The oncolytic efficacy and in vivo pharmacokinetics of [2-(4-chlorophenyl)quinolin-4-yl](piperidine-2-yl)methanol (Vacquinol-1) are governed by distinct stereochemical features

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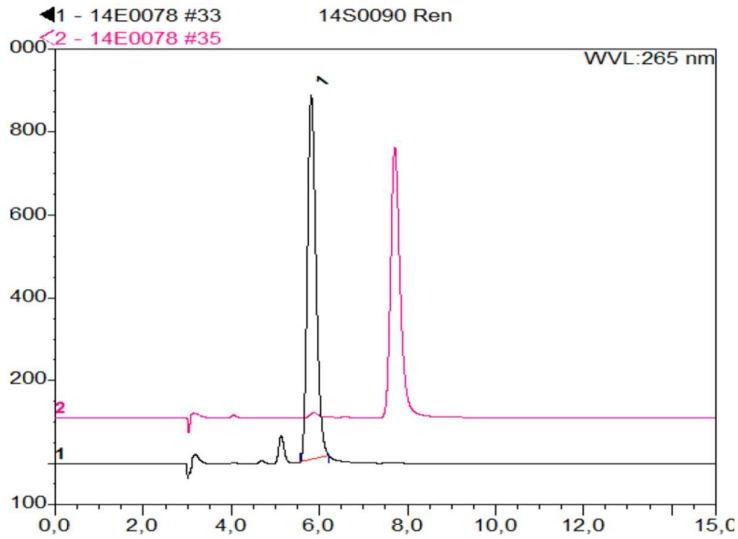
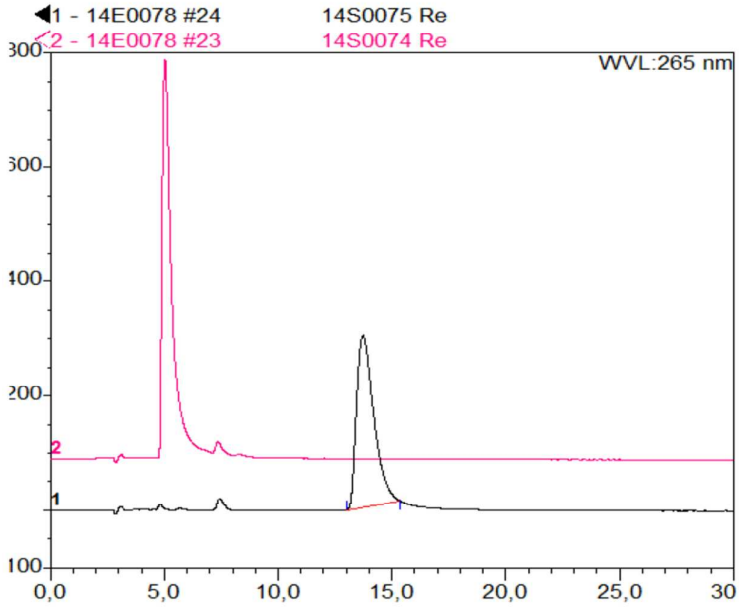
Figure S1. Purity, ee and yield of recovered fractions from chiral separation of 300 mg **1** into enantiomerically pure fractions *Ia*, *Ib*, and *IIa*, *IIb*, corresponding to [*R,R*]**3**, [*S,S*]**3** and [*R,S*]**2**, [*S,R*]**2,** respectively. Data is summarized in the table below.



Fractions *Ia* ([*R,R*]**3**, black trace) and *Ib* ([*S,S*]**3**, red trace) analyzed on a Chirapak AD-H column



Fractions *IIa* ([*R,S*]**2**, red trace) and *IIb* ([*S,R*]**2**, black trace) analyzed on a Chiracel OD-H column

Fraction ID	Column	Gradient	Purity (%)	ee (%)	$[\alpha]_D^{25}$ ($CHCl_3$)	Yield (mg)
Ia [*R,R*]**3**	Chiralcel OD-H	1:9 ethanol/heptane (0.2% diethylamine)	95.3	100	+19.3	35.4
Ib [*S,S*]**3**			99.2	97.8	-18.9	25.4
IIa [*R,S*]**2**	Chiralpak AD-H	3:7 ethanol/heptane (0.2% diethylamine)	98.7	100	+14.1	48.6
IIb [*S,R*]**2**			98.1	99.0	-14.3	48.2

Figure S2. ^{1}H- and ^{13}C-NMR of [*R*,*R*]**3** (fraction *Ia*) in $CDCl_3$.

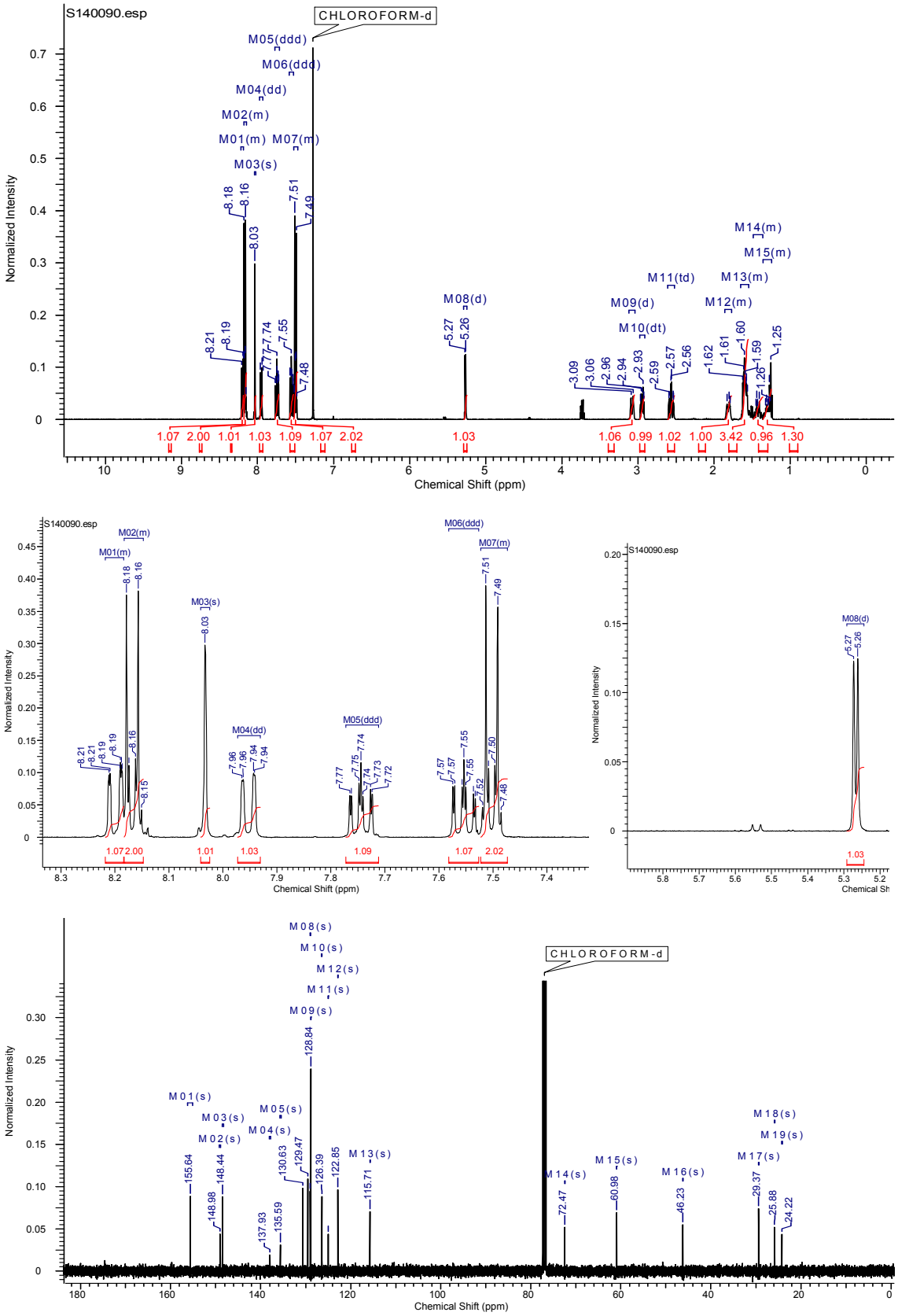


Figure S3. ^{1}H- and ^{13}C-NMR of [*S,S*]**3** (fraction *Ib*) in $CDCl_3$.

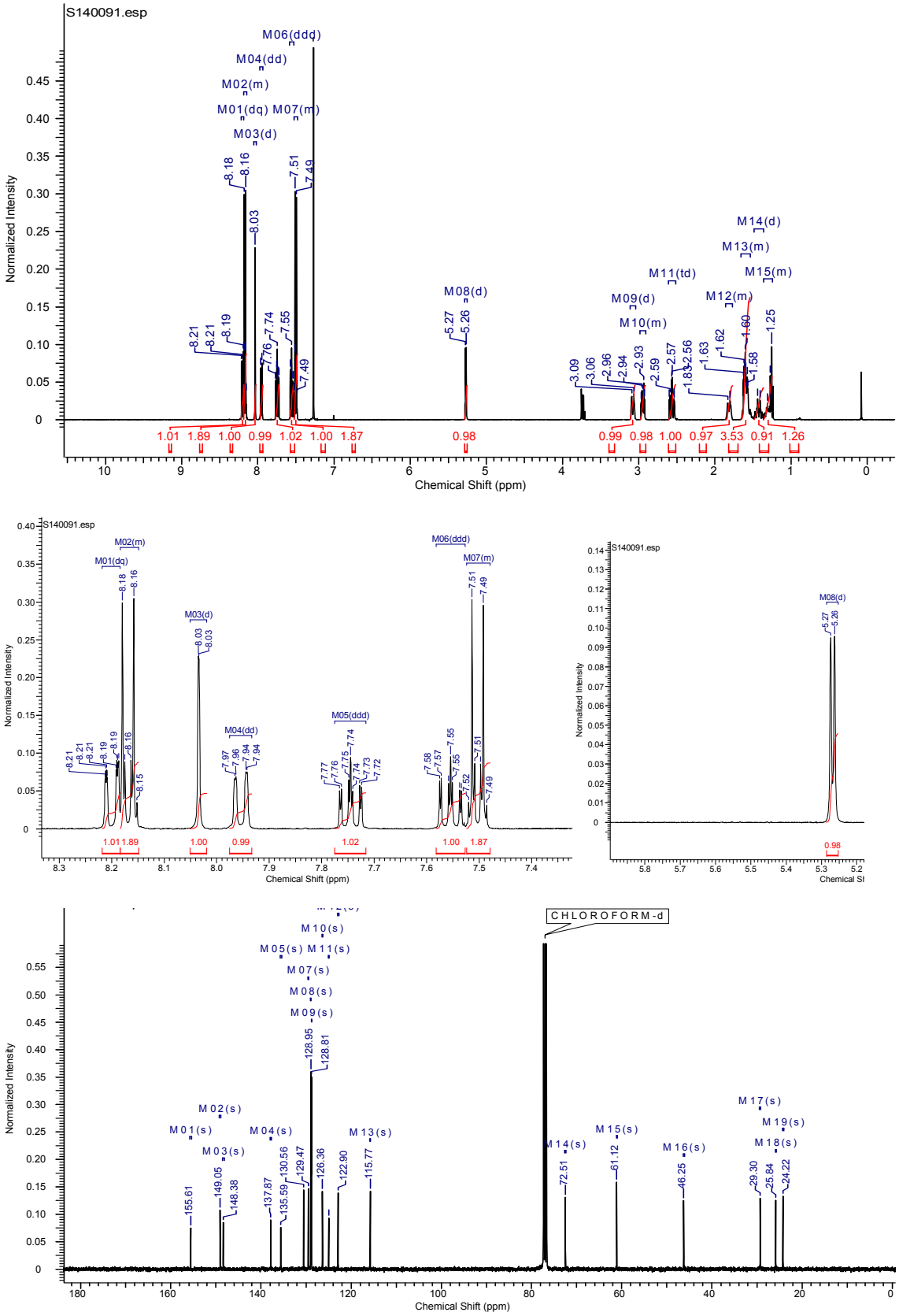


Figure S4. ^{1}H- and ^{13}C-NMR of [*R,S*]**2** (fraction *IIa*) in $CDCl_3$

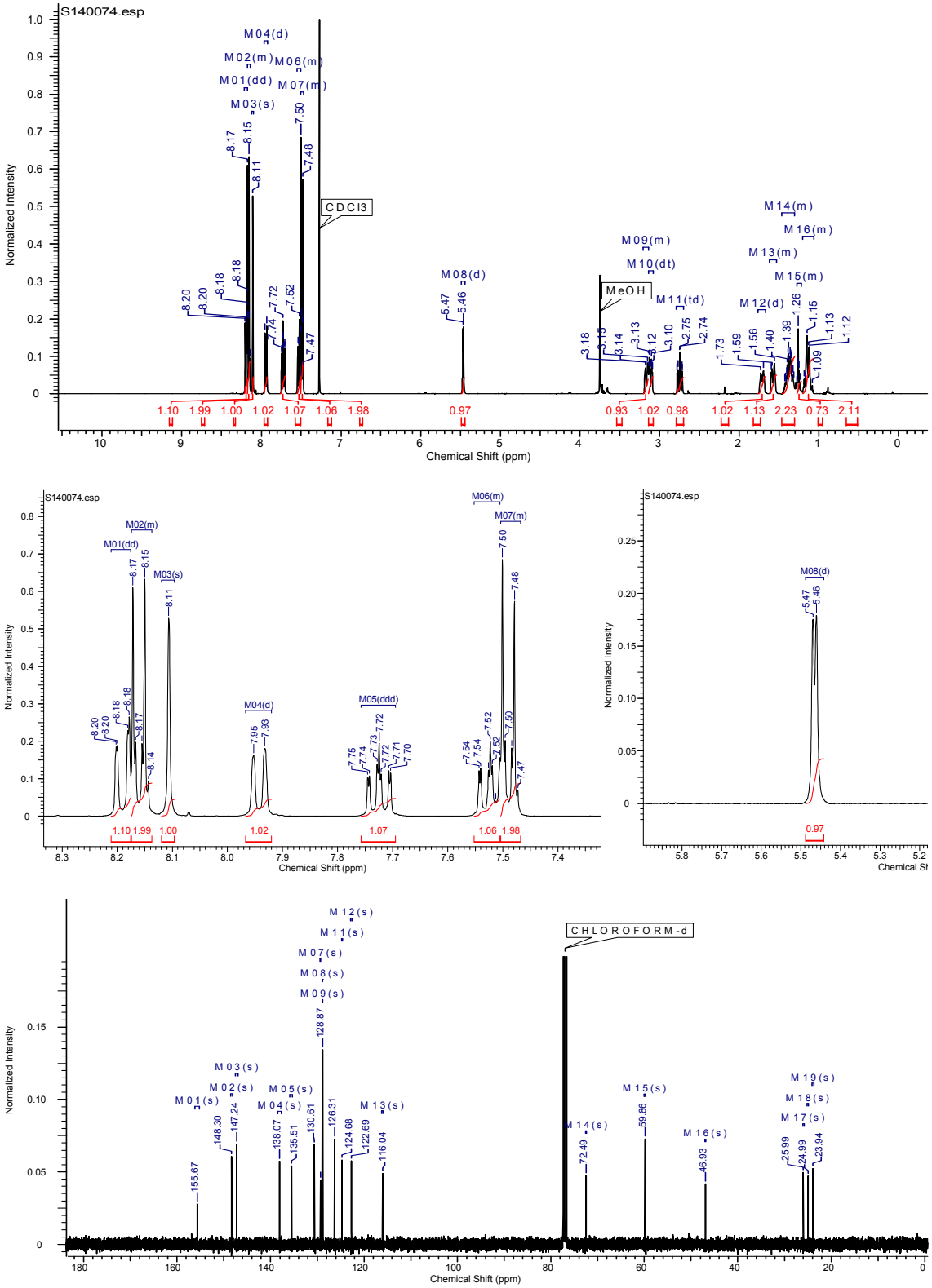


Figure S5. ^{1}H- and ^{13}C-NMR of [*S,R*]**2** (fraction *IIb*) in $CDCl_3$

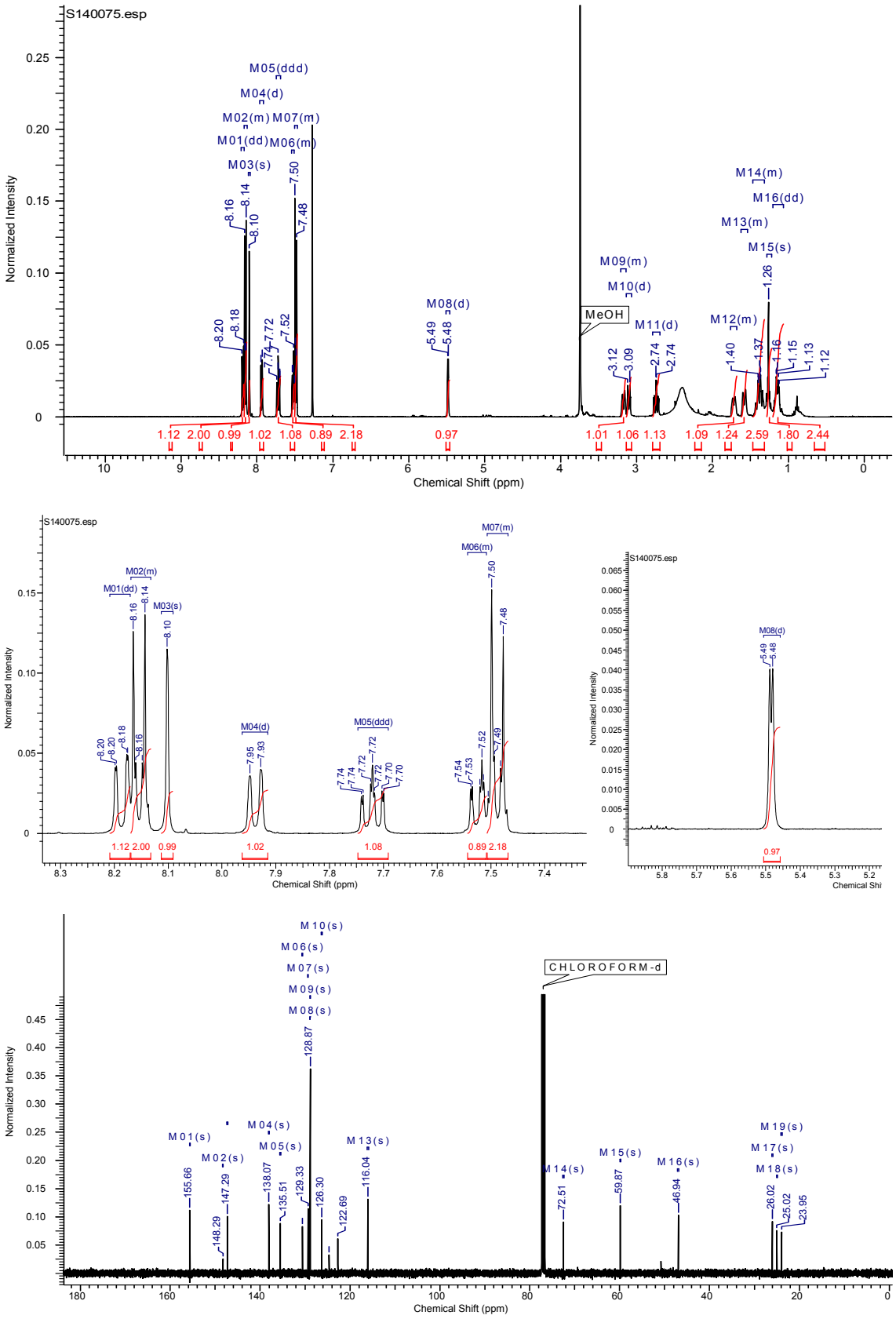


Figure S6. Analytical HPLC-UV (265 nm) chromatogram on Chiralpak AD-H column with overlaid traces of [*R,S*]**2** (blue trace, ee: 99 %) and [*S,R*]**2** (pink trace, ee: 97%), both prepared according to Scheme 1, panel B and C, respectively, compared with fraction *II* (racemic *erythro* 2, black trace), containing fraction *IIa* (Rt: 6.87 min, [*R,S*]**2**) and fraction *IIb* (Rt: 15.63 min, [*S,R*]**2**) isolated chromatographically.

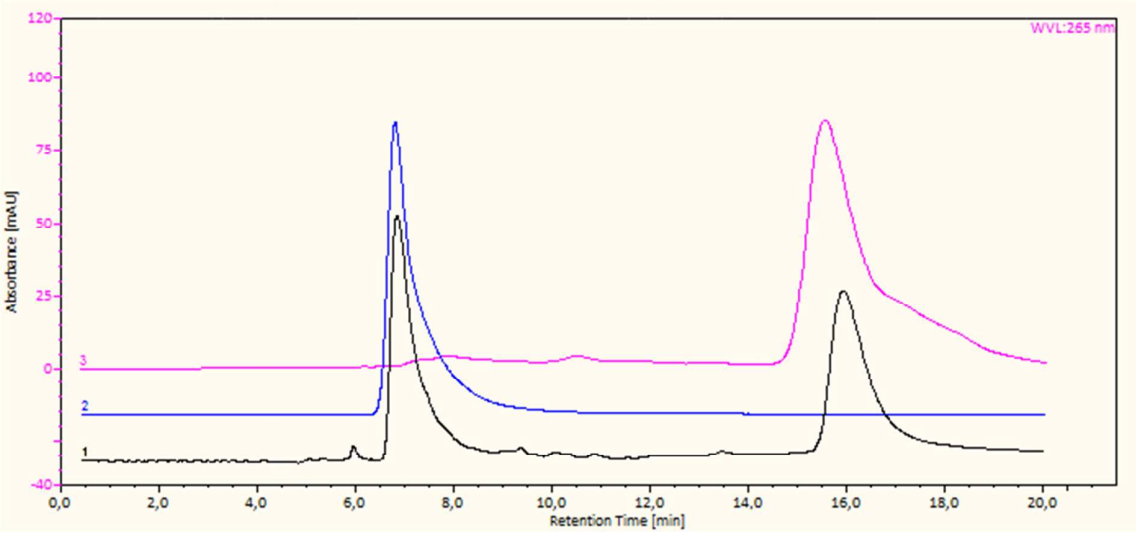


Figure S7. Chiral analytical HPLC-MS/MS (ES+) indicating the intrinsic stereoselectivity of the synthesis according to Scheme 2.
Analysis performed on a ChiralPak IC column (Diacel), 2.1 × 150 mm held 20 ˚C. Mobile phase: 7.5 mM ammonium formate in CH3CN:H2O (98:2). Flowrate: 0.17 mL/min A: [S,R]2 (Rt: 18.17 min) is produced with 6% [R,R]3 (Rt: 11.39 min) resulting from of intermediate 24. No C17(S) isomers were detected. B: [R,S]2 (Rt: 14.92 min) is produced with 9% [S,S]3 (Rt: 13.21 min) resulting from formation of intermediate 20. No C17(R) isomers were observed. C: Reference chromatogram of compound 1 including all four stereoisomers.

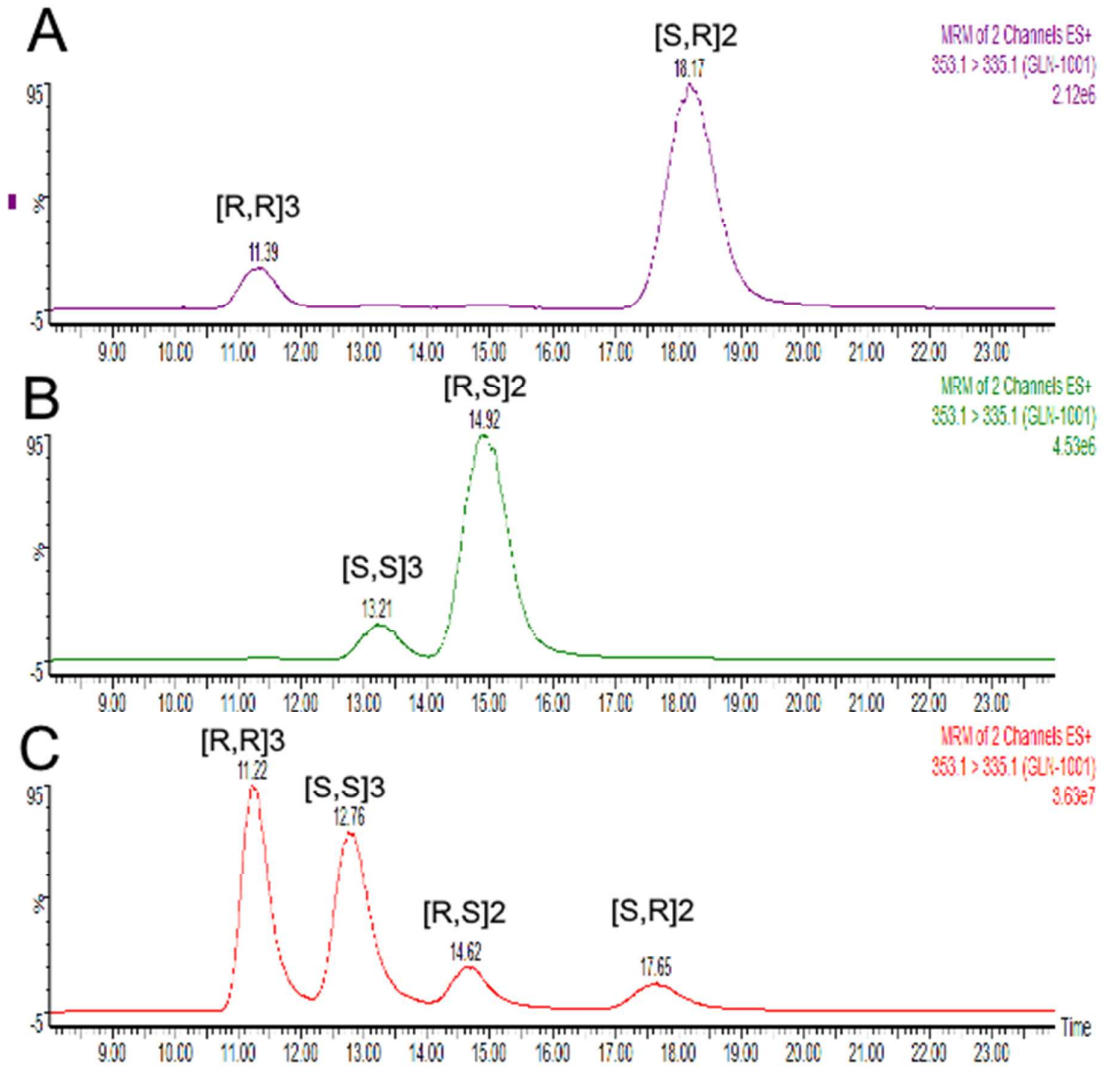


Figure S8. Compound [*S*,*R*]**2** (top) and [*R*,*S*]**2** (middle) from syntheses according to Scheme 2 prior to purification, containing 5-10% of [*R*,*R*]**3** and [*S*,*S*]**2**, respectively.
Enantiopure compound [*R*,*S*]2 (bottom) after chromatographic purification. Chiral analytical HPLC-MS/MS (ES+) analysis performed on a ChiralPak IC column (Diacel), 2.1 × 150 mm at 20 ˚C. Mobile phase: 7.5 mM ammonium formate in CH_3CN:H_2O (98:2). Flowrate: 0.17 mL/min