Supporting Information for:

Amine dehydrogenases: Efficient biocatalysts for the stereoselective reductive amination of carbonyl compounds

Tanja Knaus,[‡] Wesley Böhmer,[‡] Francesco G. Mutti*

Van 't Hoff Institute for Molecular Sciences, University of Amsterdam, Science Park 904, 1098 XH Amsterdam, Netherlands.

[‡] These authors contributed equally to this work

* Corresponding author: f.mutti@uva.nl

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1. Abbreviations

AmDH	amine dehydrogenase (variant)
Bb-PhAmDH	amine dehydrogenase variant originated from the phenylalanine dehydrogenase from *Bacillus badius*.
Rs-PhAmDH	amine dehydrogenase variant originated from the phenylalanine dehydrogenase from *Rhodoccoccus species*.
Ch1-AmDH	chimeric amine dehydrogenase generated through domain shuffling of Bb-PhAmDH variant and L-AmDH variant, the latter originated from the leucine dehydrogenase from *Bacillus stearothermophilus*.
Cb-FDH	formate dehydrogenase (variant) from *Candida boidinii*
GDH	glucose dehydrogenase from *Bacillus subtilis*
ee	enantiomeric excess
ω-TA	ω-transaminase
DCM	dichloromethane
n.a.	not applicable
n.d.	not determined
n.m.	not measureable

2. List of substrates

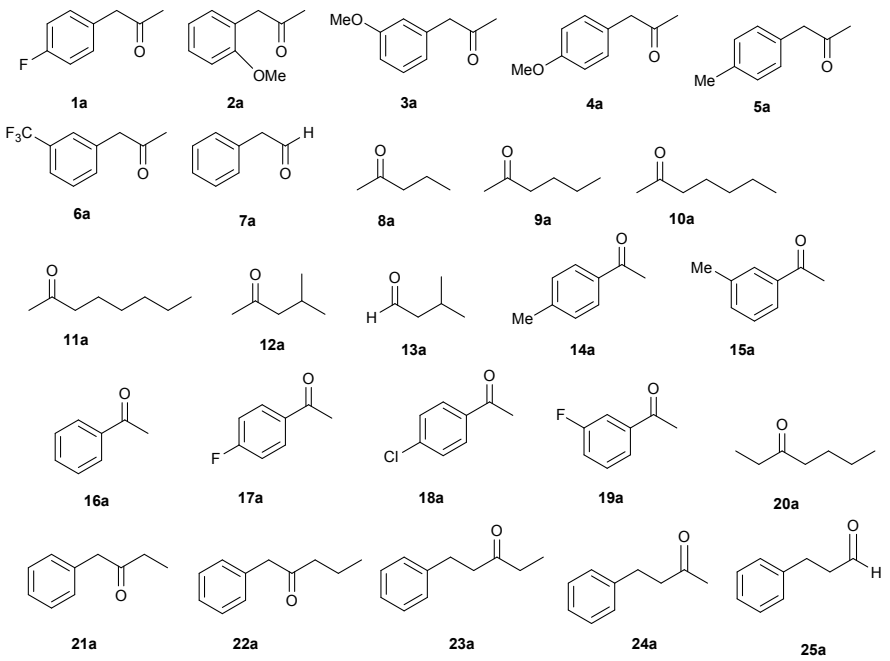


Figure S1. List of substrates used in this study.

3. General Information

Ketones **1a**, **3a**, **6a**, **8a**-**11a**, **14a**-**17a**, **19a**-**24a**, aldehydes **13a** and **25a** were purchased from Sigma-Aldrich (Steinheim, Germany). Ketone **5a** and aldehyde **7a** were purchased from Alfa Aesar (Shore Road, Heysham, UK). Ketones **2a**, **4a**, **12a**, **18a** were purchased from Acros Organics (Geel, Belgium).

Amines (*R*)-**9b,** *rac*-**16b,** (*S*)-**16b**, *rac*-**10b**, **7b** and **13b** were purchased from Sigma-Aldrich (Steinheim, Germany). Enantiopure (*S*) and (*R*)-configured amines **1b**-**6b**, **8b**, **11b**, **14b**-**24b** and terminal amine **25b** were synthesized by stereoselective amination using established enzymatic methods as reported in literature (employing commercially available stereocomplementary ω-transaminases ATA-113, ATA-117 from Codexis, Redwood City, California, US).[1]

Alcohols **8c**, **10c**-**12c** were purchased from Sigma-Aldrich (Steinheim, Germany) and alcohol **9c** was purchased from Acros Organics (Geel, Belgium).

Nicotinamide cofactor (NAD^+) was purchased from Melford Biolaboratories (Chelsworth, Ipswich, UK).

Lysozyme from chicken egg white (3.2 mg, Sigma L6876, lyophilized powder, protein 95%, >40000 U/mg protein) was purchased from Sigma-Aldrich (Steinheim, Germany). Ni^{2+} affinity columns (HisTrap FF, 5 mL) were purchased from GE Healthcare Bio-Sciences (Munich, Germany).

4. Expression and purification of recombinant proteins in *E. coli* host cells

The amine dehydrogenases Bb-PhAmDH and Ch1-AmDH were expressed and purified as described previously.[2]

For recombinant expression of the N-term His6-tagged proteins Rs-PhAmDH and Cb-FDH, 800 mL of LB medium supplemented with the appropriate antibiotic (50 µg/mL kanamycin) were inoculated with 15 mL of an overnight culture of *E. coli* BL21 DE3 cells harbouring the pET28b plasmid with the genes for the expression of the proteins. Cells were grown at 37 °C until an OD_{600} of 0.6 – 0.9 was reached and expression of protein was induced by the addition of IPTG (0.5 mM final concentration). Protein expression was carried out overnight at 20 °C and after harvesting of the cells (4 °C, 4.5 x 10^3 rpm, 15 min), the remaining cell pellet was frozen or directly resuspended in Lysis buffer (50 mM KH_2PO_4, 300 mM NaCl, 10 mM imidazole, pH 8.0) containing lysozyme 1 mg mL^{-1}) and incubated at 4 °C for 35 minutes prior to cell disruption by ultrasonication. After centrifugation (4 °C, 14 x 10^3 rpm, 1 h) the supernatant was filtered through a 0.45 µm filter and protein purification was performed by Ni-NTA affinity chromatography using pre-packed Ni-NTA HisTrap FF columns (GE Healthcare) according to the manufacturer´s instructions. After loading of the filtered lysate, the column was washed with sufficient amounts of washing buffer (50 mM KH_2PO_4, 300 mM NaCl, 25 mM imidazole,pH 8.0), and bound protein was recovered with elution buffer (50 mM KH_2PO_4, 300 mM NaCl, 200 mM imidazole, pH 8.0). The process of purification was analysed by SDS-PAGE and fractions containing sufficient purified protein were pooled and dialyzed over night against potassium phosphate buffer (50 mM, pH 8,). Protein solutions were concentrated and the their concentrations determined spectrophotometrically using an extinction coefficient at 280 nm of 26.6 x 10^3 M^{-1} cm^{-1} for Rs-PhAmDH and 51.4 x 10^3 M^{-1} cm^{-1} for Cb-FDH. A protein yield of 40 mg Rs-PhAmDH/L cell culture and 100 mg Cb-FDH/L cell culture was obtained.

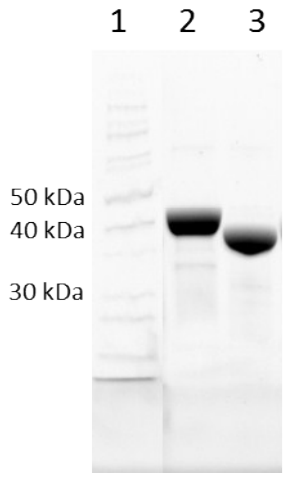


Figure S2. SDS-PAGE of purified proteins. Lane 1: PageRuler™ Unstained Protein Ladder (ThermoFisher Scientific), lane 2: Cb-FDH, lane 3: Rs-PhAmDH.

5. Biocatalytic transformations

5.1. Study of the stability of nicotinamide coenzyme at different pH values

The stability of NADH was studied under the following conditions:

(1) Potassium phosphate buffer (pH 6.5, 50 mM)

(2) Hepes buffer (pH 8.8, 50 mM)

(3) Sodium carbonate buffer (pH 10, 50 mM)

(4) Sodium hydroxide (0.1 N, pH 13)

The decomposition of NADH vs. time was monitored spectrophotometrically at a wavelength of 325 nm.

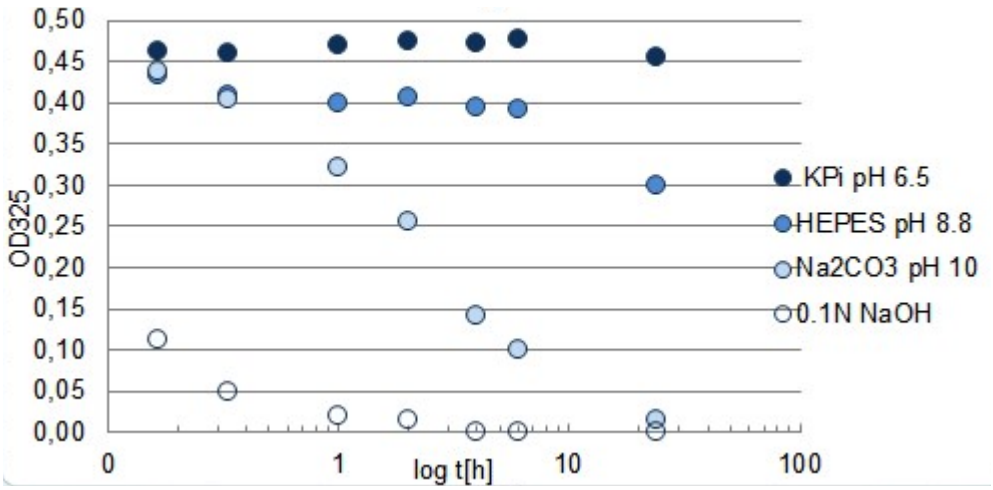


Figure S3. Stability of NADH at different pH values vs. time; x-axis: time in logarithmic scale; y-axis: absorbance of NADH at 325 nm.

5.2. *Reductive amination of **1a** by Bb-PhAmDH, **10a** by Rs-PhAmDH and **24a** by Ch1-AmDH in different buffer systems using GDH for NAD^+/NADH regeneration*

In this experiment two buffer systems were used, *i.e.* ammonium chloride buffer (final concentration 1 M, pH 8.7) and ammonium formate buffer (final concentration 1.005 M, pH 8.5).

NH_4Cl/NH_4OH buffer was prepared by dissolving NH_4Cl (2.85 g, 53 mmol) in water (45 mL). NH_4OH (1.238 mL, 6.4 M) was added and the pH was adjusted with 1 M HCl.

$HCOONH_4/NH_4OH$ buffer was prepared by dissolving $HCOONH_4$ (3.16 g, 50.1 mmol) in water (45 mL). NH_4OH (0.458 mL, 6.4 M) was added and the pH was adjusted with 1 M formic acid.

Two different stock solutions were prepared containing glucose (60 mM) and GDH (150 U per sample):

Stock solution 1

NAD^+ (3.5 mg, 0.005 mmol, final concentration 1 mM) was dissolved in NH_4Cl/NH_4OH buffer (4.84 mL). Glucose (37.6 mg, 0.209 mmol) and GDH (300 U mL^{-1}) were dissolved in ammonium chloride buffer (3.27 mL) containing the NAD^+.

Stock solution 2

NAD^+ (6.7 mg, 0.01 mmol, final concentration 1 mM) was dissolved in $HCOONH_4/NH_4OH$ buffer (9.24 mL). Glucose (36.9 mg, 0.205 mmol) and GDH (300 U mL^{-1}) were dissolved in ammonium formate buffer (3.21 mL) containing 1 mM NAD^+.

Biocatalytic reactions

Reaction samples were prepared by combining one of the stock solution above (470 µL), the substrate (20 mM), and the AmDH (*i.e.* Bb-PhAmDH (115 µM), Rs-PhAmDH (121 µM) or Ch1-AmDH (82.1 µM)). The reactions were run at 30 °C in an incubator for 21 hours (190 rpm). Work-up was performed by the addition of KOH (100 µL, 10 M), followed by the extraction with dichloromethane (1x600 µL). After centrifugation, the organic layer was dried with $MgSO_4$. Conversion and enantiomeric excess were determined as reported in the experimental section of the main manuscript. Analytical methods as well as retention times are reported in paragraph 7.

5.3. *Reductive amination of **1a** by Bb-PhAmDH, **10a** by Rs-PhAmDH and **24a** by Ch1-AmDH using formate dehydrogenase Cb-FDH as NAD^+/NADH cofactor recycling enzyme*

$HCOONH_4/NH_4OH$ buffer (1.005 M, pH 8.5) was prepared as explained in paragraph 5.2 containing NAD^+ (final concentration 1 mM).

Biocatalytic transformations were prepared by combining the buffer/NAD^+ solution (470 µL), the Cb-FDH (14 µL), the substrate (20 mM), and the AmDH (*i.e.* Bb-PhAmDH (115 µM), Rs-PhAmDH (121 µM) or Ch1-AmDH (82.1 µM)). The reactions were run at 30 °C in an incubator for 21 hours (190 rpm). Work-up was performed by the addition of KOH (100 µL, 10 M), followed by the extraction with dichloromethane (1x600 µL). After centrifugation, the organic layer was dried with $MgSO_4$. Conversion and enantiomeric excess were determined as reported in the experimental section of the main manuscript. Analytical methods as well as retention times are reported in paragraph 7 and the results are summarized in Table 1 in the main paper.

*5.4. Reductive amination of **1a** by Bb-PhAmDH, **10a** by Rs-PhAmDH and **24a** by Ch1-AmDH at varying substrate concentrations using Cb-FDH as cofactor recycling enzyme*

Biocatalytic transformations were run under the same reaction conditions as described in 5.3. Substrate concentrations were varied from 30 mM, 40 mM, to 50 mM per sample. The results are reported in Table S1.

Reaction conditions: final volume = 0.5 mL; [NAD^+] = 1 mM; [substrate] = 30-50 mM; [AmDH] = 80-120 µM; [Cb-FDH] = 14 µM; incubation at 190 rpm and 30 °C for 21 h.

Table S1. Reductive amination of **1a**, **10a**, and **24a** by Bb-PhAmDH, Rs-PhAmDH and Ch1-AmDH, respectively, using Cb-FDH for coenzyme recycling in ammonium formate buffer (1.005 M, pH 8.5) at varying substrate concentrations.

entry	enzyme	substrate	substrate [mM]	enzyme [µM]	conversion [%] *	*ee*%
1	Bb-PhAmDH	**1a**	30 mM	115	96	>99
2	Bb-PhAmDH	**1a**	40 mM	115	91	>99
3	Bb-PhAmDH	**1a**	50 mM	115	88	>99
4	Rs-PhAmDH	**24a**	30 mM	130	>99	>99
5	Rs-PhAmDH	**24a**	40 mM	130	>99	>99
6	Rs-PhAmDH	**24a**	50 mM	130	>99	>99
7	Ch1-AmDH	**10a**	30 mM	80	>99	>99
8	Ch1-AmDH	**10a**	40 mM	80	>99	>99
9	Ch1-AmDH	**10a**	50 mM	80	>99	>99

* Conversion depicted here is the average value obtained from two independent experiments.

*5.5. Reductive amination of **10a** by Rs-PhAmDH and **24a** by Ch1-AmDH at varying enzyme concentrations and Cb-FDH as cofactor recycling enzyme*

Biocatalytic transformations were run under the same reaction conditions as described in 5.4. Enzyme concentrations were varied from 33-115 µM per sample. The results are reported in Table S2.

Table S2. Reductive amination of **10a**, and **24a** by Rs-PhAmDH and Ch1-AmDH at varying enzyme concentrations using Cb-FDH for coenzyme recycling in ammonium formate buffer (1.005 M, pH 8.5).

entry	enzyme	substrate	enzyme [μM]	conversion [%] *	*ee*%
1	Rs-PhAmDH	**24a**	121	>99	>99%
2	Rs-PhAmDH	**24a**	96	>99	>99%
3	Rs-PhAmDH	**24a**	73	>99	>99%
4	Rs-PhAmDH	**24a**	50	>99	>99%
5	Ch1-AmDH	**10a**	82	99	>99%
6	Ch1-AmDH	**10a**	66	99	>99%
7	Ch1-AmDH	**10a**	49	98	>99%
8	Ch1-AmDH	**10a**	32	95	>99%

* Conversion depicted here is the average value obtained from two independent experiments.

*5.6. Time study for the reductive amination of **1a** by Bb-PhAmDH at different temperatures*

The reactions were prepared as previously described with a substrate concentration of 50 mM and an enzyme concentration of 46 μM. The progress of the reaction was monitored at different temperatures (from 30 to 60 °C) vs. time (1h, 2h, 3h, 5h, 8h, 16h, 21h, 24h, etc.). Every biocatalytic transformation was performed in duplicate and the results were averaged. At 60 °C no measurable conversion could be observed. Conversion and enantiomeric excess were determined as described in paragraph 7 and the results are summarized in Table S3.

Reaction conditions: final volume = 0.5 mL; ammonium formate buffer (1.005 M, pH 8.7); [NAD^+] = 1 mM; [substrate] = 50 mM; [Bb-PhAmDH] = 46 μM; [Cb-FDH] = 14 μM; incubation at 190 rpm.

Table S3. Conversions [%] for the reductive amination of **1a** by Bb-PhAmDH at different temperatures vs. time.

entry	substrate	time [h]	20 °C	30 °C	40 °C	50 °C
1	**1a**	1	3	6	8.4	7
2	**1a**	2	5	10	19	10
3	**1a**	3	7	14	29	12
4	**1a**	5	9	21	43	14
5	**1a**	8	10	25	46	12
6	**1a**	16	25	52	72	n.d.
7	**1a**	18	n.d.	n.d.	n.d.	19
8	**1a**	21	37		84	n.d.
9	**1a**	24	37	63	83	21
10	**1a**	end	79	94	87	n.d.

Conversion depicted here is the average value obtained from two independent experiments.

*5.7. Time study for the reductive amination of **24a** by Rs-PhAmDH at different temperatures*

The reactions were prepared as described in paragraph 5.4 using a substrate concentration of 50 mM and an enzyme concentration of 48 µM. The progress of the reaction was monitored at different temperatures (from 30 to 70 °C) vs. time (1h, 2h, 3h, 5h, 8h, 16h, 21h, 24h, etc.). Every biocatalytic transformation was performed in duplicate and the results were averaged. At 70 °C no measurable conversion could be observed. Conversion and enantiomeric excess were determined as described in paragraph 7 and the results are summarized in Table S4.

Reaction conditions: final volume = 0.5 mL; ammonium formate buffer (1.005 M, pH 8.5); [NAD^+] = 1 mM; [substrate] = 50 mM; [Rs-PhAmDH] = 48 µM; [Cb-FDH] = 14 µM; incubation at 190 rpm.

Table S4. Conversions [%] for the reductive amination of **24a** by Rs-PhAmDH at different temperatures vs. time.

entry	substrate	time [h]	20 °C	30 °C	40 °C	50 °C	60 °C
1	**24a**	1	83	80	83	83	73
3	**24a**	2	91	94	93	92	80
5	**24a**	3	98	99	94	95	83
7	**24a**	5	>99	>99	>99	96	83
9	**24a**	8	>99	>99	>99	96	86
11	**24a**	16	>99	>99	>99	n.d.	n.d.
13	**24a**	18	n.d.	n.d.	n.d.	96	76
14	**24a**	21	>99	>99	>99	n.d.	n.d.
16	**24a**	24	>99	>99	>99	98	92
18	**24a**	30	n.d.	n.d.	n.d.	98	93
19	**24a**	end	>99	>99	>99	n.d.	n.d.

Conversion depicted here is the average value obtained from two independent experiments.

*5.8. Time study for the reductive amination of **10a** by Ch1-AmDH at different temperatures*

The reactions were prepared as previously described with a substrate concentration of 50 mM substrate and an enzyme concentration of 33 µM. The conversion was monitored at different temperatures (from 30 to 60 °C) vs. time (1h, 2h, 3h, 5h, 8h, 16h, 21h, 24h, etc.). Every biocatalytic transformation was performed in duplicate and the results were averaged. A preliminary time study at 70 °C provided mediocre conversions and therefore this temperature was not thoroughly analysed (data at 18 h and 48 h are available). Conversion and enantiomeric excess were determined as described in paragraph 7 and the results are summarized in Table S5.

Reaction conditions: final volume = 0.5 mL; ammonium formate buffer (1.005 M, pH 8.5); [NAD^+] = 1 mM; [substrate] = 50 mM; [Ch1-AmDH] = 33 µM; [Cb-FDH] = 14 µM; incubation at 190 rpm.

Table S5. Conversions [%] for the reductive amination of **10a** by Ch1-AmDH at different temperatures over time.

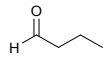
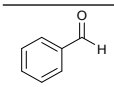
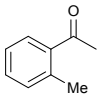
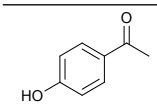
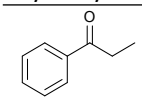
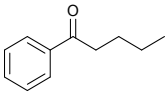
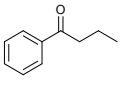
entry	substrate	time [h]	20 °C	30 °C	40 °C	50 °C	60 °C	70 °C
1	**10a**	1	23	38	58	58	61	n.d.
2	**10a**	2	38	61	80	73	74	n.d.
3	**10a**	3	56	78	90	79	82	n.d.
4	**10a**	5	69	90	94	93	82	n.d.
5	**10a**	8	80	94	97	90	84	n.d.
6	**10a**	16	86	98	98	n.d.	n.d.	n.d.
7	**10a**	18	n.d.	n.d.	n.d.	96	92	8
8	**10a**	21	98	98	99	n.d.	n.d.	n.d.
9	**10a**	24	98	97	>99	n.d.	93	n.d.
10	**10a**	30	n.d.	n.d.	n.d.	98	97	n.d.
11	**10a**	end	>99	>99	>99	>99	>99	<10

Conversion depicted here is the average value obtained from two independent experiments.

5.9. Reductive amination of a library of ketones and aldehydes by Bb-PhAmDH, Rs-PhAmDH and Ch1-AmDH

The reactions were prepared and performed following the optimal reaction conditions as described in the experimental section of the main manuscript. The main results are reported in the manuscript: tables 2-5. Substrates that were tested and found not to be accepted or only accepted poorly are reported in Table S6.

Table S6. Attempts of reductive amination on substrates that were not converted or converted at a very moderate extent using Bb-PhAmDH, Rs-PhAmDH, and Ch1-AmDH.

substrate	enzyme	enzyme [µM]	time [h]	conversion [%]	*ee*% (*R*)
 2-butanone	Ch1-AmDH	32.2	24	8	n.d.
	Rs-PhAmDH	51.4	24	4	n.d.
 butanal	Ch1-AmDH	32.2	24	n.m.	n.a.
	Rs-PhAmDH	51.4	24	n.m.	n.a.
 Benzaldehyde	Ch1-AmDH	32.2	24	n.m.	n.a.
	Rs-PhAmDH	51.4	24	n.m.	n.a.
 *o*-methylacetophenone	Ch1-AmDH	32.2	24	n.m.	n.d.
		32.2	48	n.m.	n.d.
	Rs-PhAmDH	51.4	24	n.m.	n.d.
		51.4	48	n.m.	n.d.
 *p*-hydroxyacetophenone	Ch1-AmDH	32.2	24	n.m.	n.d.
		32.2	48	n.m.	n.d.
	Rs-PhAmDH	51.4	24	n.m.	n.d.
		51.4	48	n.m.	n.m.
 propiophenone	Ch1-AmDH	91.8	48	8	n.d.
	Rs-PhAmDH	102.6	48	n.m.	n.d.
 valerophenone (butyl phenyl ketone)	Ch1-AmDH	91.8	48	2	n.d.
	Rs-PhAmDH	102.6	48	2	n.d.
 butyrophenone	Ch1-AmDH	91.8	48	n.m.	n.d.
	Rs-PhAmDH	102.6	48	1	n.d.

5.10. *Additional data set for substrates that are moderately converted or not converted by some of the amine dehydrogenases from this study*

Table S7. Reductive amination of phenyl-propanone derivatives with Bb-PhAmDH. Reaction conditions: final volume = 0.5 mL; ammonium formate buffer (1.005 M, pH 8.5); [NAD^+] = 1 mM; [substrate] = 50 mM; [Cb-FDH] = 14 µM; incubation at 190 rpm, 30 °C.

entry	substrate	enzyme [µM]	time [h]	conversion substrate [%]	*ee*% (*R*)
1	**2a**	51	48	10	>99
2	**3a**	51	48	21	>99
3	**4a**	51	48	3	n.d.
4	**5a**	51	48	7	>99
5	**6a**	51	48	no conv.	-
6	**21a**	51	48	8	>99

n.d. = not determined due to too low conversion

Table S8. Additional data for the reductive amination of substrates with Ch1-AmDH. Reaction conditions: final volume = 0.5 mL; ammonium formate buffer (1.005 M, pH 8.5); [NAD^+] = 1 mM; [substrate] = 50 mM; [Cb-FDH] = 14 µM; incubation at 190 rpm, 30 °C.

entry	substrate	enzyme [µM]	time[h]	conversion substrate[%]	*ee*% (*R*)
1	**18a**	32	48	2	n.d.
2	**21a**	32	24	14	>99
3	**22a**	92	48	4	>99
4	**23a**	92	48	1	n.d.
6	**25a**	92	48	70	n.a.

n.d. = not determined due to too low conversion
n.a. = not applicable

6. NMR spectra (recorded on a Brucker AMX 400 (400 Mhz, ^{1}H) in $CDCl_3$)

Figure S4. 1H-NMR of the preparative synthesis of (*R*)-**4j** by Rs-PhAmDH (CDCl3, 400 Mhz).

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DB1701-30m-B: constant pressure 13.5 psi, T injector 300 °C, split ratio 40:1, T initial 60 °C, hold 6.5 min; gradient 20 °C/min up to 100 °C, hold 1 min, gradient 20 °C/min up to 280 °C, hold 1 min.

DB1701-60m: constant pressure 13.5 psi, T injector 300 °C, split ratio 40:1, T initial 80 °C, hold 6.5 min, gradient 5 °C/min up to 160 °C, hold 2 min; gradient 20 °C/min up to 280 °C, hold 1 min.

DEX-CB-A: constant flow 1.4 mL/min, T injector 250 °C, split ratio 40:1, T initial 100 °C, hold 2 min; gradient 1 °C/min up to130 °C, hold 5 min; gradient 10 °C/min up to 170 °C, hold 10 min.; gradient 10 °C/min up to 180 °C, hold 1 min.

DEX-CB-B: constant flow 1.4 ml/min, T injector 250 °C, split ratio 40:1, T initial 100 °C, hold 2 min; gradient1 °C/min up to 118 °C, hold 5 min; gradient 10 °C/min up to 170 °C hold 10 min, gradient 10 °C/min up to 180 °C, hold 1 min.

7.2. Retention times

Table S9. GC retention times for measuring the conversion of substrates **1**-**25a** and **1**-**25b**.

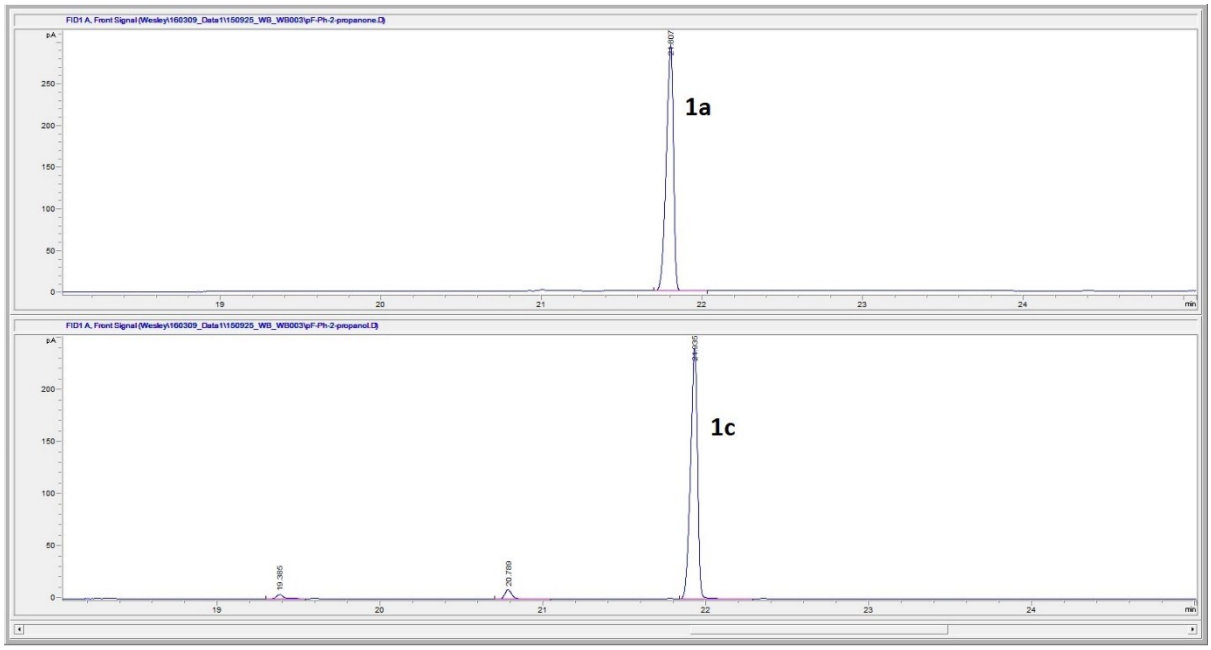
number	retention time [min]	number	retention time [min]	number	retention time [min]	GC method
1a	21.8			**1c**	21.9	DB1701-60m
2a	27.7	**2b**	26.5			DB1701-60m
3a	28.4	**3b**	27.6			DB1701-60m
4a	28.7	**4b**	27.8			DB1701-60m
5a	24.4	**5b**	22.5			DB1701-60m
6a	22.3	**6b**	20.1			DB1701-60m
7a	17.7	**7b**	18.6			DB1701-60m
8a	3.0	**8b**	2.6			DB1701-30m-B
9a	4.7	**9b**	3.7			DB1701-30m-B
10a	4.8	**10b**	3.9	**10c**	5.0	DB1701-30m-A
11a	7.7	**11b**	6.0	**11c**	8.1	DB1701-30m-A
12a	3.7	**12b**	3.1			DB1701-30m-B
13a	2.6	**13b**	2.8	**13c**	3.9	DB1701-30m-B
14a	13.3	**14b**	11.7	**14c**	13.0	DB1701-30m-A
15a	22.0	**15b**	19.7			DB1701-60m
16a	10.7	**16b**	9.3	**16c**	11.0	DB1701-30m-A
17a	10.6	**17b**	9.8	**17c**	11.7	DB1701-30m-A
18a	14.3	**18b**	14.0	**18c**	15.4	DB1701-30m-A
19a	10.4	**19b**	10.0	**19c**	11.9	DB1701-30m-A
20a	4.6	**20b**	3.8			DB1701-60m
21a	24.1	**21b**	22.6			DB1701-60m
22a	15.6	**22b**	15.0			DB1701-60m
23a	16.2	**23b**	15.5			DB1701-60m
24a	24.9	**24b**	23.3	**24c**	25.3	DB1701-60m
25a	13.0	**25b**	14.6	**25c**	14.6	DB1701-60m

Table S10. GC retention times for measuring the enantiomeric excess of substrates **1**-**24b**.

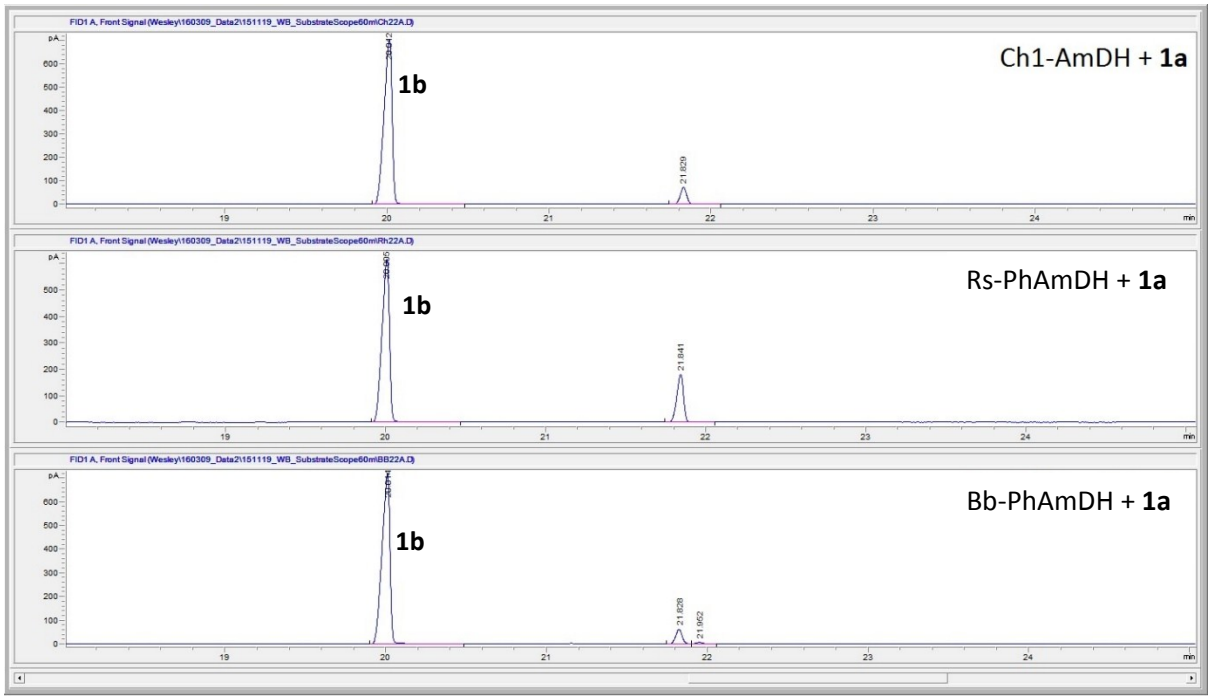
number	retention time [min]	number	retention time [min]	GC method
(*S*)-**1b**	30.8	(*R*)-**1b**	31.3	DEX-CB-A
(*S*)-**2b**	30.9	(*R*)-**2b**	32.0	DEX-CB-A
(*S*)-**3b**	36.1	(*R*)-**3b**	36.4	DEX-CB-A
(*S*)-**4b**	37.4	(*R*)-**4b**	37.7	DEX-CB-A
(*S*)-**5b**	32.2	(*R*)-**5b**	32.3	DEX-CB-A
(*S*)-**6b**	30.4	(*R*)-**6b**	30.7	DEX-CB-A
(*S*)-**8b**	13.1	(*R*)-**8b**	14.1	DEX-CB-B
(*S*)-**9b**	14.4	(*R*)-**9b**	15.6	DEX-CB-B
(*S*)-**10b**	16.3	(*R*)-**10b**	17.7	DEX-CB-A
(*S*)-**11b**	23.9	(*R*)-**11b**	25.2	DEX-CB-A
(*S*)-**12b**	14.4	(*R*)-**12b**	14.7	DEX-CB-B
(*S*)-**14b**	39.5	(*R*)-**14b**	40.0	DEX-CB-A
(*S*)-**15b**	30.6	(*R*)-**15b**	30.9	DEX-CB-A
(*S*)-**16b**	32.8	(*R*)-**16b**	34.5	DEX-CB-A
(*S*)-**18b**	43.0	(*R*)-**18b**	44.4	DEX-CB-A
(*S*)-**19b**	34.1	(*R*)-**19b**	36.2	DEX-CB-A
(*S*)-**20b**	13.7	(*R*)-**20b**	14.9	DEX-CB-A
(*S*)-**21b**	31.5	(*R*)-**21b**	31.6	DEX-CB-A
(*S*)-**22b**	42.0	(*R*)-**22b**	42.1	DEX-CB-A
(*S*)-**23b**	44.1	(*R*)-**23b**	44.3	DEX-CB-A
(*S*)-**24b**	43.1	(*R*)-**24b**	43.5	DEX-CB-A

7.3. GC chromatograms

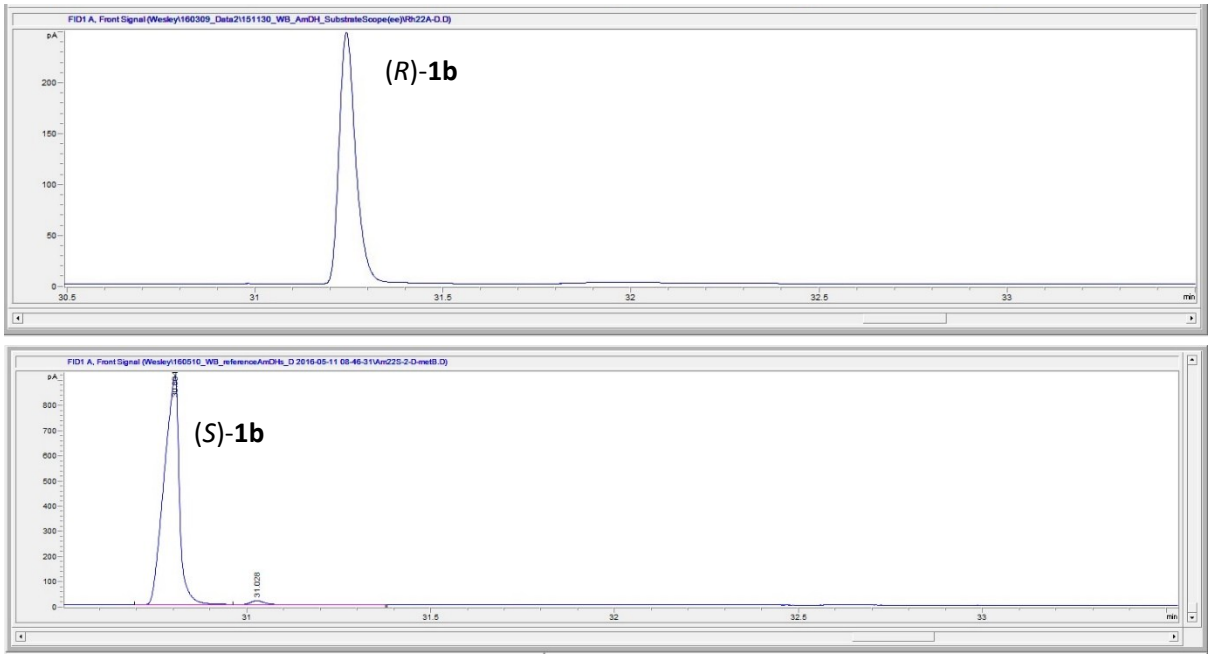
Reference compounds **1a** and **1c**



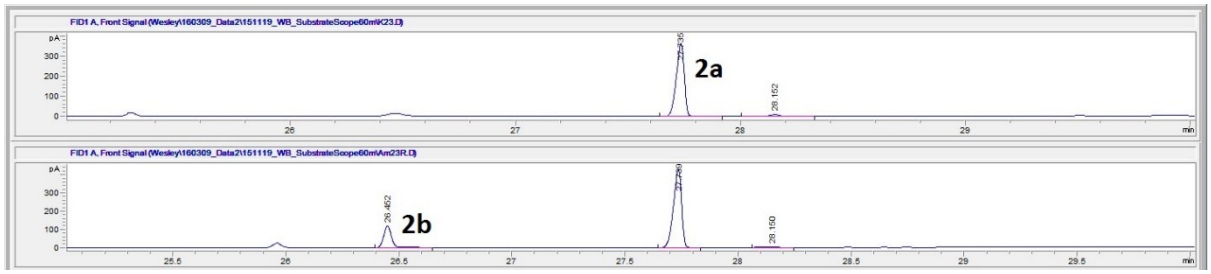
Reductive amination of **1a**



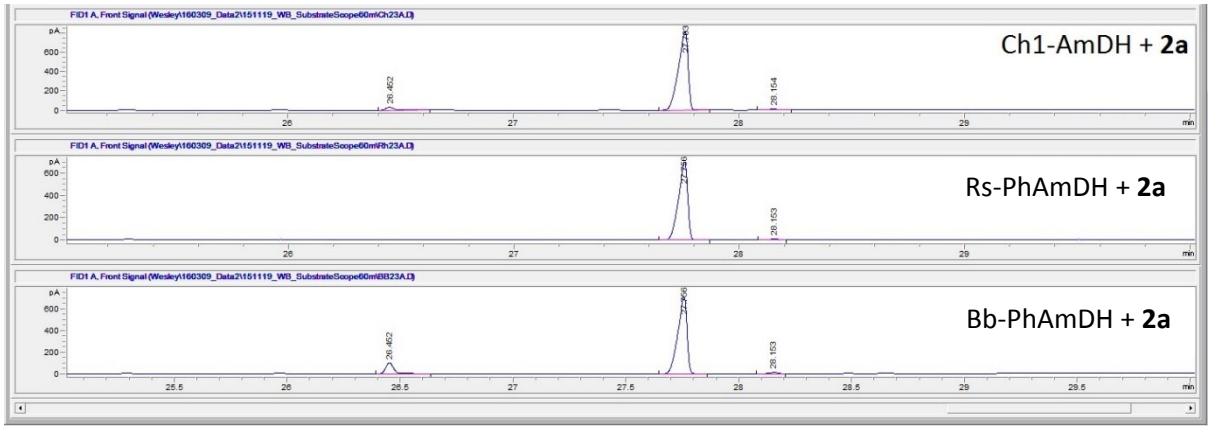
Enantiomeric excess of **1b**



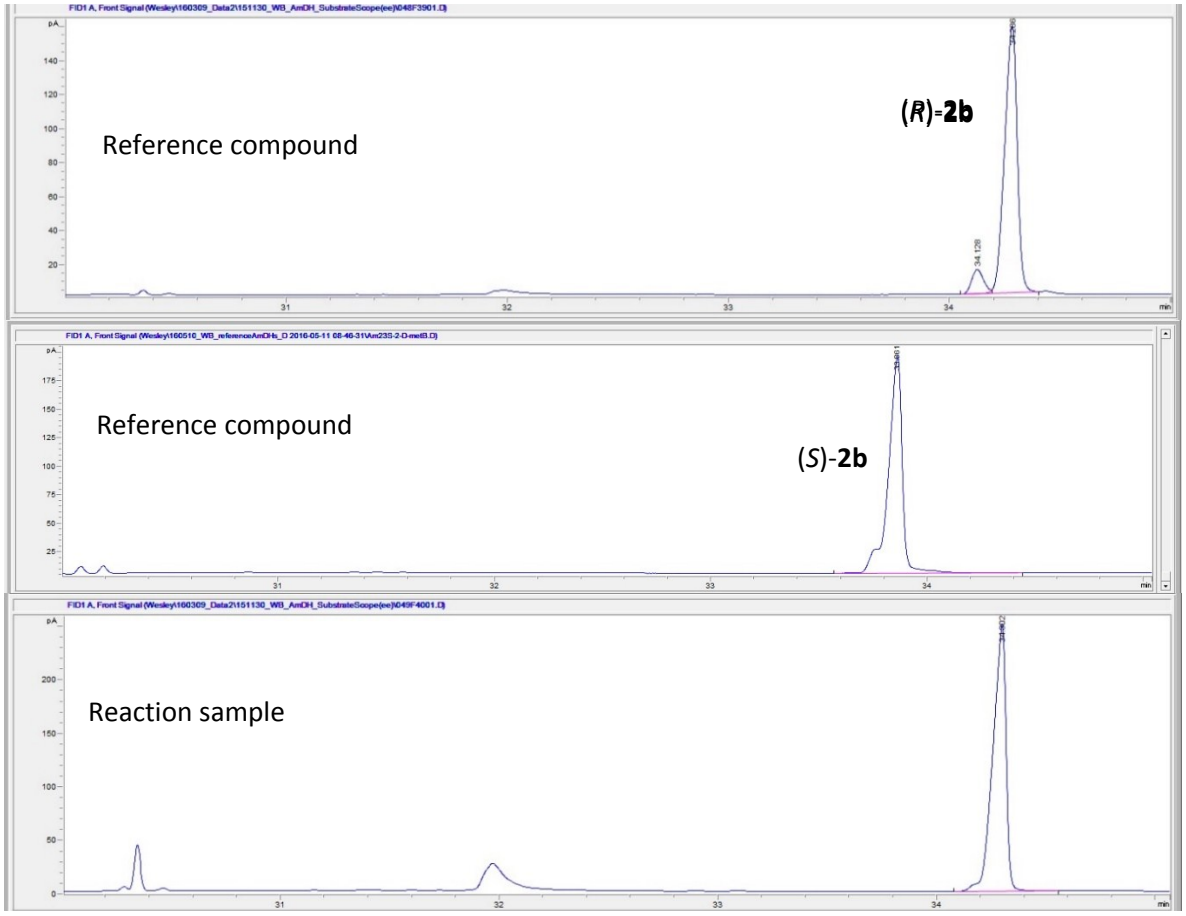
Reference compounds **2a** and **2b**



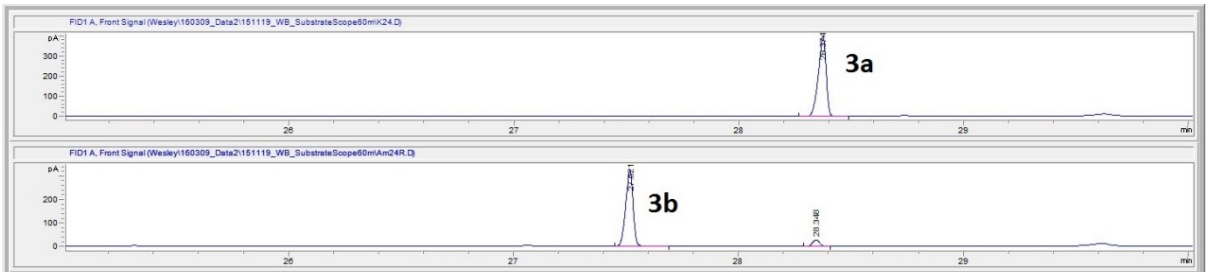
Reductive amination of **2a**



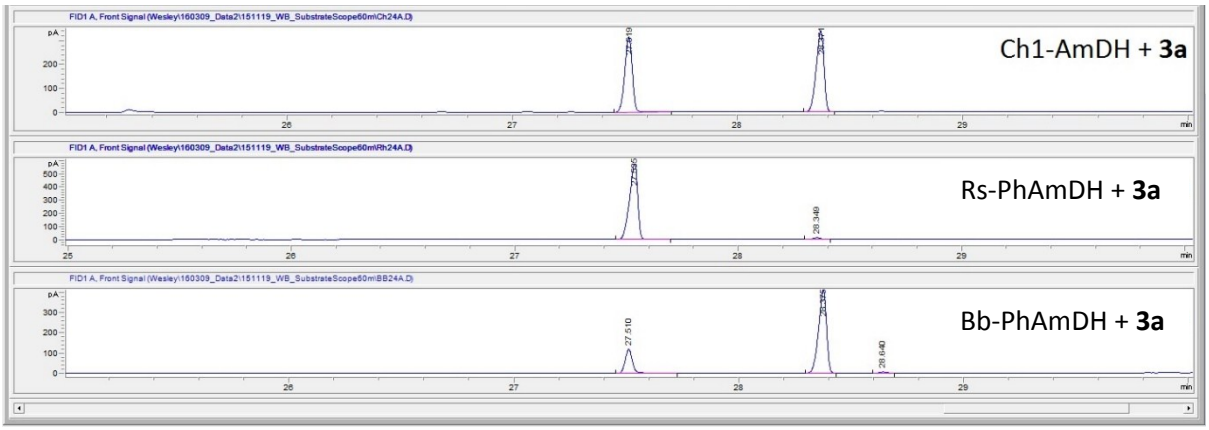
Enantiomeric excess of **2b**



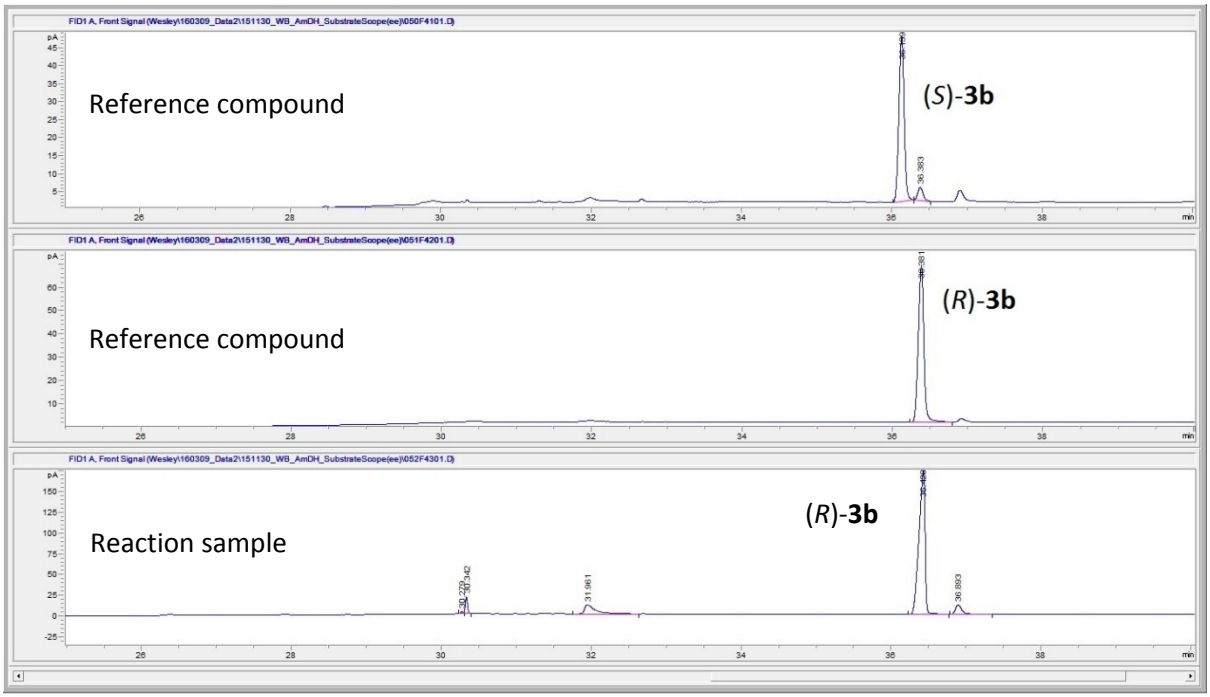
Reference compounds **3a** and **3b**



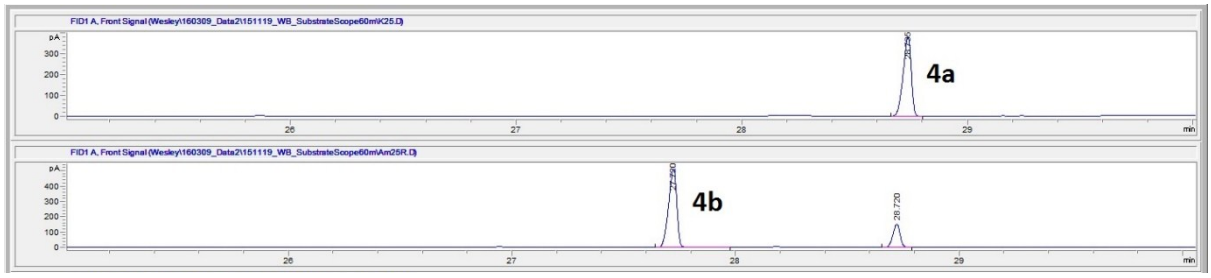
Reductive amination of **3a**



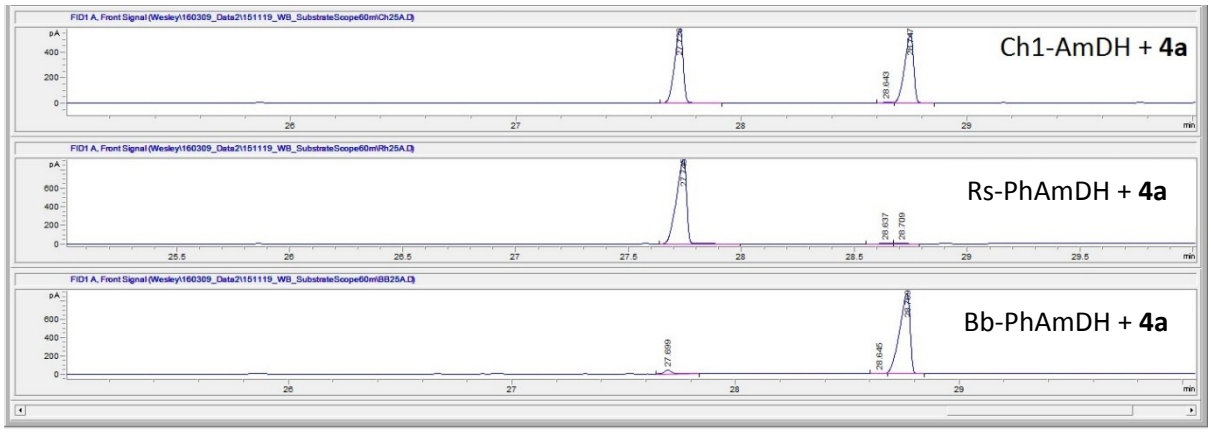
Enantiomeric excess of **3b**



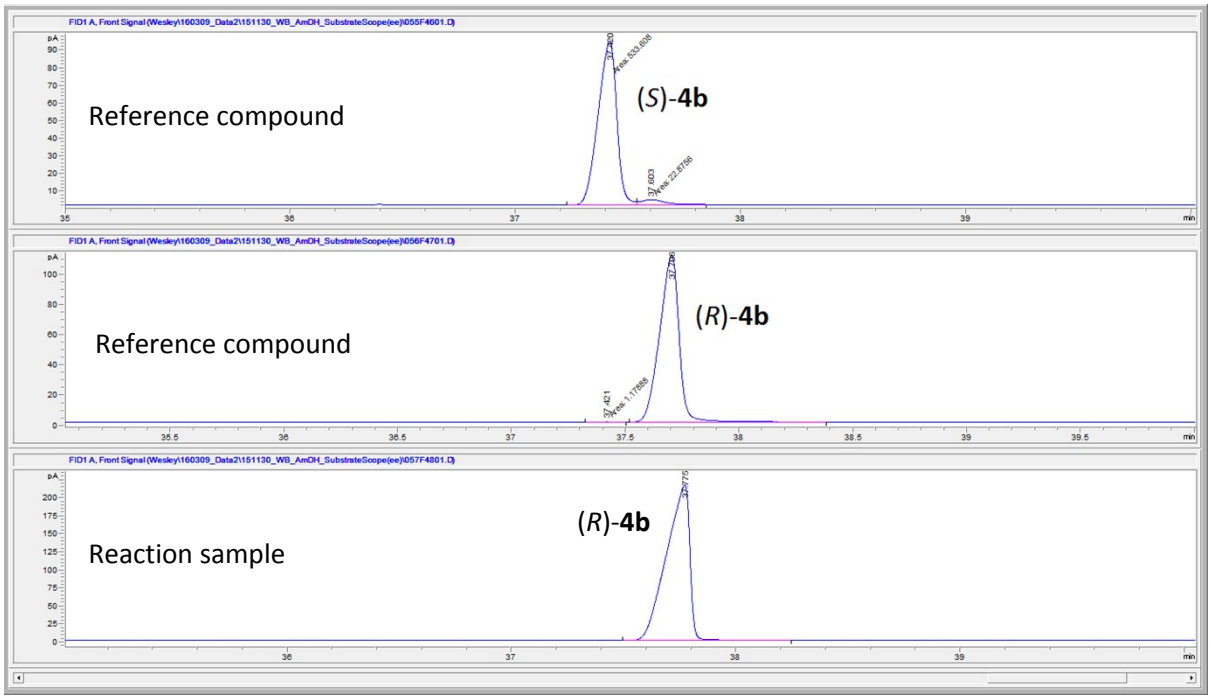
Reference compounds **4a** and **4b**



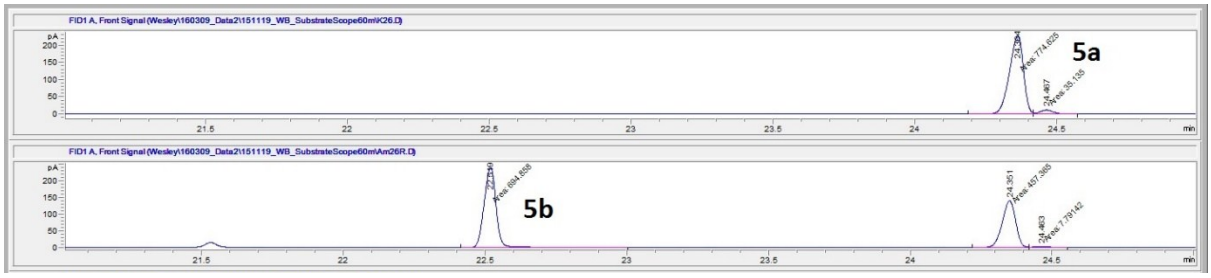
Reductive amination of **4a**



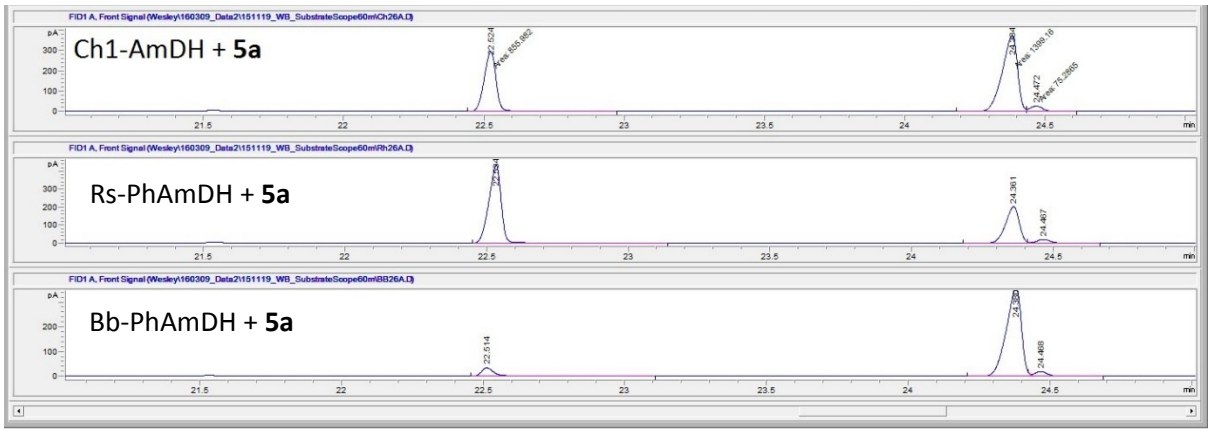
Enantiomeric excess of **4b**



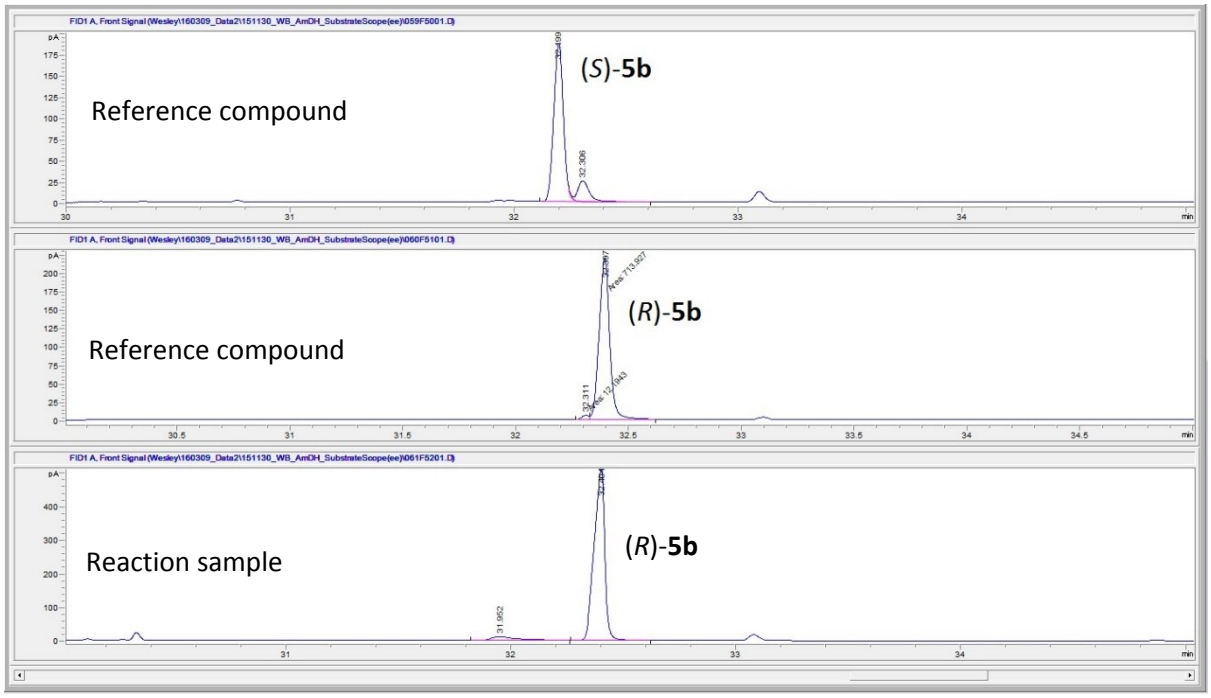
Reference compounds **5a** and **5b**



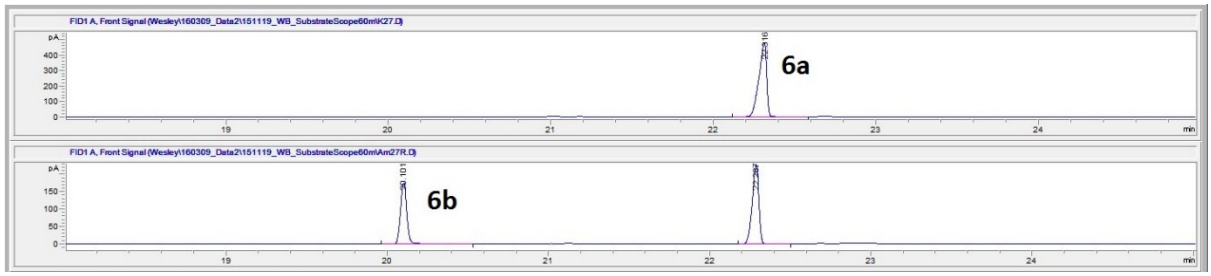
Reductive amination of **5a**



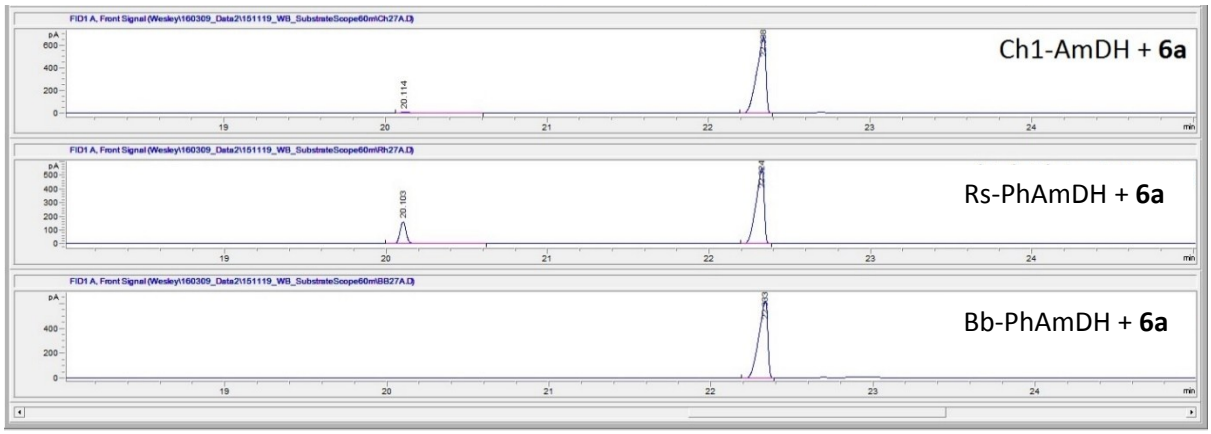
Enantiomeric excess of **5b**



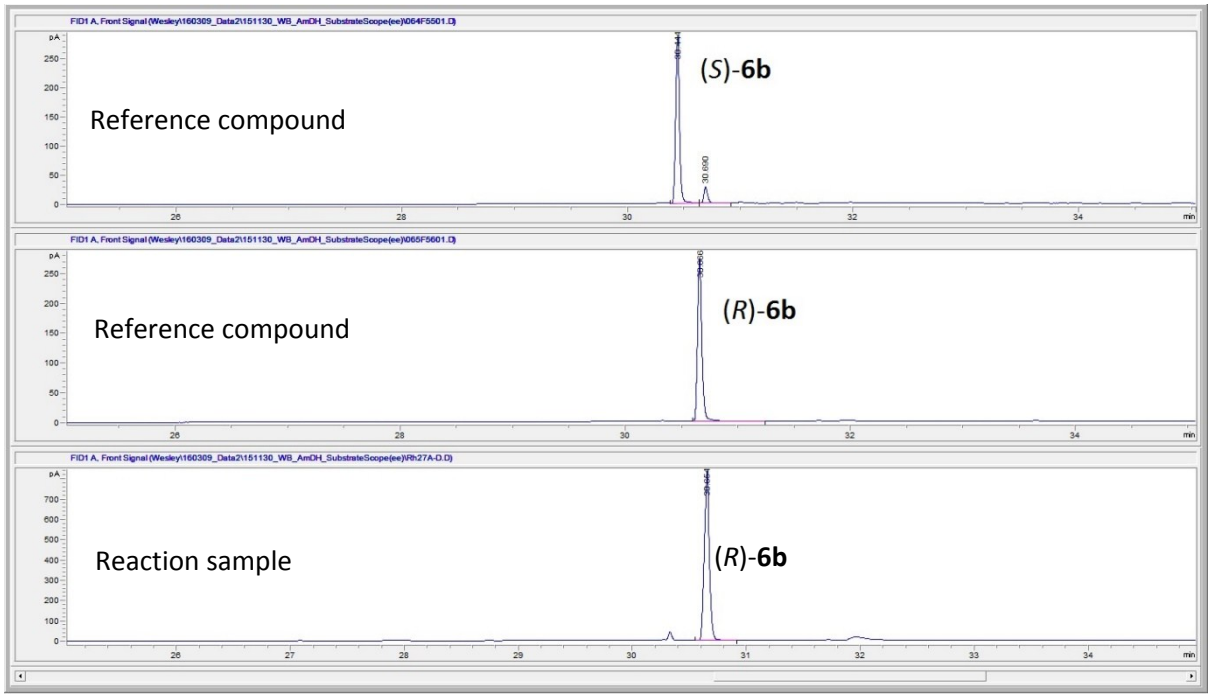
Reference compounds **6a** and **6b**



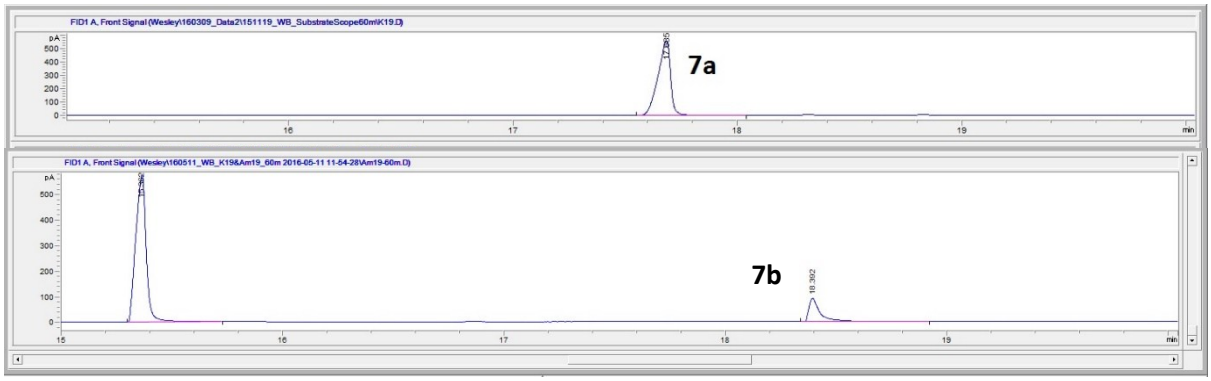
Reductive amination of **6a**



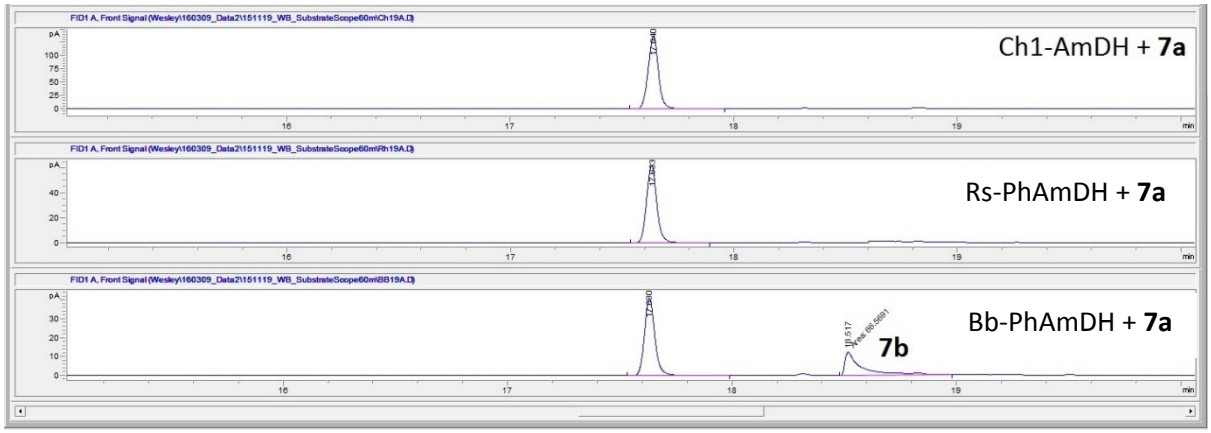
Enantiomeric excess of **6b**



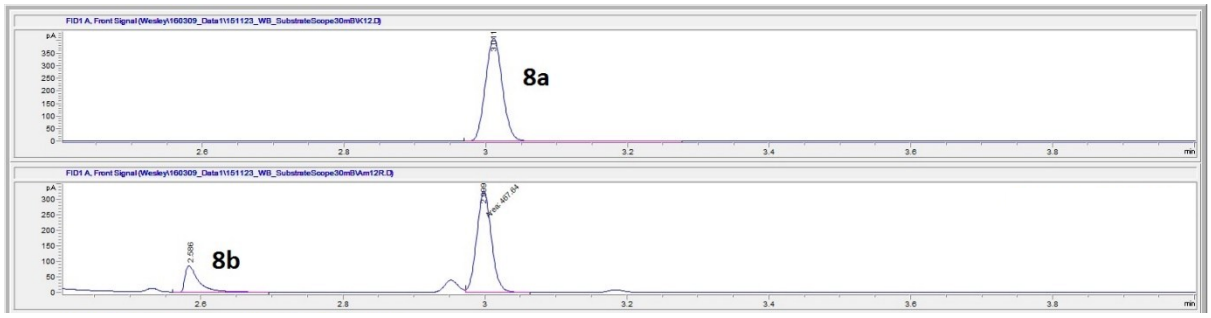
Reference compounds **7a** and **7b**



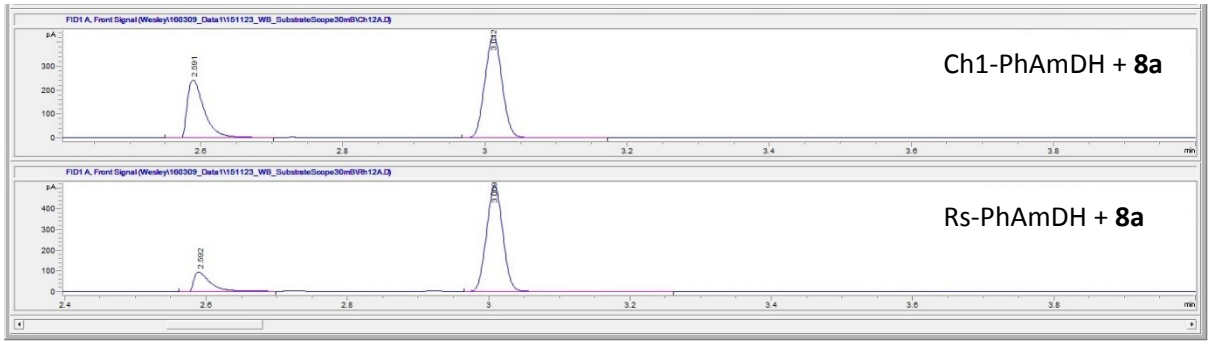
Reductive amination of **7a**



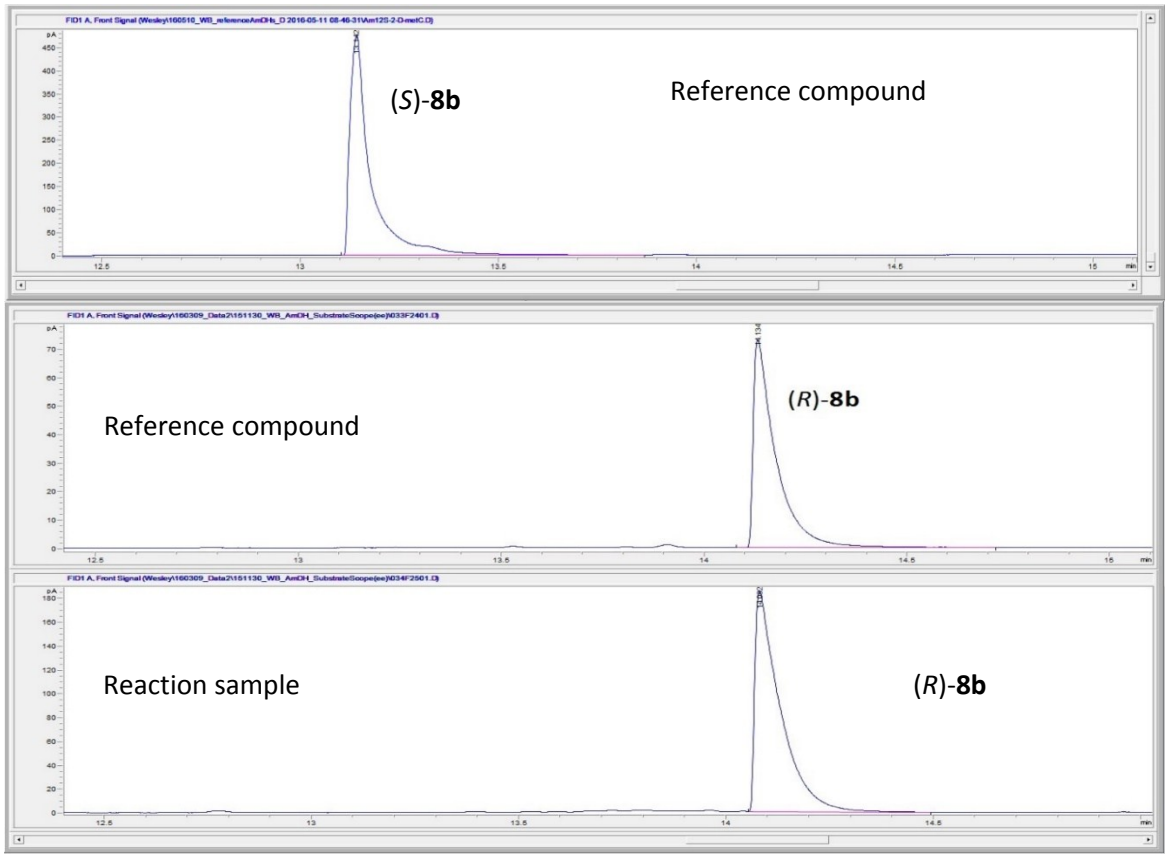
Reference compounds **8a** and **8b**



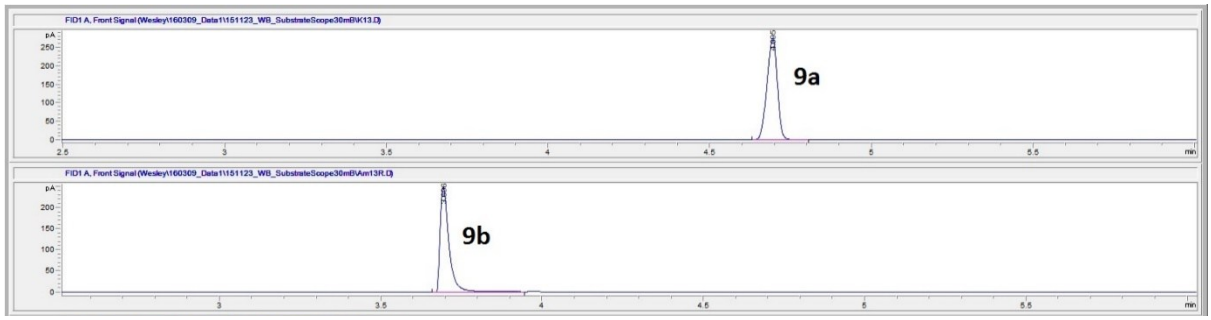
Reductive amination of **8a**



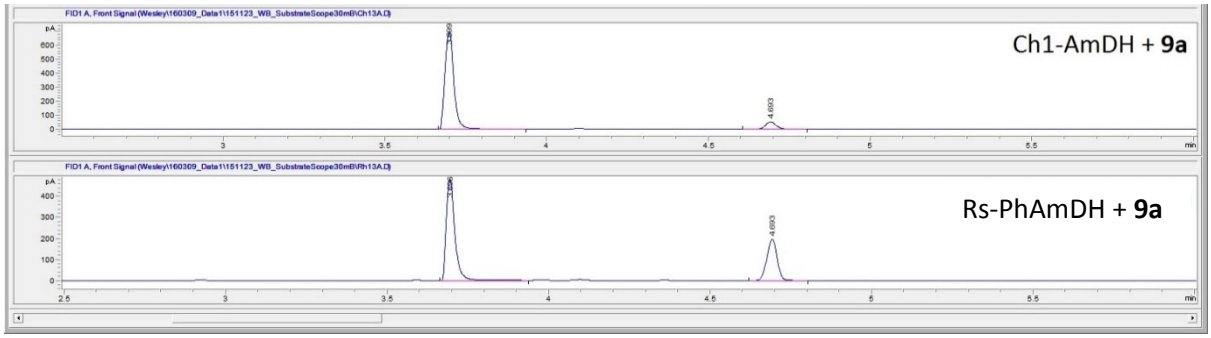
Enantiomeric excess of **8b**



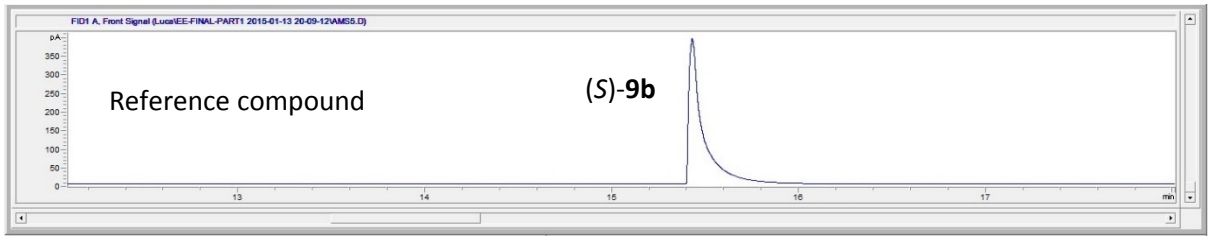
Reference compounds **9a** and **9b**

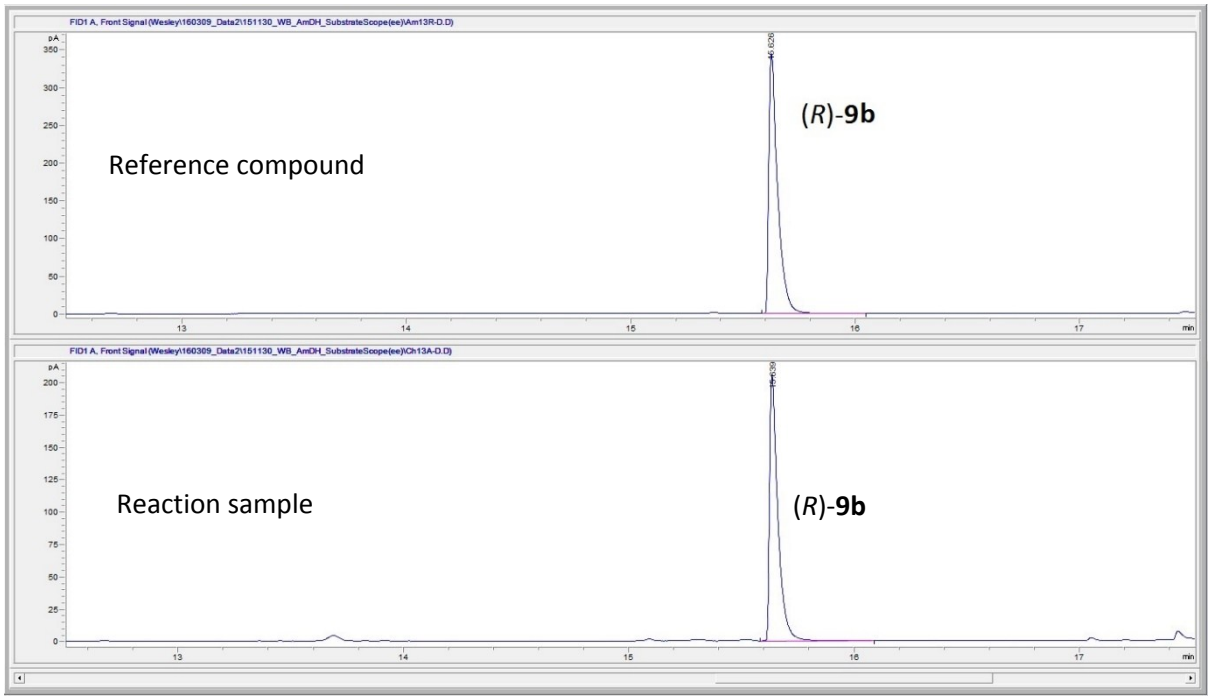


Reductive amination of **9a**

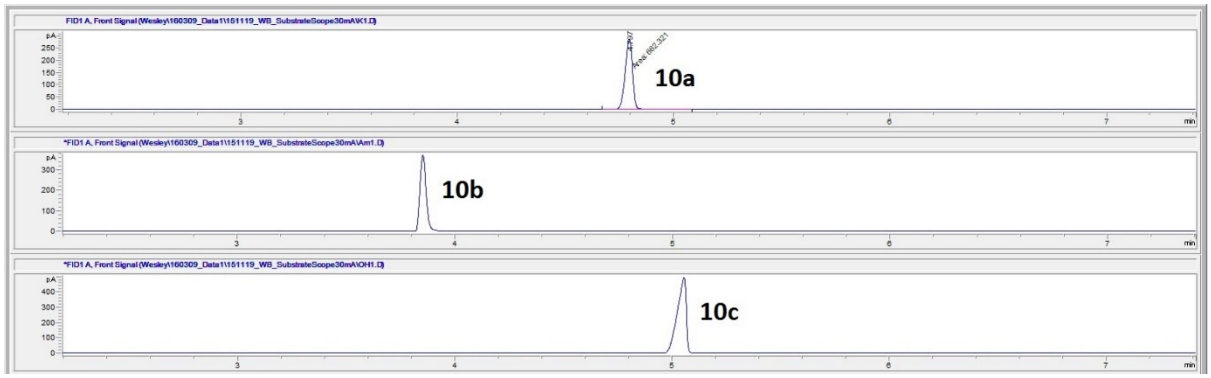


Enantiomeric excess of **9b**





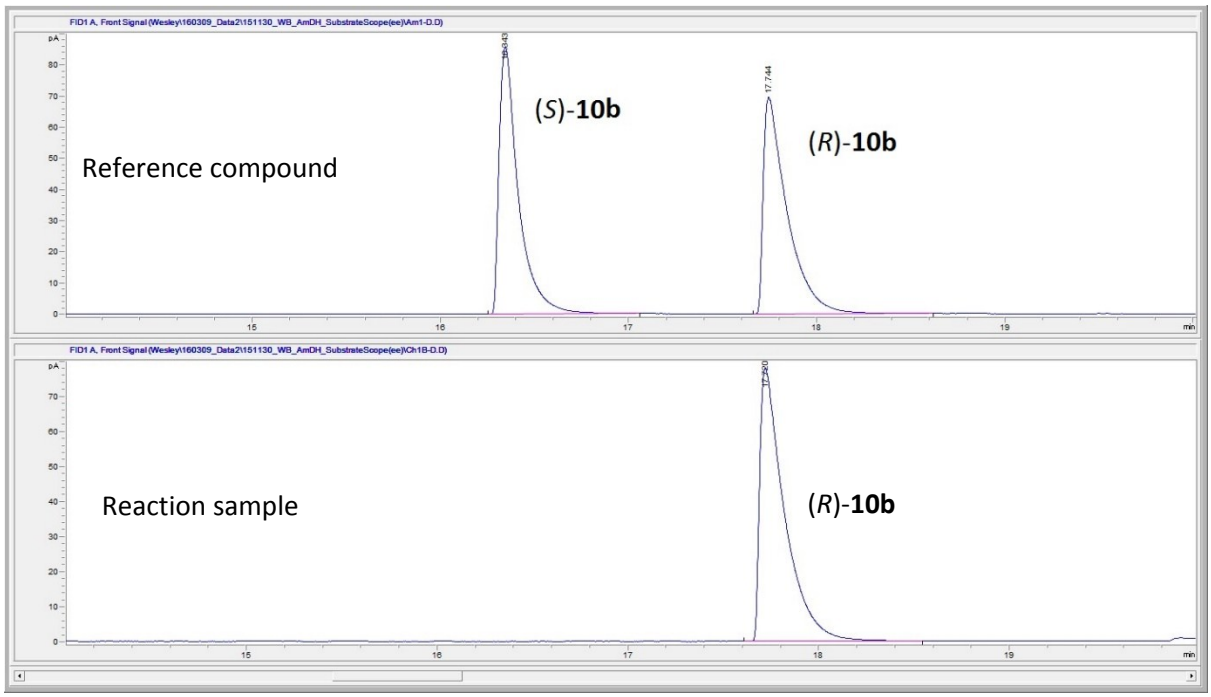
Reference compounds **10a**, **10b** and **10c**



Reductive amination of **10a**

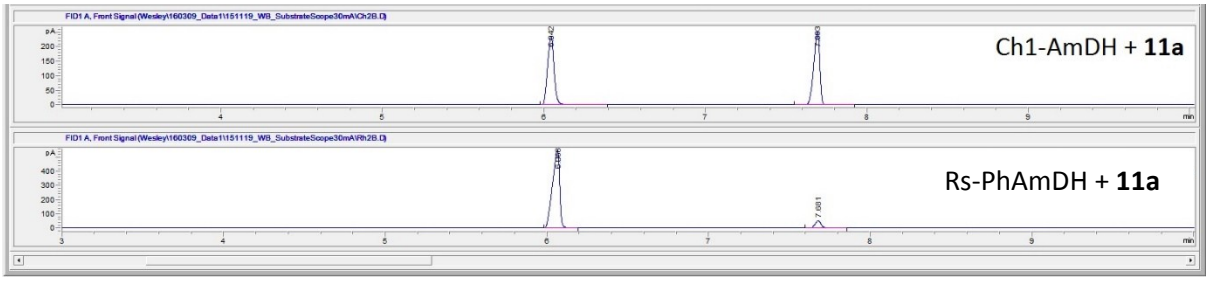
Enantiomeric excess of **10b**

(the order of the elution of (*S*)-**10b** and (*R*)-**10b** with Chromopack Chiracel Dex-CB capillary column is known from literature [1,2]).

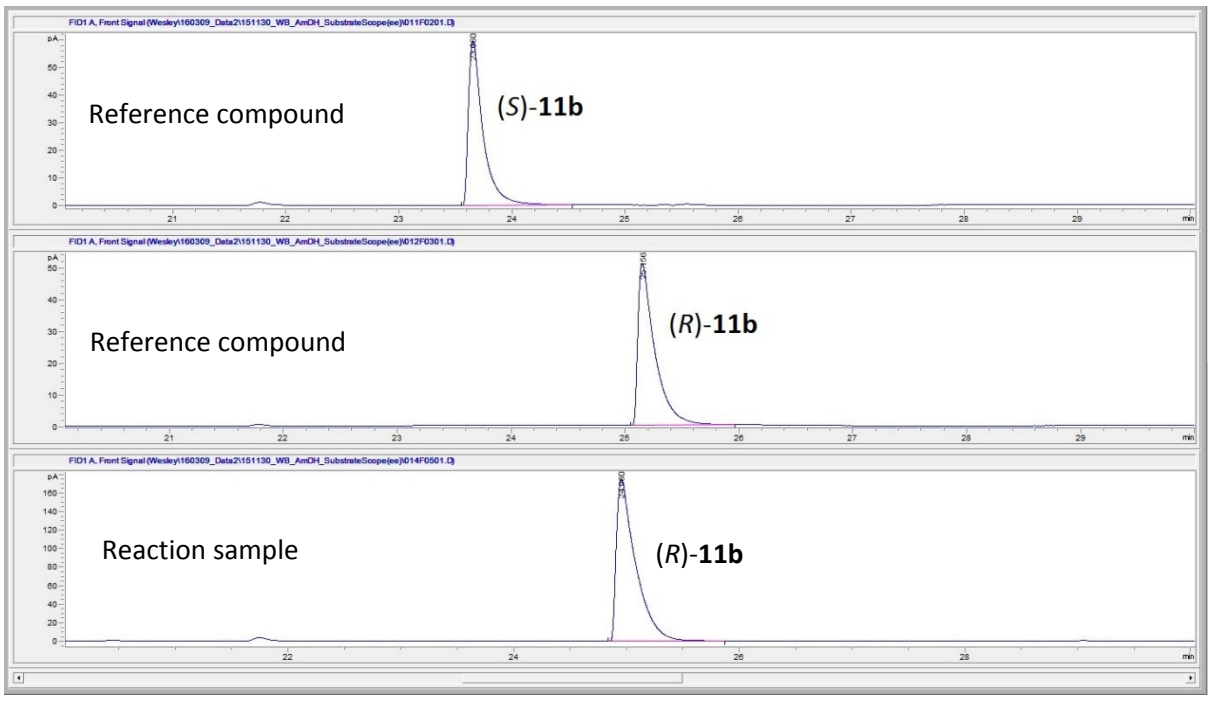


Reference compounds **11a**, **11b** and **11c**

Reductive amination of **11a**

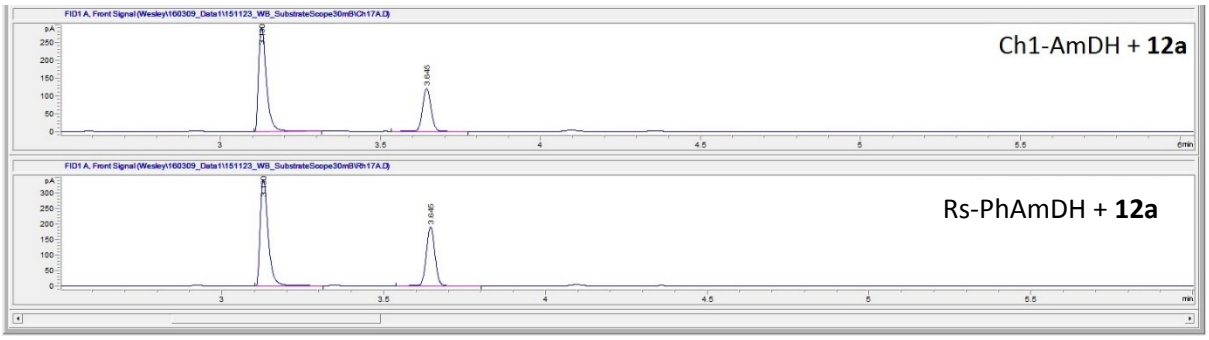


Enantiomeric excess of **11b**

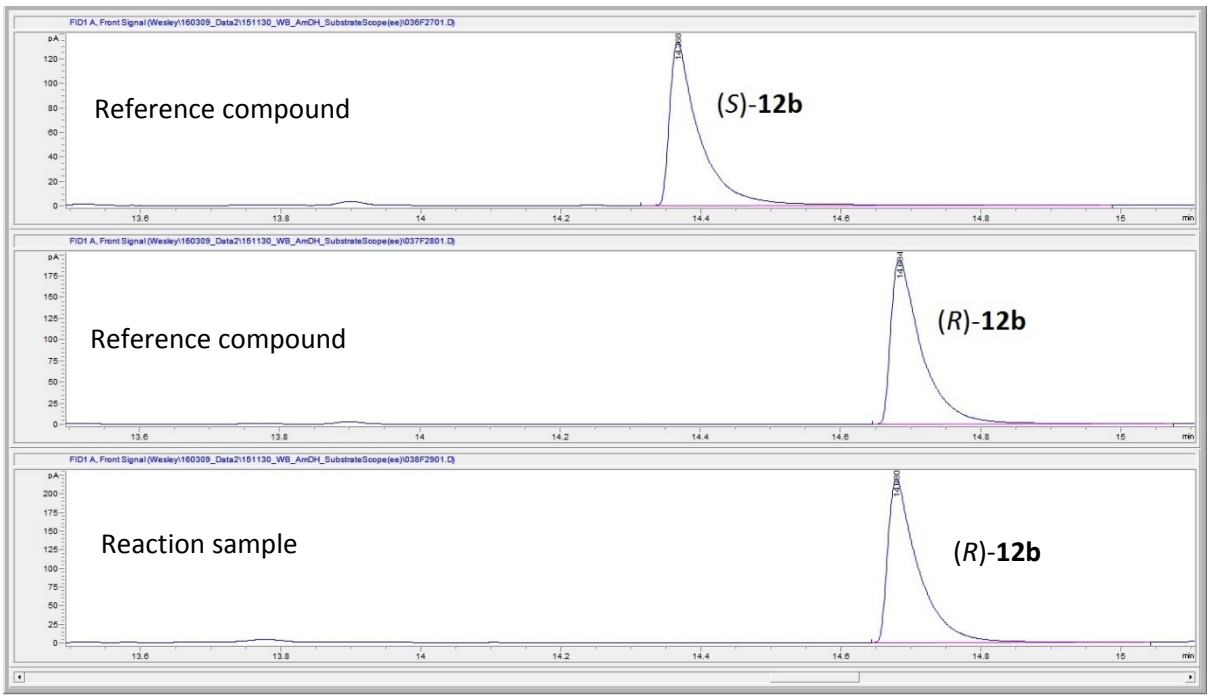


Reference compounds **12a**, **12b** and **12c**

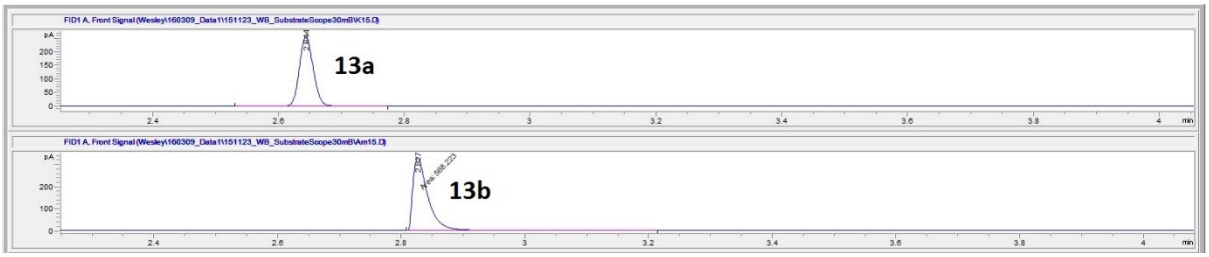
Reductive amination of **12a**



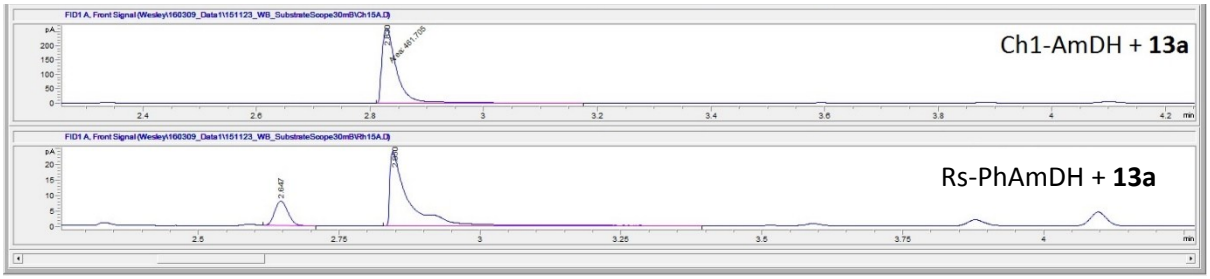
Enantiomeric excess of **12b**



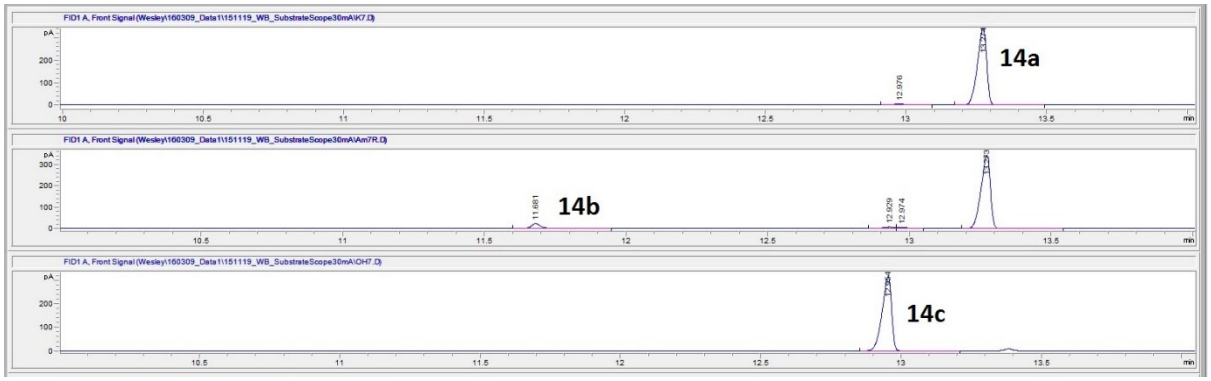
Reference compounds **13a**, **13b**



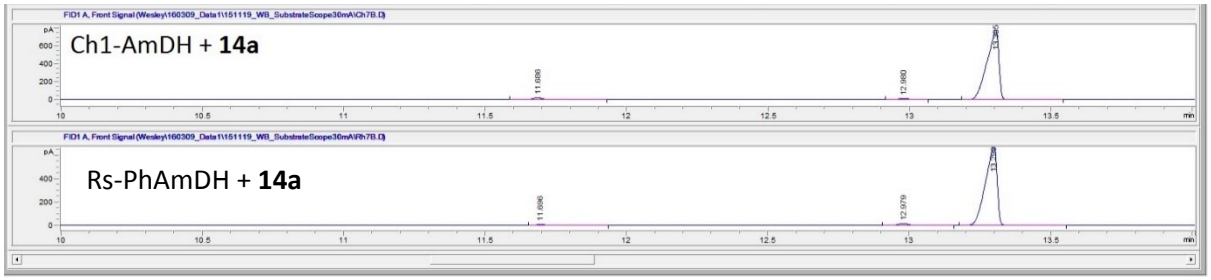
Reductive amination of **13a**



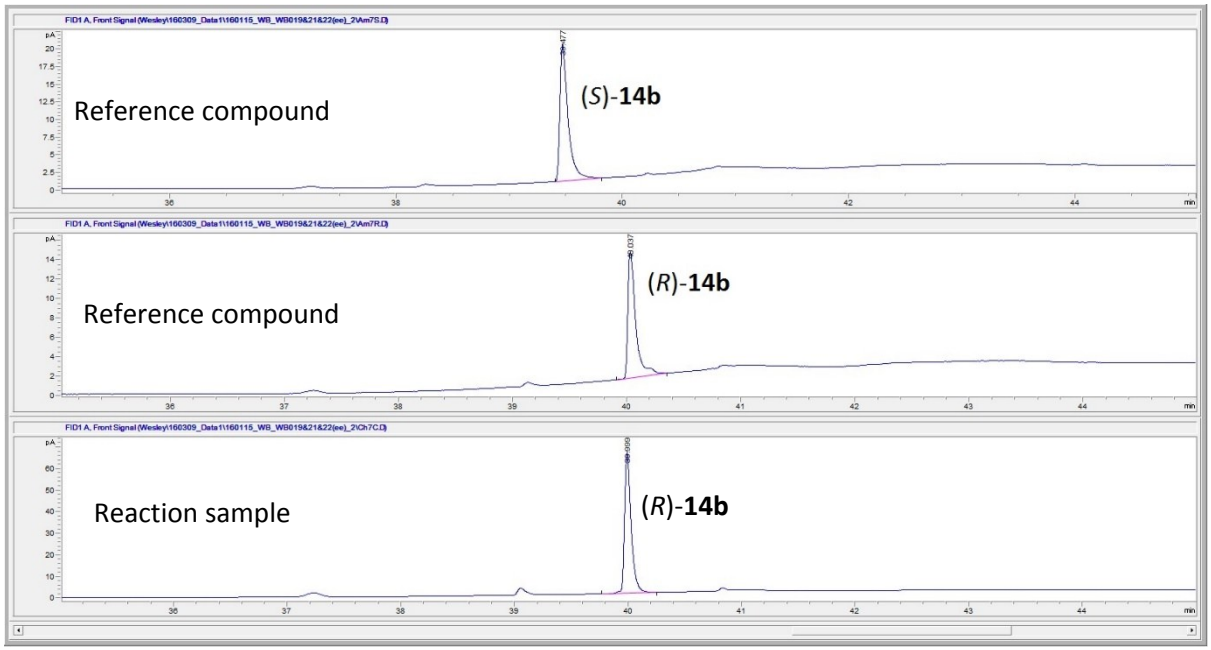
Reference compounds **14a**, **14b** and **14c**



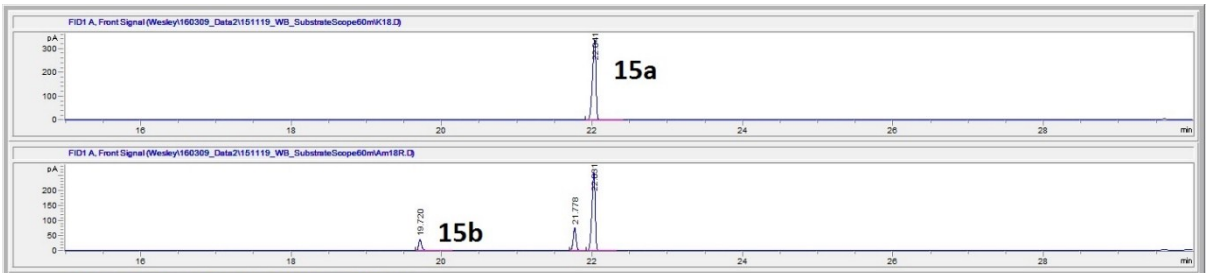
Reductive amination of **14a**



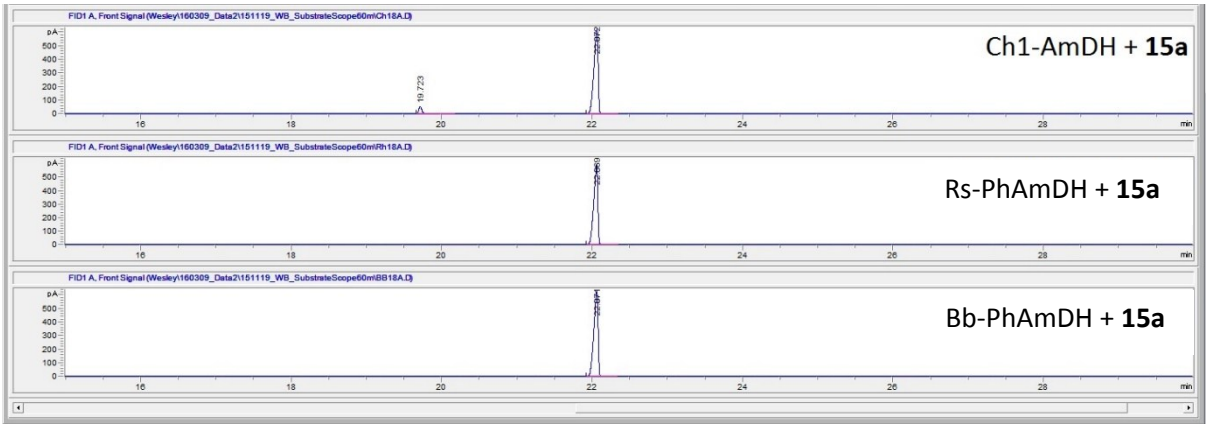
Enantiomeric excess of **14b**



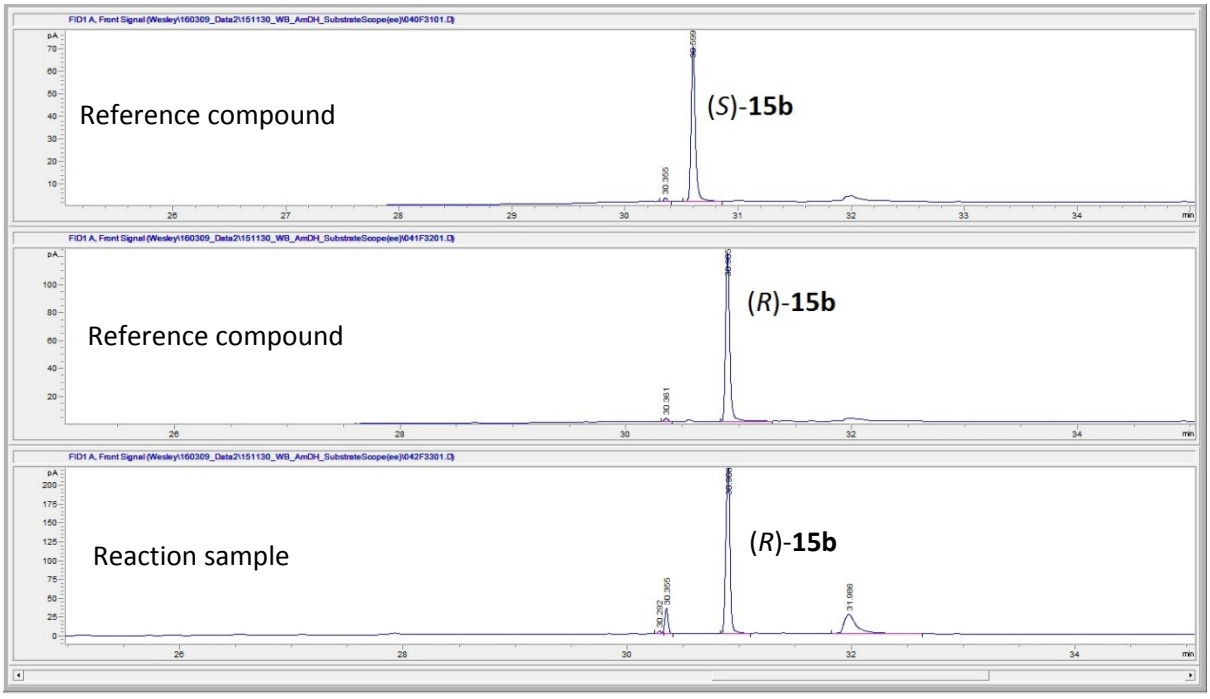
Reference compounds **15a**, **15b**



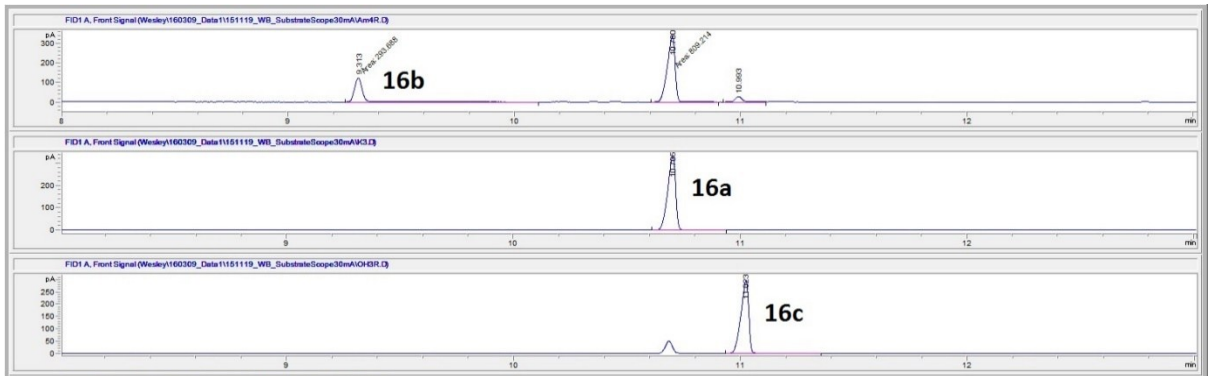
Reductive amination of **15a**



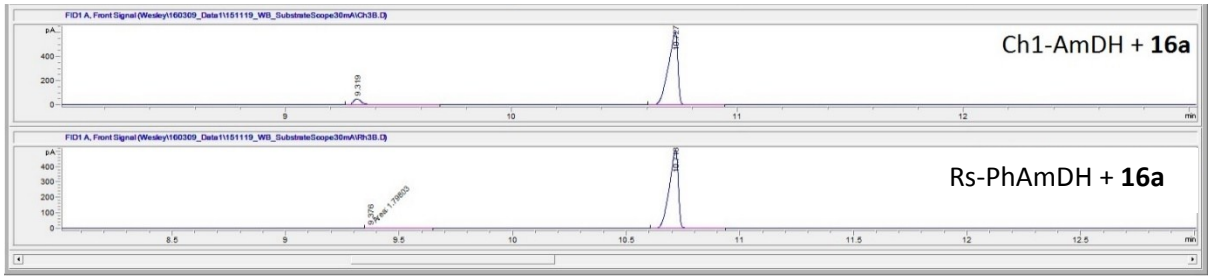
Enantiomeric excess of **15b**



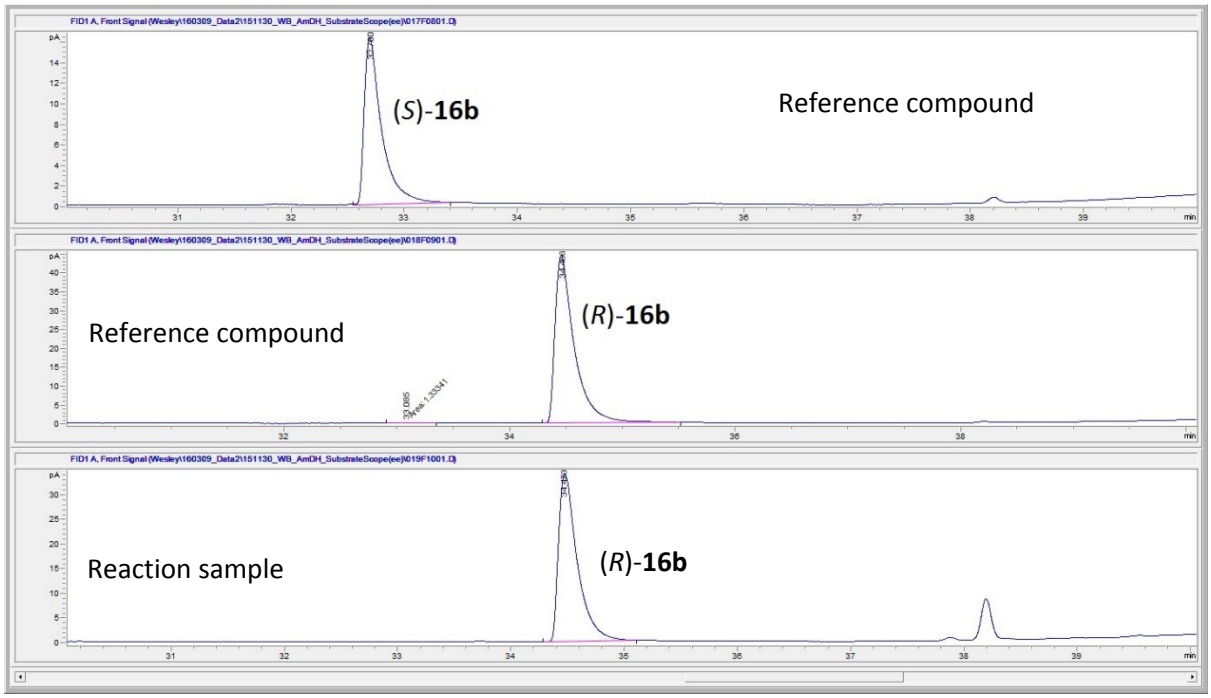
Reference compounds **16a**, **16b** and **16c**



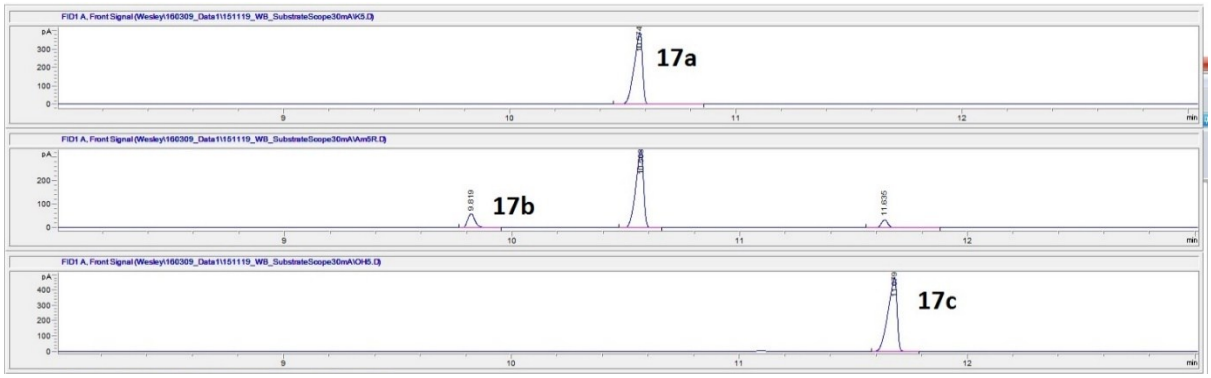
Reductive amination of **16a**



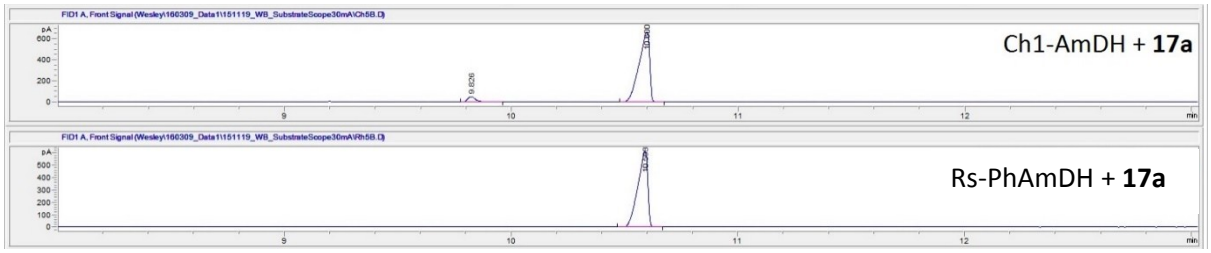
Enantiomeric excess of **16b**



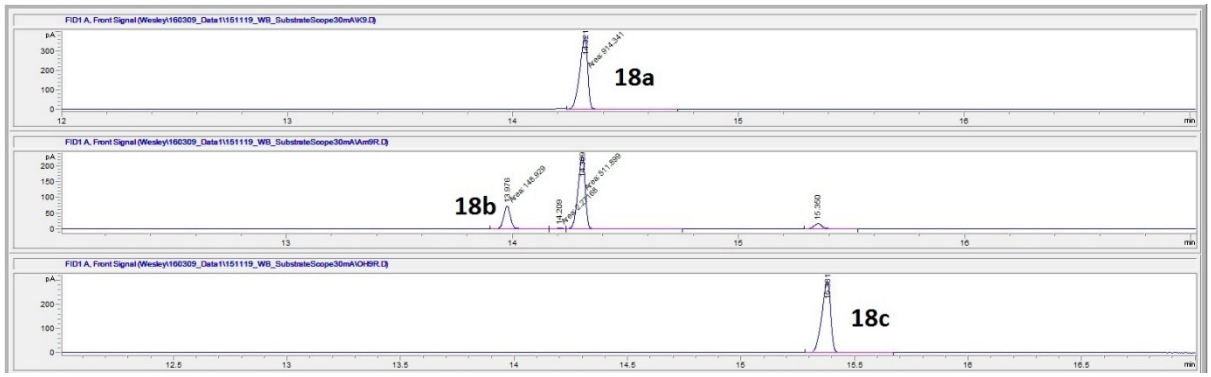
Reference compounds **17a**, **17b** and **17c**



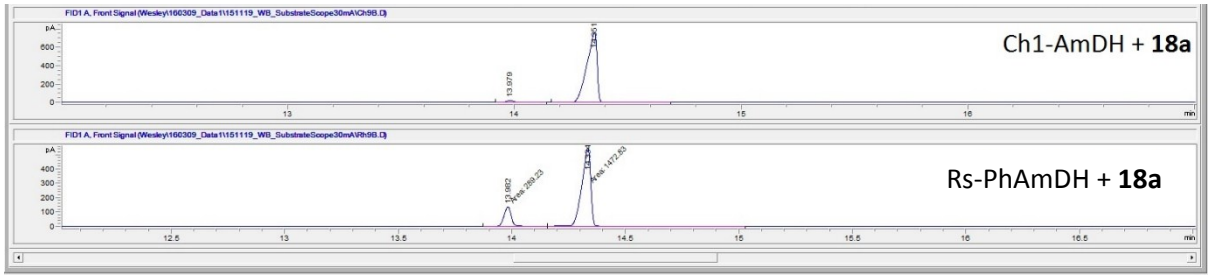
Reductive amination of **17a**



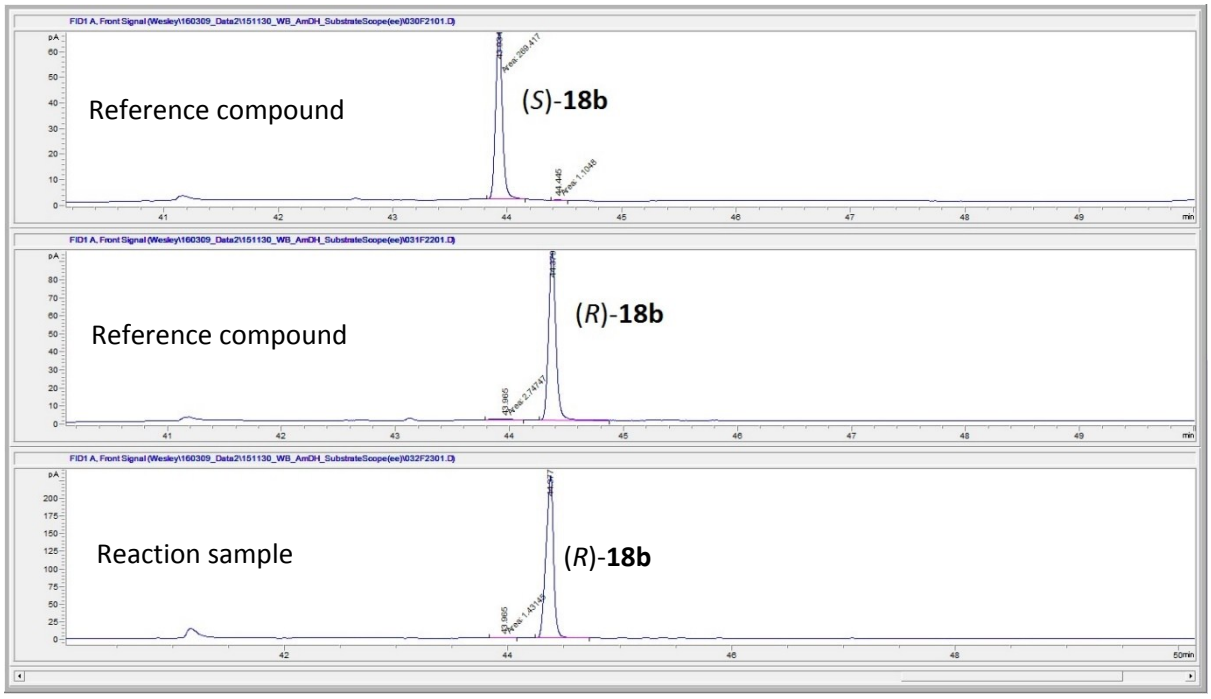
Reference compounds **18a**, **18b** and **18c**



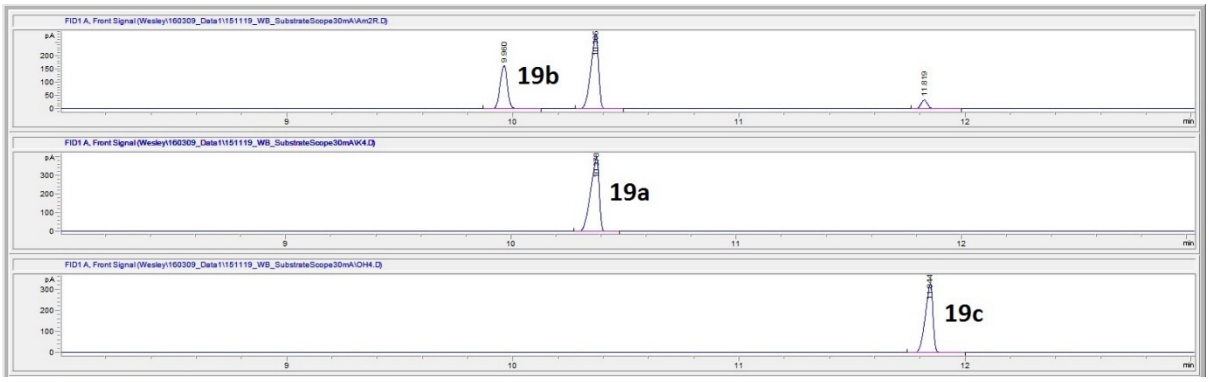
Reductive amination of **18a**



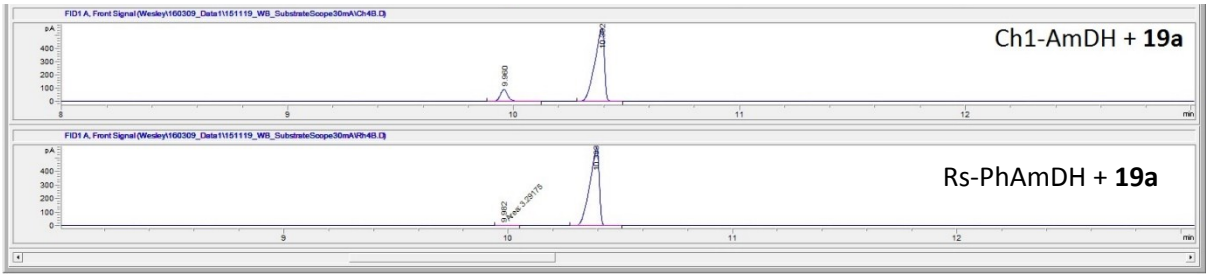
Enantiomeric excess of **18b**



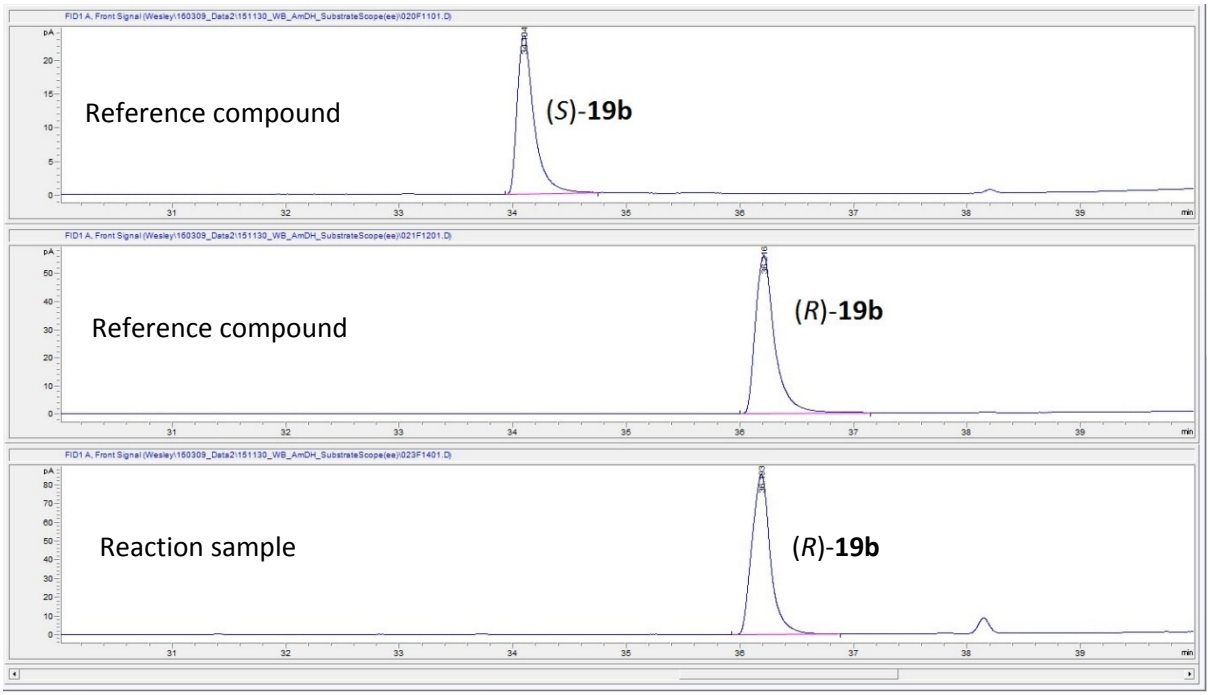
Reference compounds **19a**, **19b** and **19c**



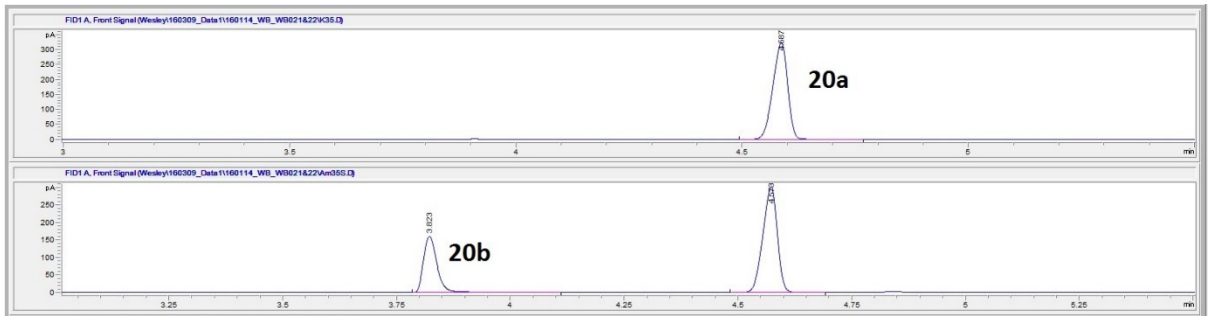
Reductive amination of **19a**



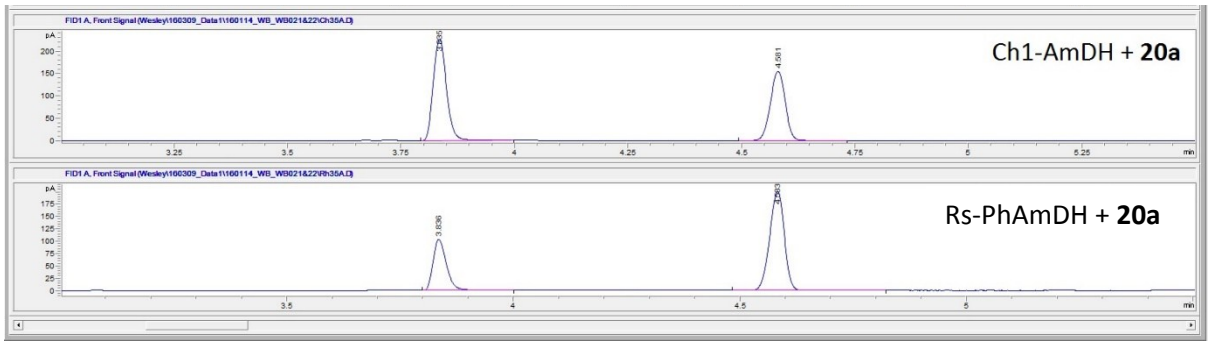
Enantiomeric excess of **19b**



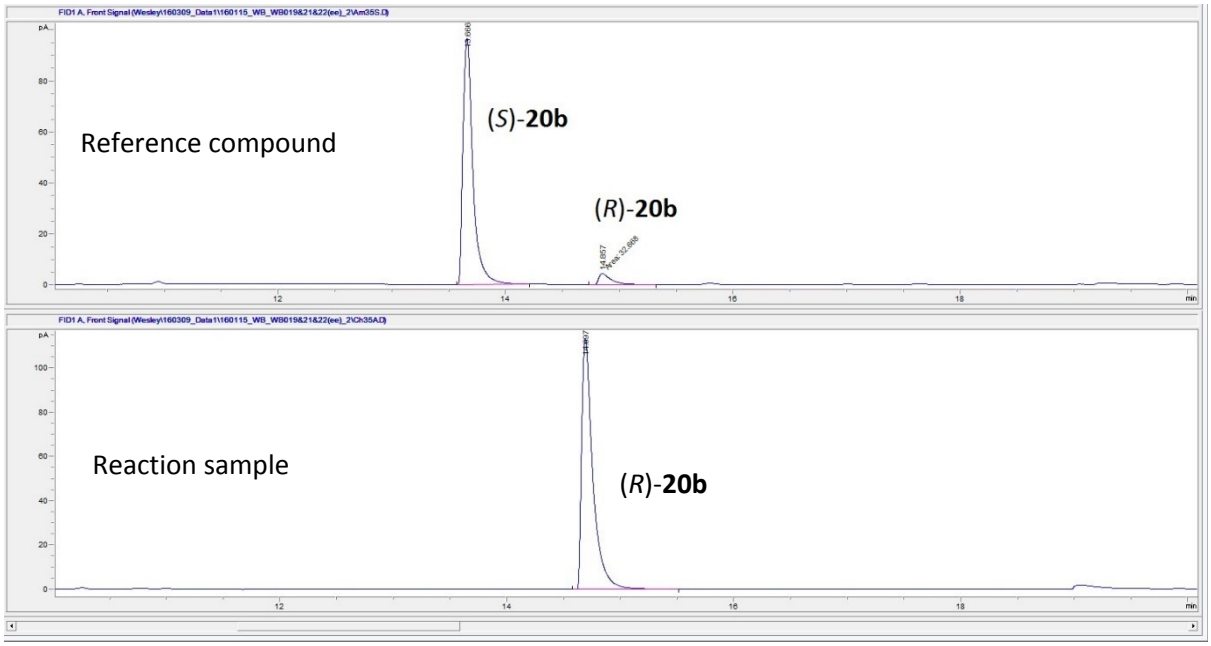
Reference compounds **20a**, **20b**



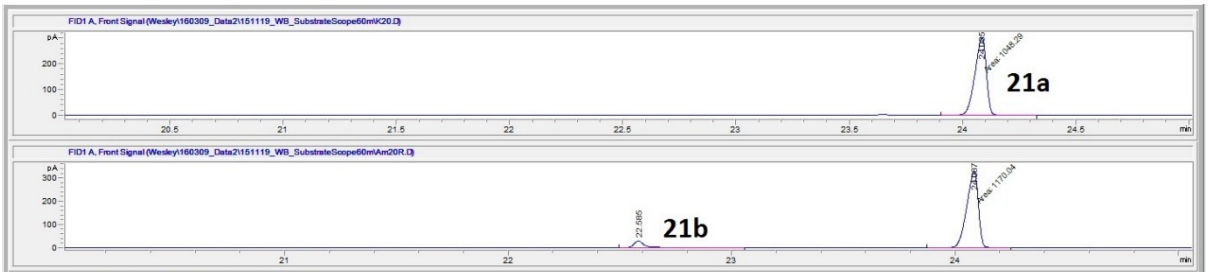
Reductive amination of **20a**



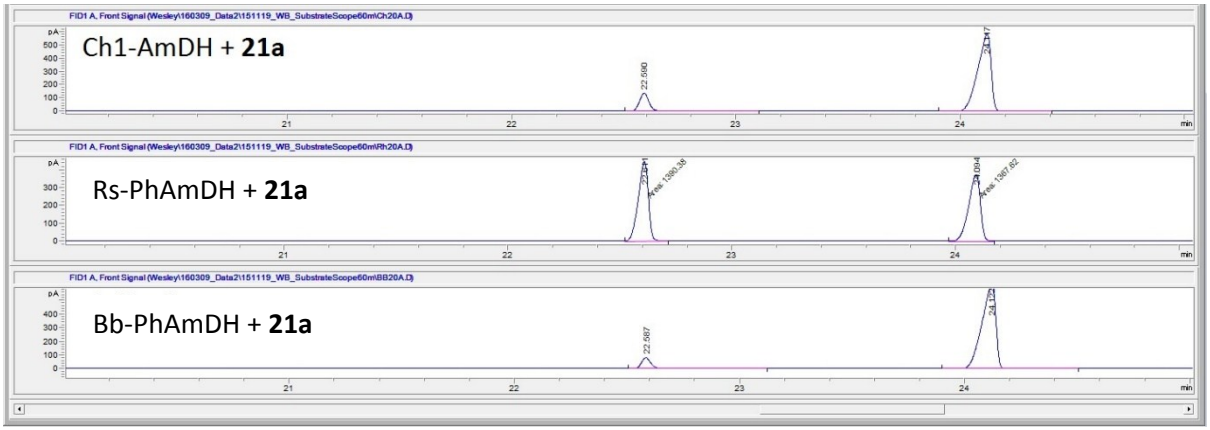
Enantiomeric excess of **20b**



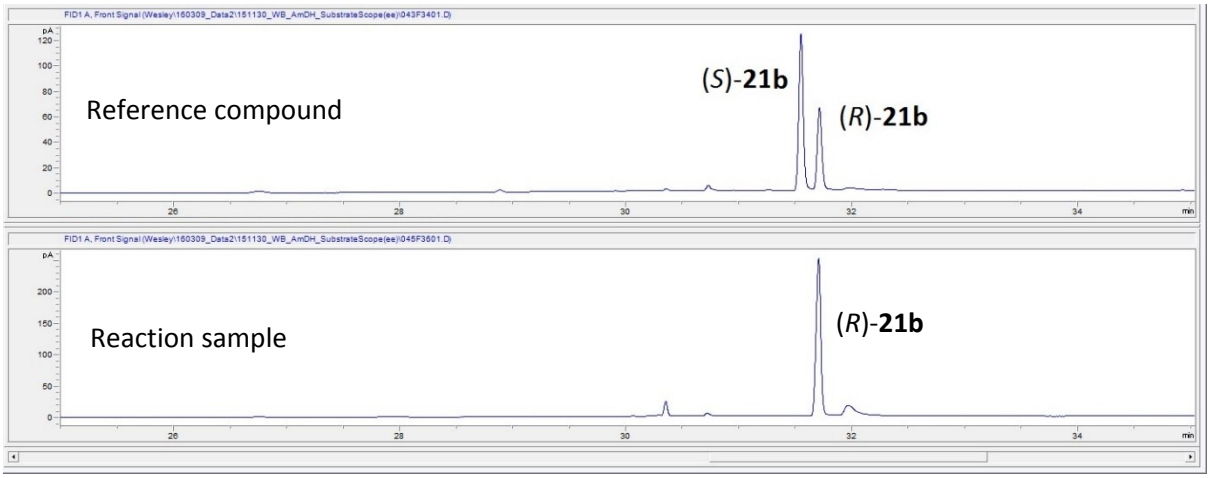
Reference compounds **21a**, **21b**



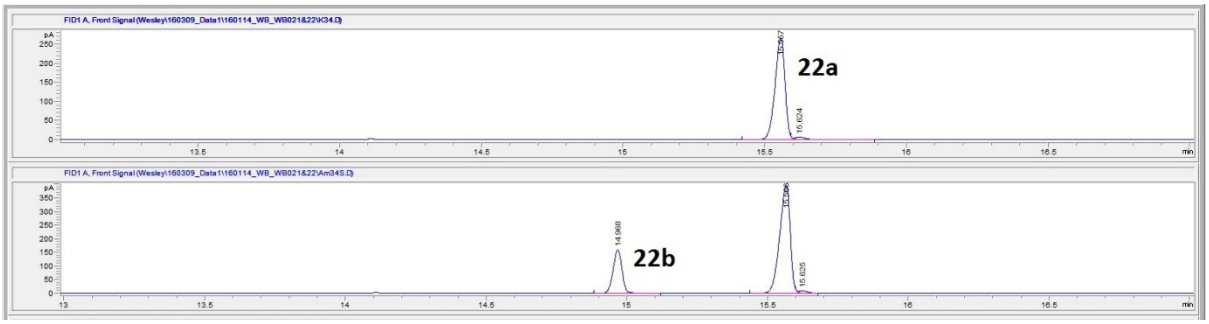
Reductive amination of **21a**



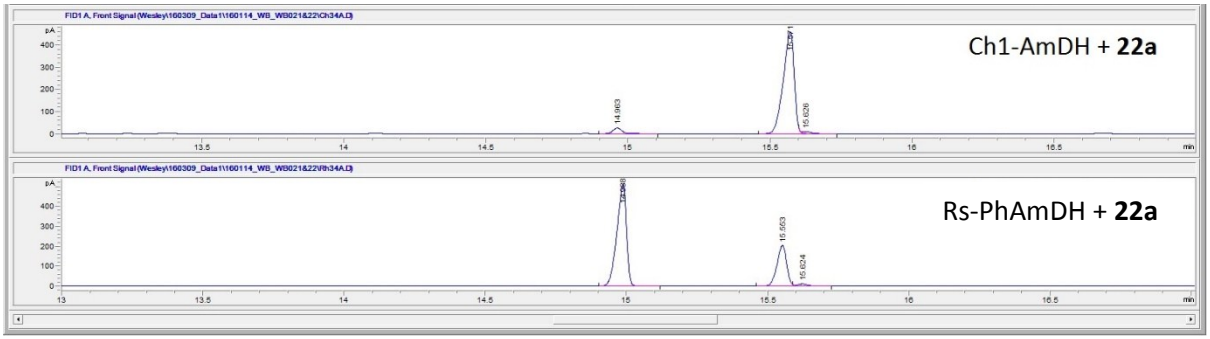
Enantiomeric excess of **21b**



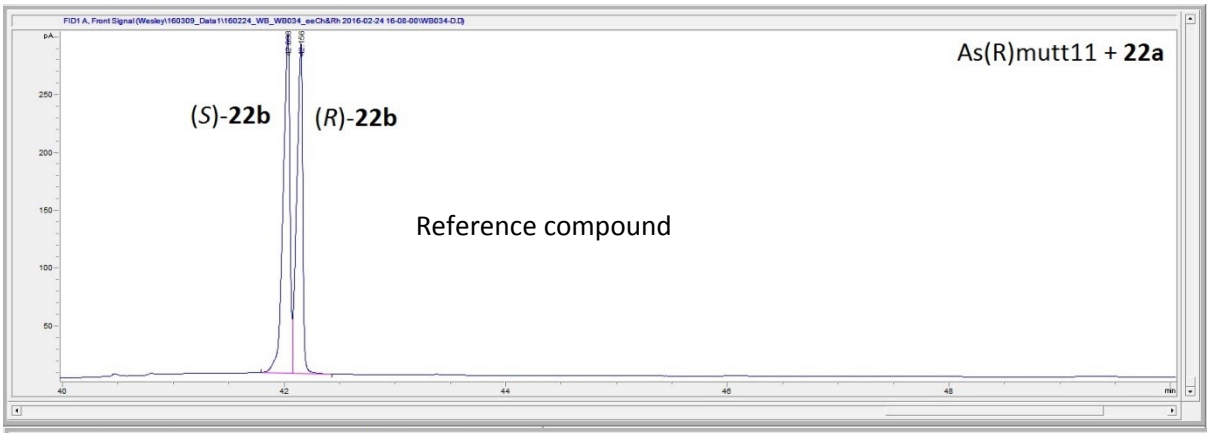
Reference compounds **22a**, **22b**

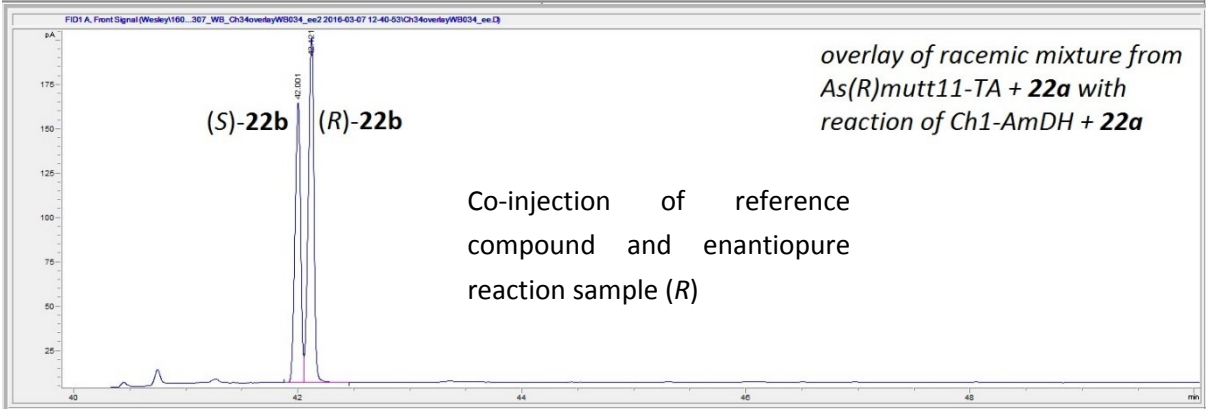


Reductive amination of **22a**

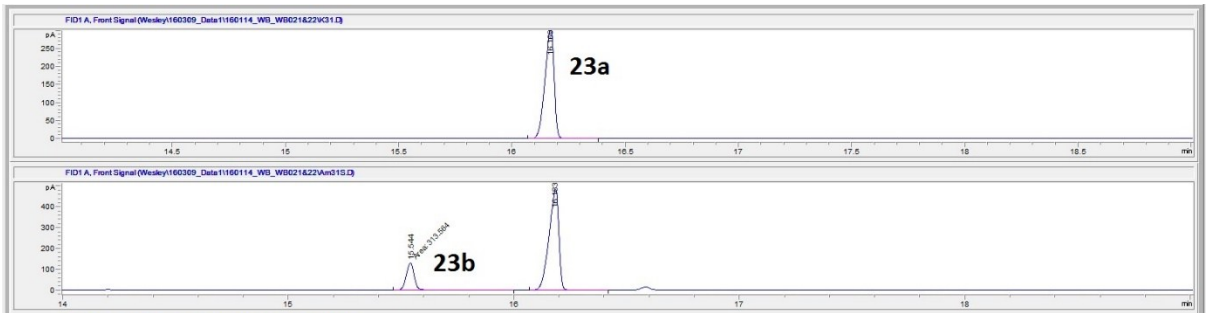


Enantiomeric excess of **22b**

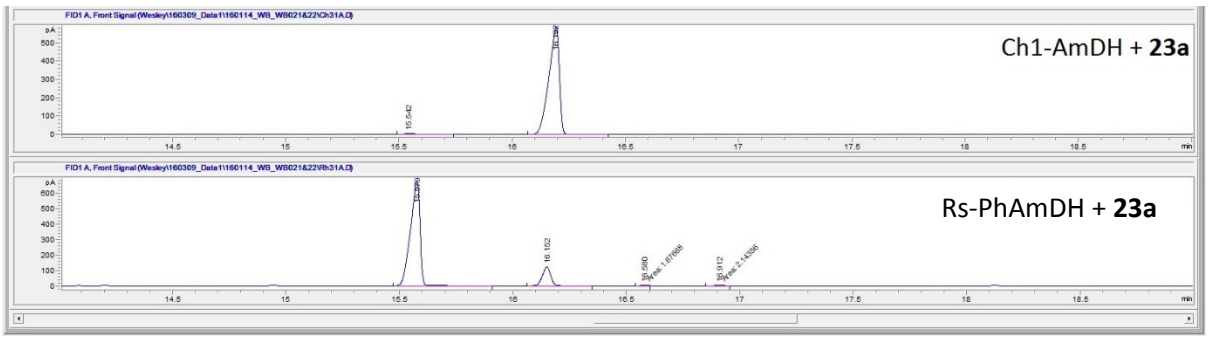




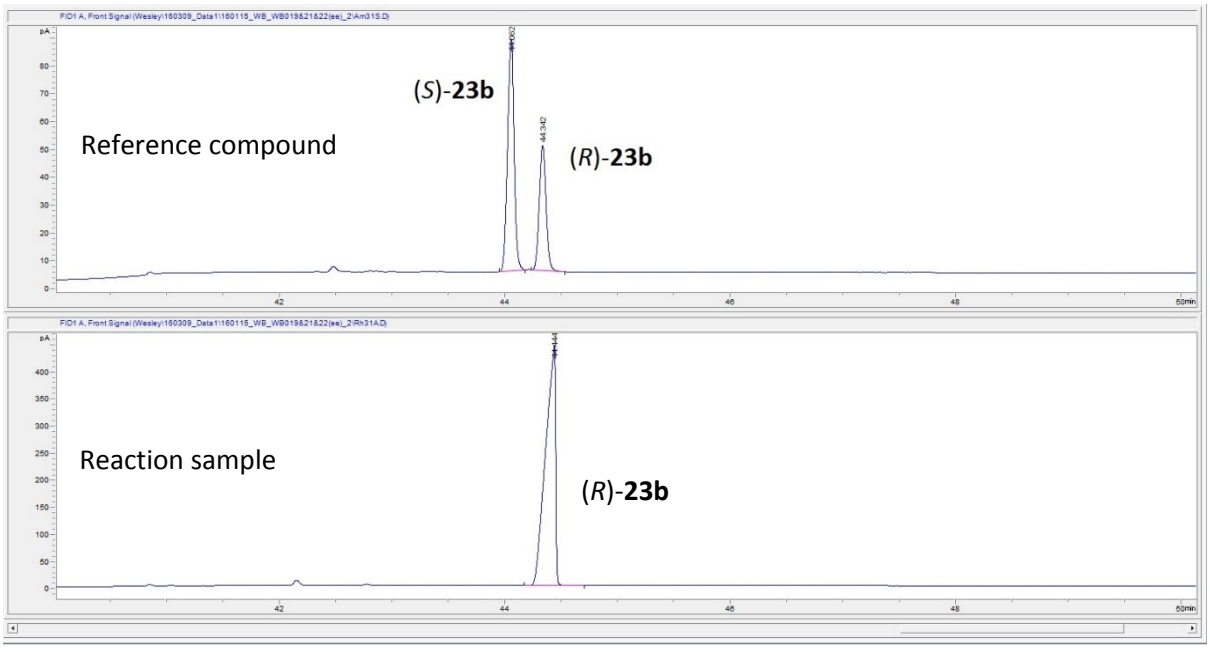
Reference compounds **23a**, **23b**



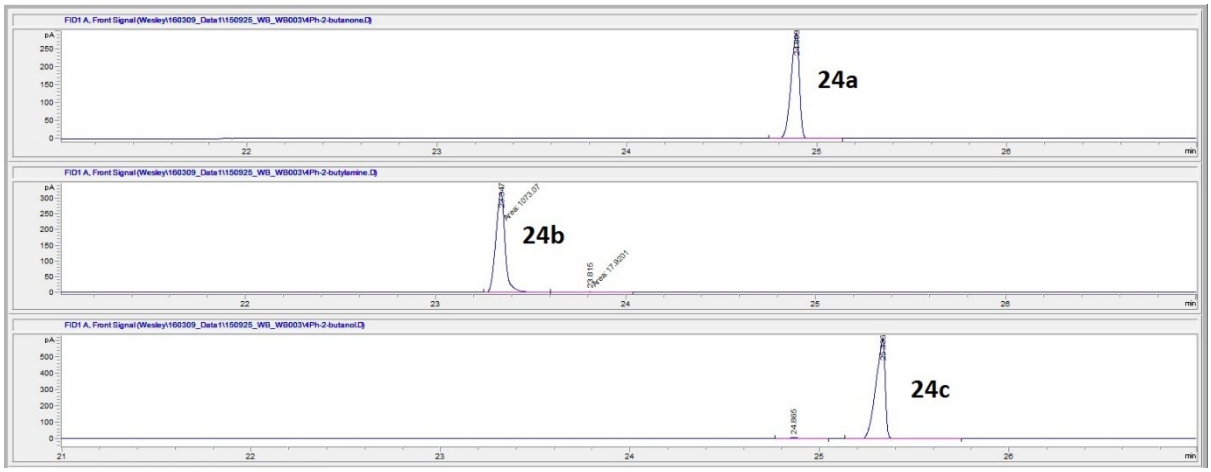
Reductive amination of **23a**



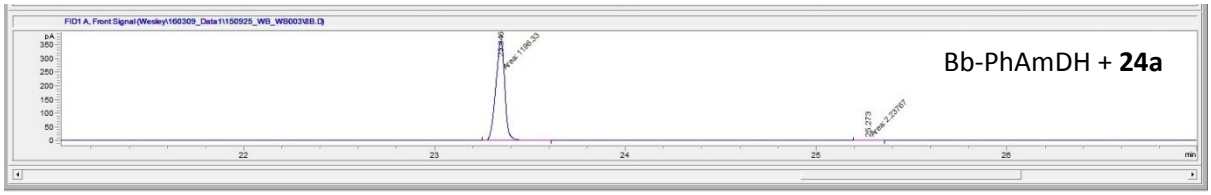
Enantiomeric excess of **23b**



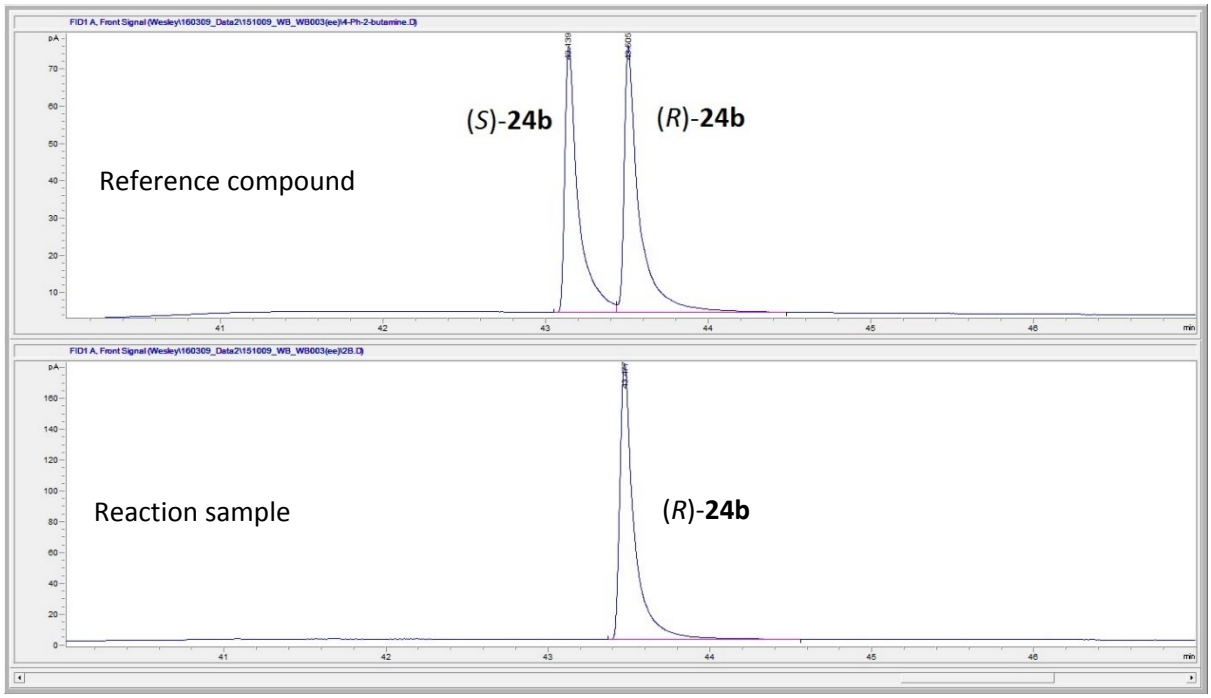
Reference compounds **24a**, **24b** and **24c**



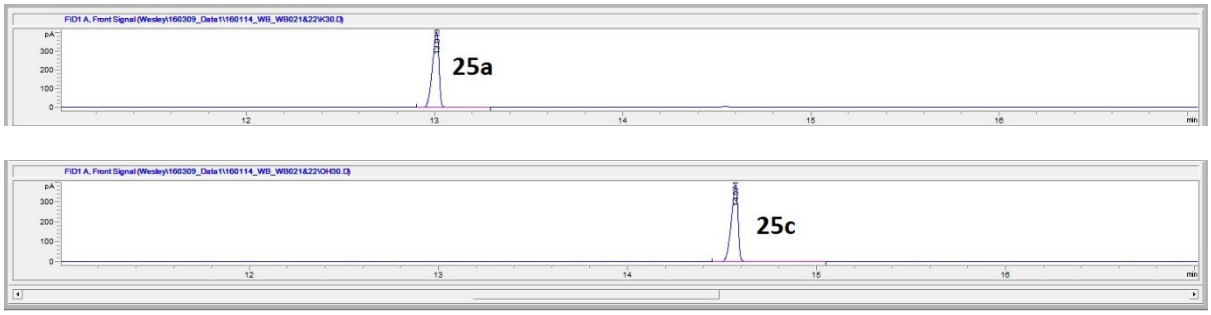
Reductive amination of **24a**



Enantiomeric excess of **24b**



Reference compounds **25a**, **25c**



Reductive amination of **25a**

8. References

1. Koszelewski, D., et al., Asymmetric Synthesis of Optically Pure Pharmacologically Relevant Amines Employing ω-Transaminases. *Adv. Synth. Catal.*, **2008**, *350*, 2761-2766.

2. Mutti, F.G., et al., Conversion of alcohols to enantiopure amines through dual-enzyme hydrogen-borrowing cascades. *Science*, **2015**, *349*, 1525-1529.