was stirred 16 h in an orbital shaker (250 rpm) in an open-to-air Erlenmeyer flask at 30°C in the absence of TEMPO and laccase, and monitored by GC. Only the starting material was detected.

## VII. Comparison with other laccase or copper/mediator systems

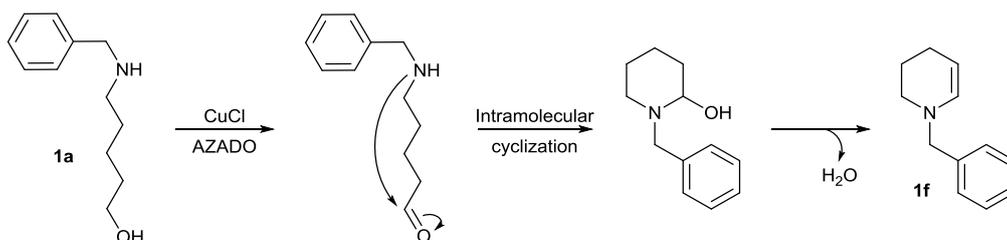
### Laccase/AZADO

In an open-to-air Erlenmeyer flask, a solution of the corresponding *N*-benzylamino alcohol (0.08 mmol, 47 mM) in previously oxygenated Tris.HCl buffer 200 mM pH 5 (1.7 mL), was treated with AZADO (33% mol, 4.01 mg), and this mixture was stirred until complete solubilization of the reactants. Then, the laccase was added (5.17 mg, 41 U/mL) and the reactions were stirred overnight in an orbital shaker at 250 rpm and 30°C. The reaction was stopped after 16 h. The extraction was carried out by adding NaOH 3N until pH 9-10 for ( $\pm$ )-*trans*-**5a** and **11a** or a saturated solution of NaHCO<sub>3</sub> until pH 7 for **1a** using dichloromethane as organic solvent (3 x 5 mL). The combined organic layers were washed with brine and dried over Na<sub>2</sub>SO<sub>4</sub>. Solvent was finally removed under vacuum.

### CuCl/AZADO

To a mixture of the corresponding *N*-benzylamino alcohols (0.5 mmol), DMAP (0.03 mmol, 3.67 mg), bpy (15  $\mu$ mol, 2.34 mg), AZADO (15  $\mu$ mol, 2.28 mg for **1a** and **11a**, and 5  $\mu$ mol, 0.76 mg for ( $\pm$ )-*trans*-**5a**), and CuCl (15  $\mu$ mol, 1.49 mg), MeCN (2.5 mL) was added. The mixture was stirred at room temperature under air atmosphere (open) for 16 h. The reaction was quenched with saturated NaHCO<sub>3</sub> and 50% Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub> (2 mL) and the mixture was vigorously stirred at room temperature for 5 min. The mixture was extracted with CH<sub>2</sub>Cl<sub>2</sub> (3 x 5 mL). The organic layer was dried over Na<sub>2</sub>SO<sub>4</sub> and evaporated under vacuum.<sup>12</sup>

When employing substrate **1a** as substrate, enamine **1f** was obtained in 18% conversion. The proposed mechanism to explain its formation is showed below (Scheme S3).



**Scheme S3** Proposed mechanism for the formation of enamine **1f**.

## VIII. Comparison with other chemical deprotection systems

**Table S3** Results obtained in the chemoselective *N*-benzyl deprotection of amino alcohol ( $\pm$ )-*trans*-**13a** using different chemical methods.

System	Substrate	Product	Solvent	Yield
H <sub>2</sub> , Pd/C			MeOH	27%
CAN			MeCN/H <sub>2</sub> O 5:1	40%
, Pd/C, CH <sub>3</sub> COOH			EtOH	<1%
DIAD			THF	80%

### H<sub>2</sub>, Pd/C

To a solution of the protected amino alcohol ( $\pm$ )-*trans*-**13a** (0.339 mmol, 100 mg) in deoxygenated MeOH (3.39 mL), Pd/C on charcoal (10% w w<sup>-1</sup>, 3.39 mg) was carefully added and the reaction was stirred for 16 h and room temperature under H<sub>2</sub> pressure (balloon). Then, the reaction was filtered over Celite and washed with MeOH. The solvent was concentrated under vacuum and the residue was purified by column chromatography on silica gel (85% EtOAc/hexane to 100% EtOAc), obtaining ( $\pm$ )-*trans*-**10a** as a yellow oil (27% yield).<sup>13</sup>

## CAN

To a stirred solution of the amino alcohol ( $\pm$ )-*trans*-**13a** (0.169 mmol, 50 mg) in MeCN/H<sub>2</sub>O (5:1, 0.93 mL), CAN (0.355 mmol, 195 mg) was added and the reaction was stirred at room temperature for 16 h. After that time, the reaction was quenched and basified by the addition of saturated aqueous sodium bicarbonate solution and stirred vigorously for ten minutes before extracting with CH<sub>2</sub>Cl<sub>2</sub> (3 x 5 mL). The combined organic extracts were dried (Na<sub>2</sub>SO<sub>4</sub>), filtered and concentrated in vacuum. The crude conversion into ( $\pm$ )-*trans*-**5a** was determined by NMR.<sup>14</sup>

## 1,4-cyclohexadiene, Pd/C

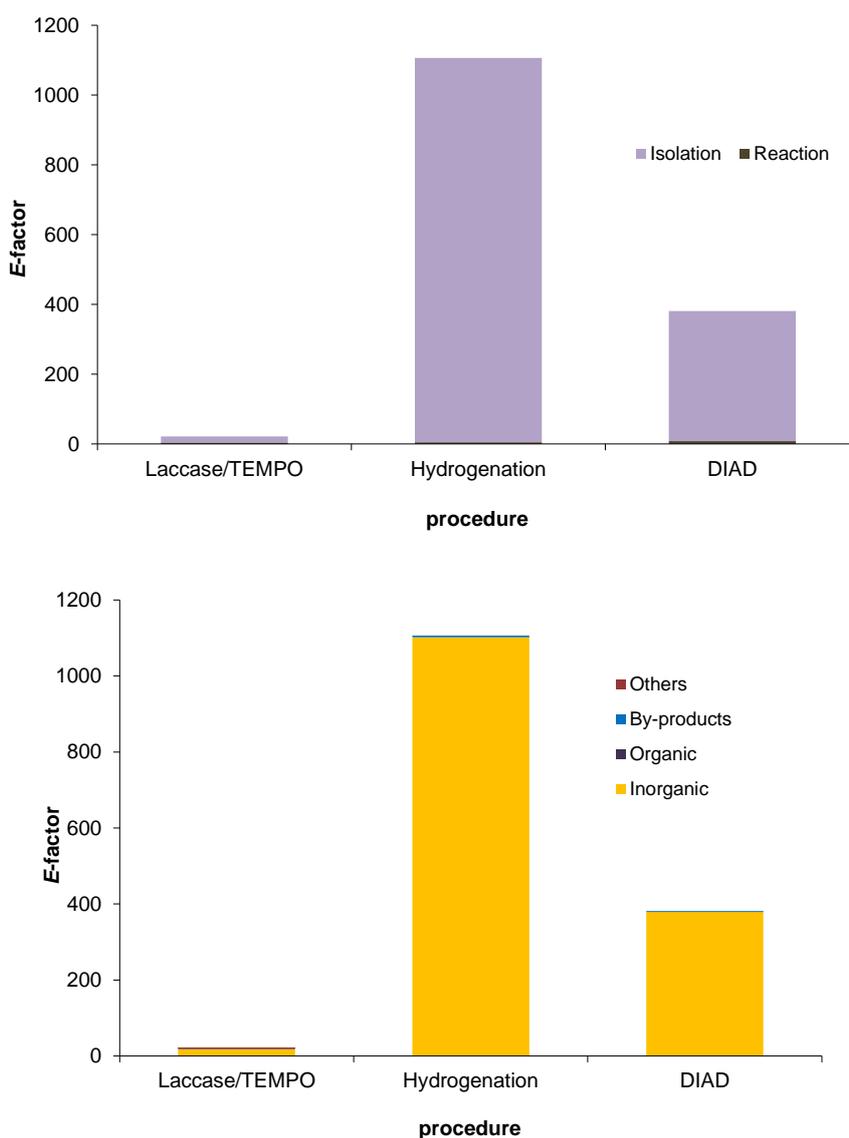
To a stirred solution of ( $\pm$ )-*trans*-**13a** (0.101 mmol, 30 mg) in absolute ethanol (1 mL) and acetic acid (0.303 mmol, 17  $\mu$ L) was added, under nitrogen atmosphere, Pd-C (10% w w<sup>-1</sup>, 30 mg) followed by 1,4-cyclohexadiene (0.505 mmol, 17  $\mu$ L). After stirring the reaction for 16 h the catalyst was removed by filtration through Celite, and the filtrate was evaporated to dryness under vacuum. The resulting crude was basified with NaOH 3N and then extracted with CH<sub>2</sub>Cl<sub>2</sub> (4 x 10 mL). The combined organic extracts were dried (Na<sub>2</sub>SO<sub>4</sub>), filtered and concentrated in vacuo. The crude conversion was determined by NMR.<sup>15</sup>

## DIAD

( $\pm$ )-*trans*-**13a** (0.169 mmol, 50 mg) was dissolved in THF (2 mL), and DIAD (0.2011 mmol, 39  $\mu$ L) was added. The solution was heated under reflux for 15 h. Reaction was monitored by TLC and stopped after consumption of the starting material. To the mixture, 5% HCl (2.5 mL) was added and the mixture was refluxed overnight. Then the solvent was removed under vacuum and the resulting crude was basified with NaOH 3N and extracted with CH<sub>2</sub>Cl<sub>2</sub> (2 x 5 mL). The combined organic extracts were dried (Na<sub>2</sub>SO<sub>4</sub>), filtered and concentrated and the residue was purified by column chromatography on silica gel (85% EtOAc/hexane to 100% EtOAc), obtaining ( $\pm$ )-*trans*-**10a** as a yellow oil (80% yield).<sup>16</sup>

## IX. Environmental assessment using EATOS

*E*-factor calculations (Fig. S5) were performed using the EATOS (v. 1.1) software tool,<sup>17</sup> while solvent demand was calculated using Microsoft Excel (determining the solvent use of each protocol to obtain 1 g of ( $\pm$ )-*trans*-**10a**). All reactions were treated as proceeding to the corresponding isolated yields, hence all losses in yield are accounted for as ‘unknown by-products’. EATOS and Excel files used for these calculations are also available as ESI material.



**Fig. S5** Contribution to *E*-factor (excluding solvents) for each procedure to synthesize *O*-benzylated amine ( $\pm$ )-*trans*-**10a**.

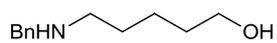
## X. Analytical data

**Table S4** Analytical separation of *N*-benzyl amines and deprotected amines.

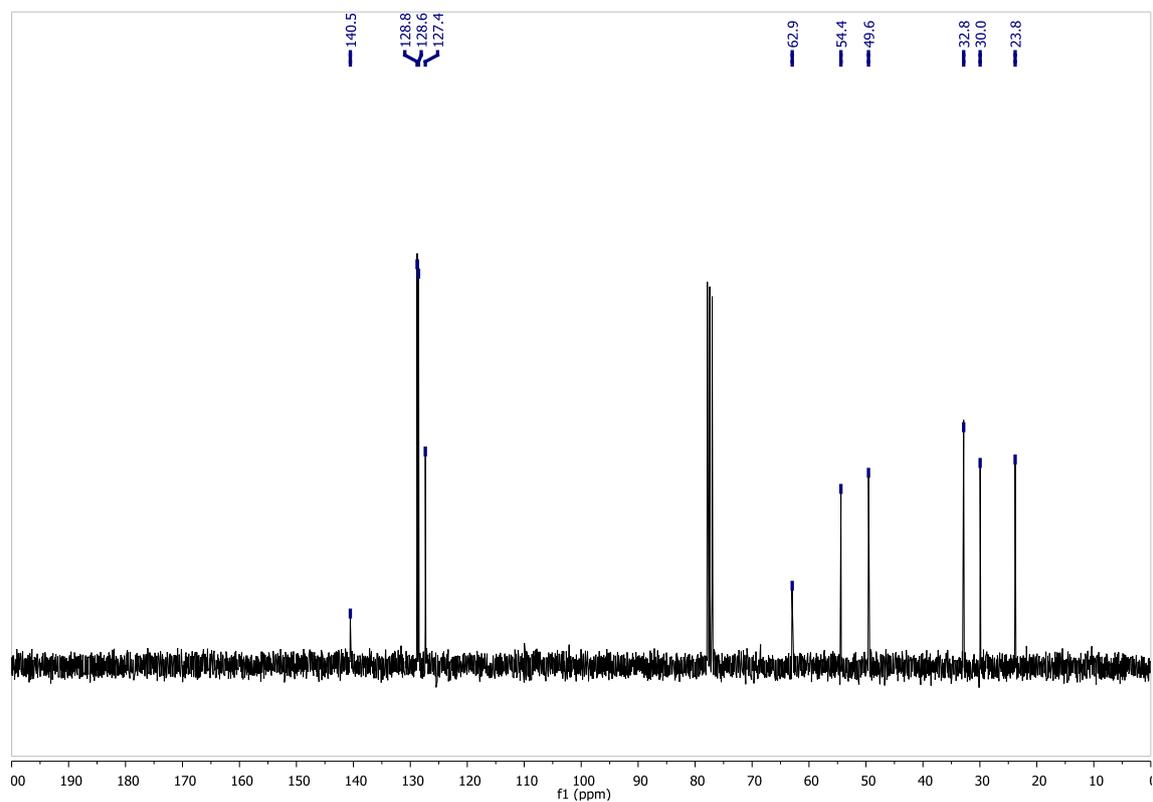
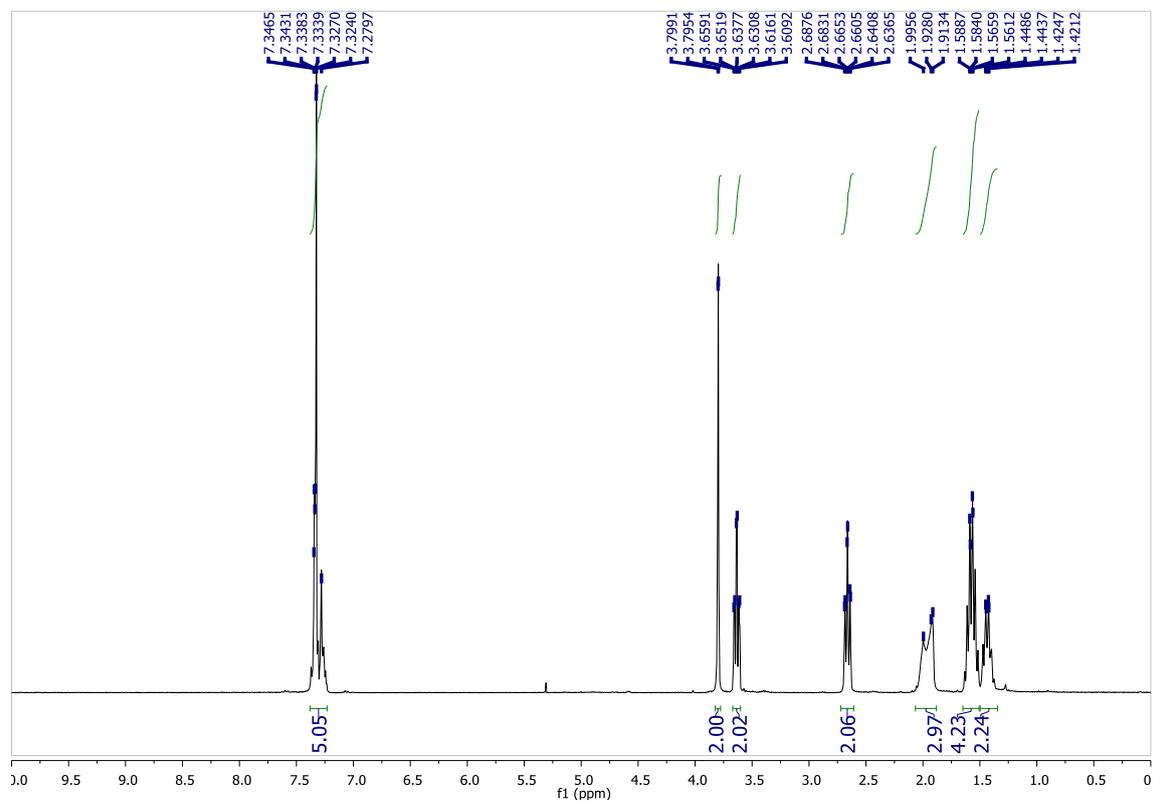
Substrate	Product	GC Method <sup>a</sup>	Retention times (min)	
			t <sub>R</sub> substrate	t <sub>R</sub> product
		90/3/10/220/2	15.4	10.0
		90/3/10/220/2	10.0	1.5
		40/3/10/160/2	12.8	2.0
		60/3/10/180/2	11.9	1.8
		60/3/10/180/2	13.8	4.2
		90/3/10/220/2	10.5	1.8
		110/3/10/220/5	9.2	1.5
		110/3/10/220/5	11.2	4.1
		60/3/10/180/2	12.2	3.2
		140/3/10/220/5	11.2	3.3
		60/3/10/180/2	13.0	4.2
		60/3/10/180/2/ 10/220/5	20.8	13.0

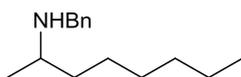
<sup>a</sup> The following GC column was used: Hewlett Packard HP-1 column (30 m x 0.32 mm x 0.25 μm, 14.5 psi). Initial T (°C) /time (min) /slope (°C/min) /T (°C) /time (min) /slope (°C/min) /final T (°C) / time (min).

# XI. Full characterization of compounds

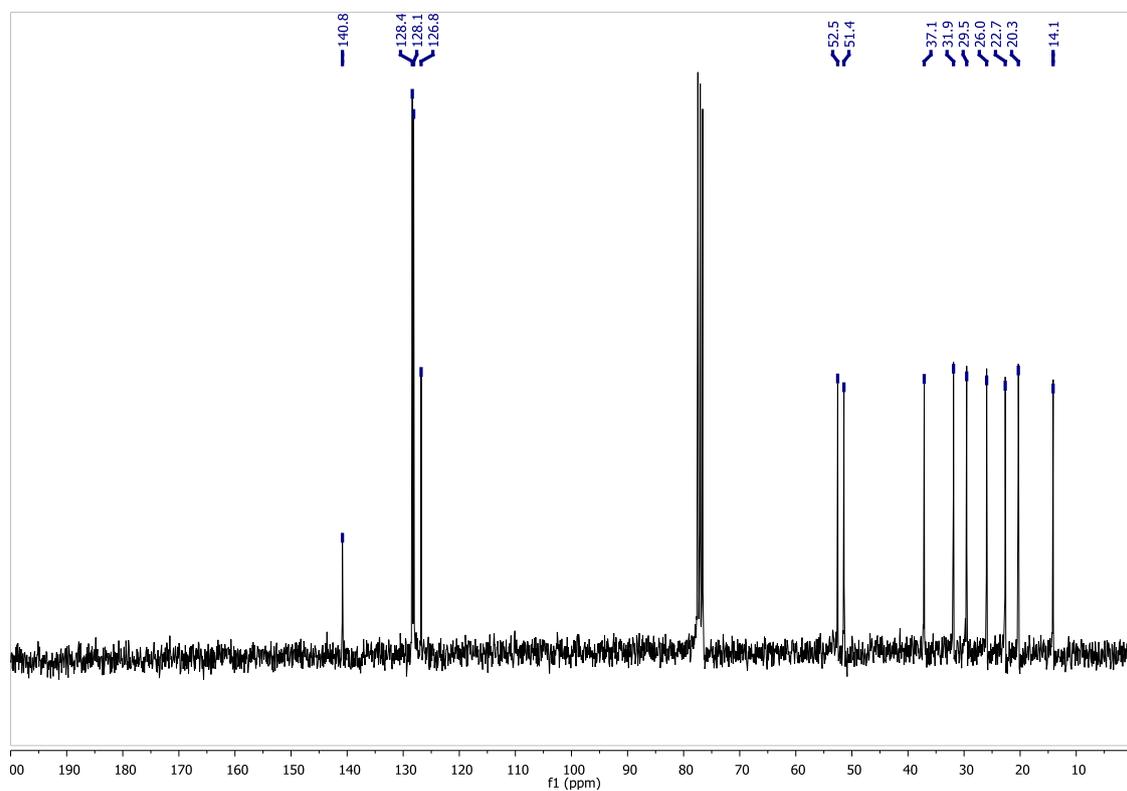
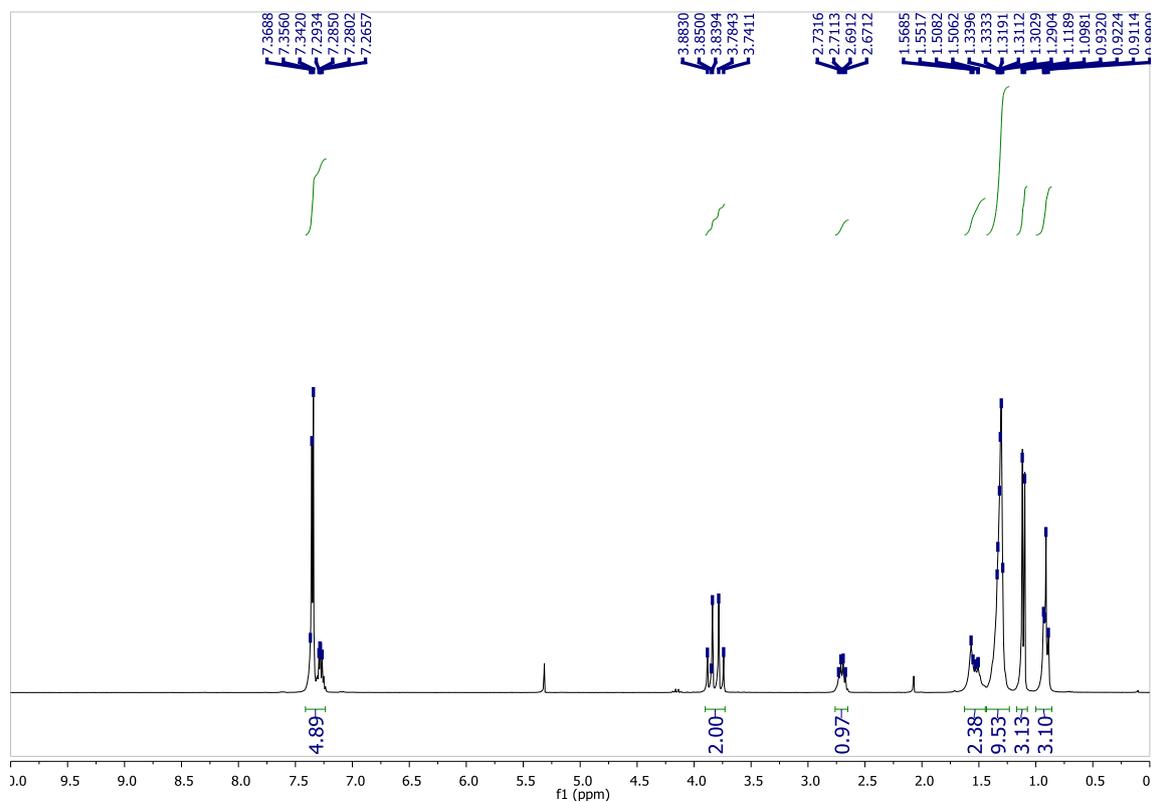


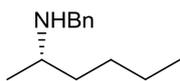
1a



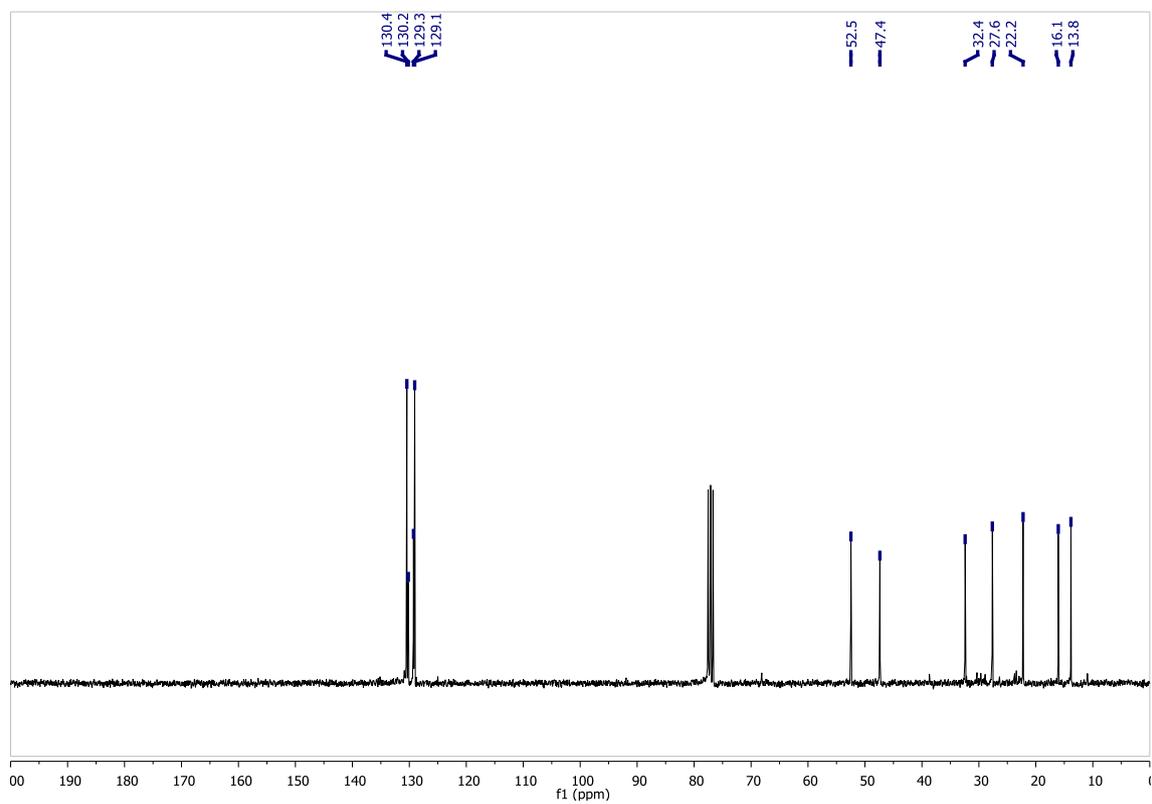
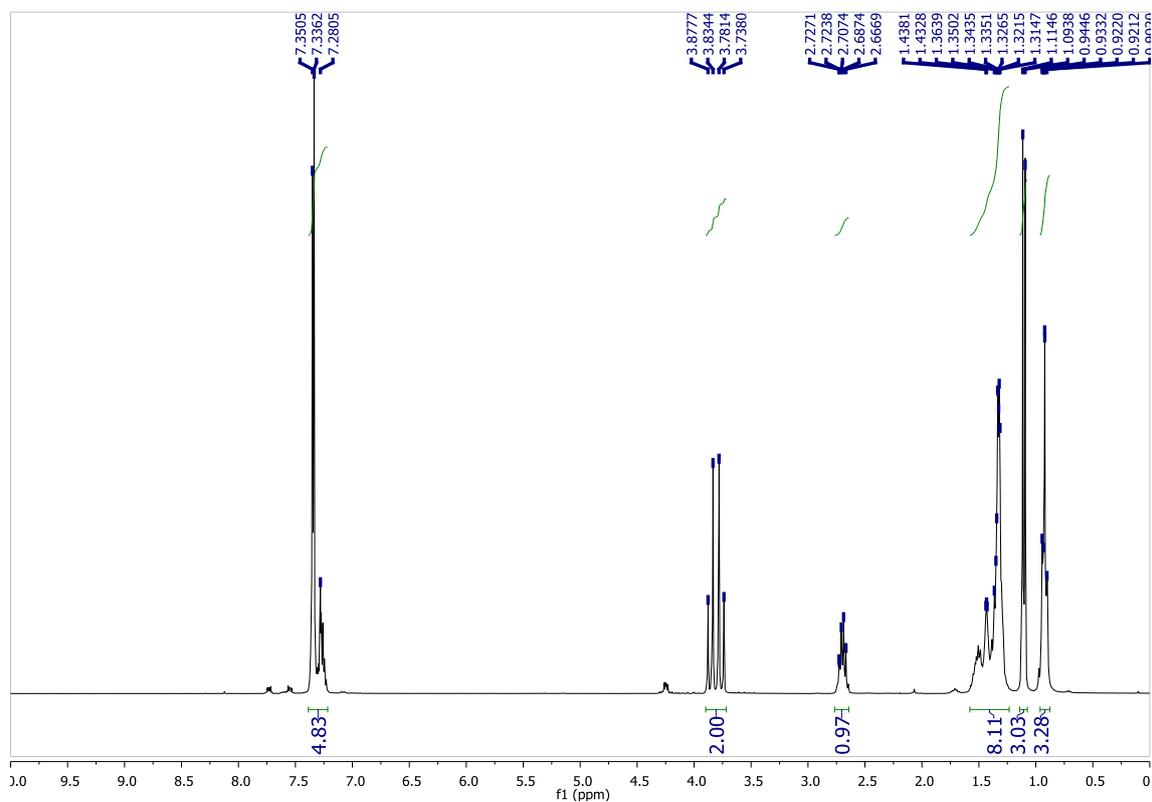


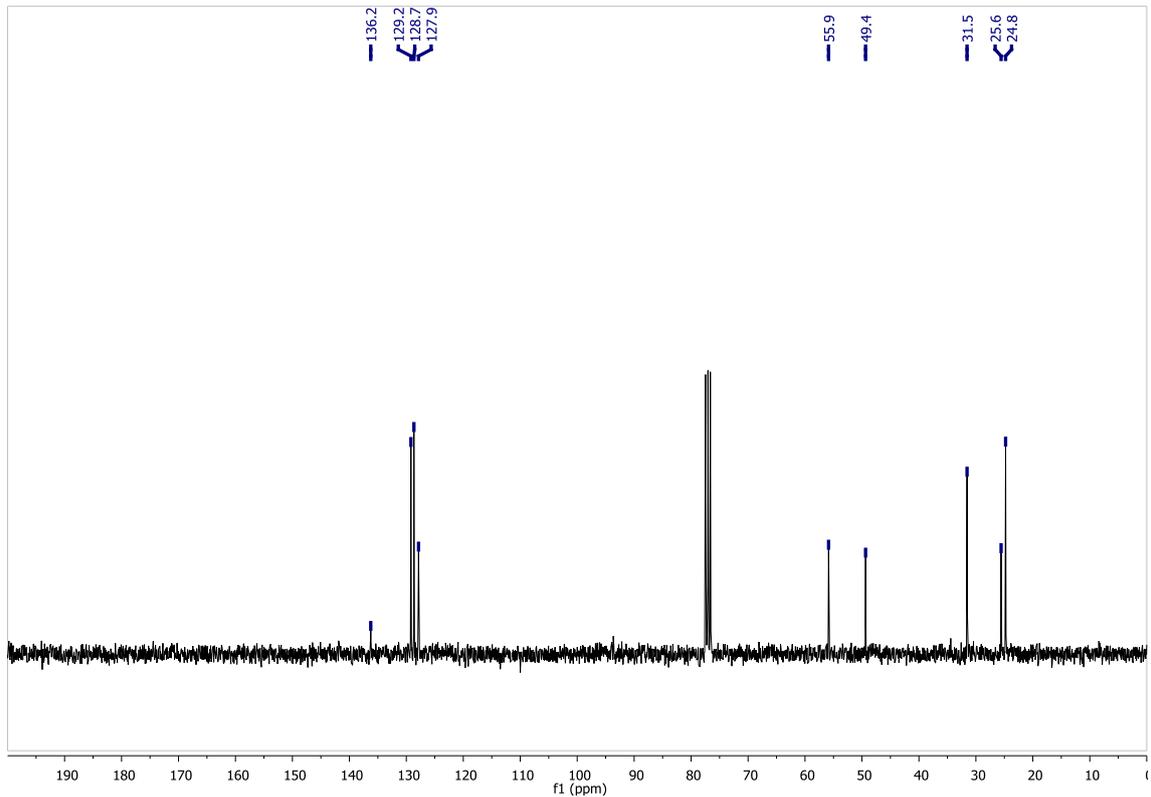
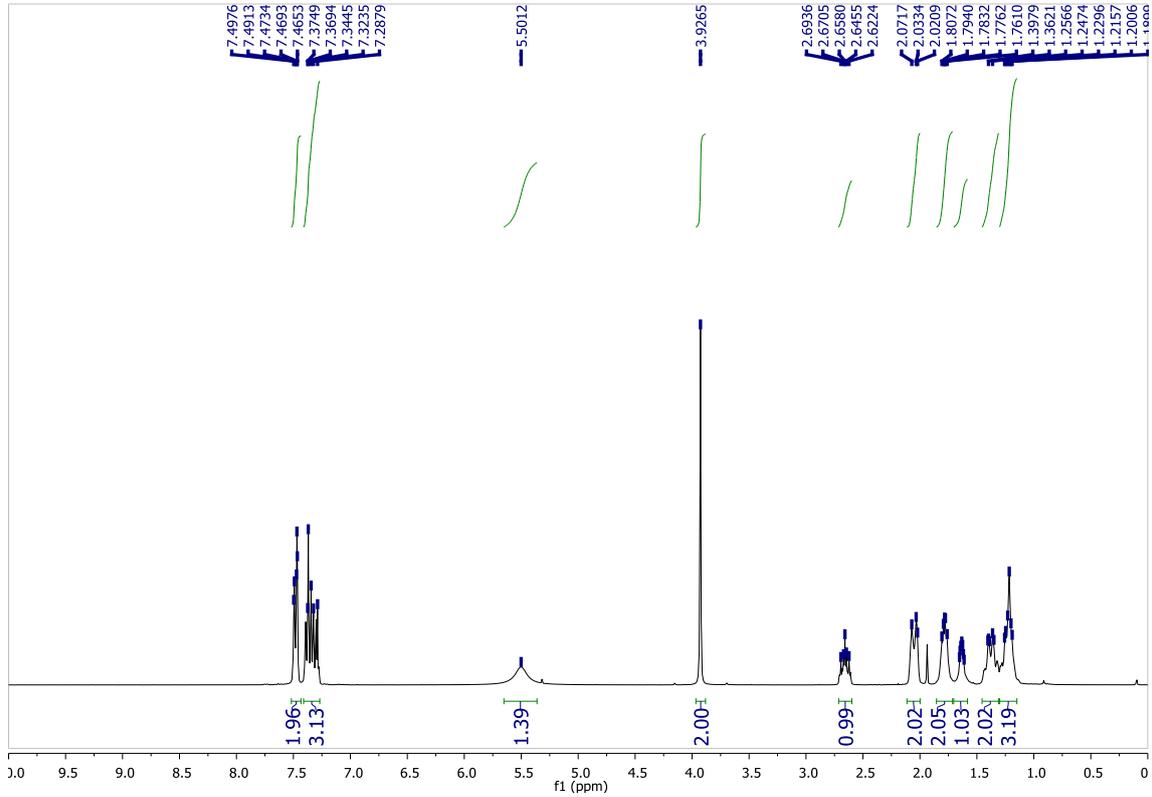
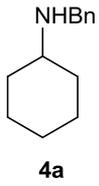
(±)-2a

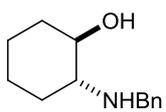




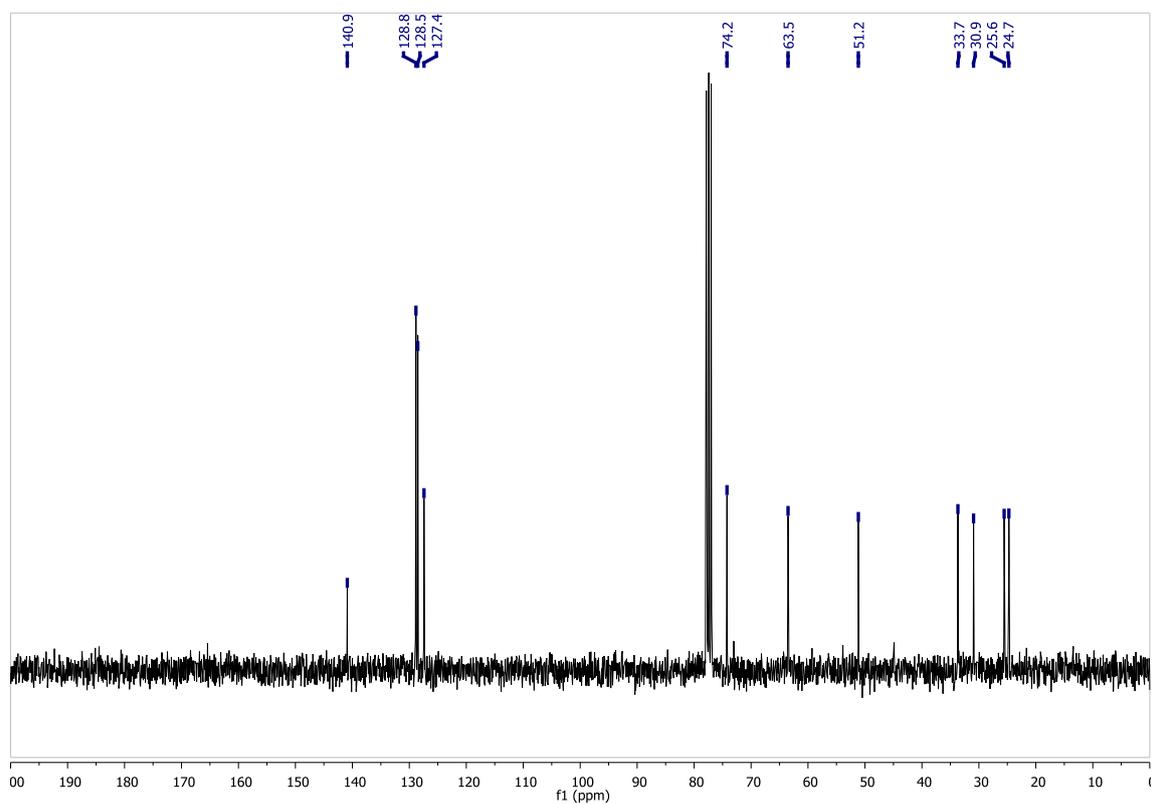
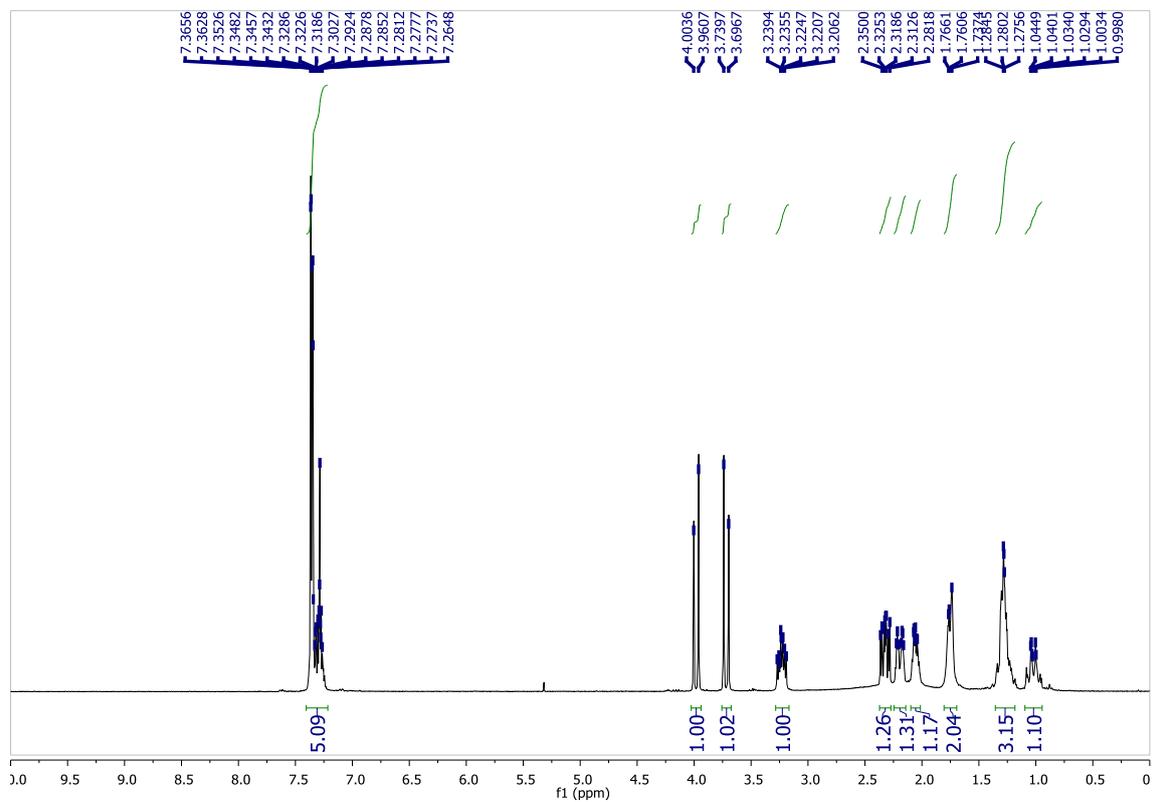
(S)-3a

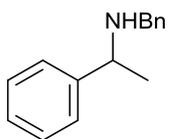




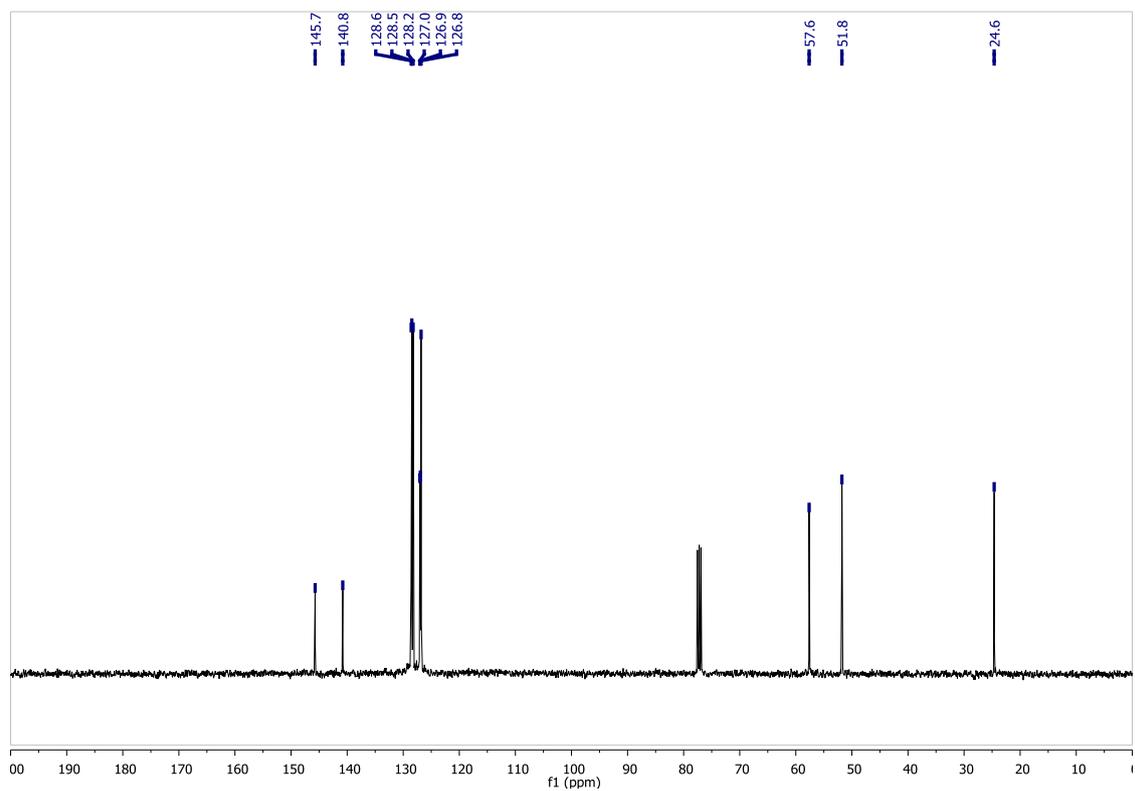
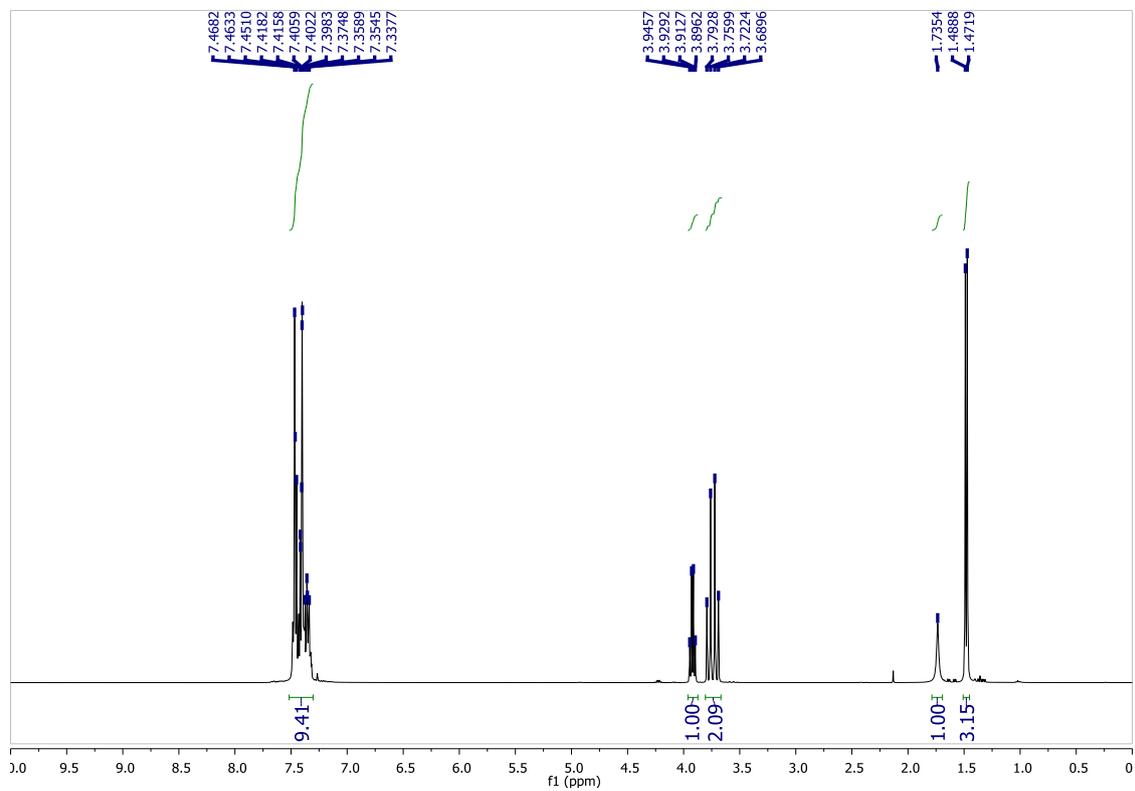


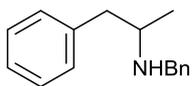
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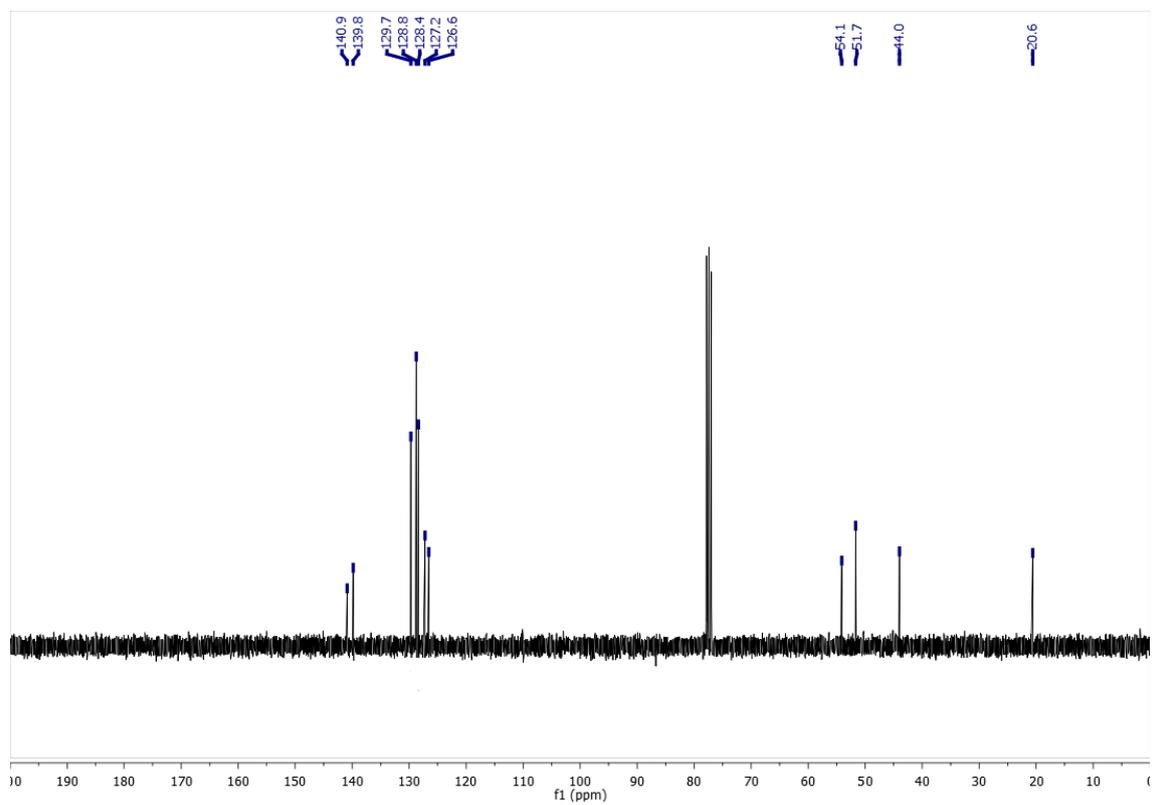
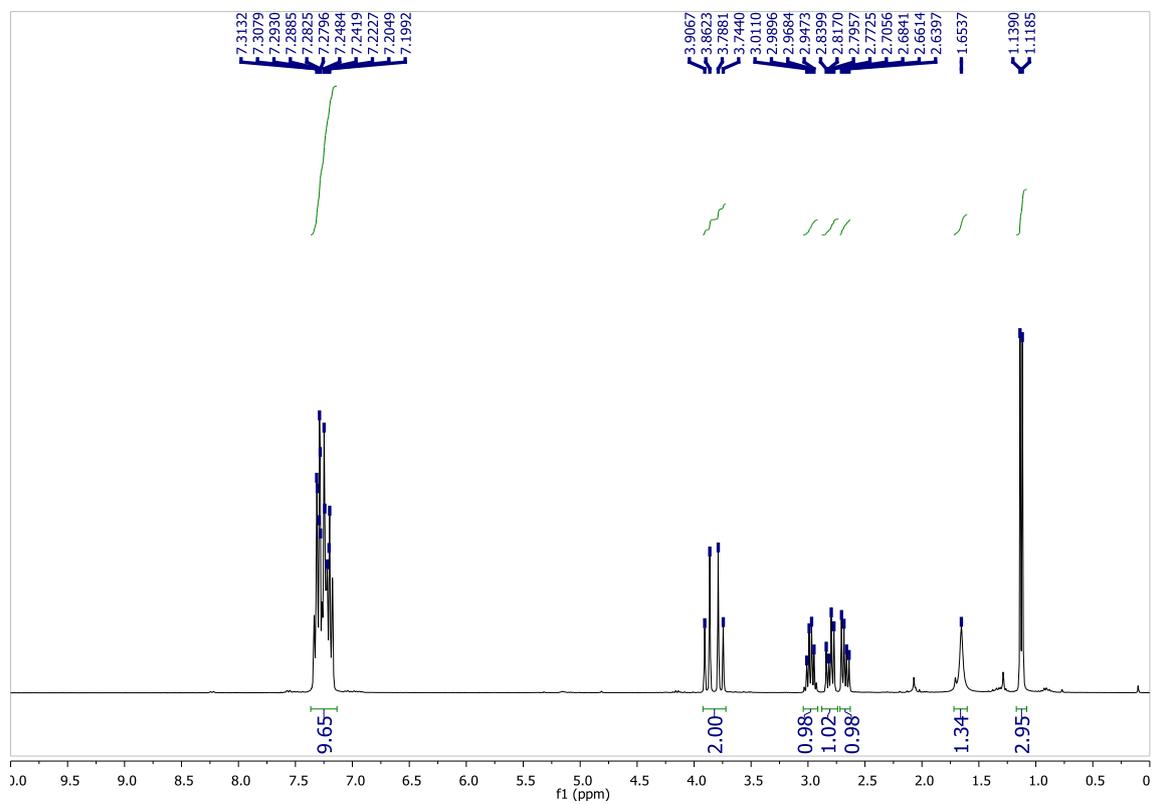


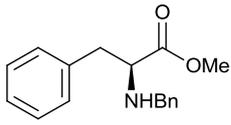
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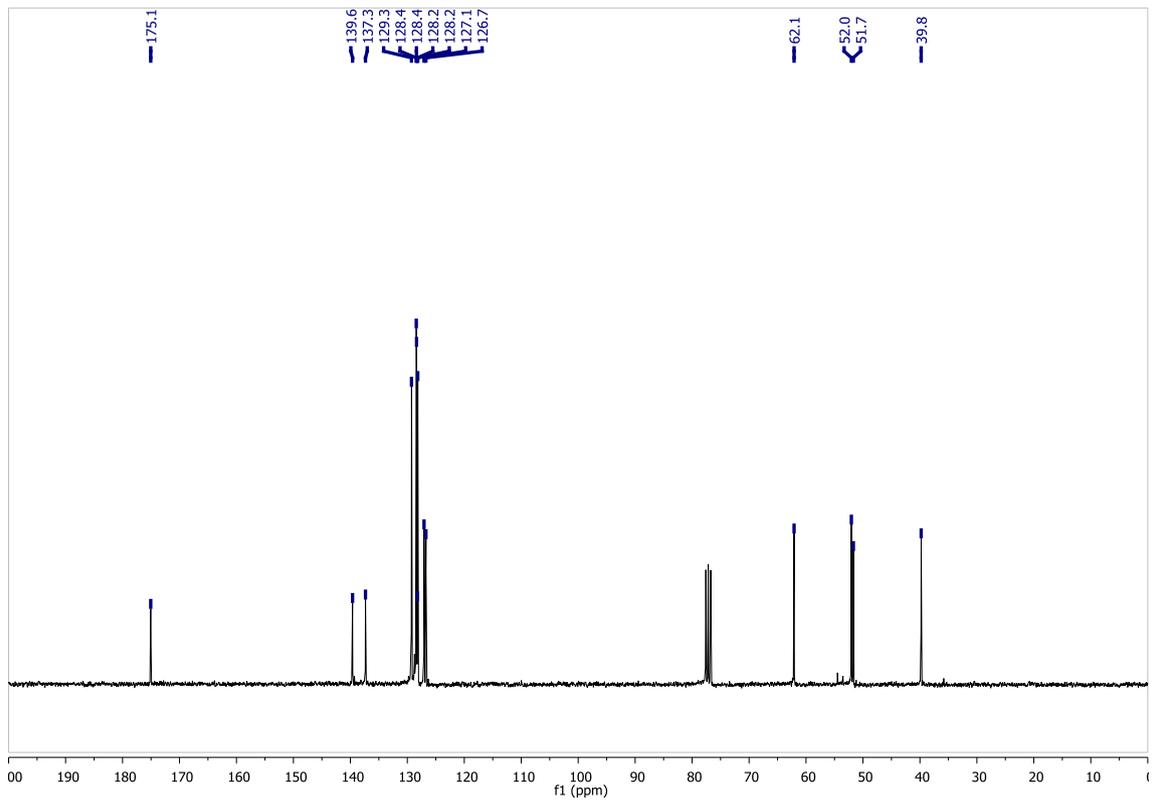
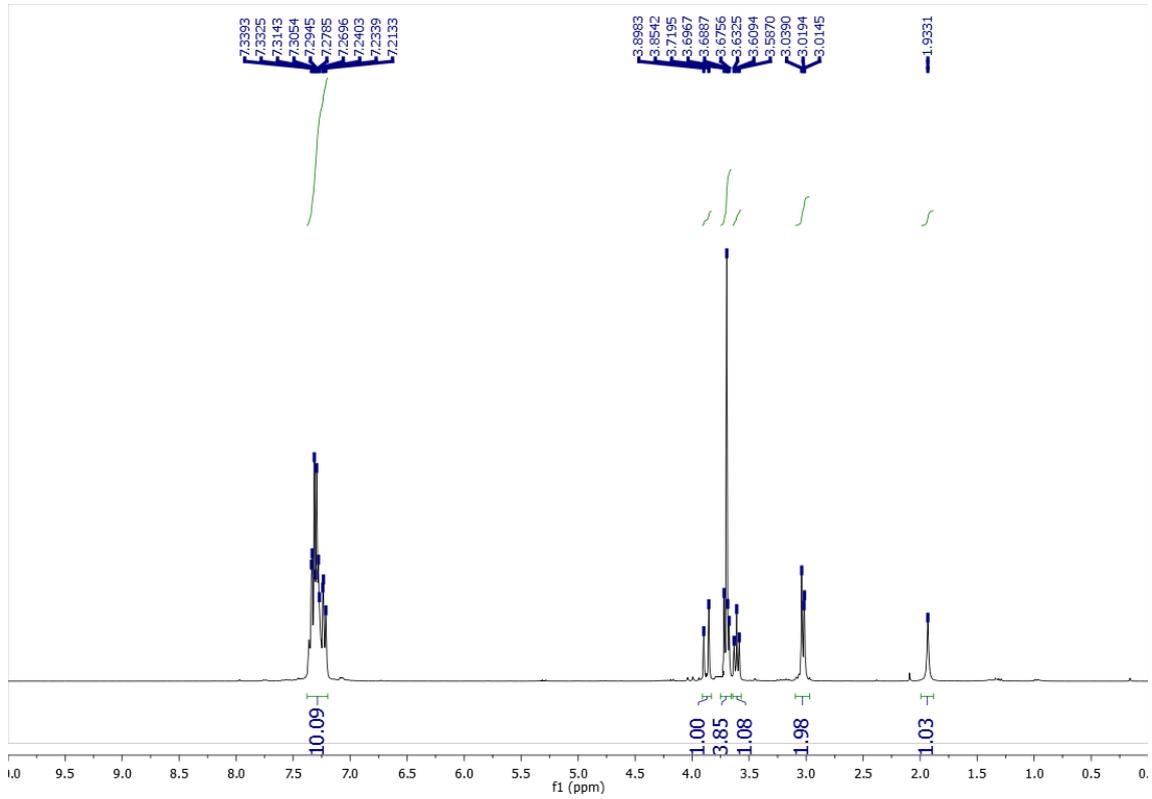


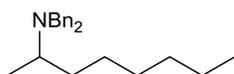
(±)-7a



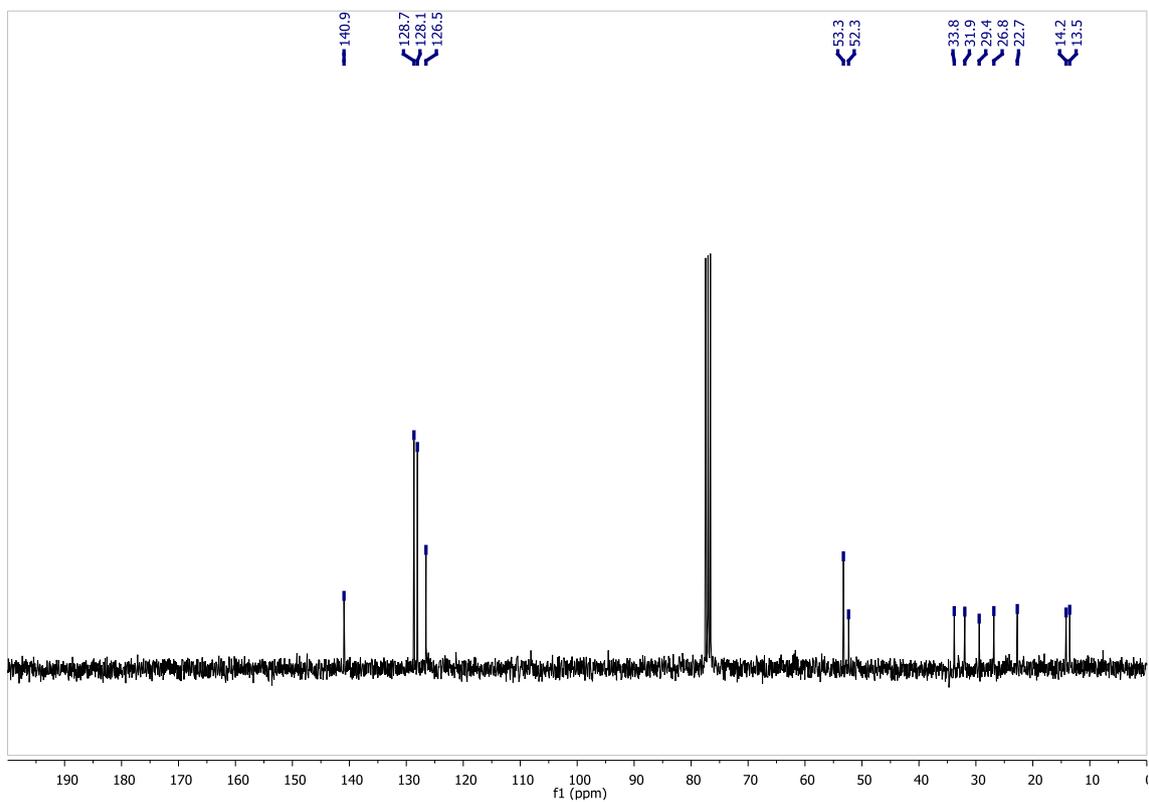
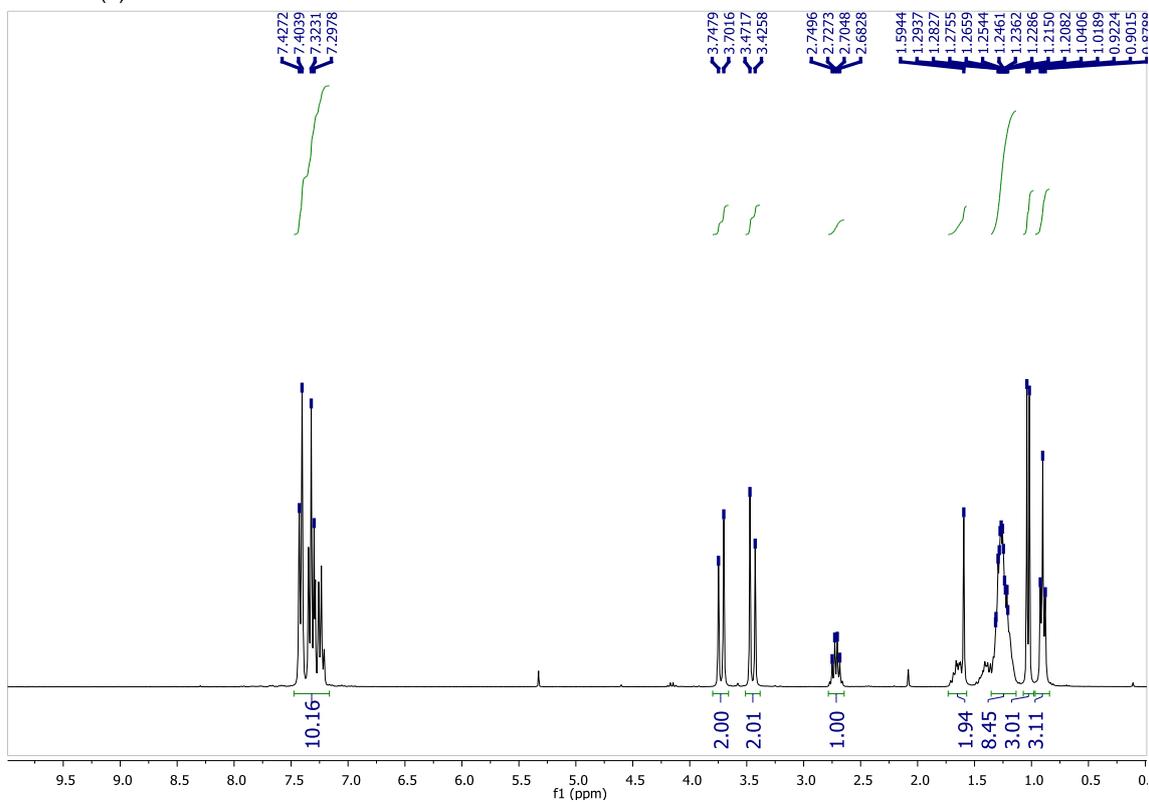


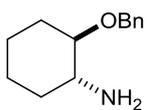
L-8a



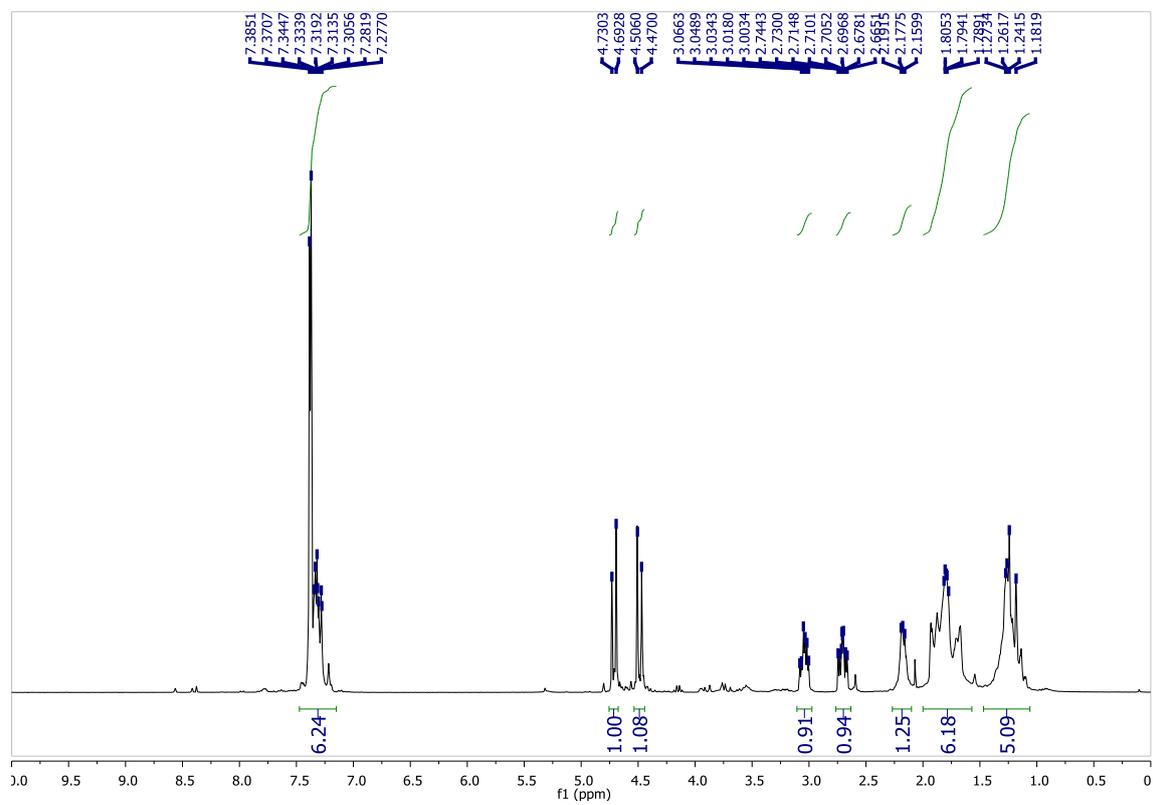


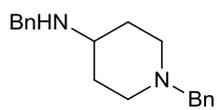
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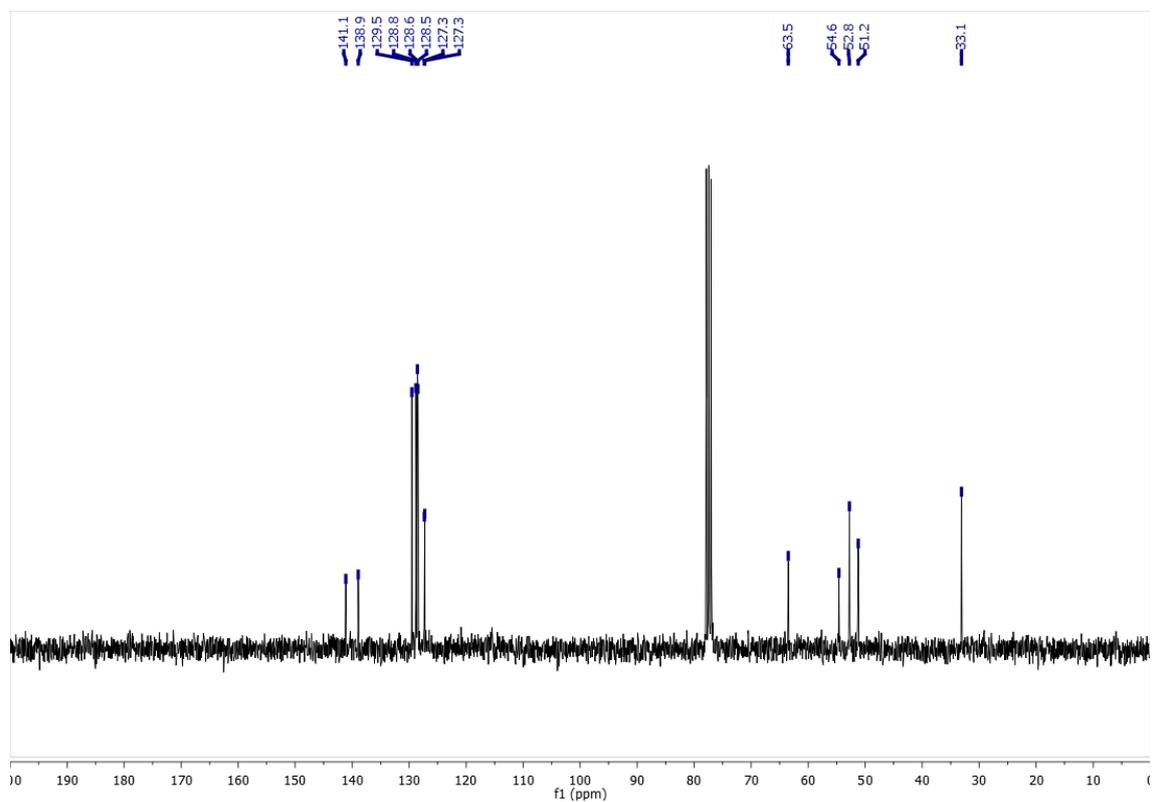
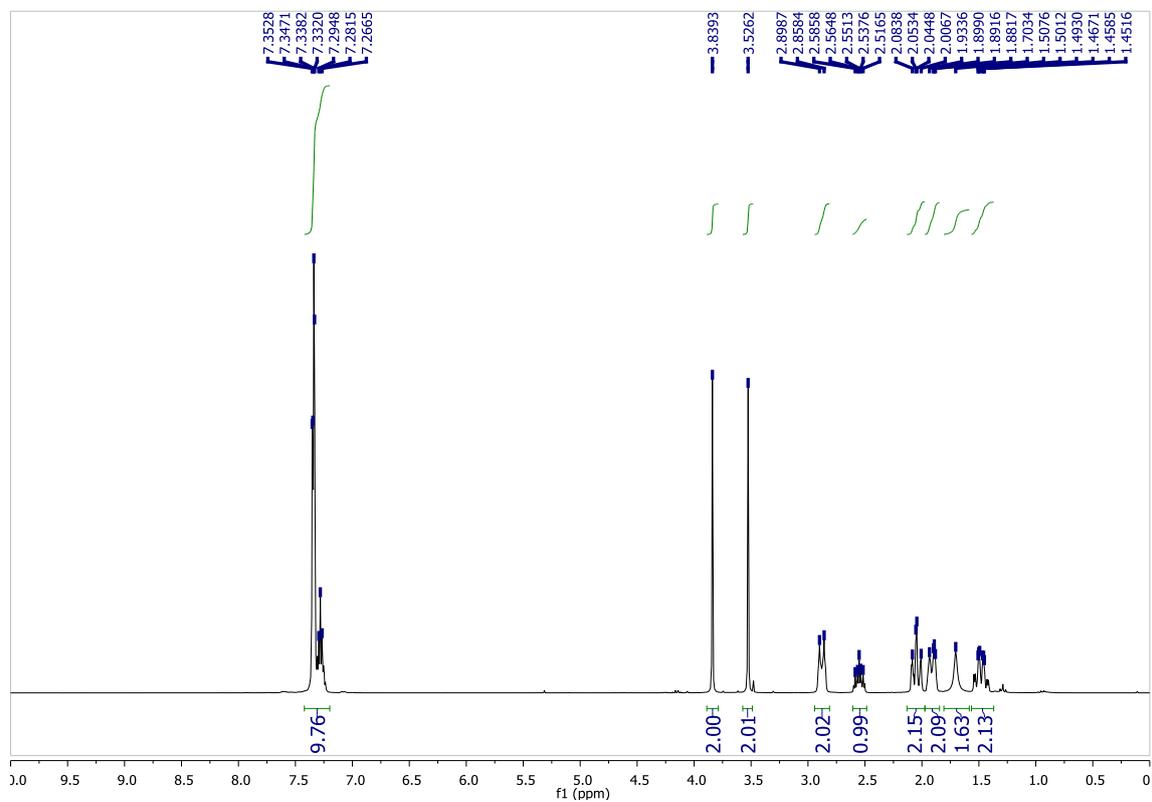


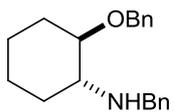
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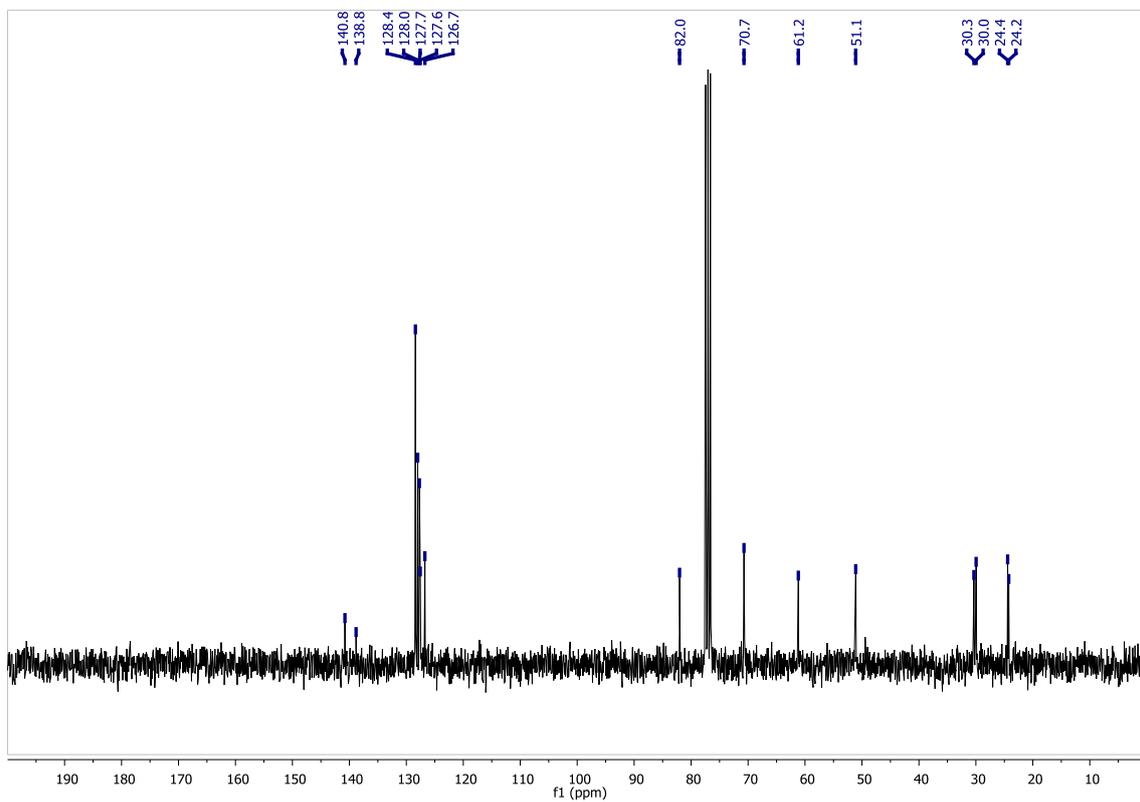
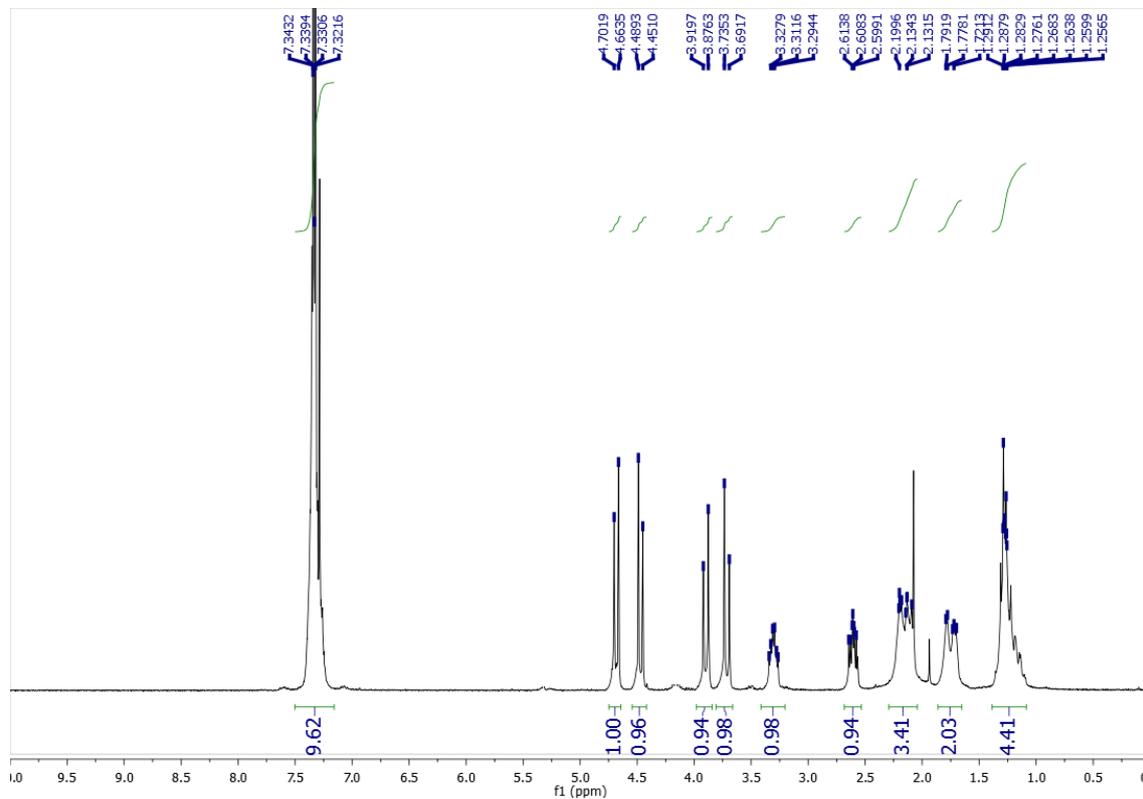


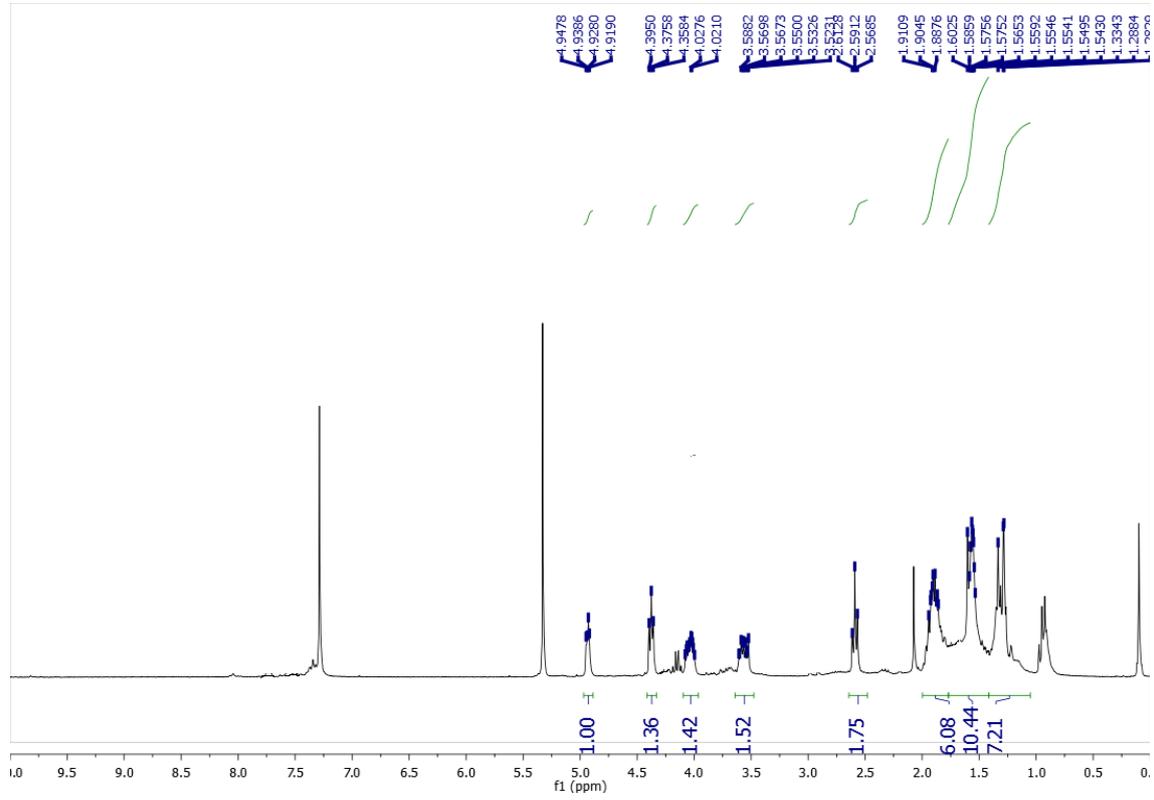
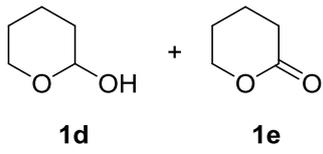
12a

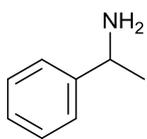




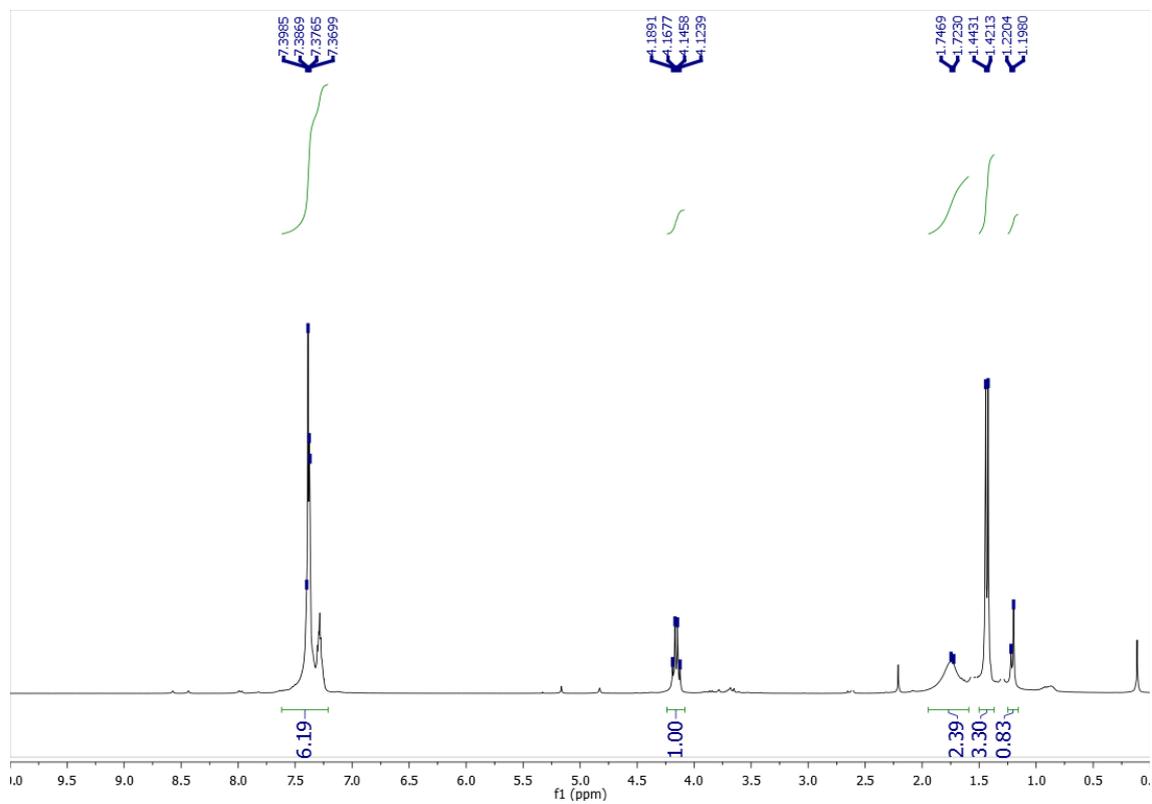
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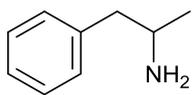




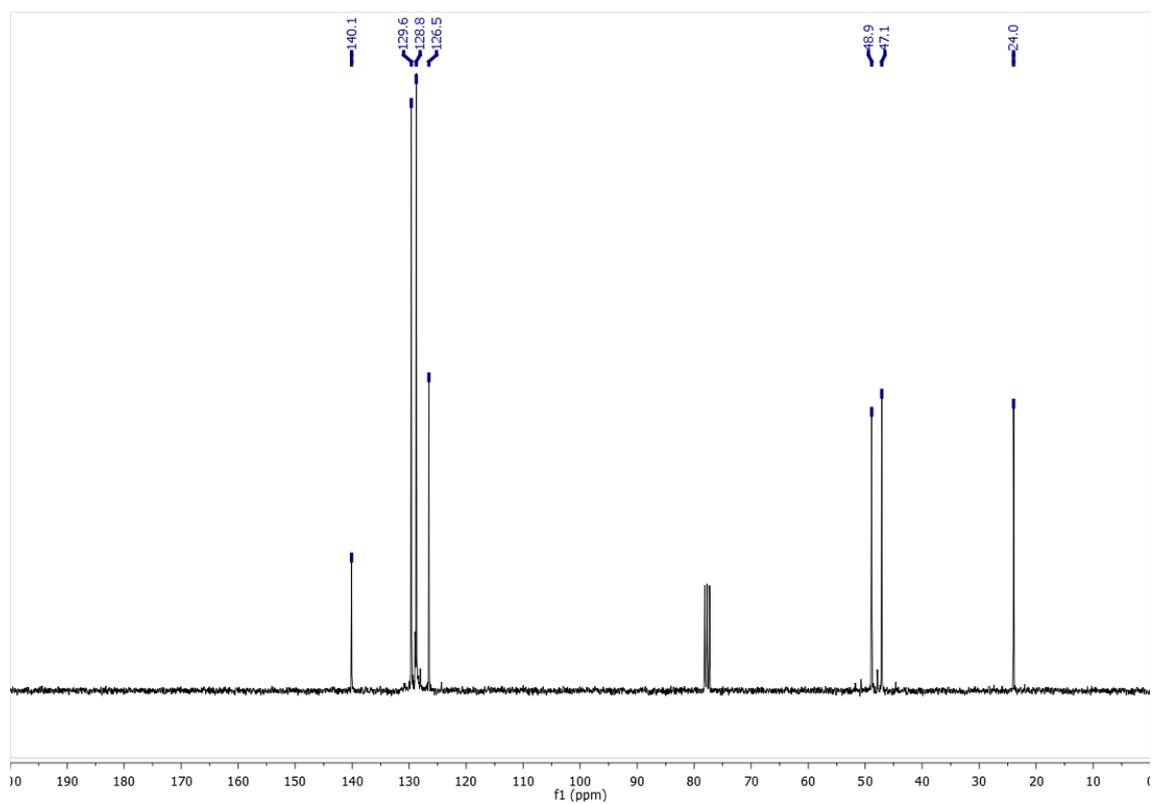
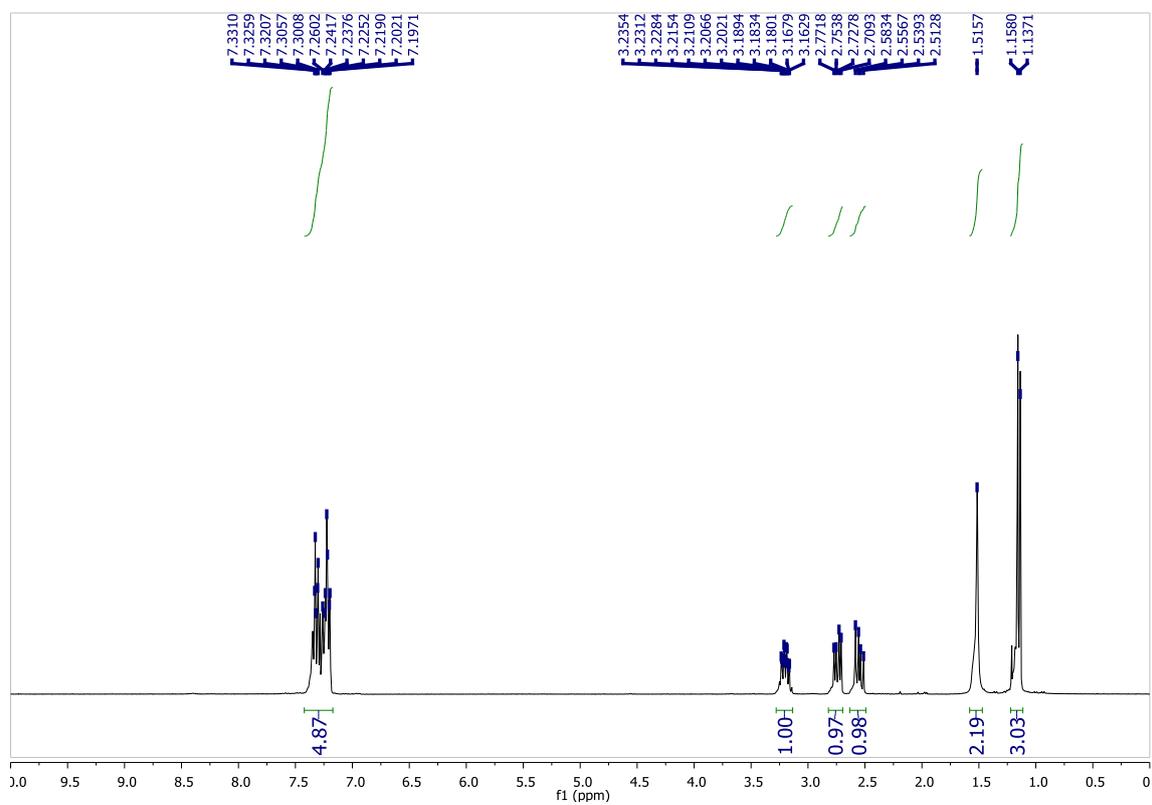


(±)-6b





(±)-7b



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