Electronic Supplementary Material (ESI) for Organic Chemistry Frontiers.

Supplementary Information

High-selective hydrogenation of aldehydes promoted by palladium-based catalyst and its application in equilibrium displacement in one-enzyme procedure using ω-transaminase

1. General information

All chemicals were commercially available and purchased Aladdin (Shanghai, China) and were used as received without further purification. All chemicals used are of analytical grade. ^{1}H- and ^{13}C-NMR of products were recorded on a Bruker 400 MHz spectrometer at 400 MHz and 100 MHz. All chemical shifts (δ) were quoted in parts per million (ppm) and reported relative to an internal tetramethylsilane (TMS, δ 0.00) standard. The following abbreviations were used to define the multiplicities: br, broad; s, singlet; d, doublet; t, triplet; q, quartet; p, pentet and m, multiplet. The conversions of products were measured by HPLC analysis equipped with a C18 column, and the ee values of the desired chiral amines were measured by HPLC analysis equipped with a coating Silica-gel Crownpak CR(+) (0.4 cm x 15 cm) column, both the conversions and ees were recorded with a Shimadzu Essentia Prep LC-16P HPLC instrument.

2. General procedure for the cloning, protein expression and purification of transaminase

The gene for Chromobacterium violaceum ω-transaminase was inserted in the plasmid pET28a(+) with an N-terminal His6-tag. After digestion (NheI, HindIII) and sequence verification the construct was transformed into *E. coli BL21 (DE)*. Expression was done by mixing a 2000 ml overnight culture with 180 ml of Luria-Bertoni (LB) medium with 50 mg/L Kanamycin and 0.4 mM IPTG and incubating for 24 h at 30°C (160 rpm, baffled flask). The cells were thereafter separated from the medium by centrifugation and resuspended in IMAC binding buffer (20 mM sodium phosphate, 500 mM sodium chloride, pH 8.2), disrupted by addition of BugBuster® 10X (Merck) and applied to a column with Chelating Sepharose FastFlow (GE Healthcare) resin treated with a saturated water solution of Cobalt(II) chloride. After washing with the binding buffer, the His6-tagged enzyme was eluted with IMAC elution buffer. An excess of cofactor (PLP) was added before desalting on a PD10 column (GE Healthcare). The simple procedure of adding PLP before buffer change ensures that the amount of cofactor is balanced with the enzyme concentration, assuming that a negligible amount of enzyme is in apo form during the desalting.

3. General procedure for chemo-selective hydrogenation of aldehydes

Ketone (2mM) and aldehyde (2mM), at equal equivalents were dissolved in 5mL H_2O, pH of the buffer (HEPES, 50mM) was made at 8.2, Pd-L8 (1.0 mmol %), were stirred at 30°C with H_2 filled in a balloon. The reaction conversions and yields were determined by HPLC analysis equipped with a C18 column. When the reaction was completed, the solution was extracted with saturated brine and ethyl acetate (3×10 ml). The organic phase was dried over anhydrous magnesium sulfate and concentrated in vacuo. The crude product was purified by column chromatography, and the pure products were characterized by NMR.

4. General procedure for the preparation of Pd-Ligands

The catalyst Pd-L1 - Pd-L9 were chemical synthesized referencing some recent publications[1,2].

5. Relative activity of enzymes exposed to air, H_2, O_2, N_2 or was sealed

2 mL of enzyme in HEPES buffer were filled in a reaction kit, air, molecular oxygen, molecular hydrogenation, or nitrogen filled in respective balloon were connected with the reaction kit and was well sealed, before this, each balloon and reaction kit were carefully tested to verify the airtightness. After incubated for 12h, the enzyme was combined with the well-prepared components ((S)-1-phenylethylamine, and pyruvate) to a final concentration of 2mM for (S)-1-phenylethylamine and 6mM for pyruvate. The initial reaction rates were measured spectrophotometrically. By detecting the consumption (reducing) of (S)-1-phenylethylamine at the wavelength of 270 nm. All continuous measurements were done on a dual beam spectrophotometer (Cary 300 UV-Vis, Varian Inc.) with appropriate blank correction.

6. General procedure for screening of amino donor

All components were dissolved separately in the buffer and the pH was corrected. These solutions were then combined to final concentration of 2 mM for acetophenone, and for 4-methylbenzylamine, benzylamine, 4-chlorobenzylamine, 3-chlorobenzylamine, 4-fluorobenzylamine was 10mM, the solutions were incubated in shaker for 24h at 30°C. ω-Transaminase of wild-type from *Chromobacterium violaceum* (Cv-ω-TA WT) was used as the model biocatalyst at the loading of 0.8U, pH of the reaction medium was set at 8.2 in HEPES buffer at 50mM on a 5 ml scale. After reactions were completed, samples were taken, treated and subject to HPLC to determine the yields of products (the desired chiral amines)

7. General procedure for the optimization of the loading of amino donor

Similar to the screening of amino donors, all the components were dissolved separately in the buffer and the pH was corrected. These solutions were then combined to final concentration of 2 mM for acetophenone, varied concentrations of amino donor from 1 to 10 equivalents were emloyed, Pd-L8 (1.0 mol %), were reacted in 30°C shaker for 24 h, Cv-ω-TA WT was used at 0.8U, after reacted for 24h, samples were taken and the yields of chiral amine were measured on a HPLC to determine the suitable loading of amino donors.

8. General procedure for one-enzyme cascades reactions using ω-TA

The reactions were performed in 50 mM HEPES buffer solution on a 5 ml scale, the pH of the buffer was 8.2 for ω-transaminases. All components were dissolved separately in the buffer and the pH was corrected. These solutions were then combined to final concentration of 2 mM acetophenone or other substituted-acetophenones (4-fluoroacetophenone, 3-

fluoroacetophenone, 4-methylacetophenone, 3-methylacetophenone), benzylamine and p-methylbenzylamine, 6mM, Pd-L8, 1.0 mol% , and transaminase (purified ω-transaminases, $[E]_{Cv\text{-}\omega\text{-}TA\ W60C}$ = 25.0 μM, ATA-117, and ω-TA-001 from Codexis, 0.8 U), were reacted in 30°C shaker for 24 h.

9. Determination of product yields

The reactions were performed as described in the text enzyme reactions. When reactions were completed, perchloric acid of 10 uL at the concentration of 70% was added into the reaction solution, and the mixed solution was centrifuged at 3000 r/min for 5 min. After that, 300 uL solution of the supernatant fluid was taken and the yield was measured using High Performance Liquid Chromatography (HPLC) equipped with a C18 column.

10. Determination of enantiomeric excess of chiral amines

The samples were subjected to High Performance Liquid Chromatography (HPLC) with a CrownpakCR(+) column (Daicel), after filtration (0.22 μm) and acidification by perchloric acid. High Performance Liquid Chromatography using ultrapure water with pH=1.0 and acetonitrile as mobile phase at the flow rate of 0.5 ml/min.

$$ee_S = \frac{[S]-[R]}{[S]+[R]} \quad or \quad ee_R = \frac{[R]-[S]}{[R]+[S]} \qquad \textbf{(Equ.1)}$$

11. General procedure for isolation of pure products

To obtain the desired pure chiral amines, reactions were performed on a 5 mL scale, reaction parameters were identical with which described in the main text of the manuscript. When reactions were completed, 100 uL perchloric (70%) was added into the reaction solution to acidate the solution, and the reaction solution was centrifuged for 5 min to remove the precipitate. The supernatant was added into with 5 mL ethyl acetate, mixed and extracted three times. The organic solution was isolated and combined together. The combined organic solution was then distilled under evacuation to remove the organic solvent, and the residue is subjected to thin layer chromatography to afford pure products. (Due to the small scales of the reactions, we combined the products of several runs to get higher concentration samples.)

12. ^{1}H NMR and ^{13}C NMR spectrum of alcohols and chiral amines

1.1 phenylmethanol

^{1}H NMR (400MHz, DMSO-d6): δ (ppm) 7.35-7.20(m, 5H); 5.24(t, J = 4.0Hz 1H); 4.52(d, J = 4.0Hz, 2H); ^{13}C NMR (100MHz, DMSO-d6): δ (ppm) 143.01, 128.52, 127.11, 126.92, 63.47.

1.2 *p*-tolylmethanol

^{1}H NMR (400MHz, DMSO-d6): δ (ppm) 7.20 (d, J = 8.0Hz, 2H); 7.12 (d, J = 8.0Hz, 2H); 5.10 (q, J = 4.0Hz, 1H); 4.46 (d, J = 4.0Hz, 2H); 2.27 (s, 3H); ^{13}C NMR (100MHz, DMSO-d6): δ (ppm) 139.98, 136.05, 129.05, 126.96, 63.26, 21.17.

1.3 *m*-tolylmethanol

^{1}H NMR (400MHz, $CDCl_3$): δ (ppm) 7.22-7.05 (m, 4H); 4.53 (s, 2H); 2.85 (brs, 1H); 2.31 (s, 3H); ^{13}C NMR (100MHz, $CDCl_3$): δ (ppm) 140.92, 138.18, 128.46, 128.32, 127.81, 124.10, 65.08, 21.41.

1.4 (*p*-fluorophenyl)methanol

^{1}H NMR (400MHz, DMSO-d6): δ (ppm) 7.39-7.36 (m, 2H); 7.16-7.11 (m, 2H); 5.25 (brs, 1H); 4.52 (s, 2H); ^{13}C NMR (100MHz, DMSO-d6): δ (ppm) 162.63, 139.10, 128.79, 115.14, 62.73.

1.5 *p*-cyanobenzyl alcohol

^{1}H NMR (400 MHz, DMSO-d6) δ 7.79 (d, J=8.0Hz, 2H), 7.53 (d, J=8.0Hz, 2H) 5.49 (t, J=4.0Hz, 1H), 4.61 (d, J=4.0Hz, 2H)；^{13}C NMR (100MHz, DMSO-d6) δ 148.97, 132.50, 127.37, 119.50, 109.78, 62.67.

1.6 (*p*-methoxyphenyl)methanol

^{1}H NMR (400MHz, DMSO-d6): δ (ppm) 7.25 (d, J = 8.0Hz, 2H); 6.88 (d, J = 8.0Hz, 2H); 5.09 (t, J = 4.0Hz, 1H); 4.44 (d, J = 4.0Hz, 2H); 3.72(s, 3H); ^{13}C NMR (100MHz, DMSO-d6): δ (ppm) 158.67, 134.99, 128.41, 113.90, 63.11, 55.40.

1.7 *m*-hydroxybenzyl alcohol

^{1}H NMR (400MHz, DMSO-d6): δ (ppm) 9.28 (brs, 1H); 7.10 (t, J = 8.0Hz, 1H); 6.76-6.61 (m, 3H); 5.11 (brs, 1H); 4.42 (s, 2H); ^{13}C NMR (100MHz, DMSO-d6): δ (ppm) 157.72, 144.54, 129.45, 117.39, 113.99, 113.72, 63.33.

1.8 (3,4,5-trimethoxyphenyl)methanol

^{1}H NMR (400MHz, DMSO-d6): δ (ppm) 6.65 (s, 2H); 5.20 (t, J = 4.0Hz, 1H); 4.46 (d, J = 4.0Hz, 2H); 3.78 (s, 6H); 3.66 (s, 3H); ^{13}C NMR (100MHz DMSO-d6): δ (ppm) 153.20, 138.78, 136.56, 103.86, 63.50, 60.39, 56.10.

1.9 (*S*)- or (*R*)- 1-phenylethanamine

^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 7.29-7.21 (m, 4H), 7.20-7.16 (m, 1H), 4.04 (q, J = 8.0 Hz, 1H), 1.66 (brs, 2H), 1.33 (d, J = 4 Hz, 3H). ^{13}C NMR (100 MHz, $CDCl_3$) δ(ppm) 147.79, 128.49, 126.81, 125.71, 51.31, 25.70.

1.10 (*S*)- or (*R*)- 1-(4-flurophenyl)ethylamine

^{1}H NMR (400MHz, $CDCl_3$) δ (ppm) 7.37-6.99 (m, 4H), 4.02 (q, *J* = 8.0 Hz, 1H), 1.92 (brs, 2H), 1.26 (d, *J* = 4.0 Hz, 3H). ^{13}C NMR (100 MHz, $CDCl_3$) δ(ppm) 161.69, 143.36, 127.25, 127.18, 115.23, 115.02, 50.65, 25.81.

1.11 (*S*)- or (*R*)- 1-(3-flurophenyl)ethylamine

^{1}H NMR (400MHz, DMSO-d6) δ (ppm) 7.33-7.28 (m, 2H), 7.02-6.96 (m, 2H), 4.10 (q, *J* = 8.0 Hz, 1H), 1.63 (brs, 2H), 1.34 (d, *J* = 4 Hz, 3H). ^{13}C NMR (100 MHz, DMSO-d6) δ(ppm) 162.76, 152.63, 130.28, 122.34, 113.21, 112.85, 50.73, 26.51.

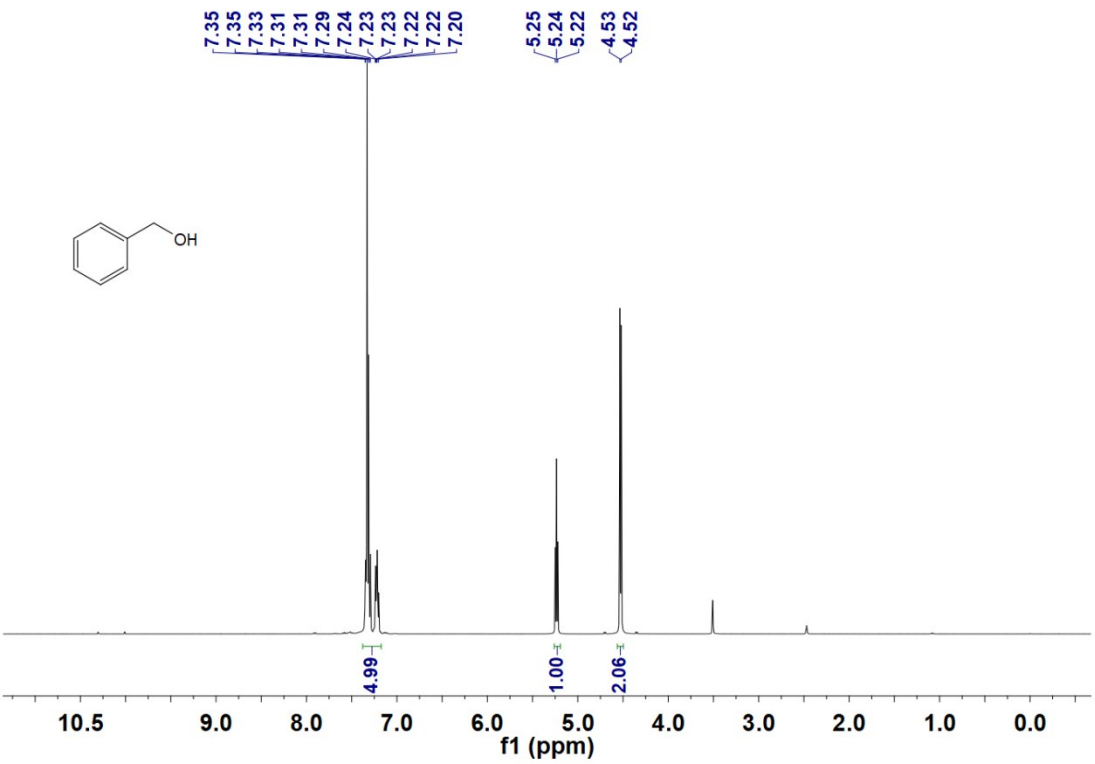
1.12 (*S*)- or (*R*)- 1-(4-methylphenyl)ethylamine

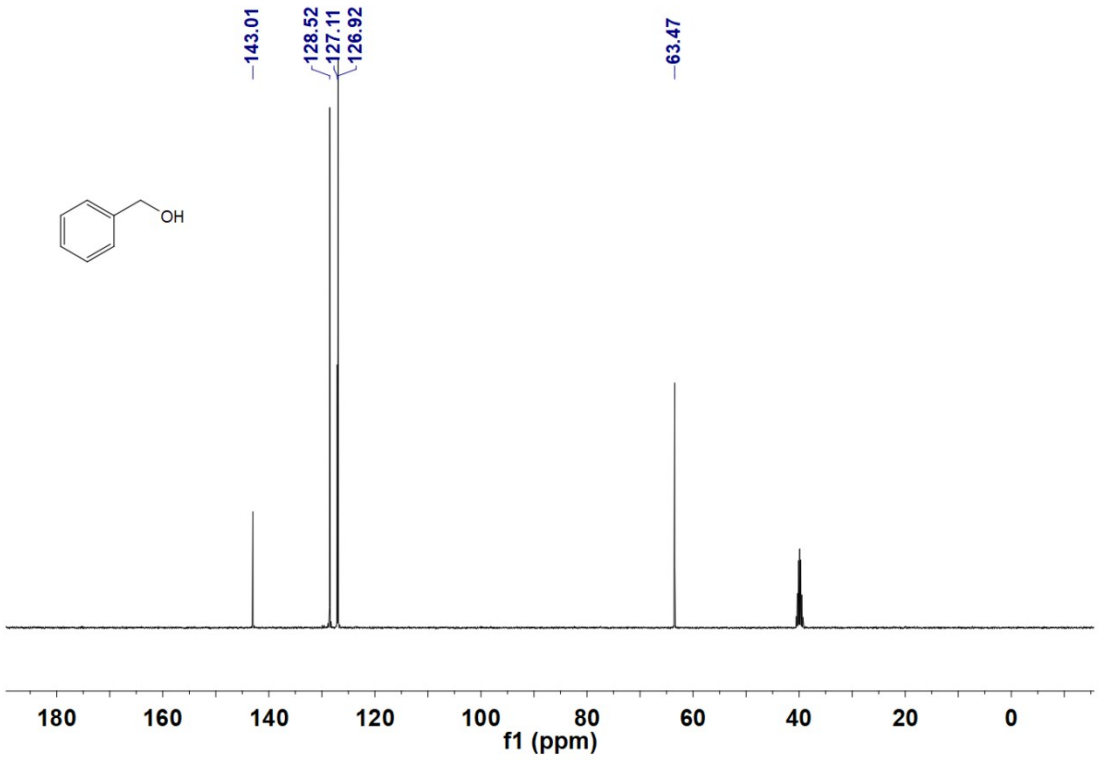
^{1}H NMR (400 MHz, $CDCl_3$) δ(ppm) 7.23-7.21 (d, 2H), 7.14-7.12 (d, 2H), 4.06 (q, *J* = 8.0 Hz, 1H), 2.32 (s, 3H), 1.77 (brs, 2H), 1.36 (d, *J* = 8 Hz, 3H). ^{13}C NMR (100 MHz, $CDCl_3$) δ(ppm) 144.76, 136.37, 129.17, 125.61, 51.04, 25.63, 21.03 .

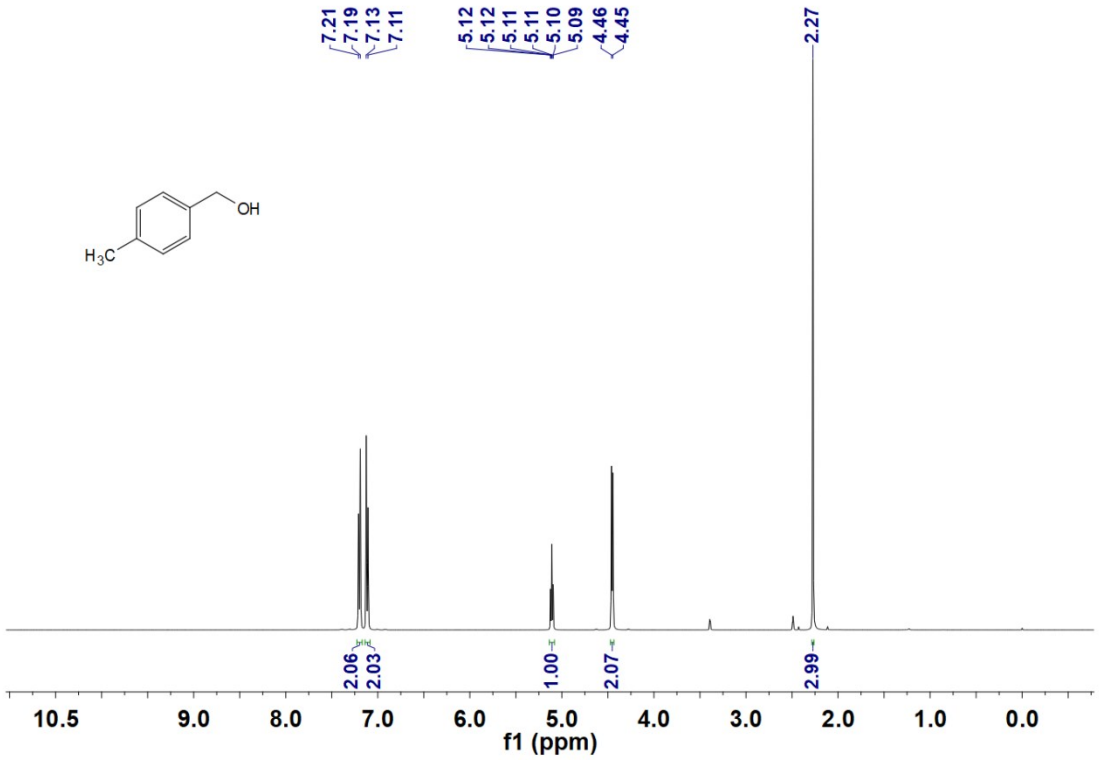
1.13 (*S*)- or (*R*)- 1-(3-methylphenyl)ethylamine

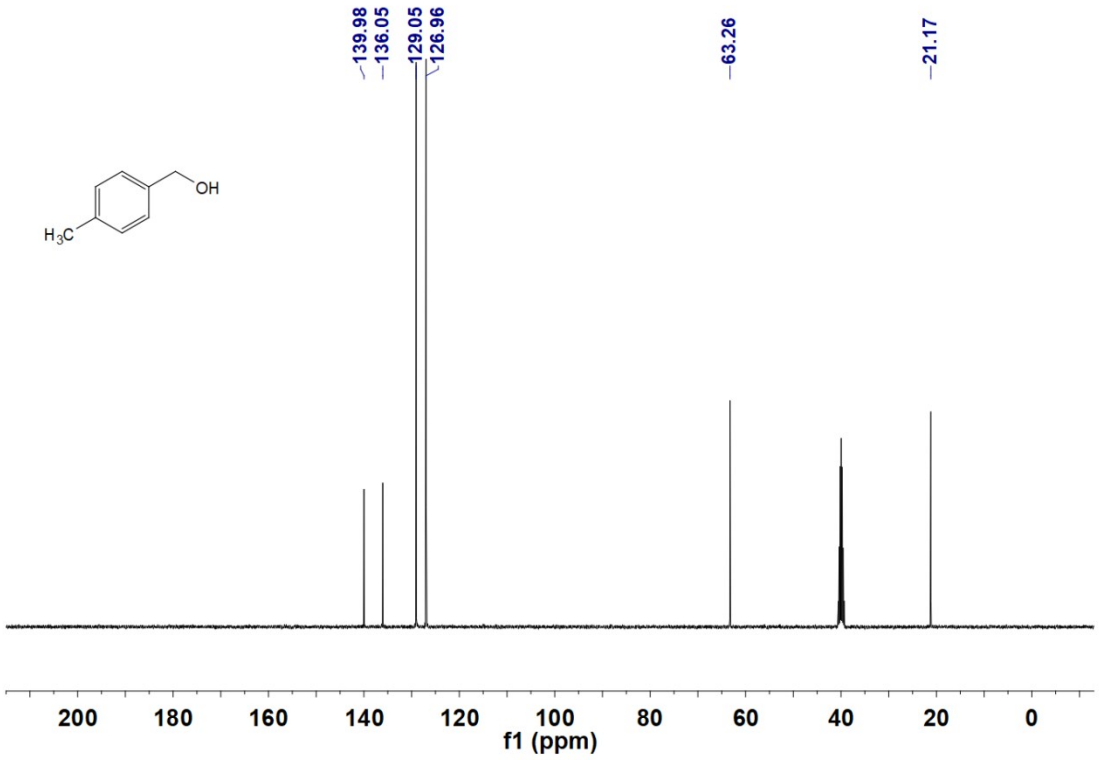
^{1}H NMR (400 MHz, $CDCl_3$) δ(ppm) 7.26-7.04 (m, 4H), 4.07 (q, *J* = 8.0 Hz, 1H), 2.35 (s, 3H), 1.82 (brs, 2H), 1.38 (d, *J* = 8 Hz, 3H). ^{13}C NMR (100 MHz, $CDCl_3$) δ(ppm) 147.61, 138.11, 128.42, 127.59, 126.44, 122.73, 51.30, 25.55, 21.47.

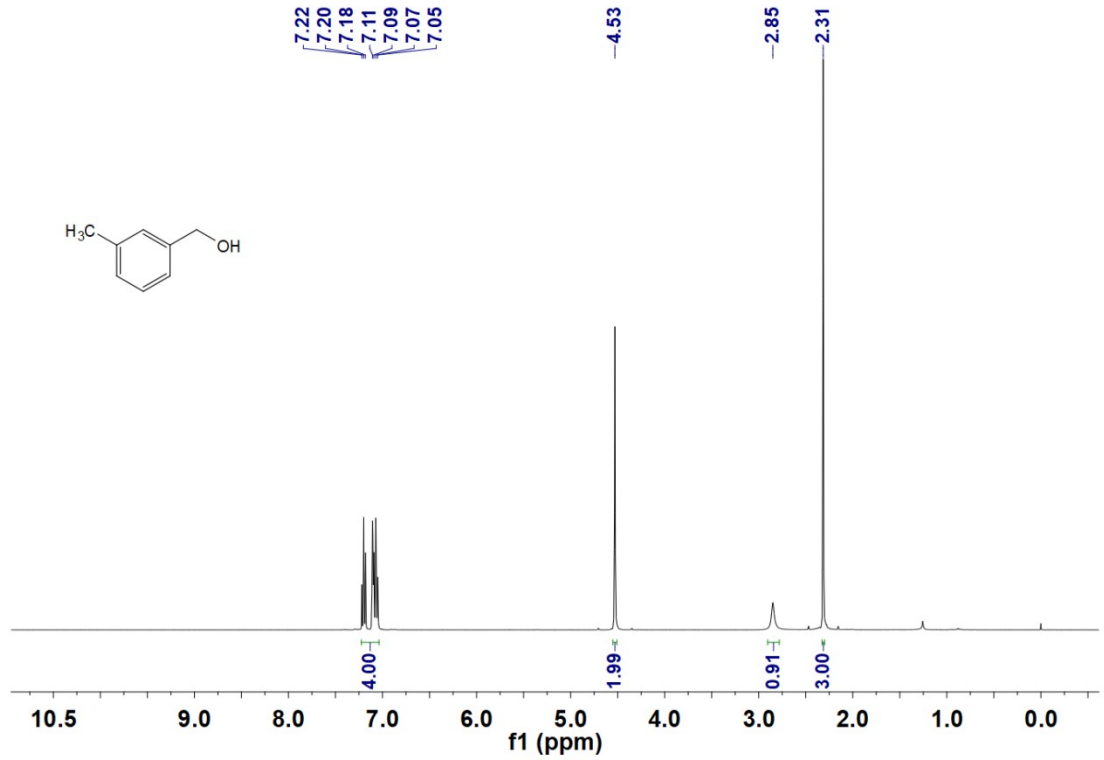
13. *^{1}H NMR and ^{13}C NMR spectra of product alcohols and chiral amines*

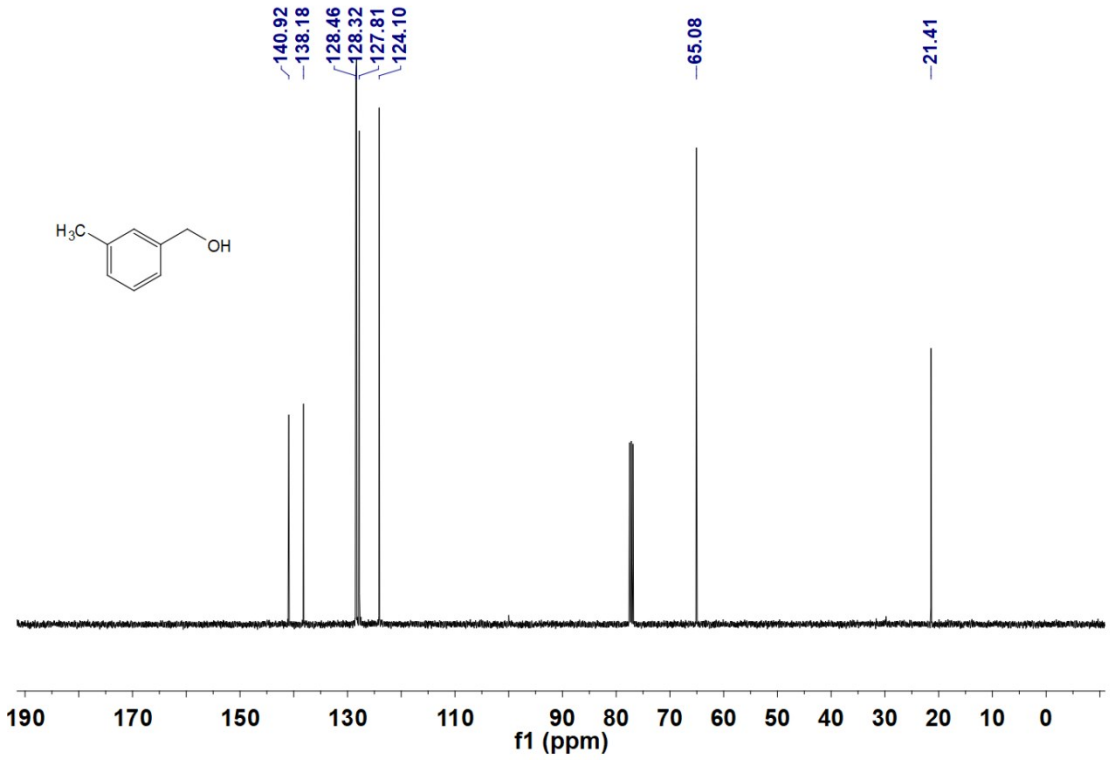

7.35
7.35
7.33
7.31
7.31
7.29
7.24
7.23
7.23
7.22
7.22
7.20
5.25
5.24
5.22
4.53
4.52
OH
4.99
1.00
2.06
10.5
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)

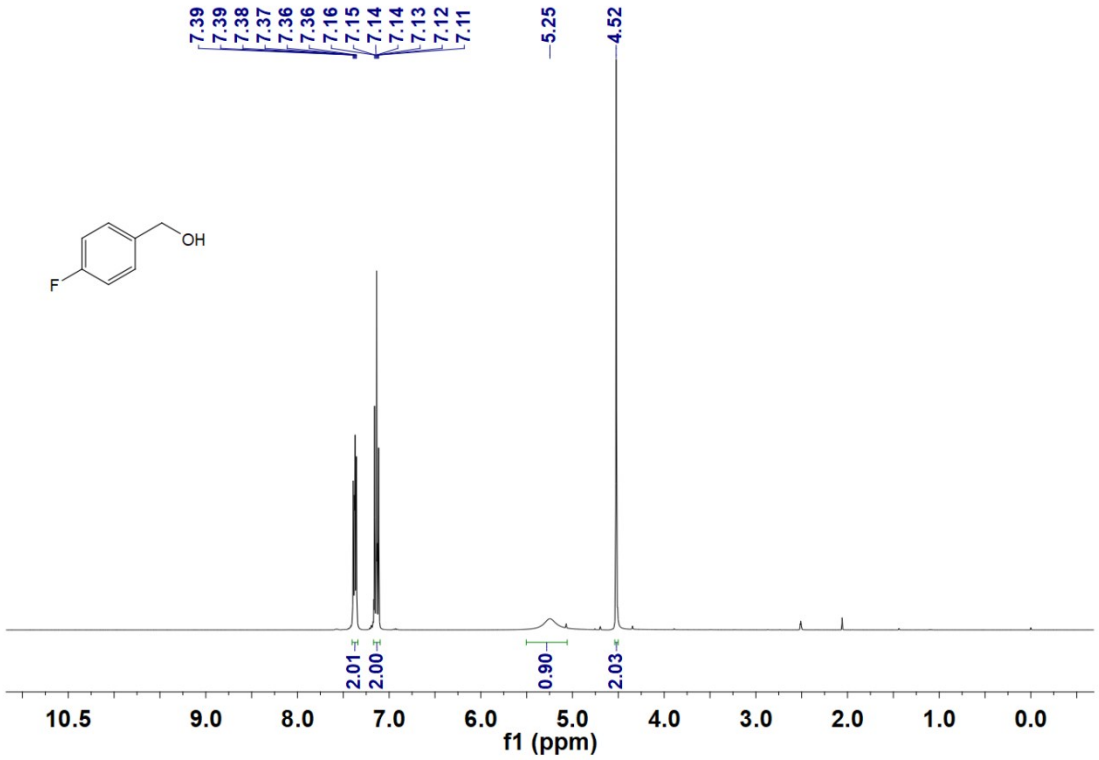

143.01
128.52
127.11
126.92
63.47
OH
180
160
140
120
100
80
60
40
20
0
f1 (ppm)

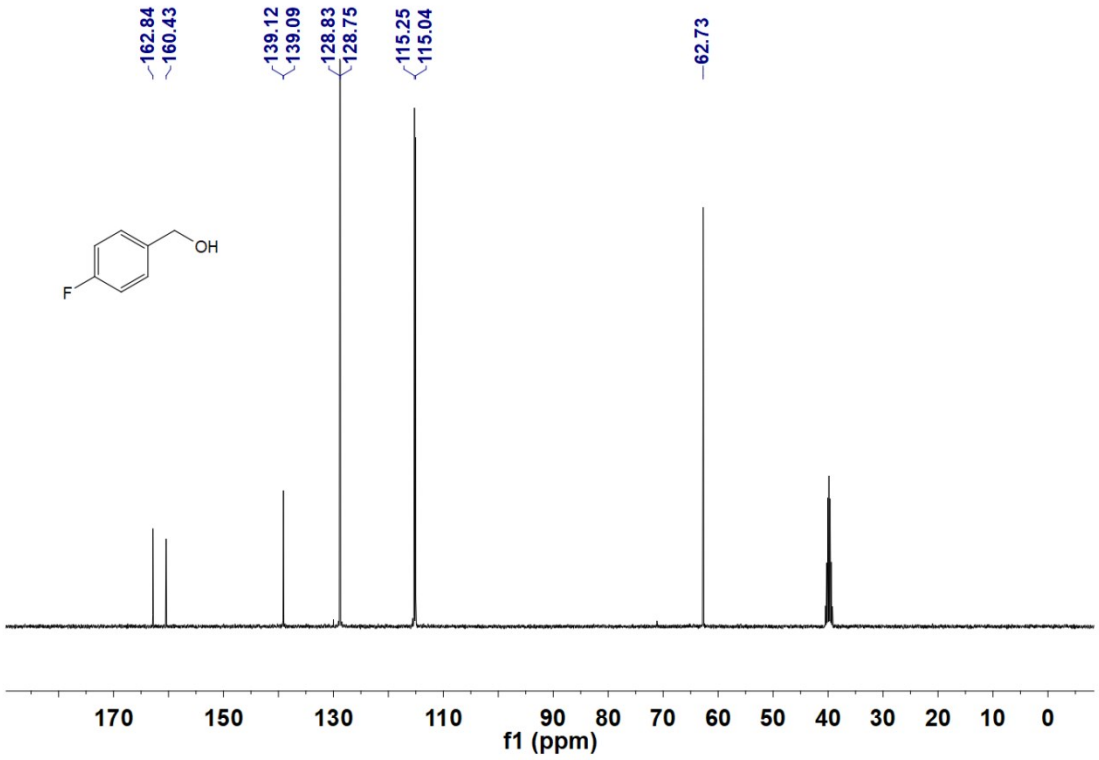

7.21
7.19
7.13
7.11
5.12
5.12
5.11
5.11
5.10
5.09
4.46
4.45
2.27
OH
H3C
2.06
2.03
1.00
2.07
2.99
10.5
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)

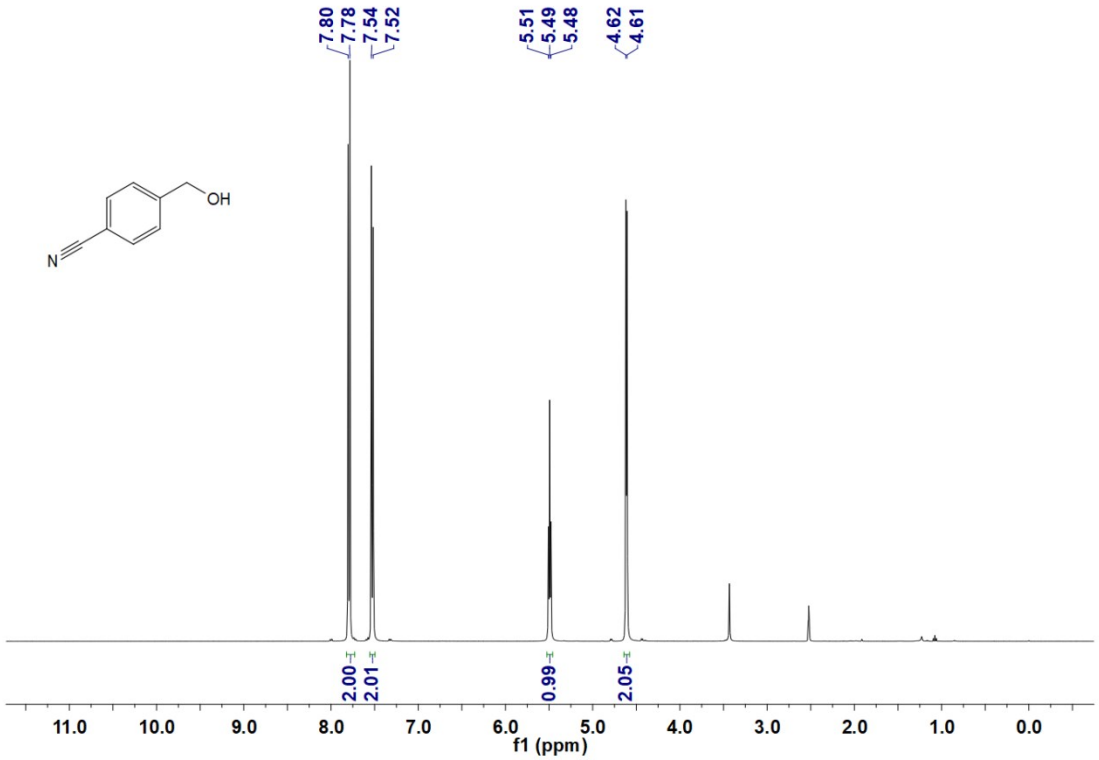

139.98
136.05
129.05
126.96
63.26
21.17
OH
H3C
200
180
160
140
120
100
80
60
40
20
0
f1 (ppm)

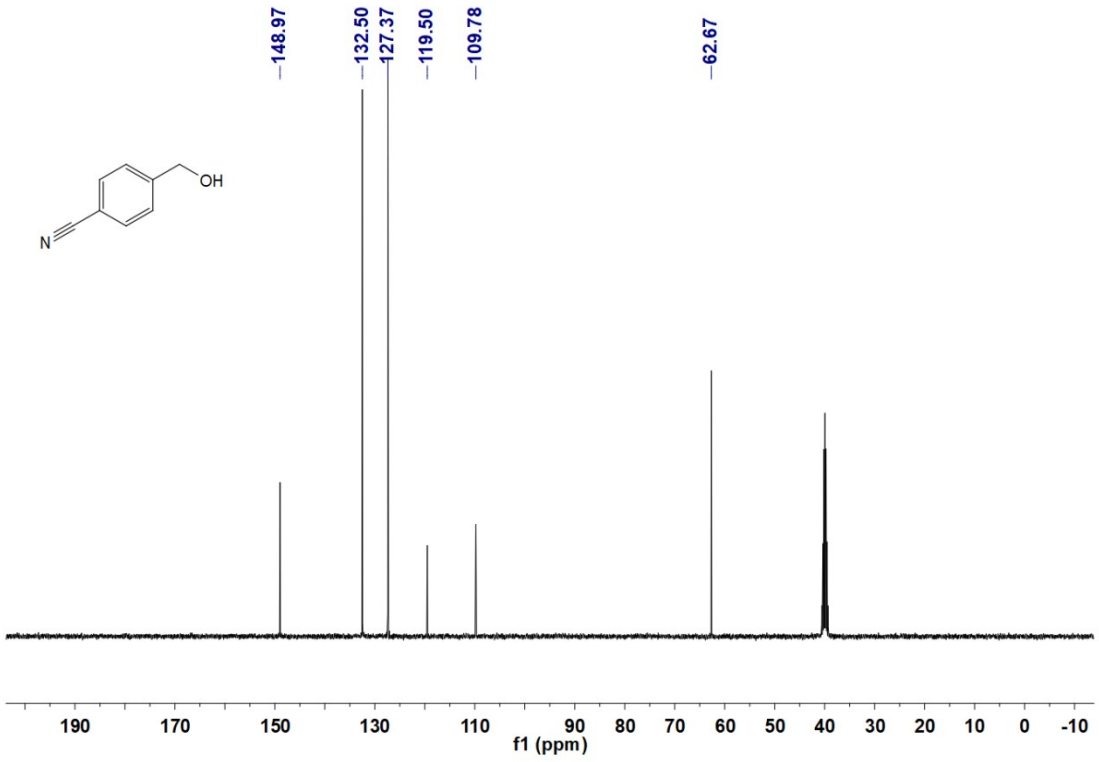

7.22
7.20
7.18
7.11
7.09
7.07
7.05
4.53
2.85
2.31
H3C
OH
4.00
1.99
0.91
3.00
10.5
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)

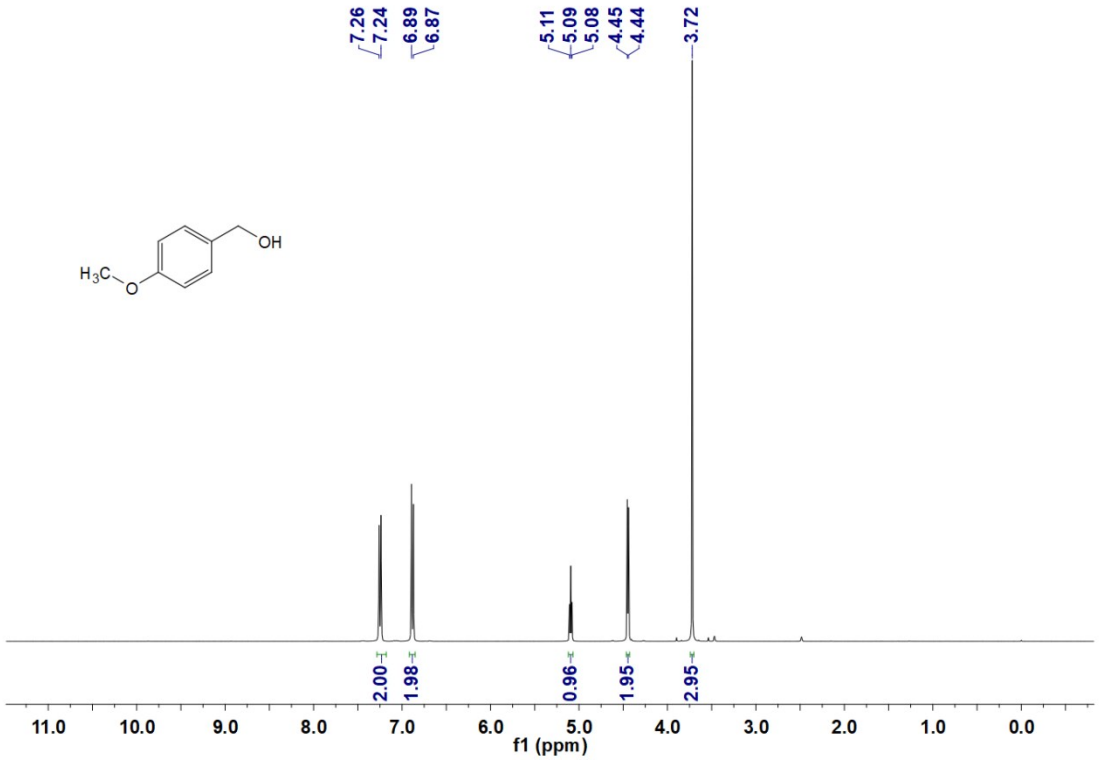

140.92
138.18
128.46
128.32
127.81
124.10
65.08
21.41
H3C
OH
190
170
150
130
110
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

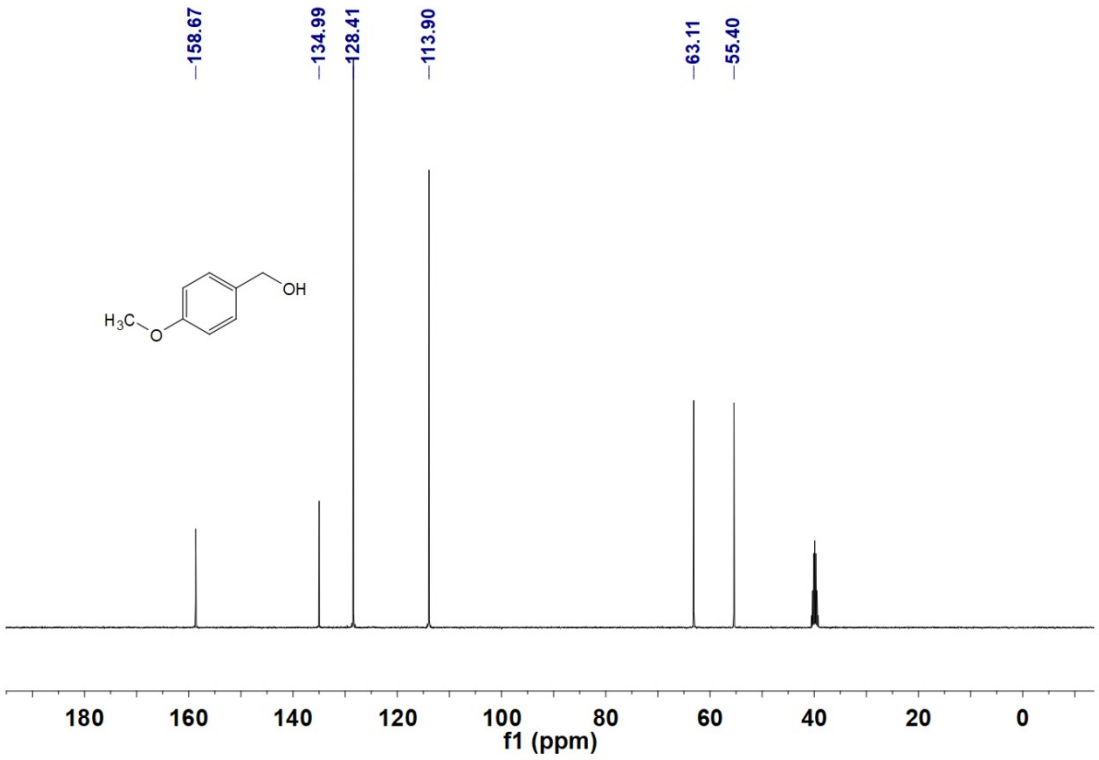

7.39
7.39
7.38
7.37
7.36
7.36
7.16
7.15
7.14
7.14
7.13
7.12
7.11
5.25
4.52
OH
F
2.01
2.00
0.90
2.03
10.5
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)

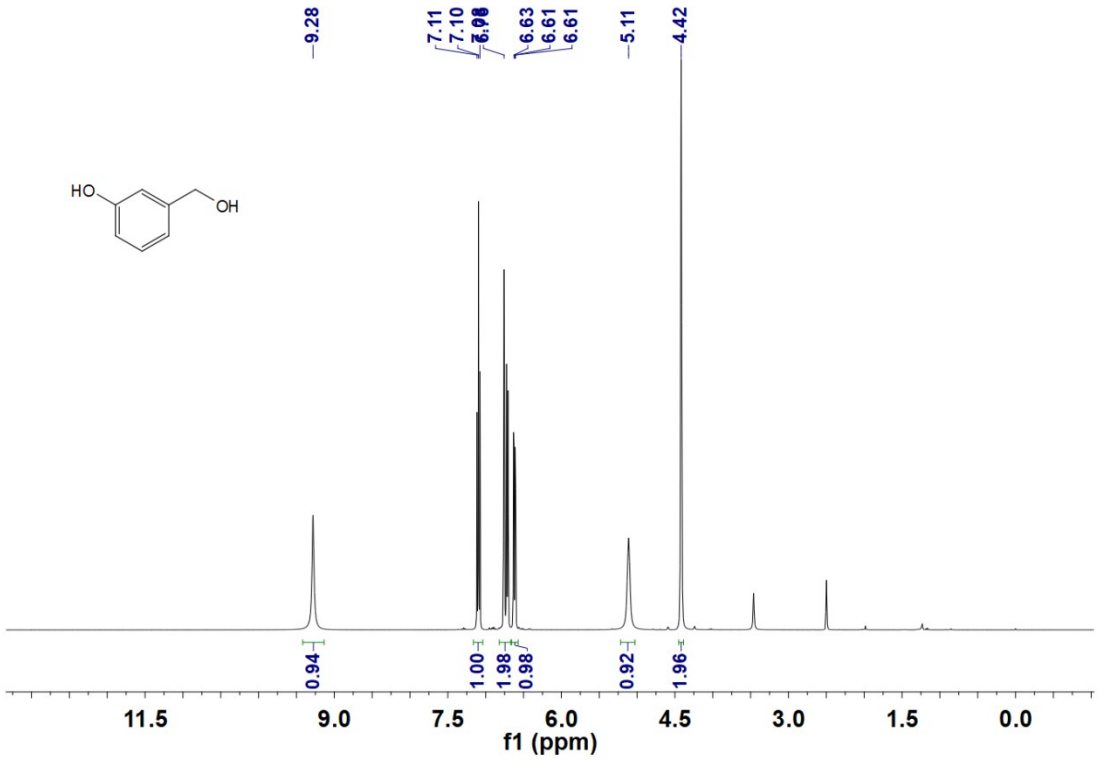

162.84
160.43
139.12
139.09
128.83
128.75
115.25
115.04
62.73
OH
F
170
150
130
110
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

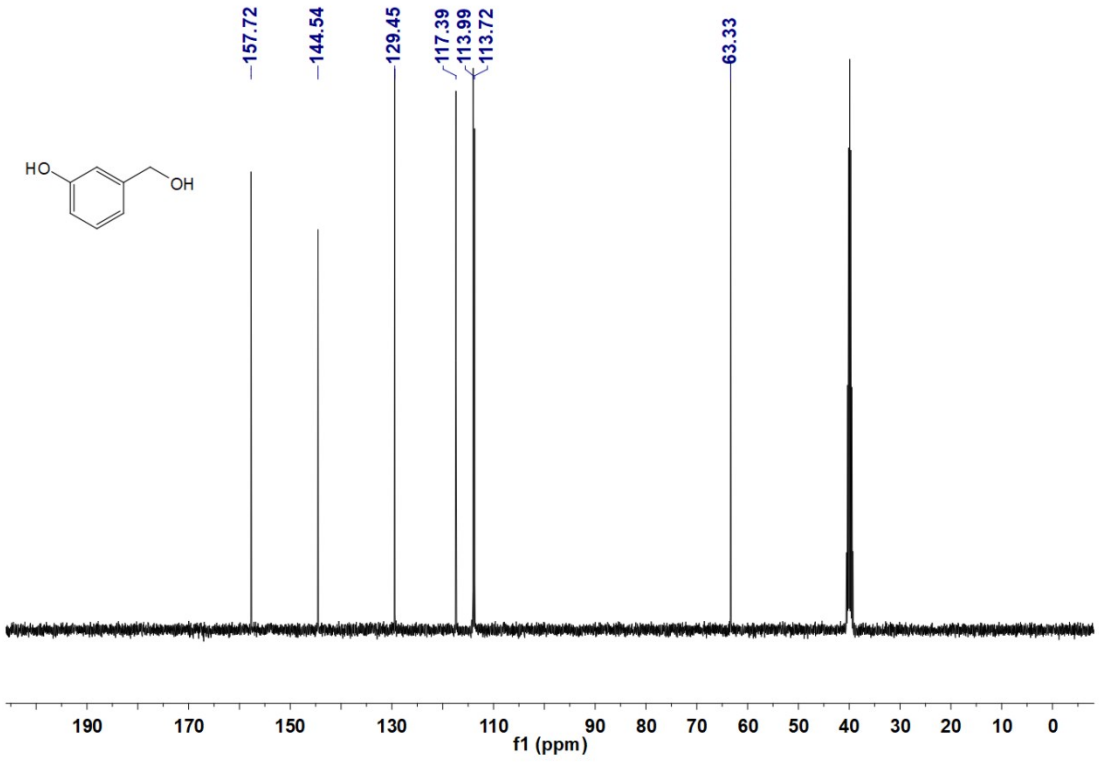

7.80
7.78
7.54
7.52
5.51
5.49
5.48
4.62
4.61
OH
N
2.00
2.01
0.99
2.05
11.0
10.0
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)

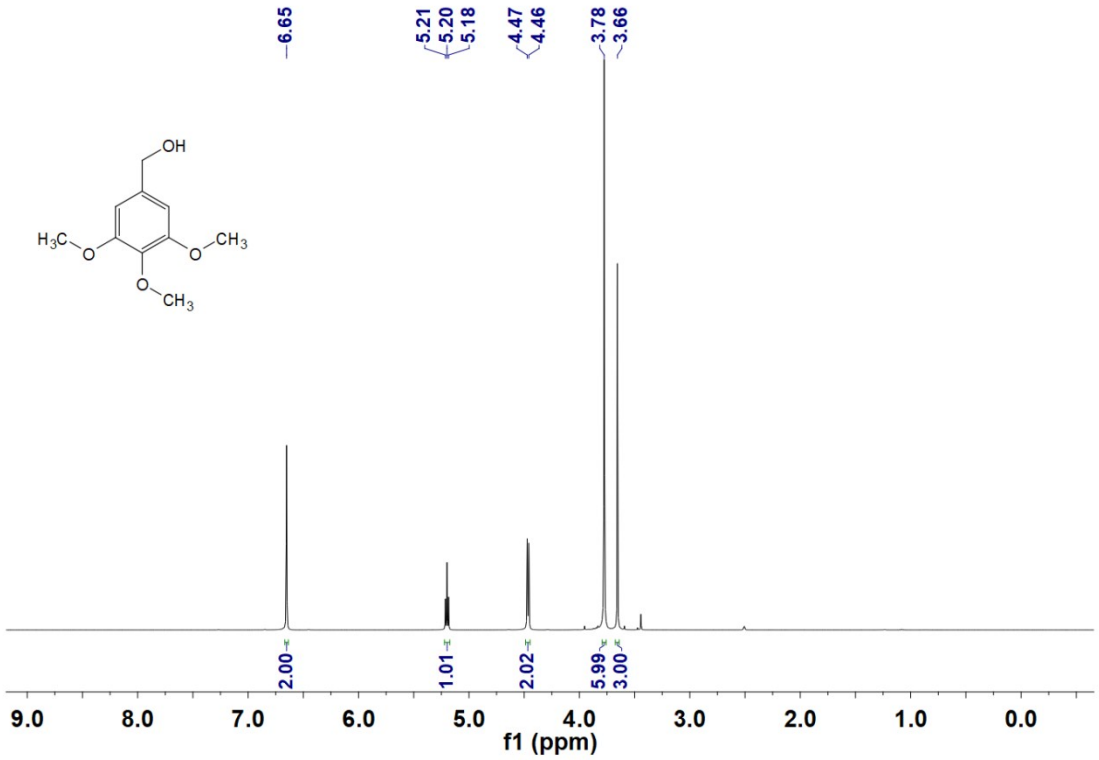

148.97
132.50
127.37
119.50
109.78
62.67
OH
N
190
170
150
130
110
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

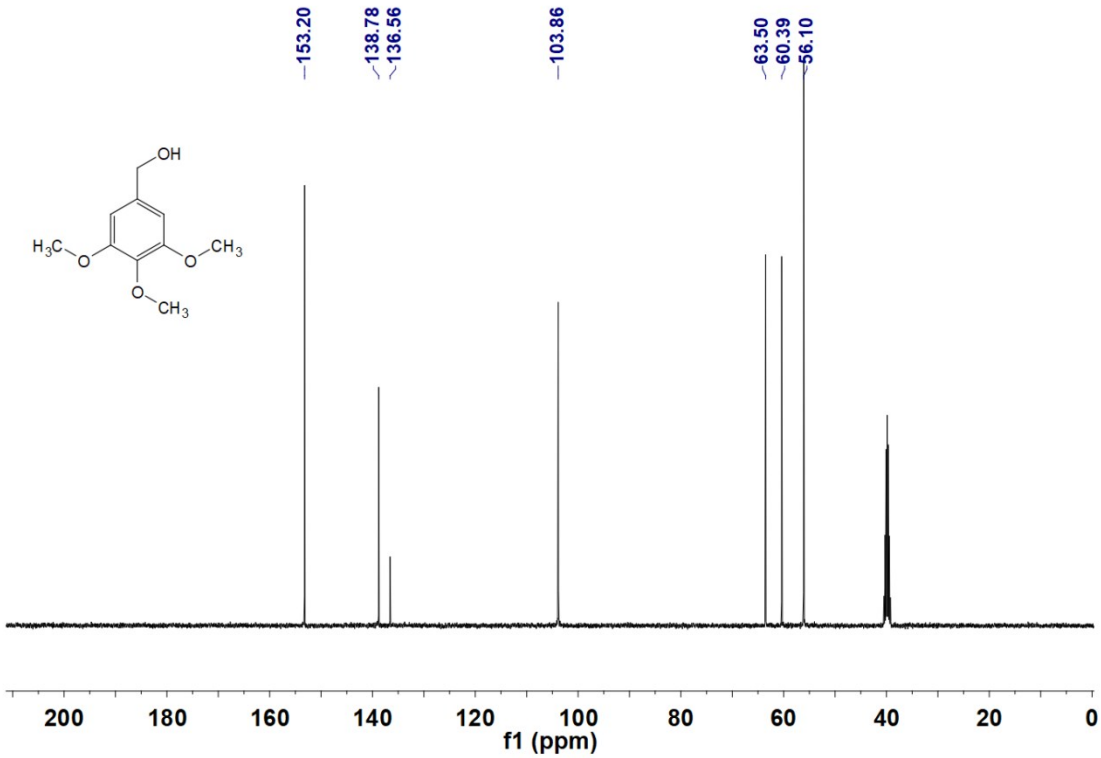

7.26
7.24
6.89
6.87
5.11
5.09
5.08
4.45
4.44
3.72
OH
H3C
O
2.00
1.98
0.96
1.95
2.95
11.0
10.0
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)

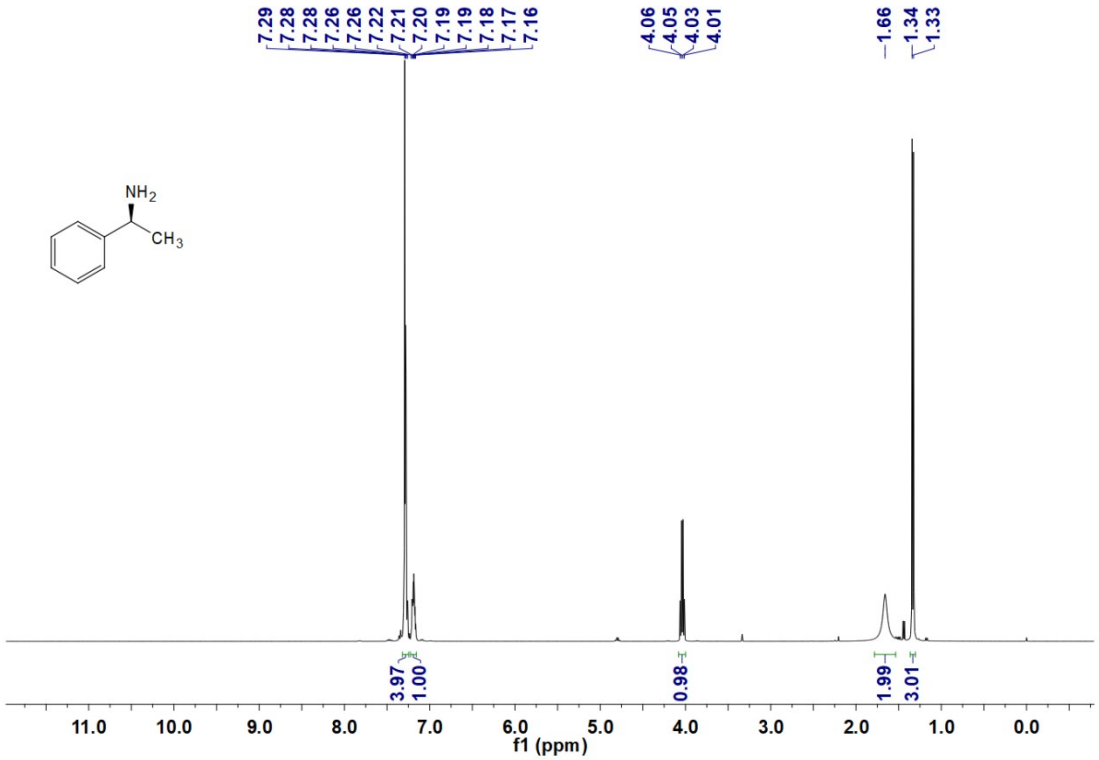

158.67
134.99
128.41
113.90
63.11
55.40
OH
H3C
O
180
160
140
120
100
80
60
40
20
0
f1 (ppm)

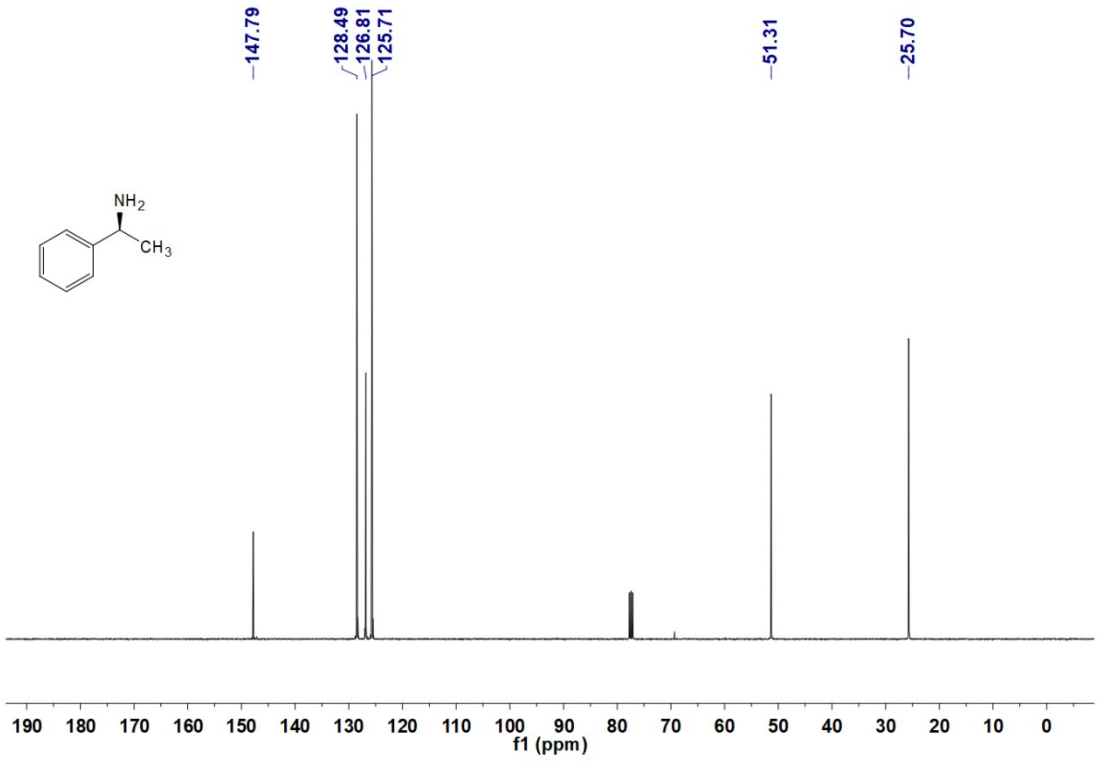

9.28
7.11
7.10
6.63
6.61
6.61
5.11
4.42
HO
OH
0.94
1.00
1.98
0.98
0.92
1.96
11.5
9.0
7.5
6.0
4.5
3.0
1.5
0.0
f1 (ppm)

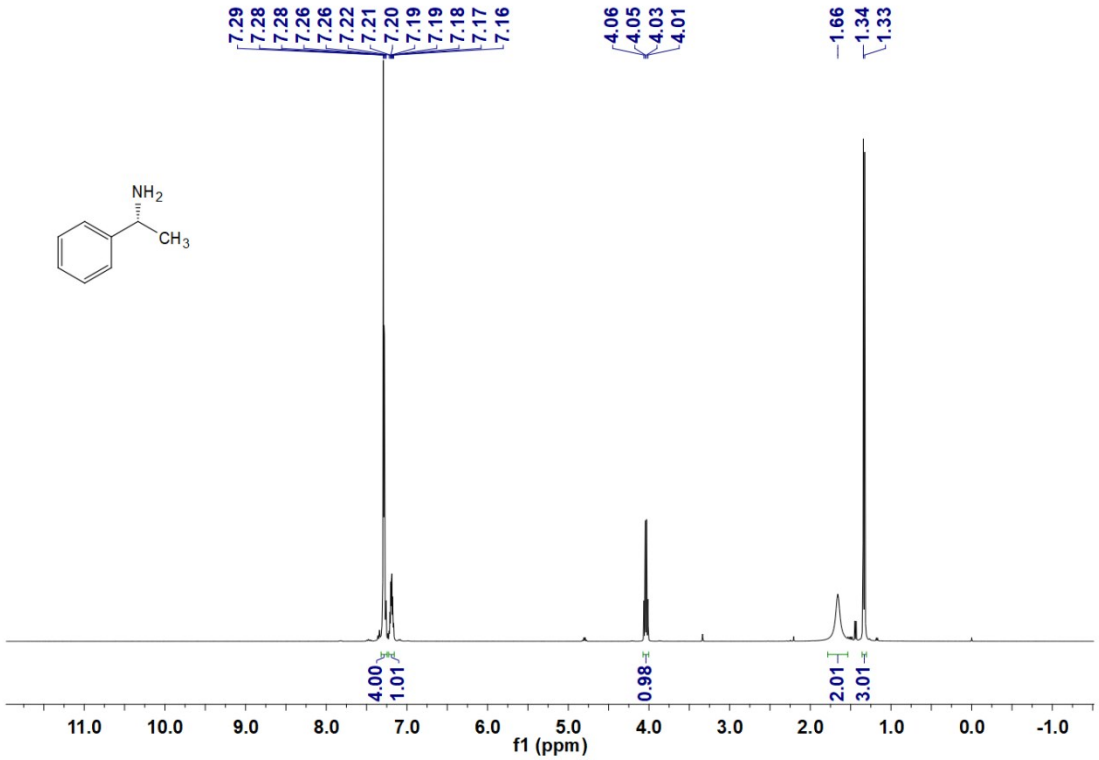

157.72
144.54
129.45
117.39
113.99
113.72
63.33
HO
OH
190
170
150
130
110
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

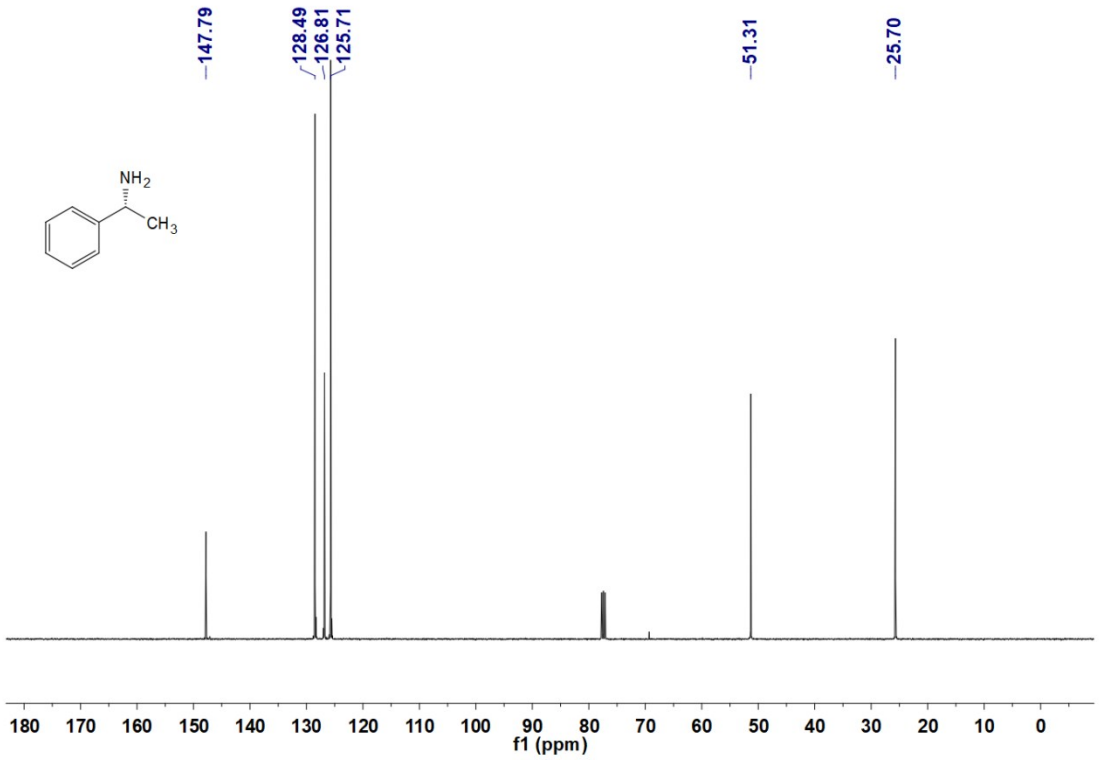

6.65
5.21
5.20
5.18
4.47
4.46
3.78
3.66
OH
H3C
O
O
CH3
O
CH3
2.00
1.01
2.02
5.99
3.00
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)

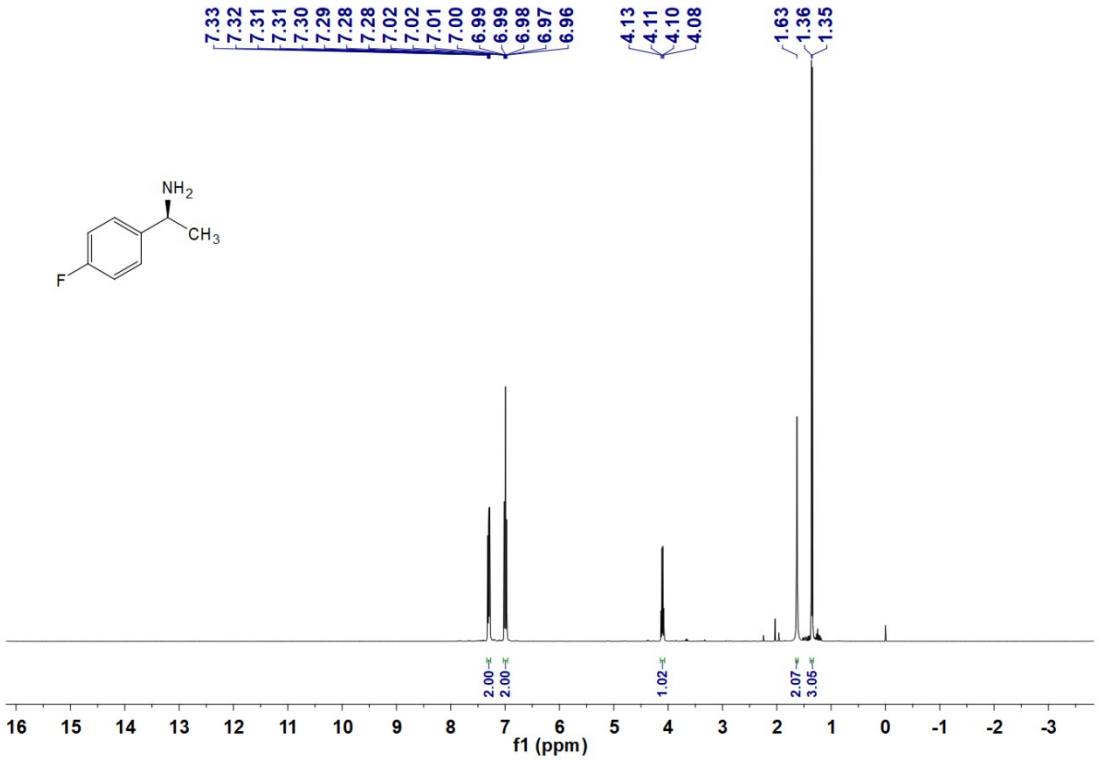

153.20
138.78
136.56
103.86
63.50
60.39
56.10
OH
H3C
O
O
CH3
O
CH3
200
180
160
140
120
100
80
60
40
20
0
f1 (ppm)

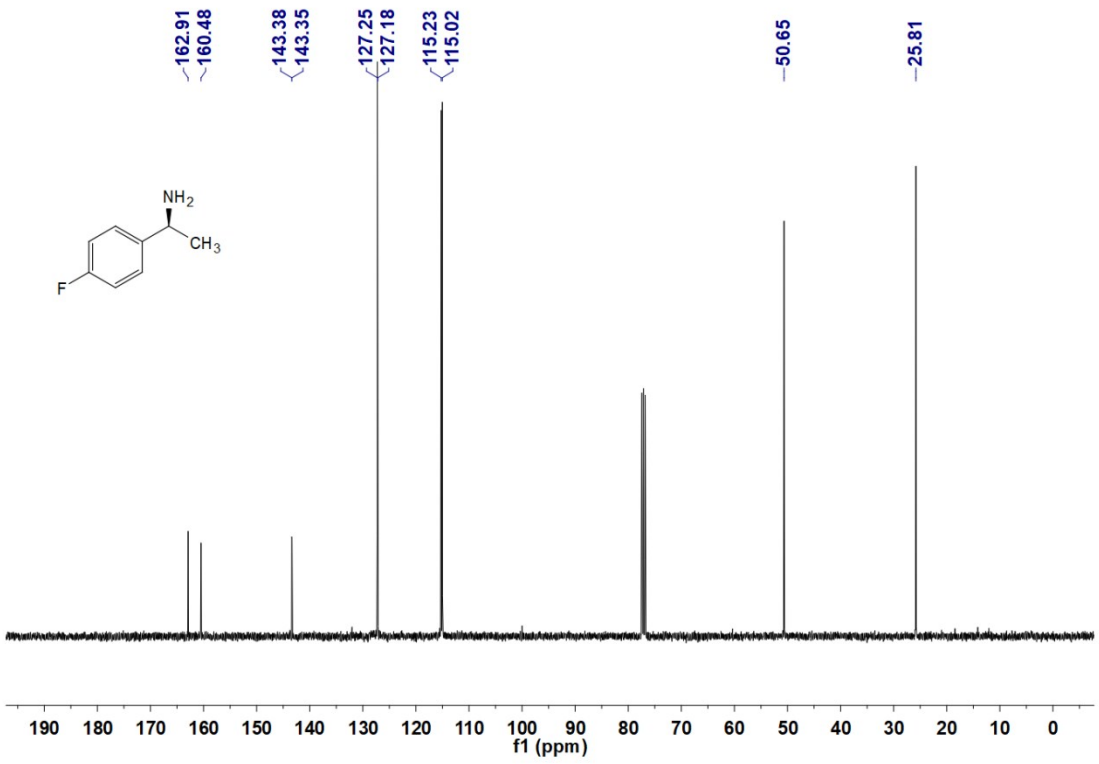

7.29
7.28
7.28
7.26
7.26
7.22
7.21
7.20
7.19
7.19
7.18
7.17
7.16
4.06
4.05
4.03
4.01
1.66
1.34
1.33
NH2
CH3
3.97
1.00
0.98
1.99
3.01
11.0
10.0
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)

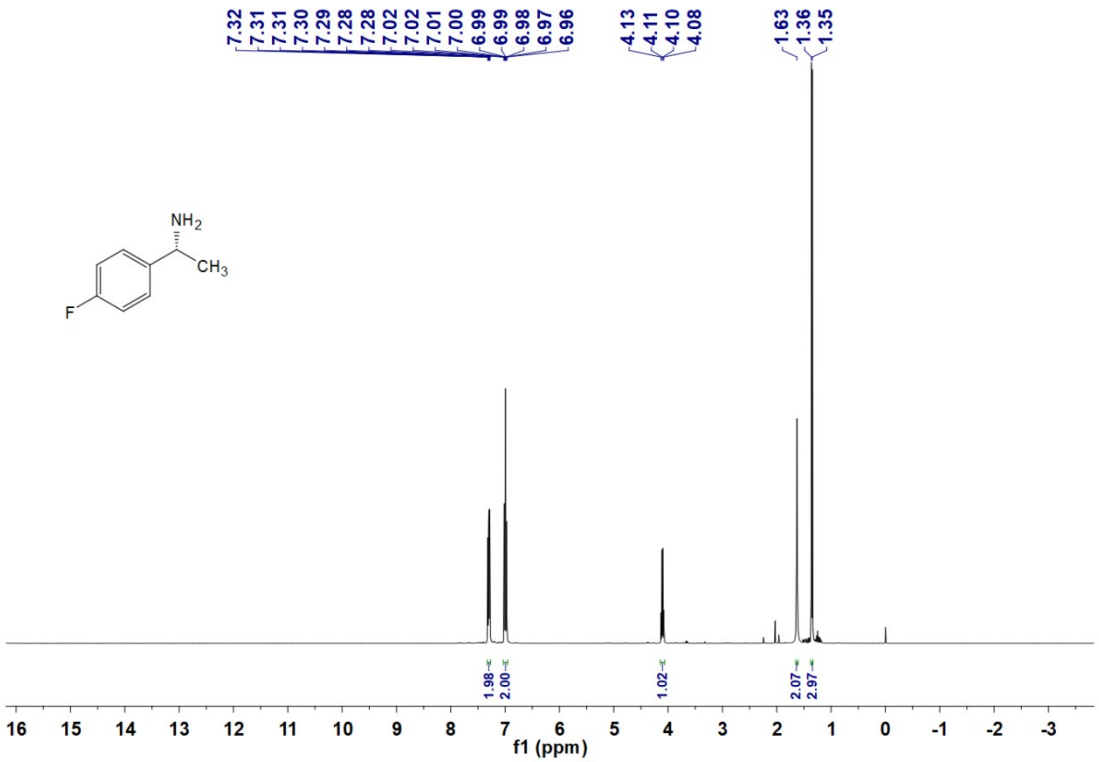

147.79
128.49
126.81
125.71
51.31
25.70
NH2
CH3
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

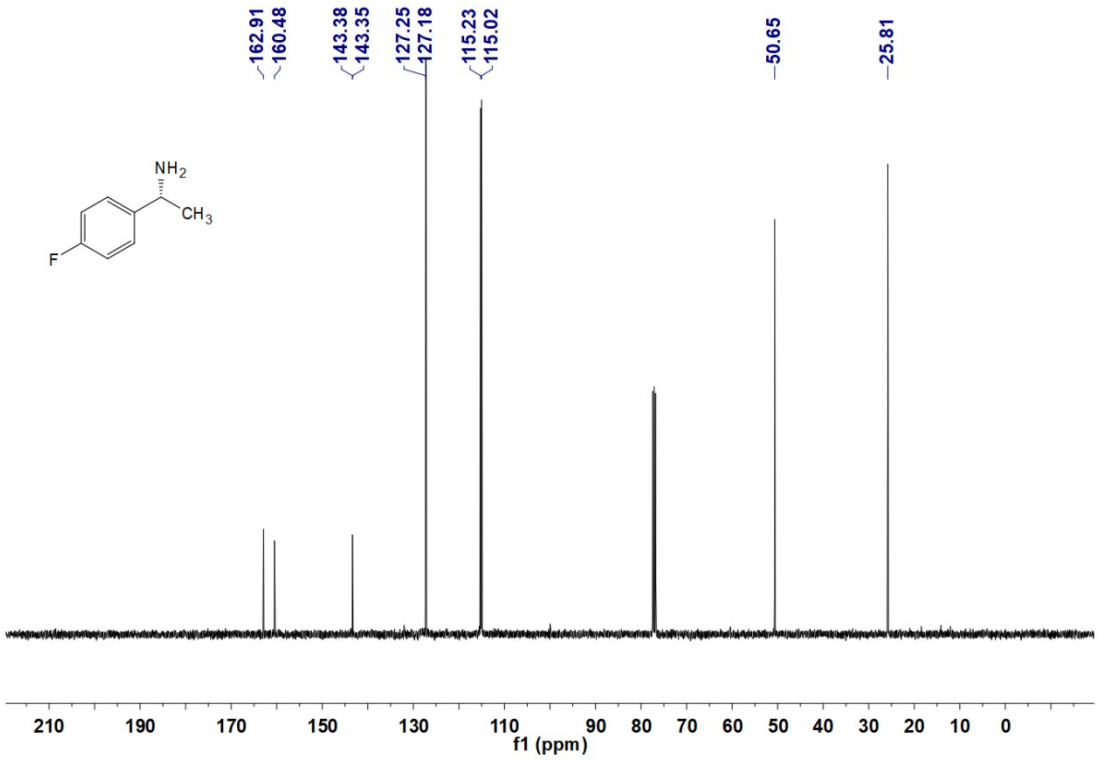

7.29
7.28
7.28
7.26
7.26
7.22
7.21
7.20
7.19
7.19
7.18
7.17
7.16
4.06
4.05
4.03
4.01
1.66
1.34
1.33
NH2
CH3
4.00
1.01
0.98
2.01
3.01
11.0
10.0
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
-1.0
f1 (ppm)

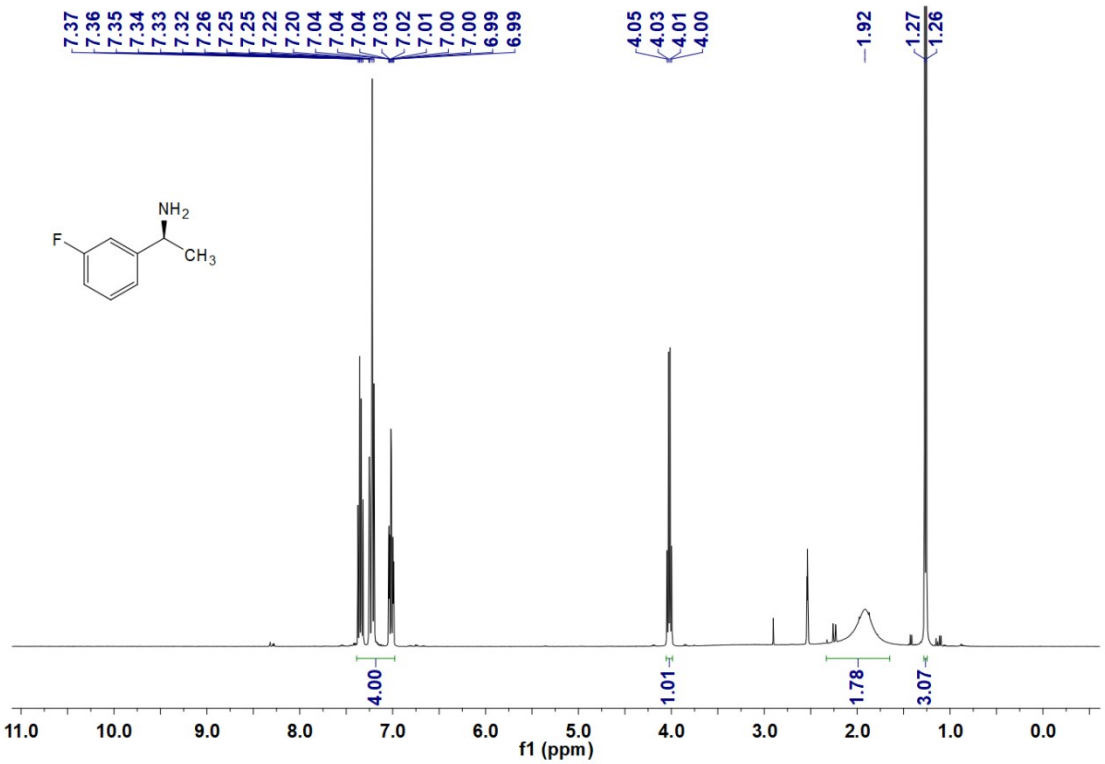

147.79
128.49
126.81
125.71
51.31
25.70
NH2
CH3
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

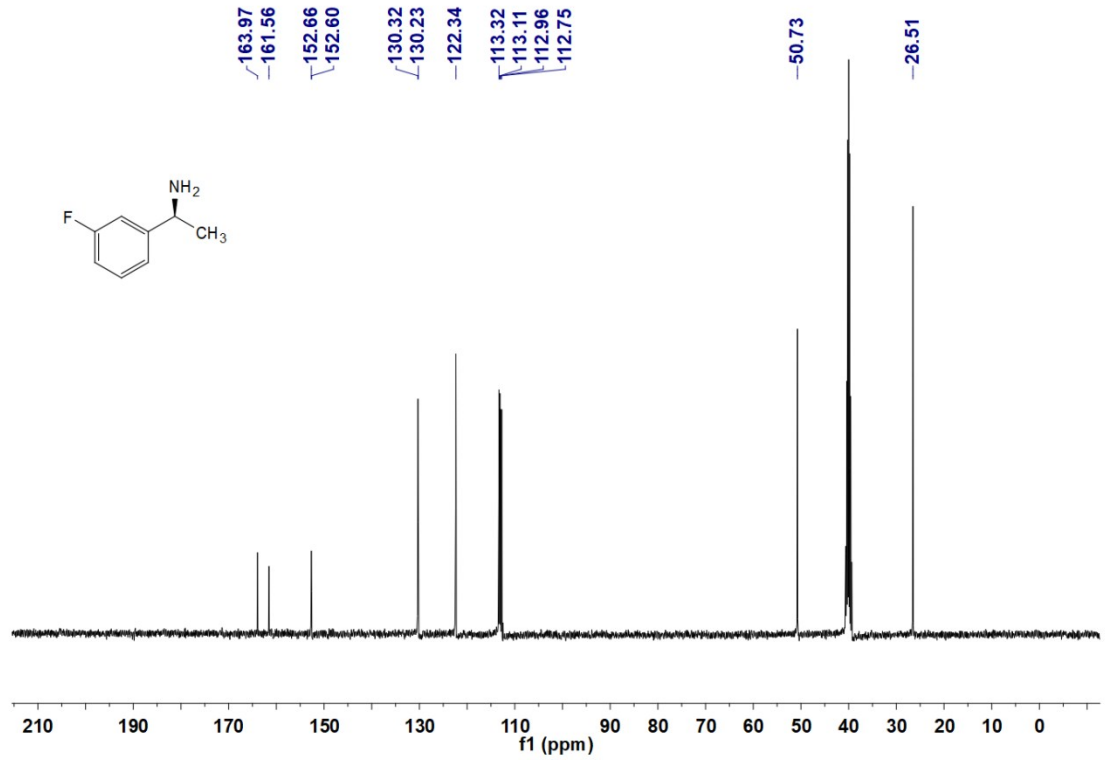

7.33
7.32
7.31
7.31
7.30
7.29
7.28
7.28
7.02
7.02
7.01
7.00
6.99
6.99
6.98
6.97
6.96
4.13
4.11
4.10
4.08
1.63
1.36
1.35
NH2
CH3
F
2.00
2.00
1.02
2.07
3.05
16 15 14 13 12 11 10 9 8 7 6 5 4 3 2 1 0 -1 -2 -3
f1 (ppm)

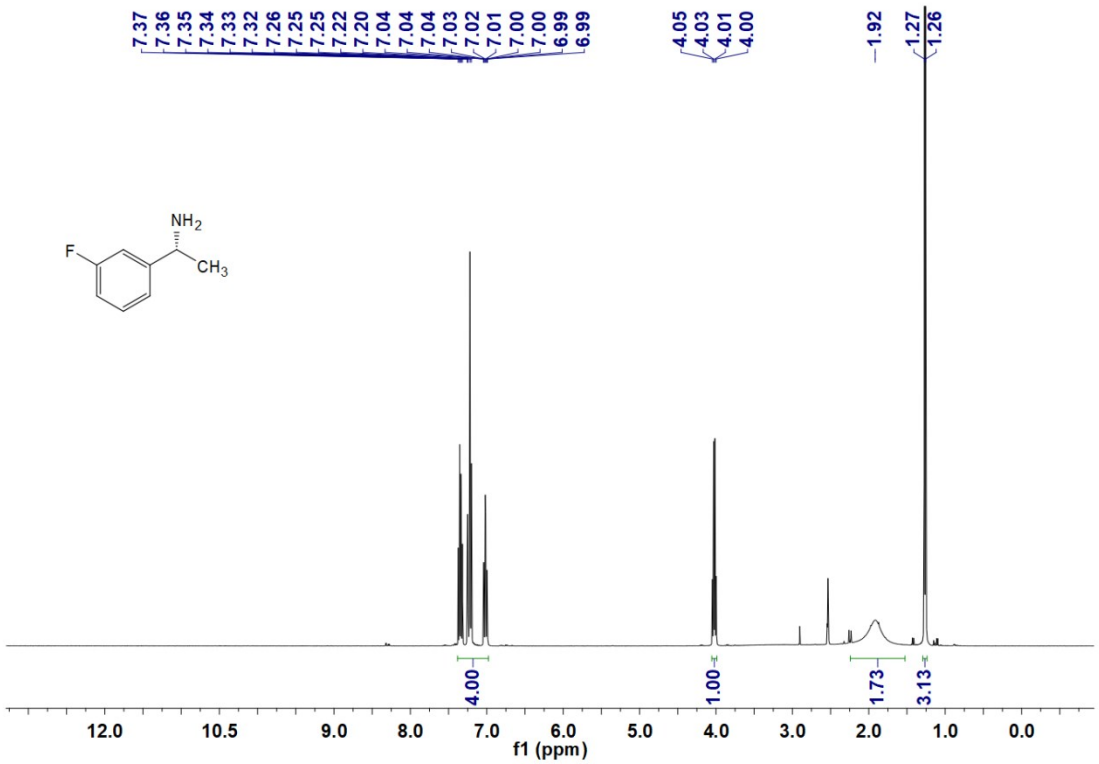

162.91
160.48
143.38
143.35
127.25
127.18
115.23
115.02
50.65
25.81
NH2
CH3
F
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
f1 (ppm)

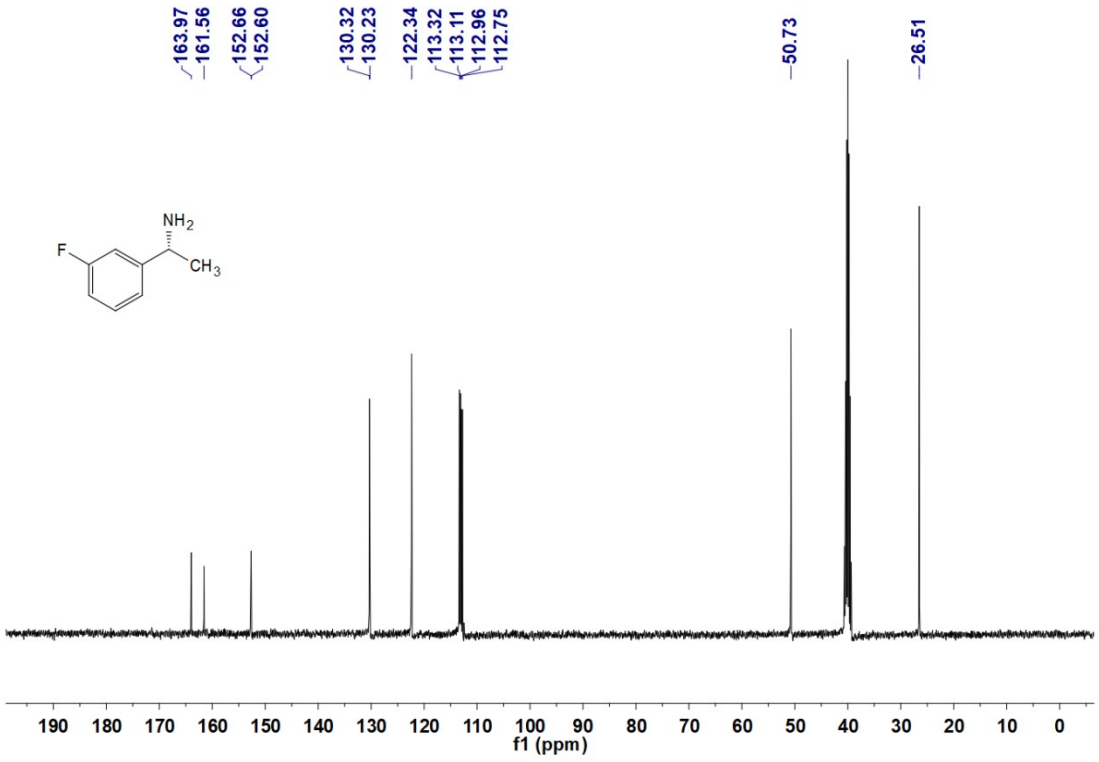

7.32
7.31
7.31
7.30
7.29
7.28
7.28
7.02
7.02
7.01
7.00
6.99
6.99
6.98
6.97
6.96
4.13
4.11
4.10
4.08
1.63
1.36
1.35
NH2
CH3
F
1.98
2.00
1.02
2.07
2.97
16 15 14 13 12 11 10 9 8 7 6 5 4 3 2 1 0 -1 -2 -3
f1 (ppm)

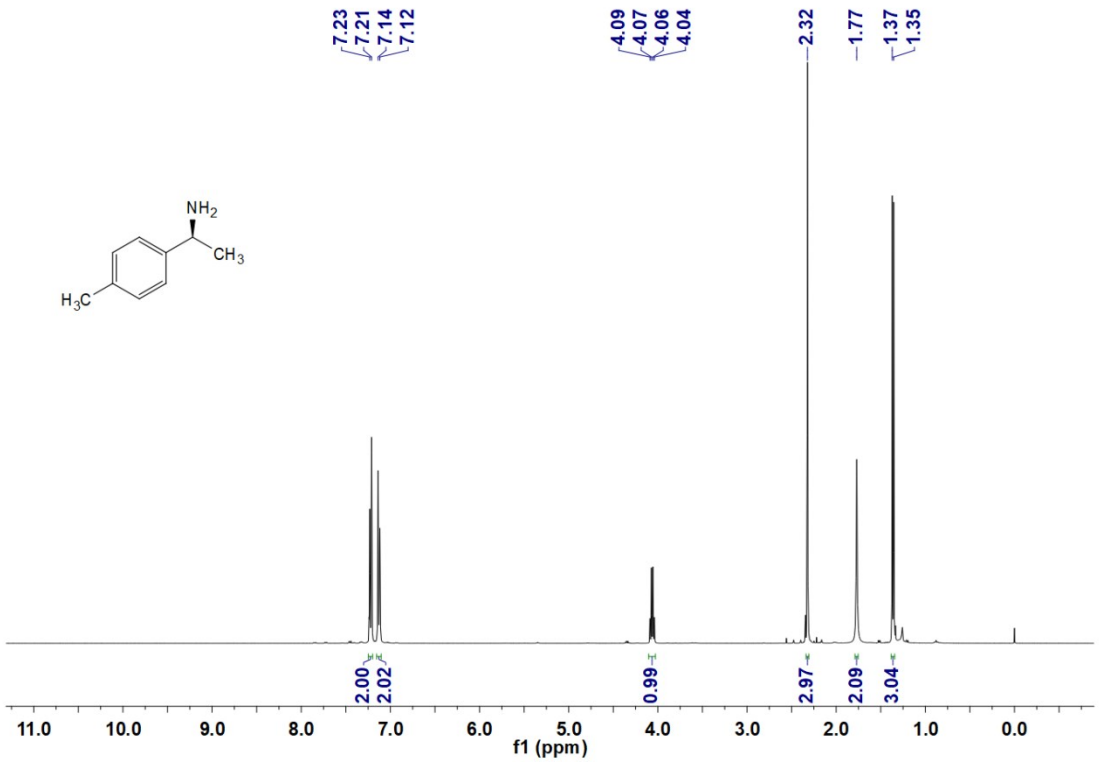

162.91
160.48
143.38
143.35
127.25
127.18
115.23
115.02
50.65
25.81
NH2
CH3
F
210 190 170 150 130 110 90 80 70 60 50 40 30 20 10 0
f1 (ppm)

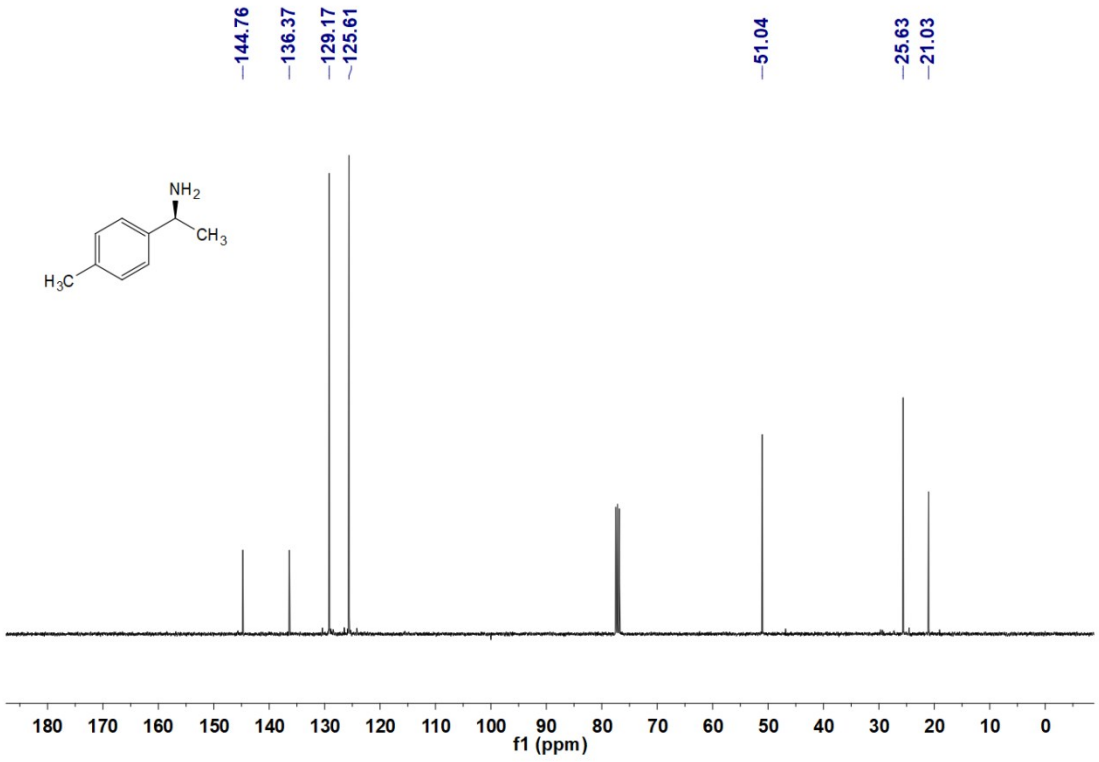

7.37
7.36
7.35
7.34
7.33
7.32
7.26
7.25
7.25
7.22
7.20
7.04
7.04
7.04
7.03
7.02
7.01
7.00
7.00
6.99
6.99
4.05
4.03
4.01
4.00
1.92
1.27
1.26
NH2
F
CH3
4.00
1.01
1.78
3.07
11.0
10.0
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)

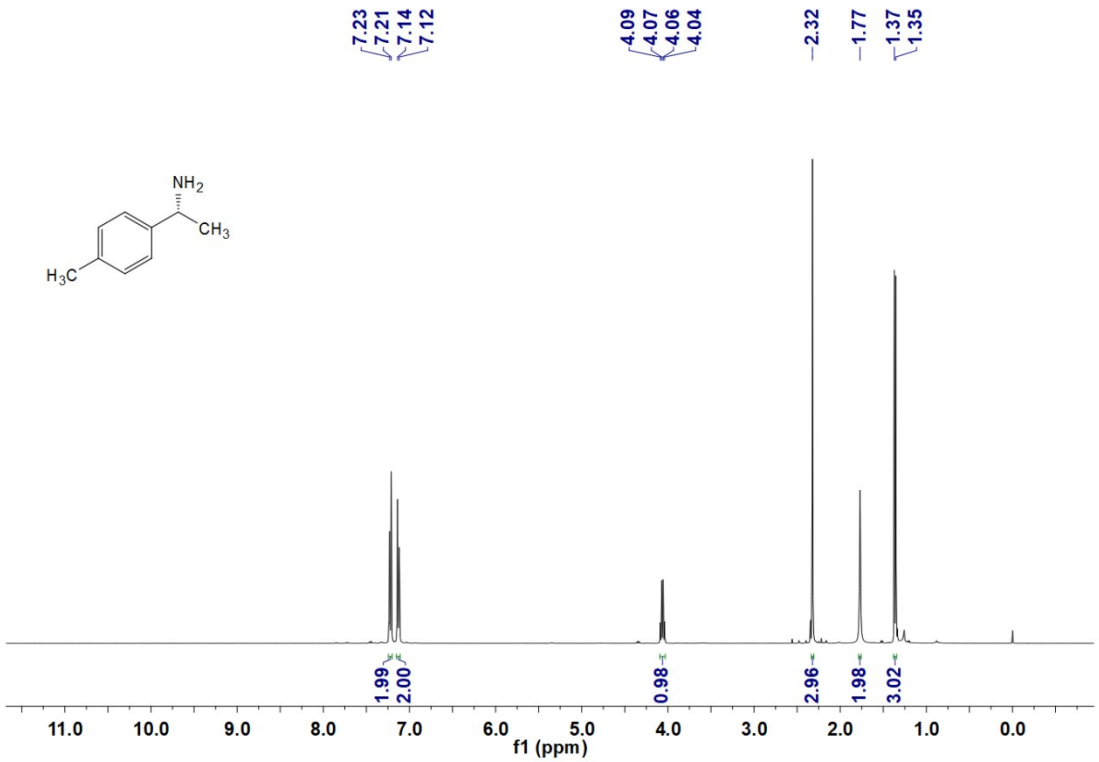

163.97
161.56
152.66
152.60
130.32
130.23
122.34
113.32
113.11
112.96
112.75
50.73
26.51
NH2
F
CH3
210
190
170
150
130
110
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

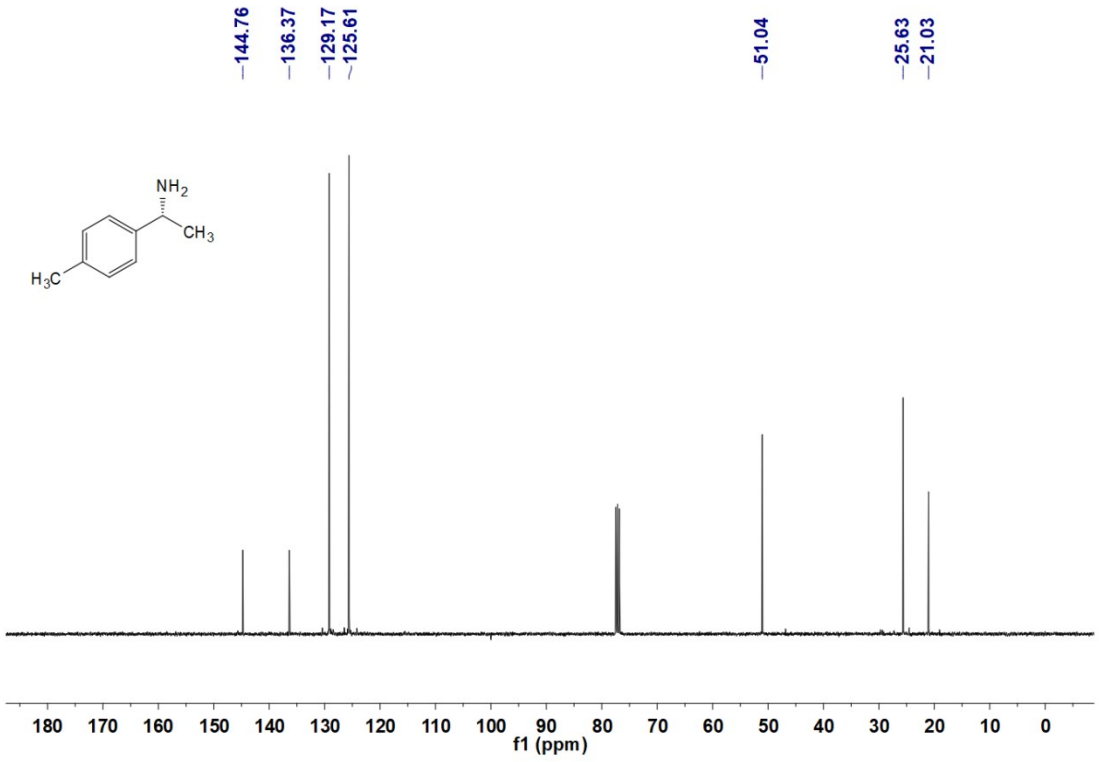

7.37
7.36
7.35
7.34
7.33
7.32
7.26
7.25
7.25
7.22
7.20
7.04
7.04
7.04
7.03
7.02
7.01
7.00
7.00
6.99
6.99
4.05
4.03
4.01
4.00
1.92
1.27
1.26
NH2
F
CH3
4.00
1.00
1.73
3.13
12.0
10.5
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)

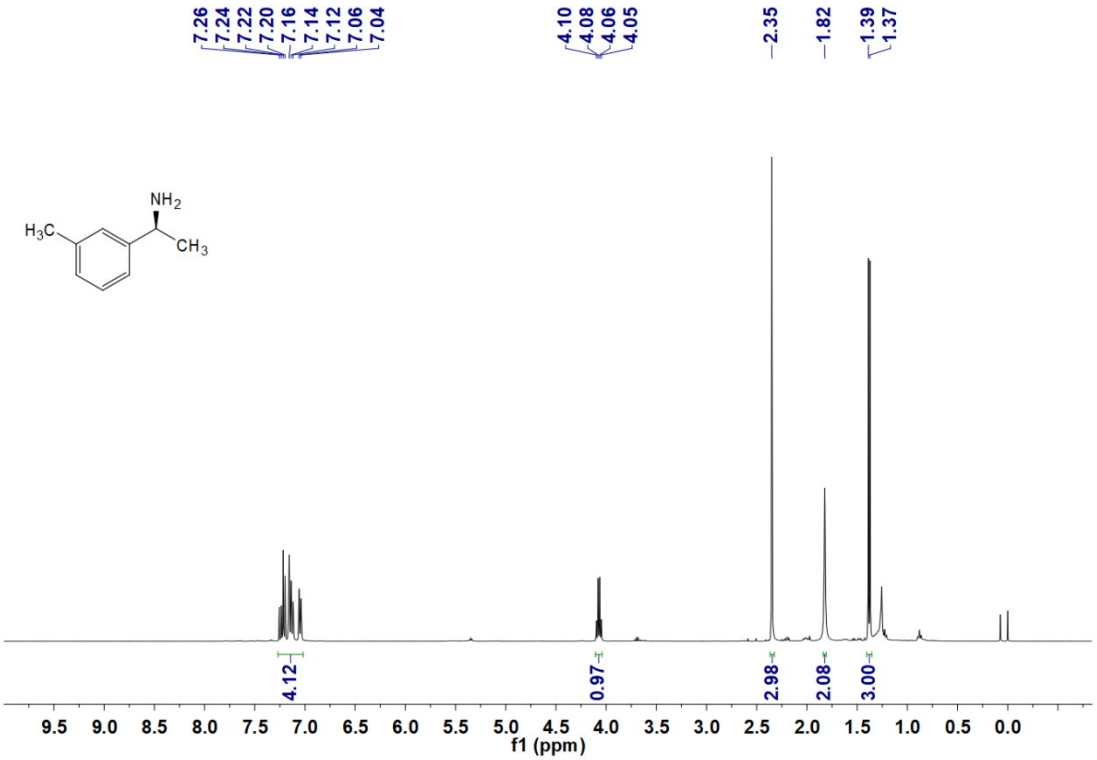

163.97
161.56
152.66
152.60
130.32
130.23
122.34
113.32
113.11
112.96
112.75
50.73
26.51
NH2
F
CH3
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

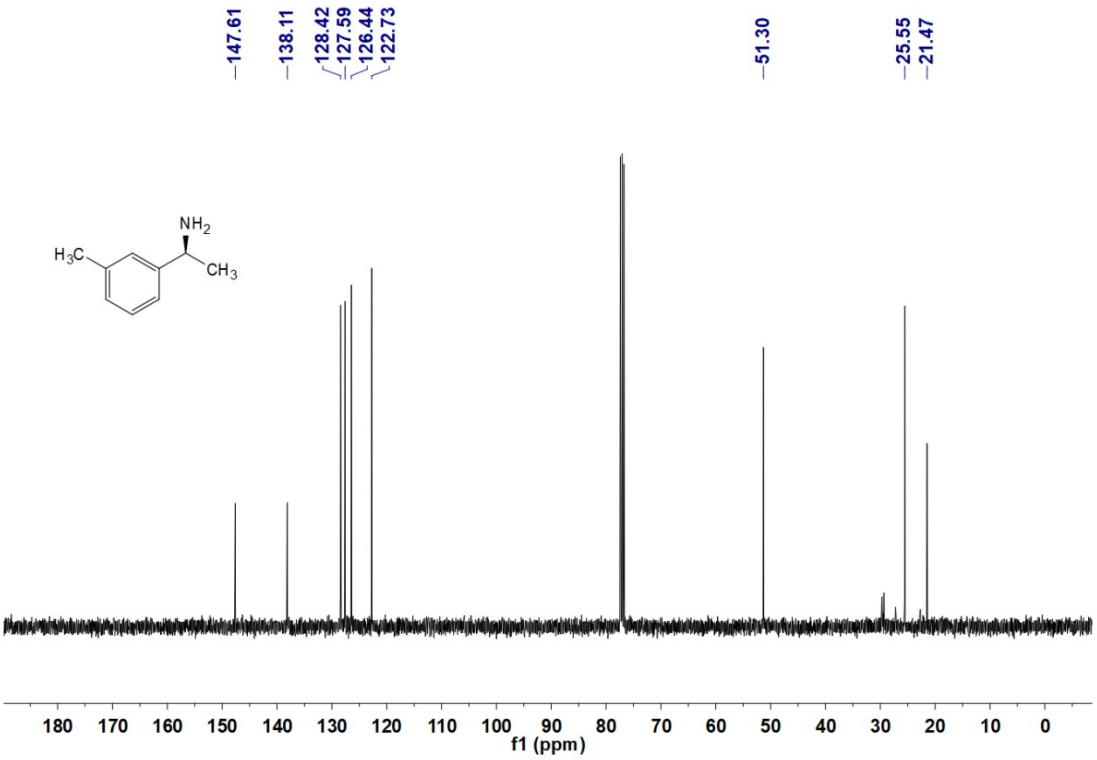

7.23
7.21
7.14
7.12
4.09
4.07
4.06
4.04
2.32
1.77
1.37
1.35
NH2
CH3
H3C
2.00
2.02
0.99
2.97
2.09
3.04
11.0
10.0
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)

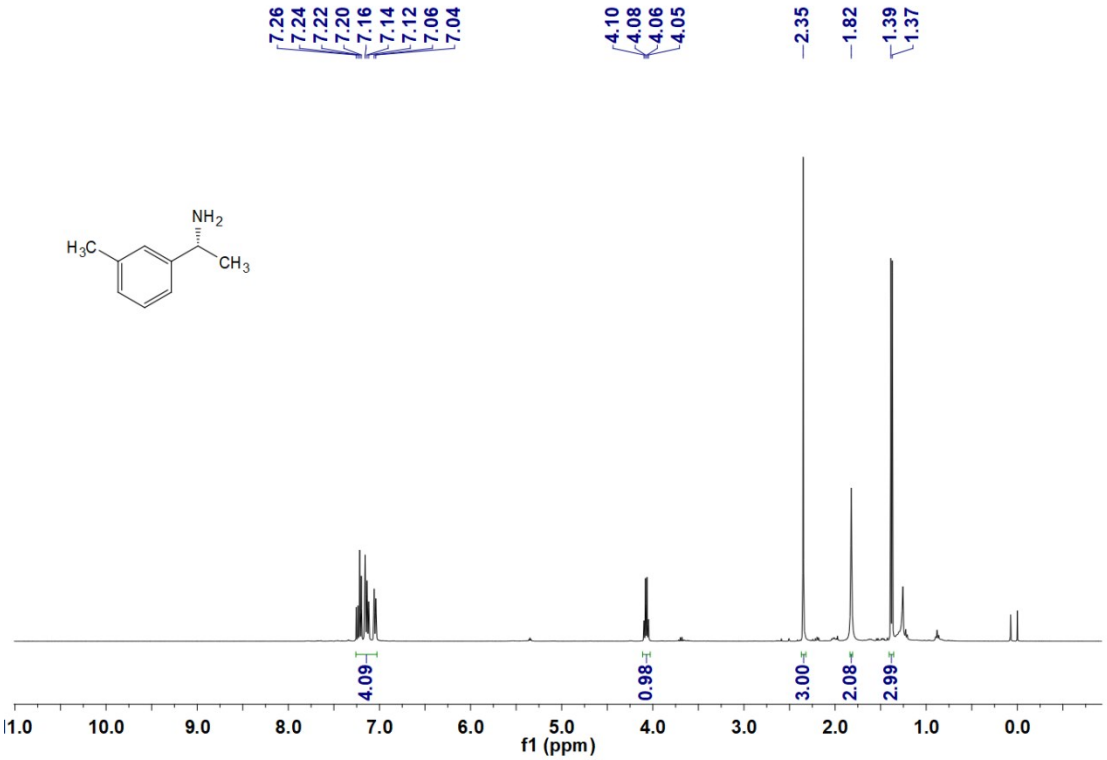

144.76
136.37
129.17
125.61
51.04
25.63
21.03
NH2
CH3
H3C
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

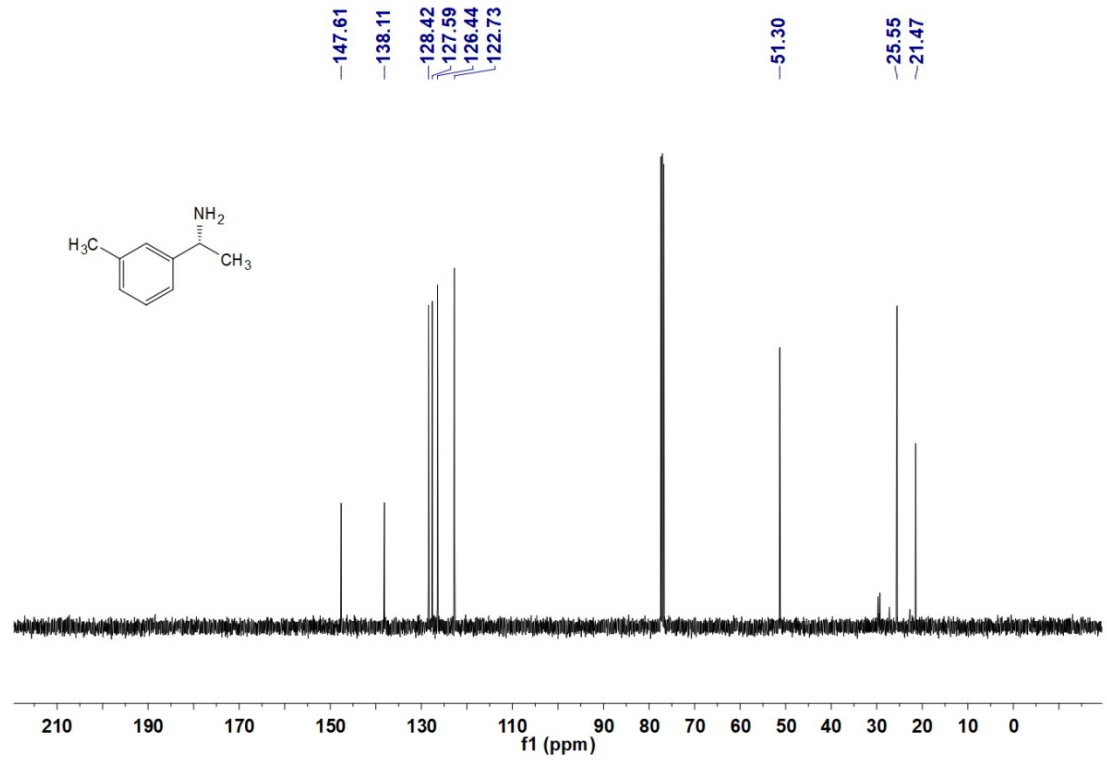

7.23
7.21
7.14
7.12
4.09
4.07
4.06
4.04
2.32
1.77
1.37
1.35
NH2
CH3
H3C
1.99
2.00
0.98
2.96
1.98
3.02
11.0
10.0
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)


144.76
136.37
129.17
125.61
51.04
25.63
21.03
NH2
CH3
H3C
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

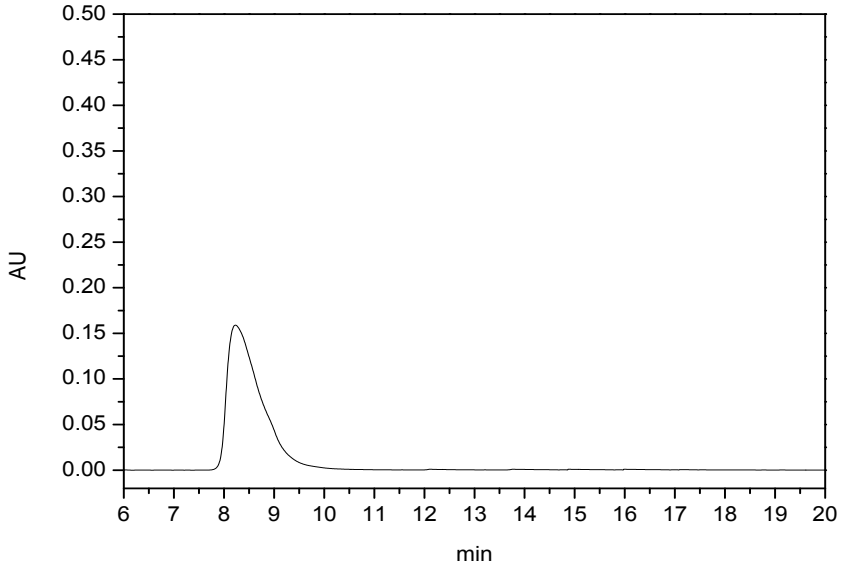

7.26
7.24
7.22
7.20
7.16
7.14
7.12
7.06
7.04
4.10
4.08
4.06
4.05
2.35
1.82
1.39
1.37
NH2
H3C
CH3
4.12
0.97
2.98
2.08
3.00
9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0
f1 (ppm)


147.61
138.11
128.42
127.59
126.44
122.73
51.30
25.55
21.47
NH2
H3C
CH3
180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
f1 (ppm)

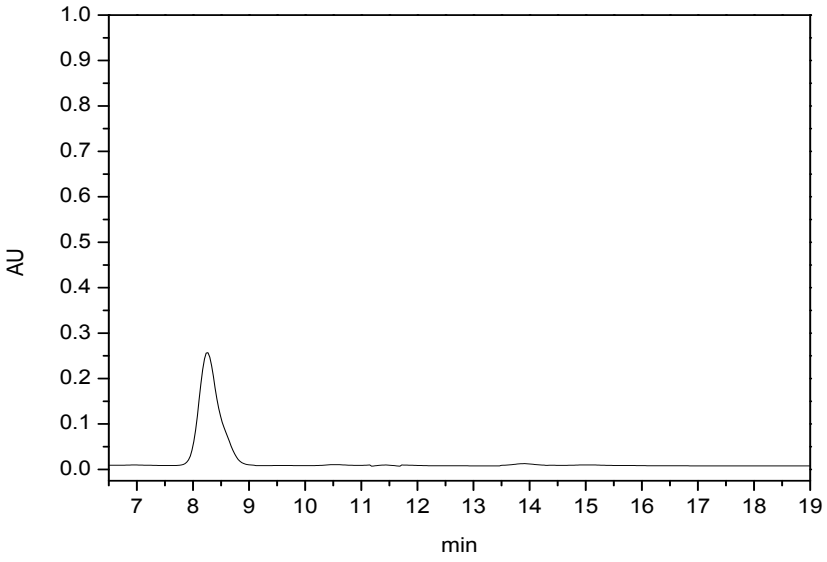

7.26
7.24
7.22
7.20
7.16
7.14
7.12
7.06
7.04
4.10
4.08
4.06
4.05
2.35
1.82
1.39
1.37
NH2
H3C
CH3
4.09
0.98
3.00
2.08
2.99
f1 (ppm)

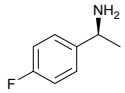

147.61
138.11
128.42
127.59
126.44
122.73
51.30
25.55
21.47
NH2
H3C
CH3
f1 (ppm)

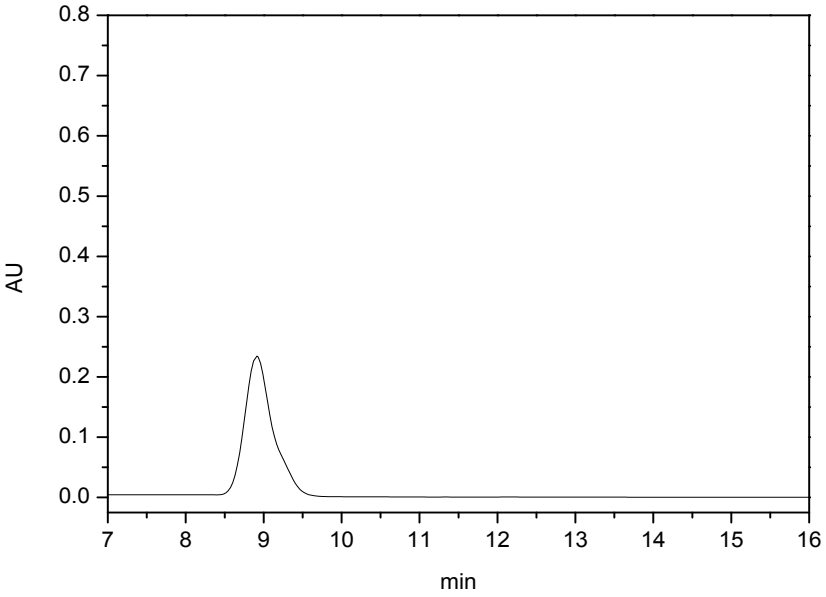
14. HPLC chromatogram

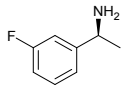


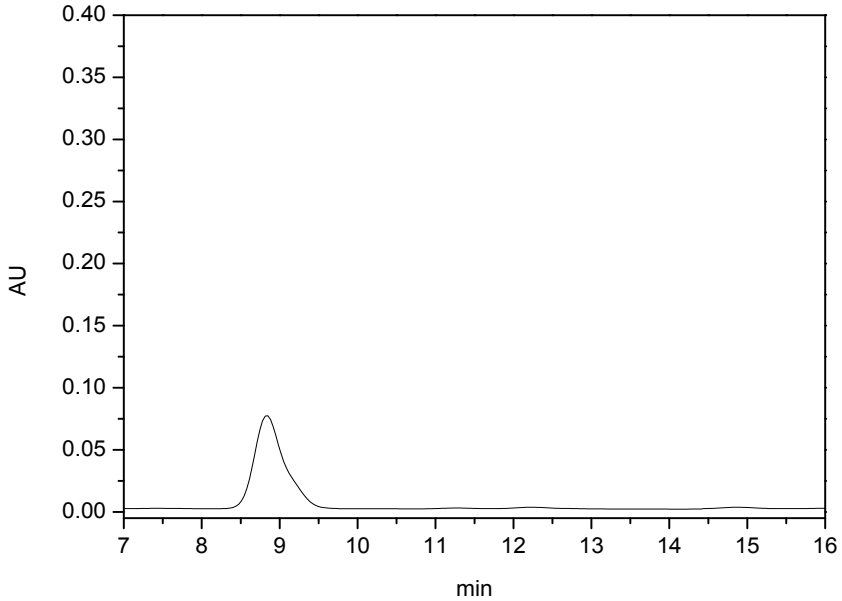
Entry	Product	Area		Retention time (min)		ee (%)
1		*S*	4223481	*S*	8.229	99.5
		R	11103	*R*	12.367	

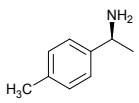


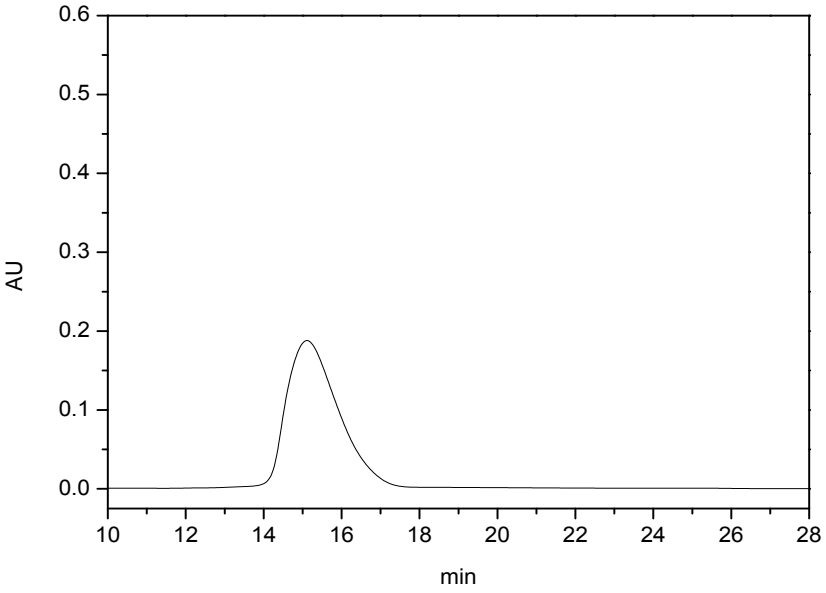
Entry	Product	Area		Retention time (min)		ee (%)
2		*S*	6791623	*S*	8.268	99.8
		R	7399	*R*	13.907	

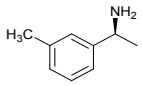


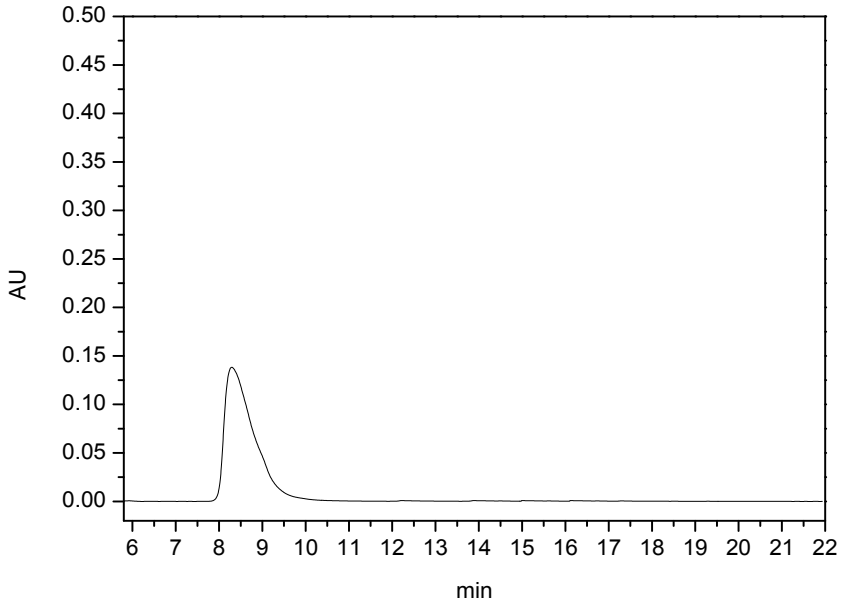
Entry	Product	Area		Retention time (min)		ee (%)
3		*S*	6620197	*S*	8.913	99.2
		R	27469	*R*	11.332	



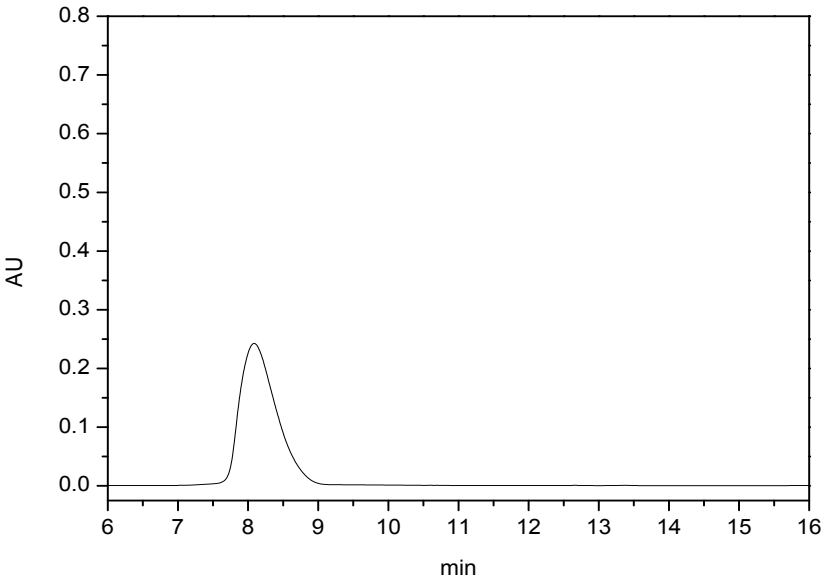
4		*S*	8898793	*S*	13.454	99.7
		R	15544	*R*	21.236	

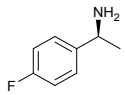


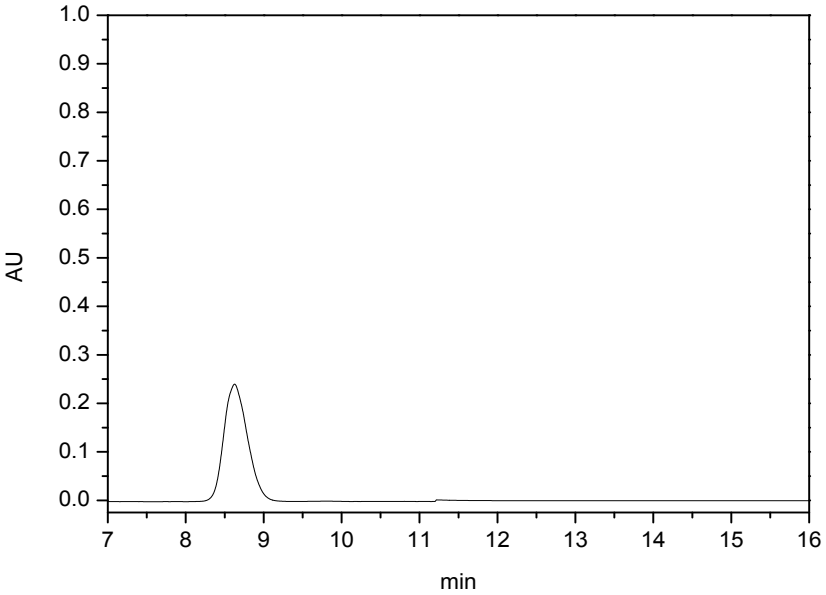
Entry	Product	Area		Retention time (min)		ee (%)
5		*S*	6514110	*S*	15.108	99.8
		R	65823	*R*	18.380	

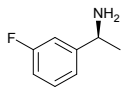


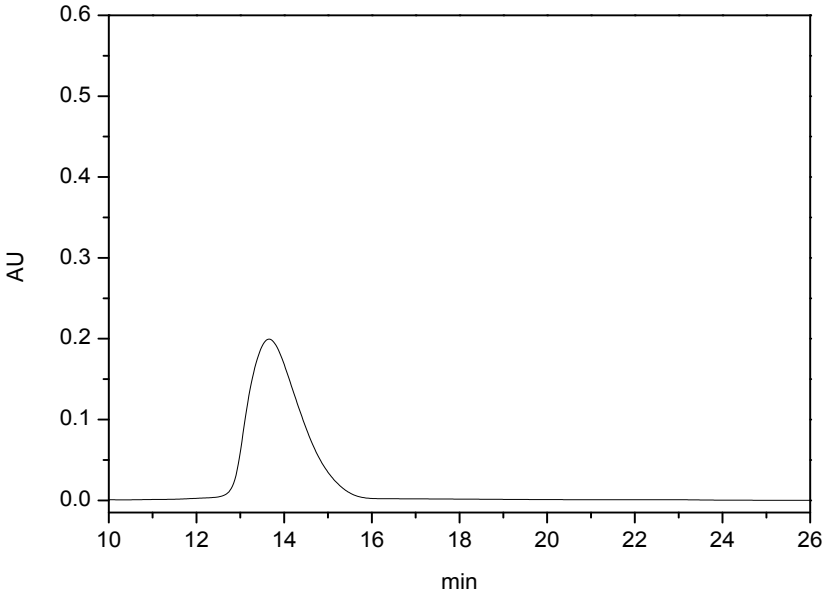
Entry	Product	Area		Retention time (min)		ee (%)
6		*S*	3672638	*S*	8.295	99.9
		R	601	*R*	12.691	

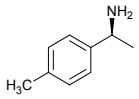


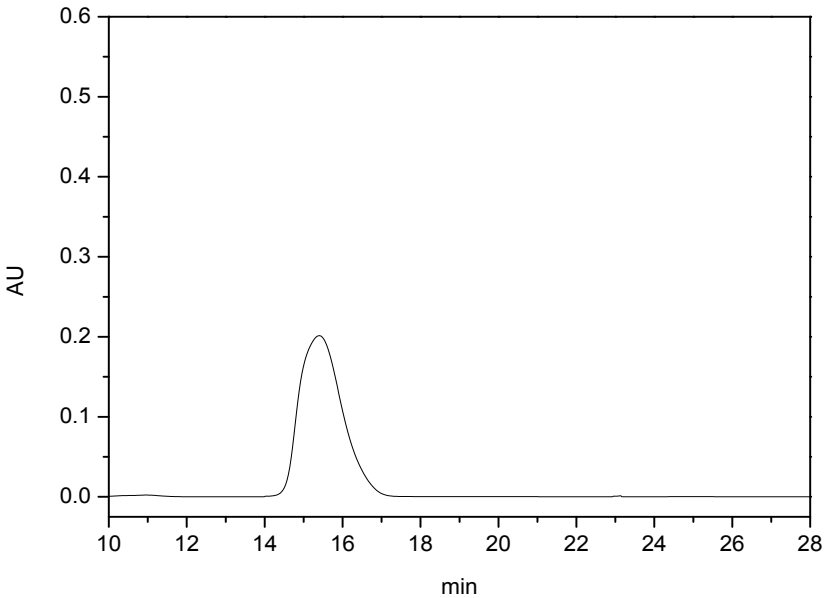
Entry	Product	Area		Retention time (min)		ee (%)
7		*S*	6413724	*S*	8.085	99.8
		R	7927	*R*	13.833	

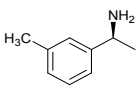


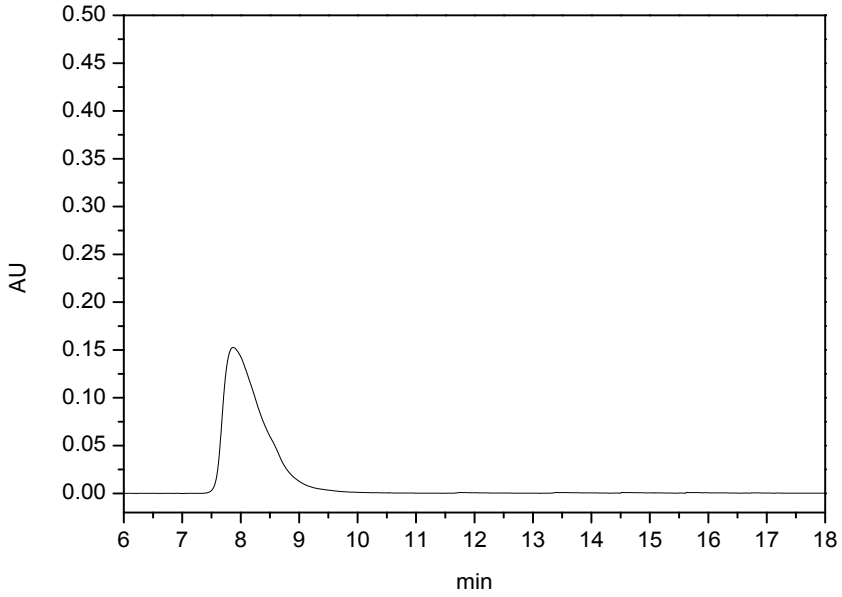
Entry	Product	Area		Retention time (min)		ee (%)
8		*S*	6760692	*S*	8.627	99.2
		R	27929	*R*	11.216	



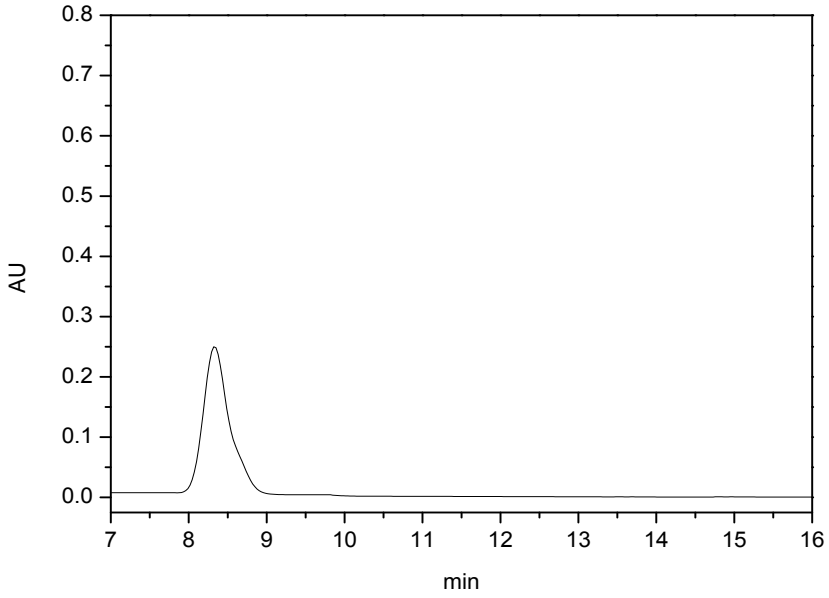
Entry	Product	Area		Retention time (min)		ee (%)
9		*S*	8616401	*S*	13.653	99.2
		R	32816	*R*	21.474	

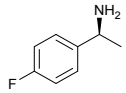


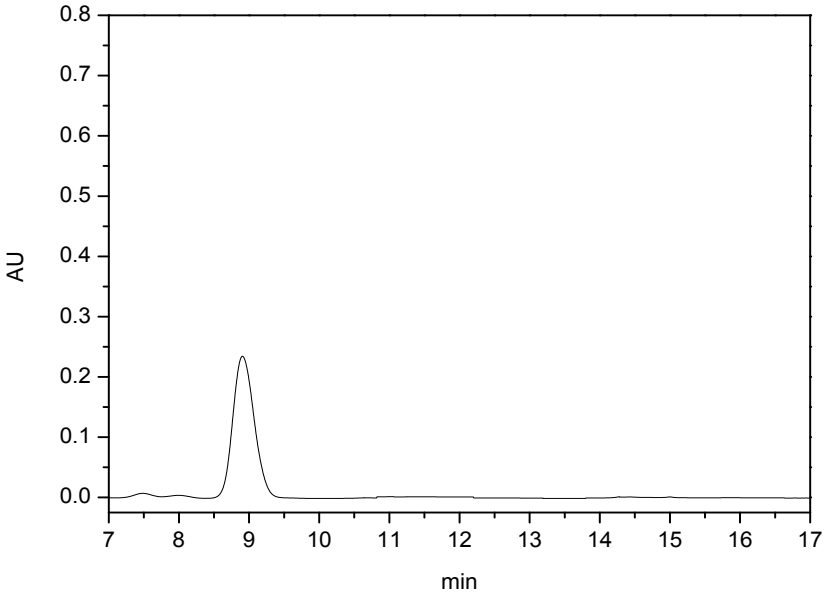
Entry	Product	Area		Retention time (min)		ee (%)
10		*S*	6980418	*S*	15.410	99.7
		R	9665	*R*	18.576	

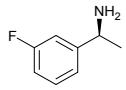


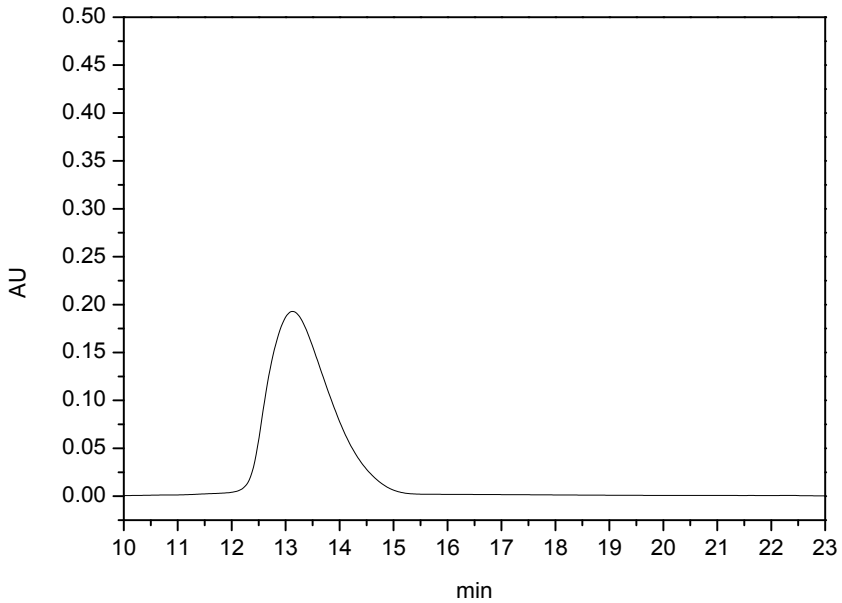
Entry	Product	Area		Retention time (min)		ee (%)
11		*S*	4054619	*S*	7.872	99.2
		R	15937	*R*	11.743	

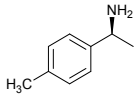


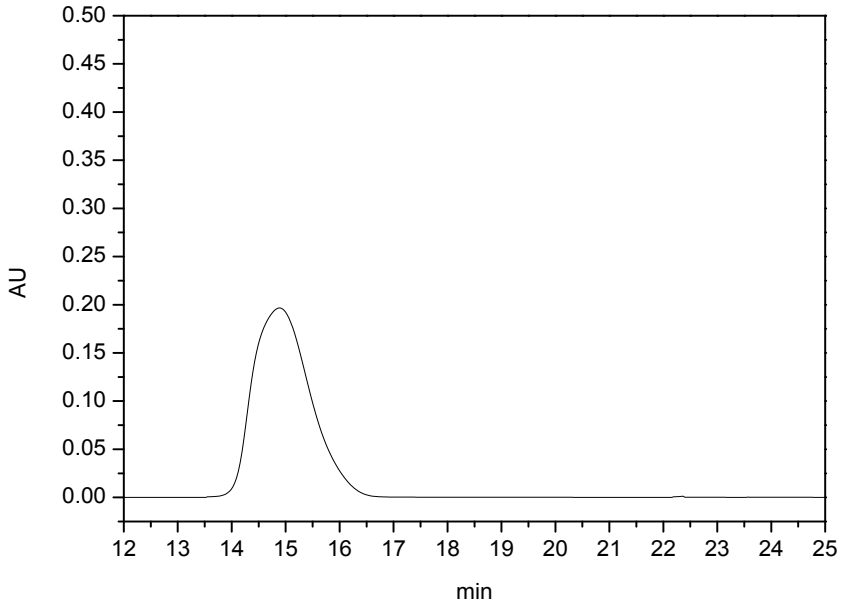
Entry	Product	Area		Retention time (min)		ee (%)
12		*S*	6606374	*S*	8.3216	99.5
		R	17943	*R*	14.0635	

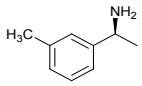


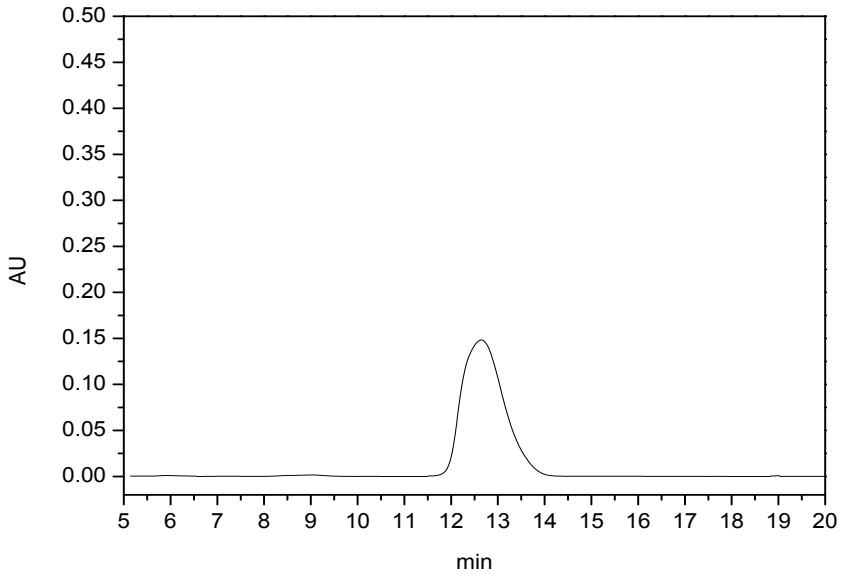
Entry	Product	Area		Retention time (min)		ee (%)
13		*S*	6610887	*S*	8.906	99.1
		R	29905	*R*	11.531	



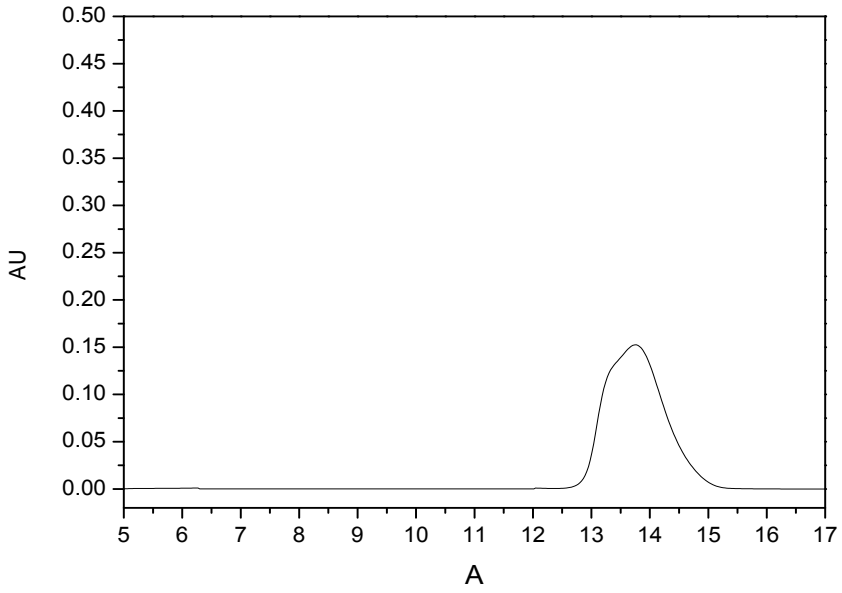
Entry	Product	Area		Retention time (min)		ee (%)
14		*S*	8330987	*S*	13.126	99.1
		R	37004	*R*	20.981	

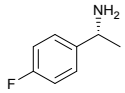


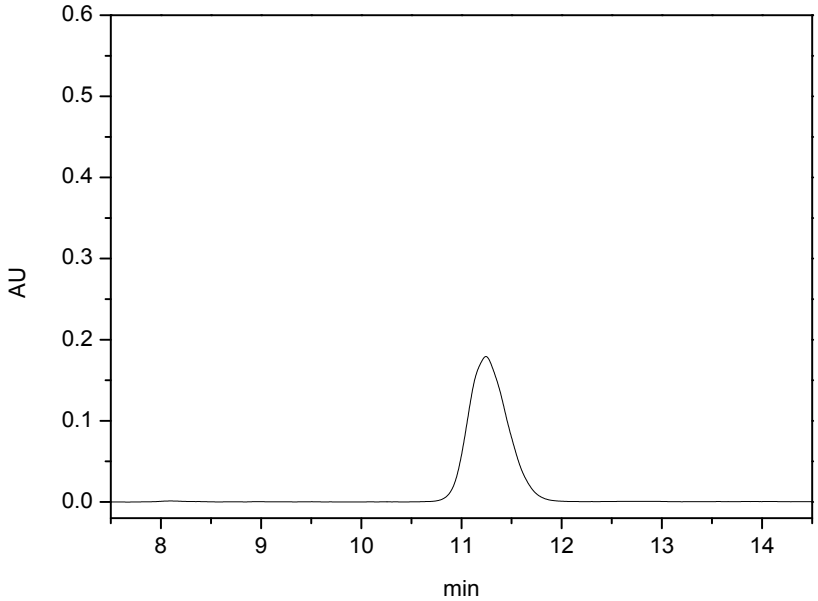
Entry	Product	Area		Retention time (min)		ee (%)
15		*S*	6812394	*S*	14.896	99.7
		R	9354	*R*	17.923	

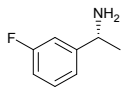


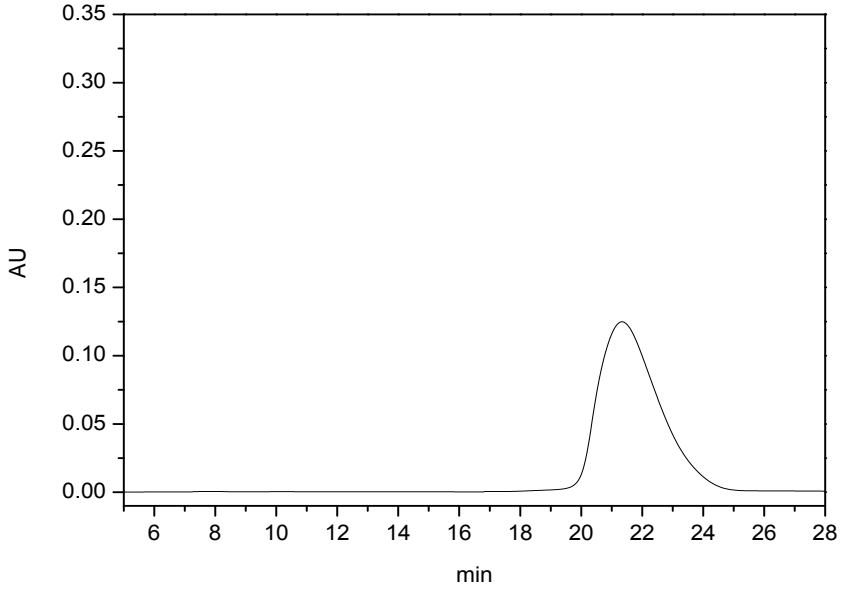
Entry	Product	Area		Retention time (min)		ee (%)
16		*S*	15937	*S*	8.31	99.2
		R	3984205	*R*	12.65	

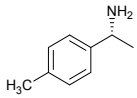


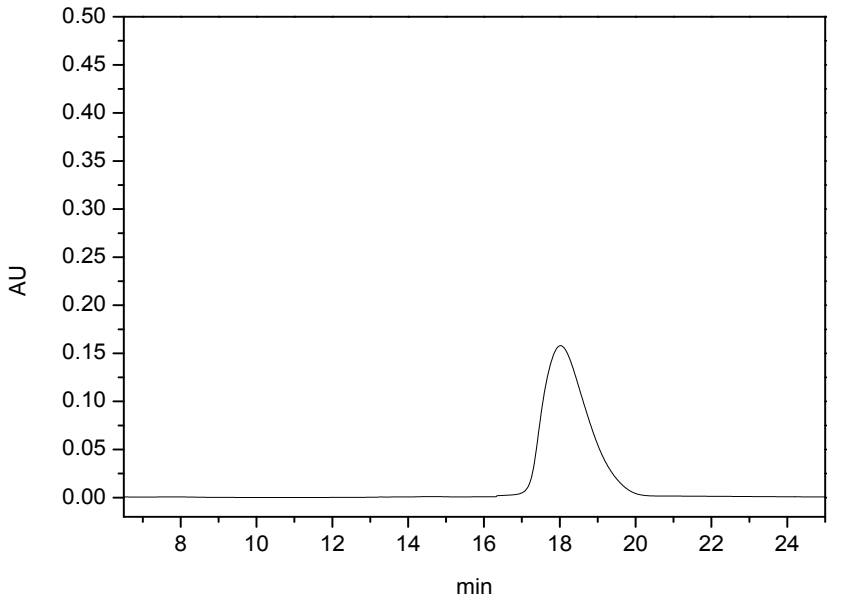
Entry	Product	Area		Retention time (min)		ee (%)
17		*S*	6614	*S*	7.962	99.8
		R	7009854	*R*	13.767	

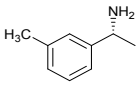


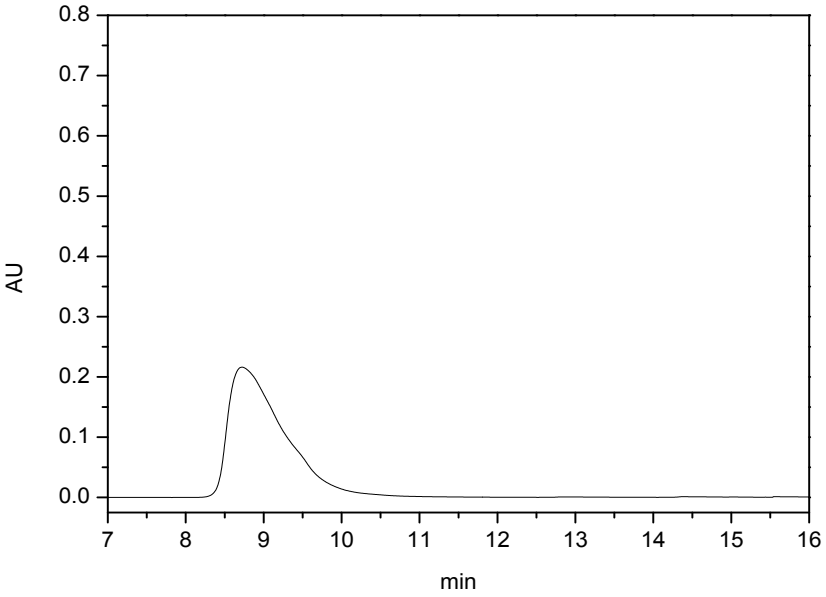
Entry	Product	Area		Retention time (min)		ee (%)
18		*S*	8360	*S*	8.831	99.7
		R	6653613	*R*	11.244	



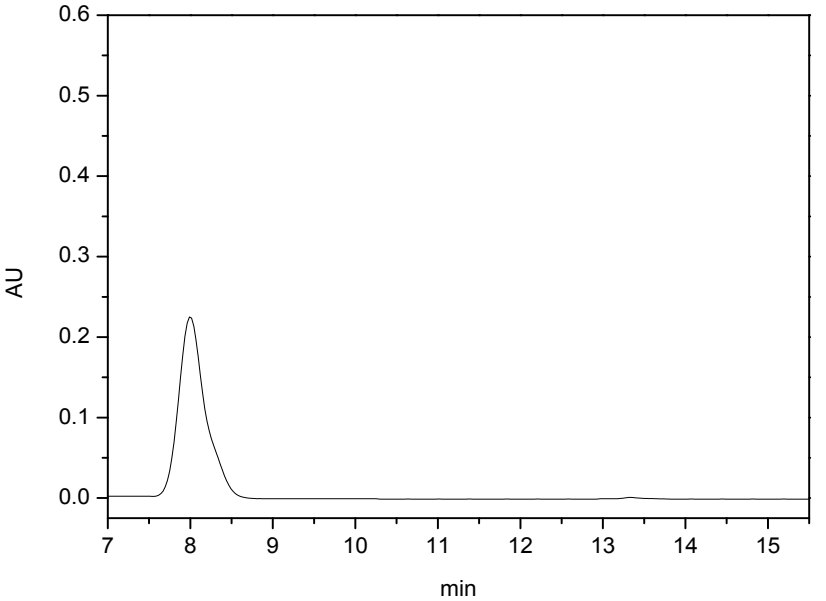
Entry	Product	Area		Retention time (min)		ee (%)
19		*S*	24076	*S*	13.289	99.4
		R	8689637	*R*	21.333	

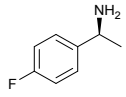


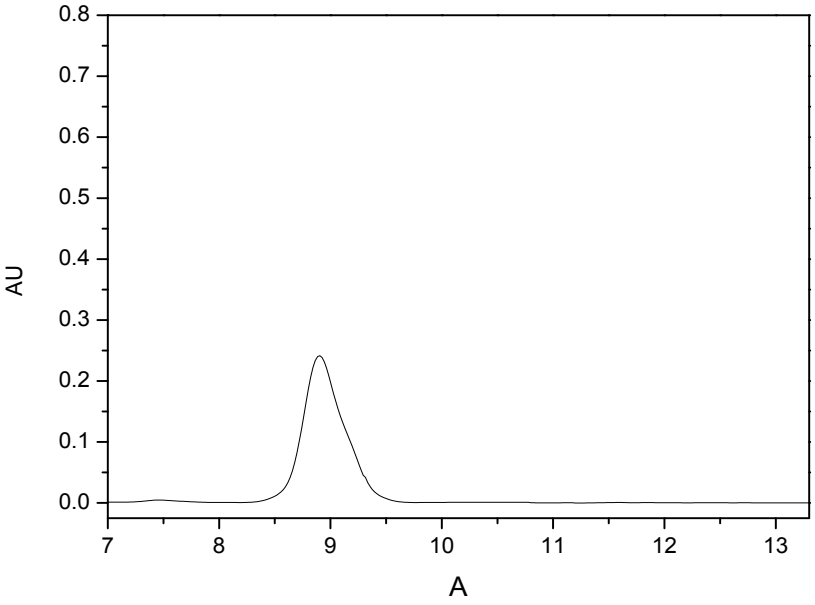
Entry	Product	Area		Retention time (min)		ee (%)
20		*S*	36906	*S*	14.726	99.1
		R	8541265	*R*	18.017	

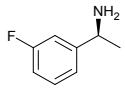


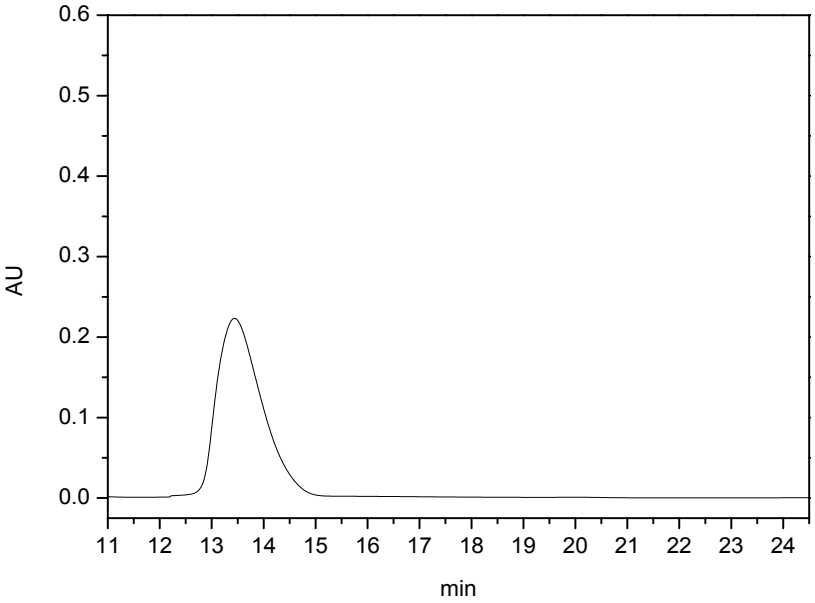
Entry	Product	Area		Retention time (min)		ee (%)
21		*S*	5739527	*S*	8.723	99.2
		R	22897	*R*	12.87	

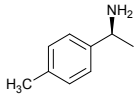


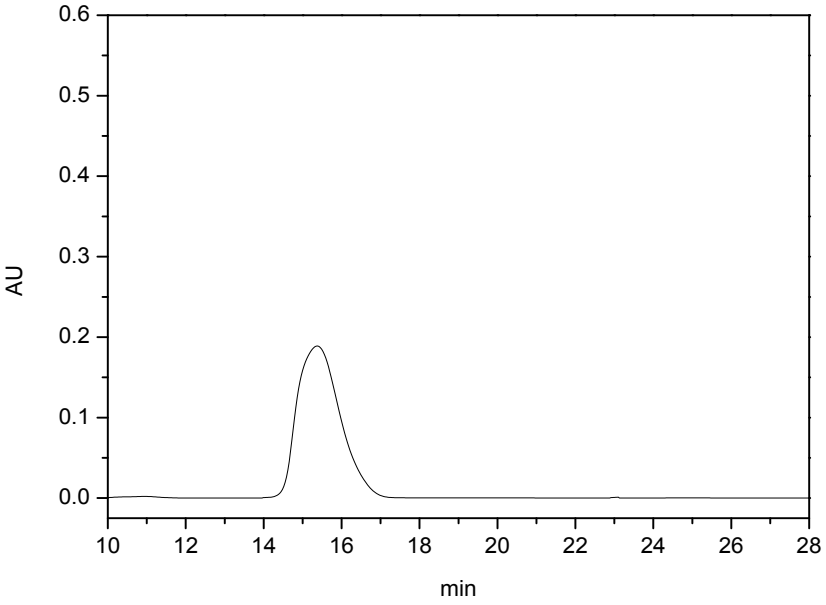
Entry	Product	Area		Retention time (min)		ee (%)
22		*S*	6077843	*S*	7.989	99.3
		R	21141	*R*	13.501	

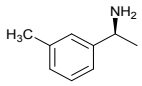


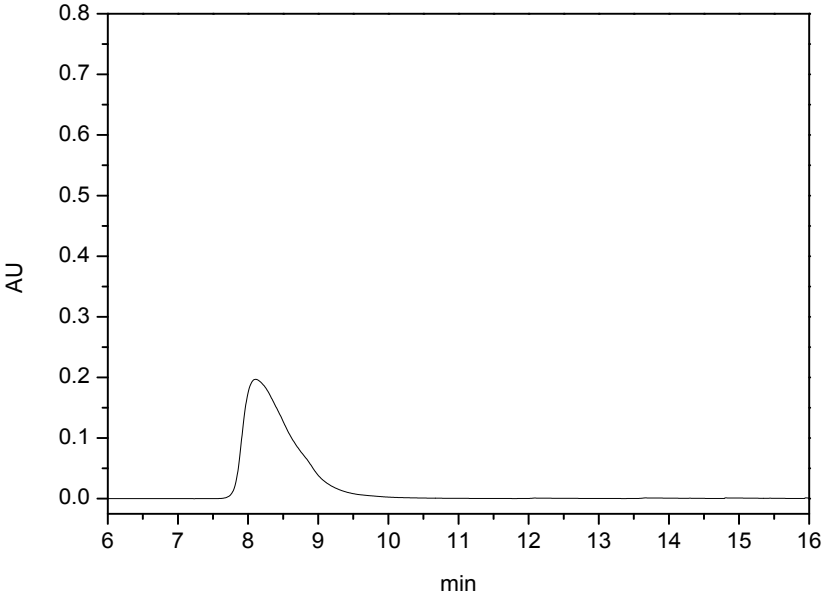
Entry	Product	Area		Retention time (min)		ee (%)
23		*S*	6816269	*S*	8.904	99.5
		R	15516	*R*	11.756	



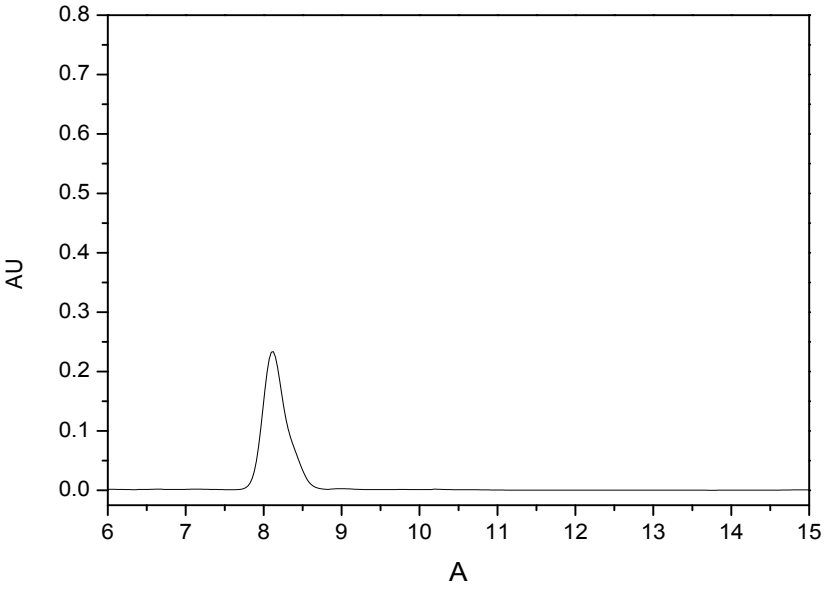
Entry	Product	Area		Retention time (min)		ee (%)
24		*S*	9644929	*S*	13.436	99.2
		R	38429	*R*	21.433	

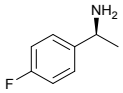


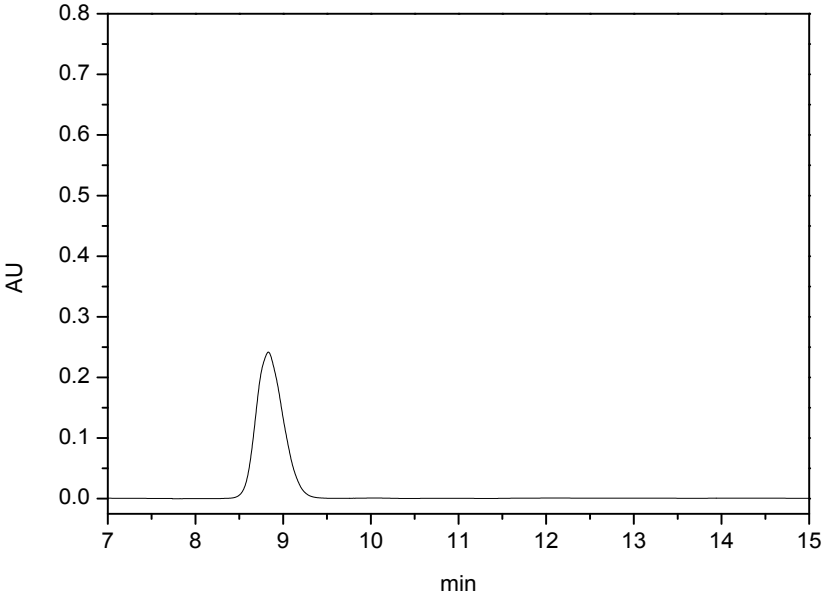
Entry	Product	Area		Retention time (min)		ee (%)
25		*S*	6546329	*S*	15.383	99.7
		R	9076	*R*	18.508	

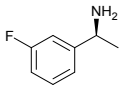


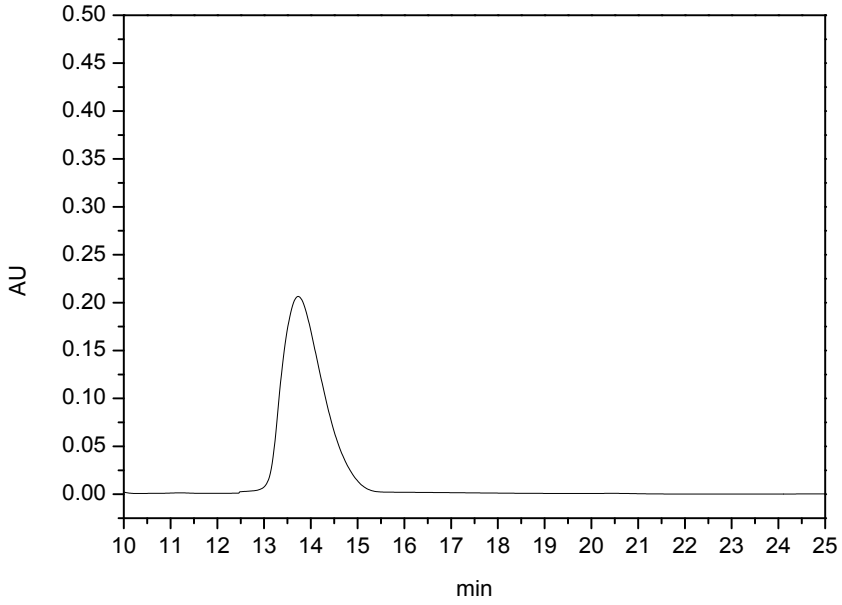
Entry	Product	Area		Retention time (min)		ee (%)
26		*S*	5862781	*S*	8.032	99.1
		R	26297	*R*	12.065	

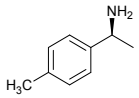


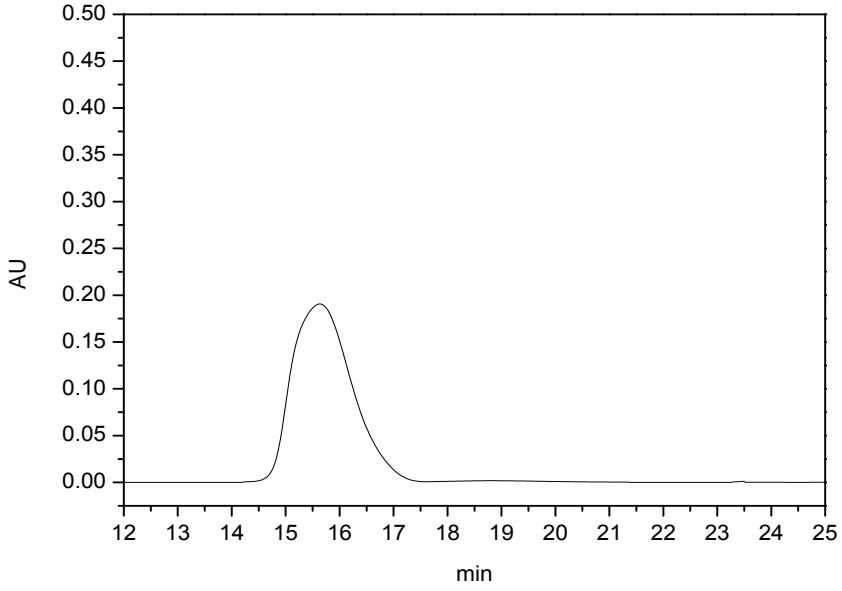
Entry	Product	Area		Retention time (min)		ee (%)
27		*S*	6174564	*S*	8.124	99.7
		R	7928	*R*	13.486	

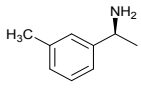


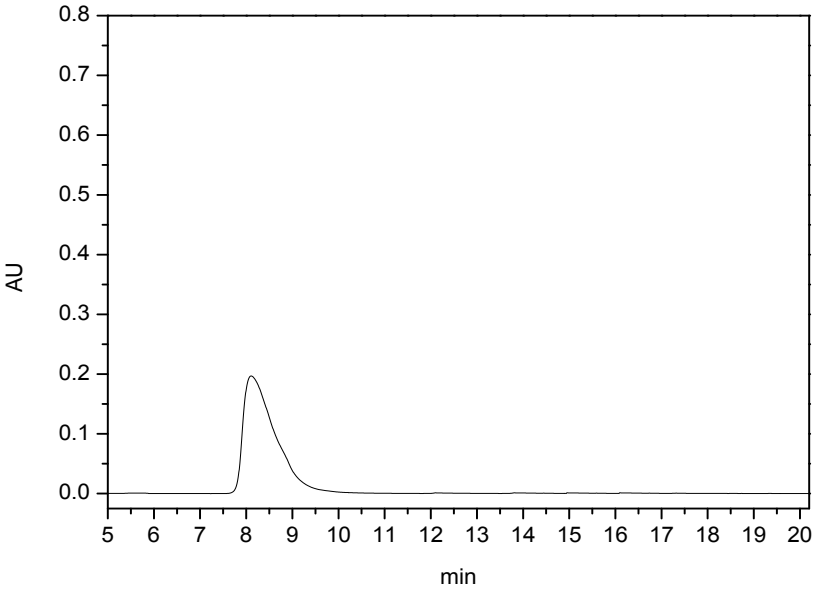
Entry	Product	Area		Retention time (min)		ee (%)
28		*S*	6820783	*S*	8.831	99.6
		R	12695	*R*	11.282	



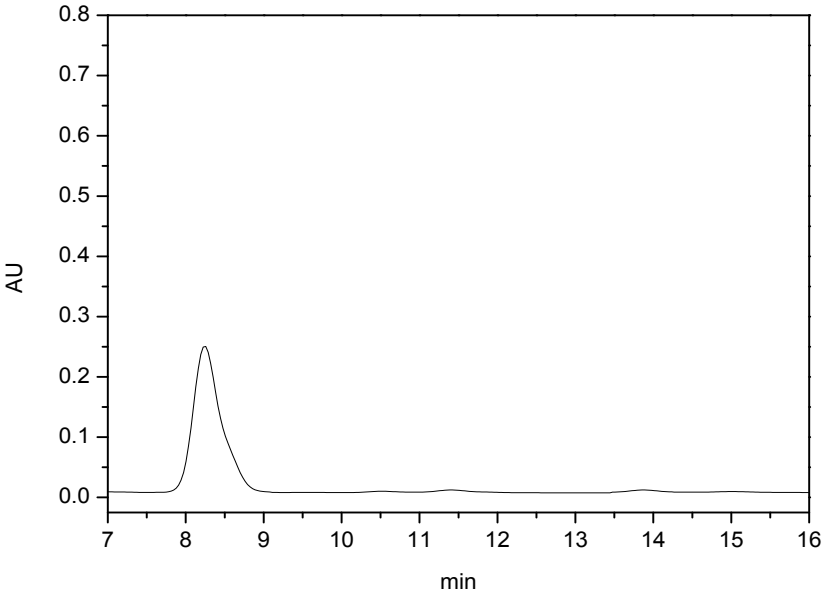
Entry	Product	Area		Retention time (min)		ee (%)
29		*S*	8904406	*S*	13.708	99.8
		R	10233	*R*	21.692	

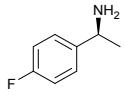


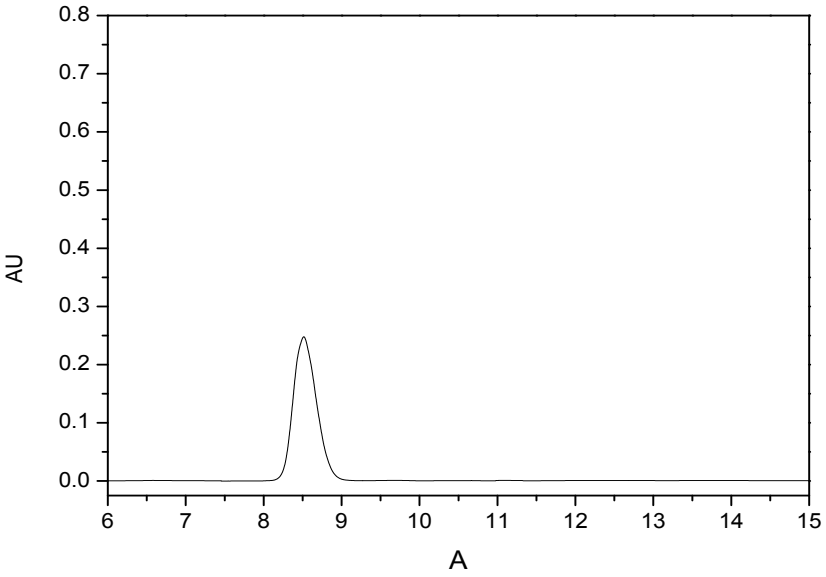
Entry	Product	Area		Retention time (min)		ee (%)
30		*S*	6604877	*S*	15.643	99.8
		R	6340	*R*	18.891	

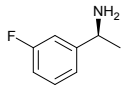


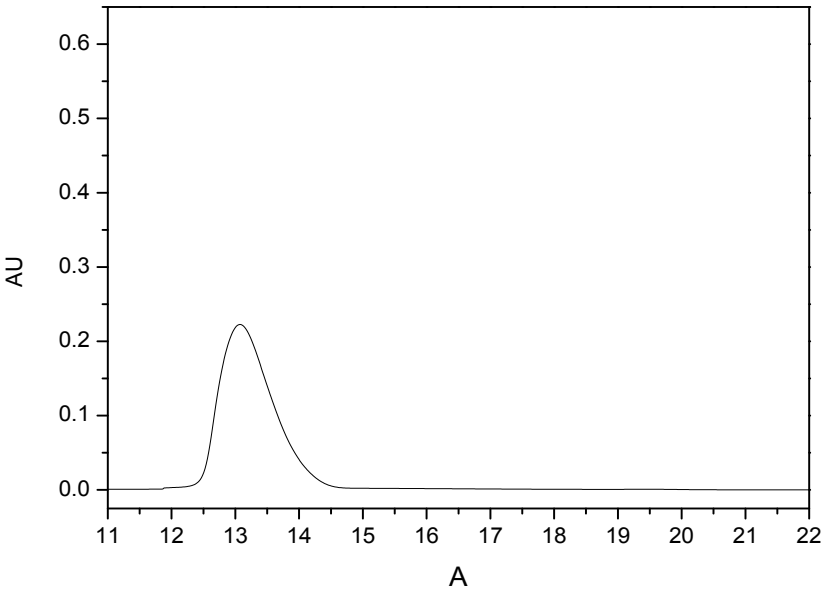
Entry	Product	Area		Retention time (min)		ee (%)
31		*S*	5232965	*S*	8.106	99.0
		R	26032	*R*	12.236	

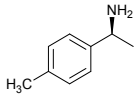


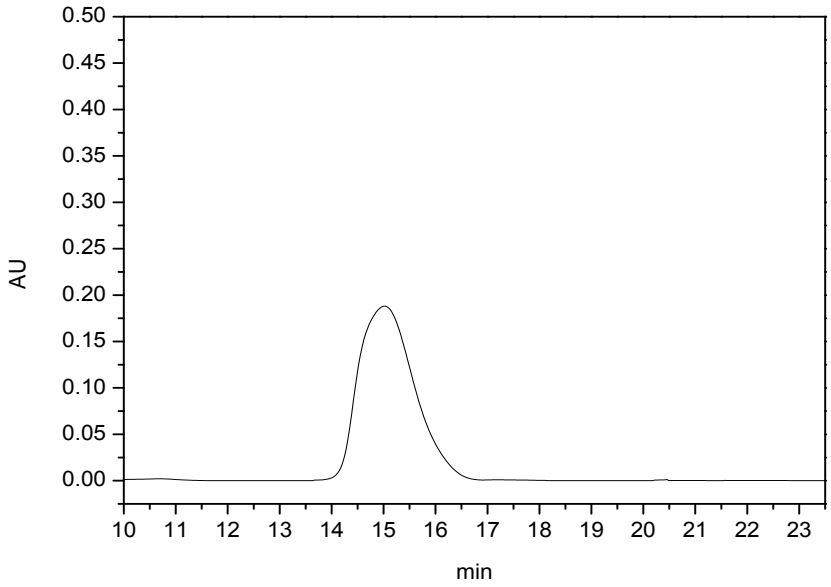
Entry	Product	Area		Retention time (min)		ee (%)
32		*S*	6621965	*S*	8.256	99.0
		R	32610	*R*	13.887	

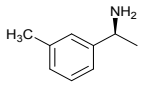


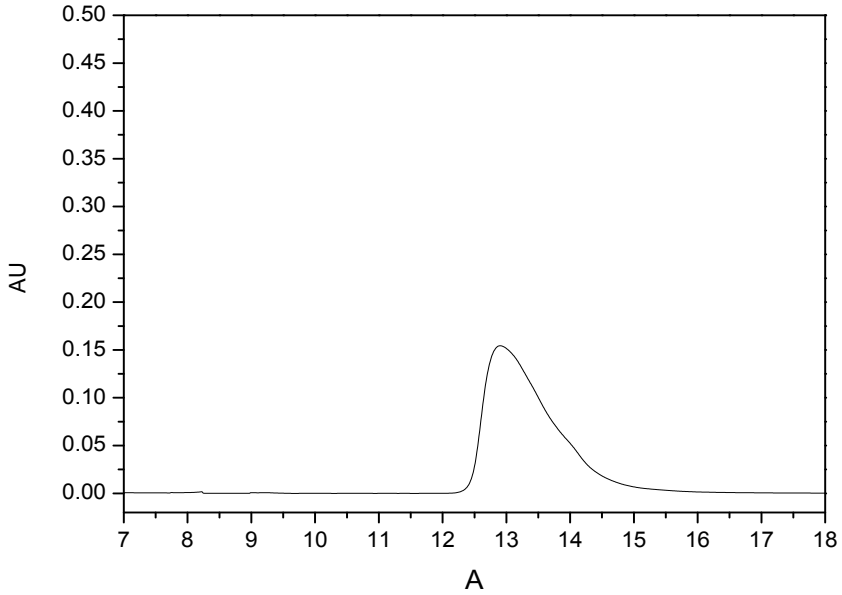
Entry	Product	Area		Retention time (min)		ee (%)
33		*S*	6984412	*S*	8.521	99.2
		R	28212	*R*	11.054	



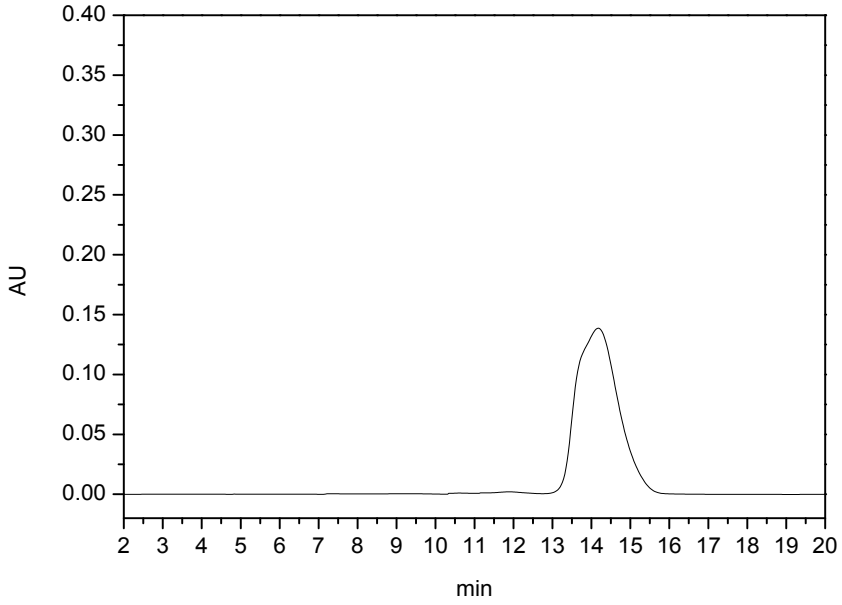
Entry	Product	Area		Retention time (min)		ee (%)
34		*S*	9617726	*S*	13.071	99.9
		R	4750	*R*	21.106	

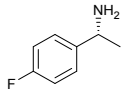


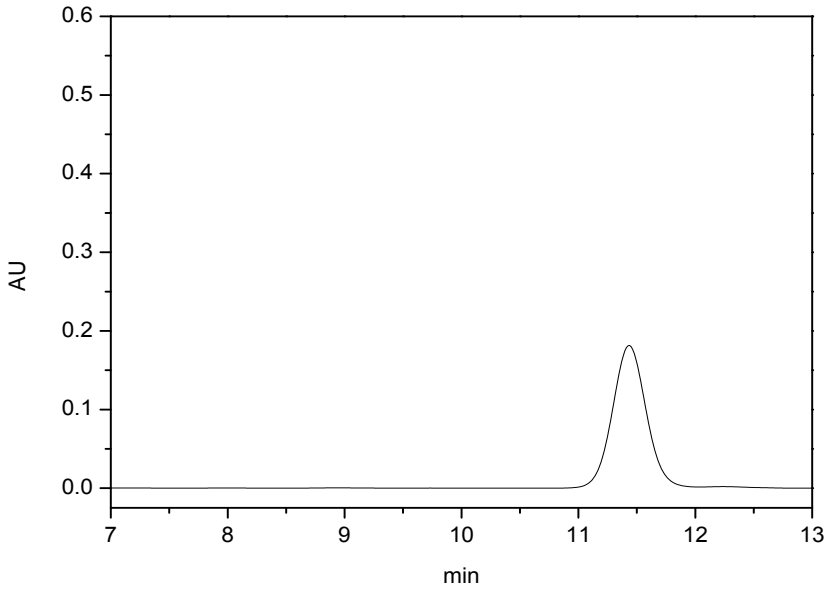
Entry	Product	Area		Retention time (min)		ee (%)
35		*S*	6518267	*S*	15.027	99.1
		R	30833	*R*	18.148	

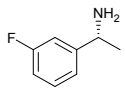


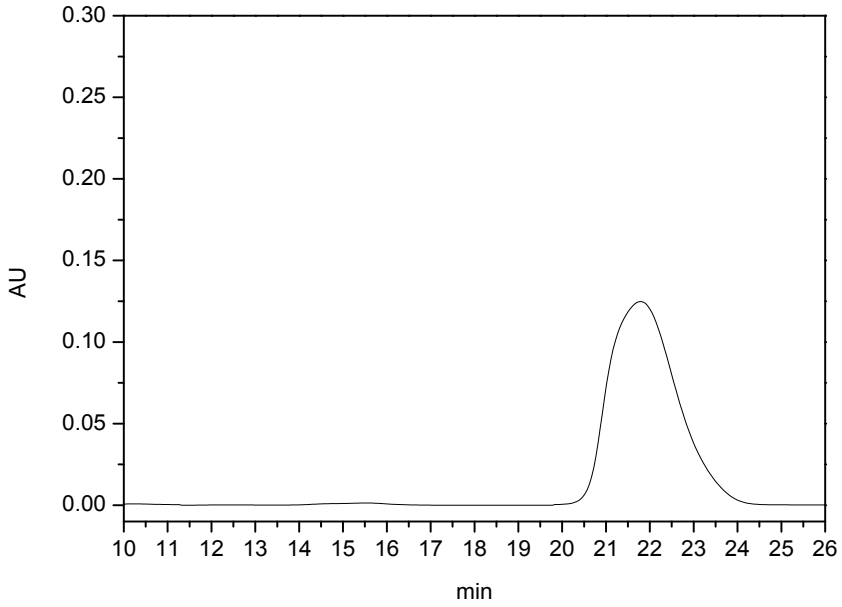
Entry	Product	Area		Retention time (min)		ee (%)
36		*S*	5329	*S*	8.748	99.6
		R	2902719	*R*	12.907	

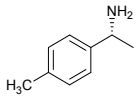


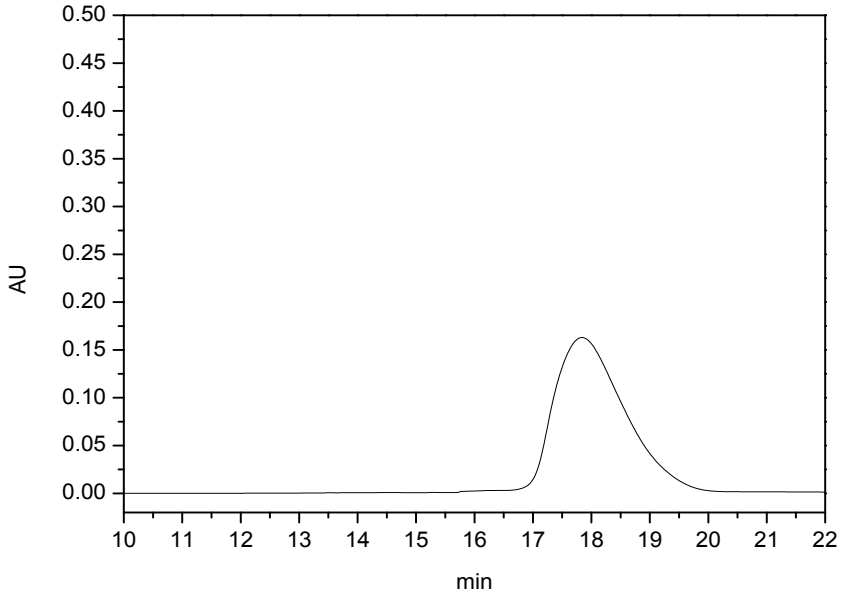
Entry	Product	Area		Retention time (min)		ee (%)
37		*S*	14079	*S*	8.597	99.5
		R	6371842	*R*	14.188	

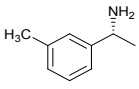


Entry	Product	Area		Retention time (min)		ee (%)
38		*S*	18189	*S*	8.947	99.5
		R	6735651	*R*	11.438	



Entry	Product	Area		Retention time (min)		ee (%)
39		*S*	41750	*S*	13.732	99.0
		R	8686853	*R*	21.803	



Entry	Product	Area		Retention time (min)		ee (%)
40		*S*	40640	*S*	15.299	99.1
		R	8812030	*R*	17.836	

15. References

1. S. S. Kotha, N. Sharma. G. Sekar, *Adv. Synth. Catal*. 2016, 358, 1695.

2. D. Ganapathy, G. Sekar, *Catal. Commun*. 2013, 39, 50.