

Electronic Supplementary Information

Development of Novel PP2A Activators For Use in the Treatment of Acute Myeloid Leukaemia

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1. General Experimental

Melting points were obtained on OptiMelt Automated Melting Point System with Digital Image Processing Technology and are uncorrected. ^1H NMR and ^{13}C NMR were recorded at the Nuclear Magnetic Resonance Facility within the Mark Wainwright Analytical Centre at The University of New South Wales on a Bruker Avance III 300 (300 MHz), Bruker DPX 300 (300 MHz), Bruker Avance III 400 (400 MHz), Bruker Avance III 500 (500 MHz) or Bruker Avance III 600 (600 MHz), with data acquired and processed using TopSpin 3.0 software. Chemical shifts are expressed in parts per million (PPM) on the δ scale. Chemical shifts in (a) CDCl_3 were referenced relative to CHCl_3 (7.26 ppm) for ^1H NMR and CHCl_3 (77.16 ppm) for ^{13}C NMR, (b) MeOD were referenced relative to CH_3OH (3.31 ppm) for ^1H NMR and CD_3OD (49.00 ppm) for ^{13}C NMR, and (c) $(\text{CD}_3)_2\text{SO}$ were referenced relative to $(\text{CH}_3)_2\text{SO}$ (2.50 ppm) for ^1H NMR and $(\text{CD}_3)_2\text{SO}$ (39.52 ppm) for ^{13}C NMR.¹ Infrared spectra were obtained on a ThermoNicolet Avatar 370 FT-IR spectrometer and are reported in wavenumbers (cm^{-1}). Spectra were recorded from thin films using NaCl plates. HRMS were performed at the Bioanalytical Mass Spectrometry Facility within the Mark Wainwright Analytical Centre at The University of New South Wales on an Orbitrap LTQ XL (Thermo Fisher Scientific, San Jose, Ca, USA) ion trap mass spectrometer using a nanospray (nano-electrospray) ionization source to generate ions from the analyte in solution. The instrument was calibrated with a standard calibration solution (as outlined in the instrument manual) on the day of analysis using direct infusion with the nanospray source. The instrument conditions were optimized for sensitivity on each compound of interest using LC tune software. The analysis was carried out in positive ion mode using the orbitrap FTMS analyser at a resolution of 100000. Samples, 5 μL , (1 $\mu\text{g}/\text{mL}$ in methanol or acetonitrile), were injected into a glass needle and inserted into the nanospray source. Ions generated were measured over the mass range 150 to 2000. Data was acquired in full scan mode over 60 seconds. Data was analyzed using the Qual Browser feature in Xcaliber 2.1 (Thermo Fisher Scientific, San Jose, Ca, USA). Optical rotations (α) were recorded on Rudolph Research Analytical Autopol 1 Automatic Polarimeter. Samples were prepared in 10 or 5 mL volumetric flasks at stated concentration ($\text{g}/100 \text{ mL}$) in chloroform. Measurements were taken at 589 nm (sodium D line), at the stated temperature in a 1.0 or 0.5 dm path length optical cell. Values are reported as specific rotations ($[\alpha]$).

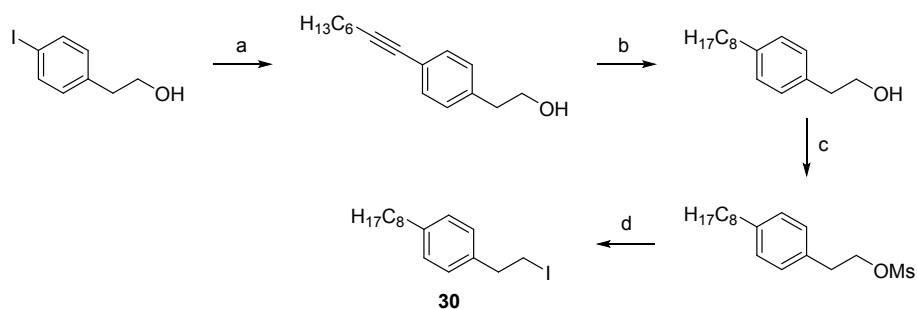
Unless otherwise stated all reactions were performed in flame dried glassware under an atmosphere of dry argon. Reaction temperatures refer to the external bath temperature.

Concentration of solvents was performed under reduced pressure on a rotary evaporator after which, residual solvent was removed under high vacuum (~0.1 mm/Hg).

Reagents and solvents were purchased from commercial sources and used without further purification, unless stated below. Reagents and solvents used in reactions were purified according to well established procedures.² In particular, tetrahydrofuran (THF) was freshly distilled from sodium and benzophenone under an inert atmosphere of argon. *N,N*-Dimethylformamide (DMF) was dried sequentially over three batches of 4Å molecular sieves (3 × 24 h), before finally being stored over a fourth batch of 4Å molecular sieves, under argon. To remove residual *N,N*-dimethylamine from DMF, the solvent was evacuated (~0.1 mm/Hg) for at least 30 min prior to use. Methanol was distilled from magnesium and stored over 3Å molecular sieves, under argon. Triethylamine and dichloromethane were distilled from calcium hydride immediately prior to use. (*S*)-Schöllkopf reagent **29** was distilled immediately before use (53 – 55°C at 0.1 mm/Hg). *n*-Butyllithium in hexanes was purchased from Sigma Aldrich and titrated using menthol and 2,2'-bipyridine in THF as described by Eastham.³ Tetrakis(triphenylphosphine)palladium(0) was synthesised following the reported method.⁴ The following compounds were synthesised in a previous communication: TBS-ether **21**, phenol **27**, analogs **4 - 11**, **12**, **16** and **17**.⁵

Analytical thin layer chromatography was conducted on Merck, aluminium-backed silica plates 60 F₂₅₄ or silica gel 60 RP-C₁₈ F₂₅₄ plates and visualised using UV light and stained with a dip of either a potassium permanganate, vanillin or phosphomolybdic acid. Flash chromatography was routinely performed using Grace Davison Discovery Sciences, Davisil LC60A 40 – 63 micron silica gel, following published guidelines.⁶ Solvent was eluted using a Thomson SINGLE StEP pump at the flow rate recommended by the manufacturer (Thomson Instrument Company, Oceanside, Ca, USA). Deactivated silica gel was prepared by washing a column packed with silica gel with neat triethylamine (5 column volumes). After drying, the column was washed with *n*-hexane to remove any residual triethylamine.

2. Synthesis of 1-(2-iodoethyl)-4-octylbenzene (30)



(a) Tetrakis(triphenylphosphine)palladium(0) (48 mg, 0.072 mmol), copper(I) iodide (136 mg, 0.71 mmol), 2-(4-iodophenyl)-1-ethanol (**S6**) (1.46 g, 7.15 mmol), 1-octyne (1.7 mL, 11.52 mmol) and triethylamine (35.8 mL) were placed in a Youngs tube. The flask was freeze-pump-thawed ($\times 3$) then backfilled with argon. The solution was stirred at room temperature for 17 h. The mixture was poured onto 1M aqueous hydrochloric acid solution and extracted with ethyl acetate ($\times 3$). The organic extracts were combined and washed with water and brine, then dried (Na_2SO_4). The solvent was removed under reduced pressure and the crude material purified by flash chromatography on silica gel, eluting with 15 % ethyl acetate/*n*-hexane, to afford 2-(4-(oct-1-yn-1-yl)phenyl)-1-ethanol (**S7**) as a clear colourless oil (0.84 g, 51 %). ^1H NMR (300 MHz; CDCl_3) δ 0.90 (t, $J = 6.9$ Hz, 3H), 1.29 – 1.37 (m, 4H), 1.40 – 1.50 (m, 2H), 1.55 – 1.65 (m, 3H), 2.39 (t, $J = 7.0$ Hz, 2H), 2.84 (t, $J = 6.6$ Hz, 2H), 3.81 – 3.87 (m, 2H), 7.13 – 7.16 (m, 2H), 7.33 – 7.35 (m, 2H); ^{13}C NMR (75 MHz; CDCl_3) δ 14.2, 19.6, 22.7, 28.7, 28.9, 31.5, 39.2, 63.6, 80.5, 90.4, 122.4, 129.0, 131.9, 138.0; IR (NaCl, neat) 2227, 3350 cm^{-1} ; HRMS (ESI-MS): m/z calcd for $\text{C}_{16}\text{H}_{22}\text{ONa}$ $[\text{M}+\text{Na}]^+$ 253.2568, found 253.1552.

(b) A balloon filled with hydrogen gas was attached to a flask containing 2-(4-(oct-1-yn-1-yl)phenyl)-1-ethanol (**S7**) (0.38 g, 1.63 mmol) and 10 wt. % Pd/C (0.19 g) in ethyl acetate (8 mL). The flask was evacuated and purged with hydrogen gas ($\times 3$) then allowed to stir at room temperature for 17 h. The reaction mixture was filtered through a short pad of Celite, eluting with ethyl acetate. The solvent was removed under reduced pressure to afford 2-(4-octylphenyl)-1-ethanol (**S8**) as a clear colourless oil which did not require further purification (0.37 g, 98 %). With all analytical data matching that reported in the literature.⁸ ^1H NMR (300 MHz; CDCl_3) δ 0.88 (t, $J = 6.9$ Hz, 3H), 1.27 – 1.39 (m, 12H), 1.62 (s, 1H), 2.57 (t, $J = 7.0$ Hz, 2H), 2.84 (t, $J = 6.5$ Hz, 2H), 3.82 – 3.88 (m, 2H), 7.12 – 7.14 (m, 4H).

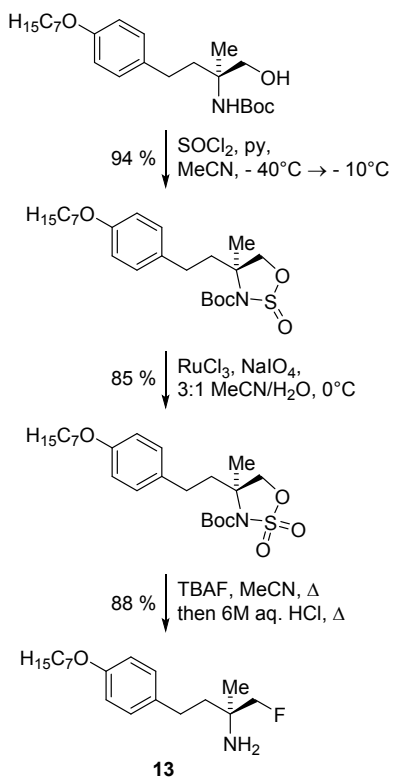
(c) Methanesulfonyl chloride (0.2 mL, 2.58 mmol) was added dropwise to a solution of 2-(4-octylphenyl)-1-ethanol (**S8**) (0.40 g, 1.72 mmol) and triethylamine (0.72 mL, 5.12 mmol) in dichloromethane (17 mL) at 0°C . The solution was allowed to stir at 0°C for 15 min. then, the cold bath was removed and the solution stirred at room temperature for 3 h.

The reaction mixture was poured onto brine and extracted with dichloromethane ($\times 2$). The organic extracts were combined and washed with water and brine, then dried (Na_2SO_4). The solvent was removed under reduced pressure to afford a light orange residue of mesylate **S9** which was used in the next step without further purification.

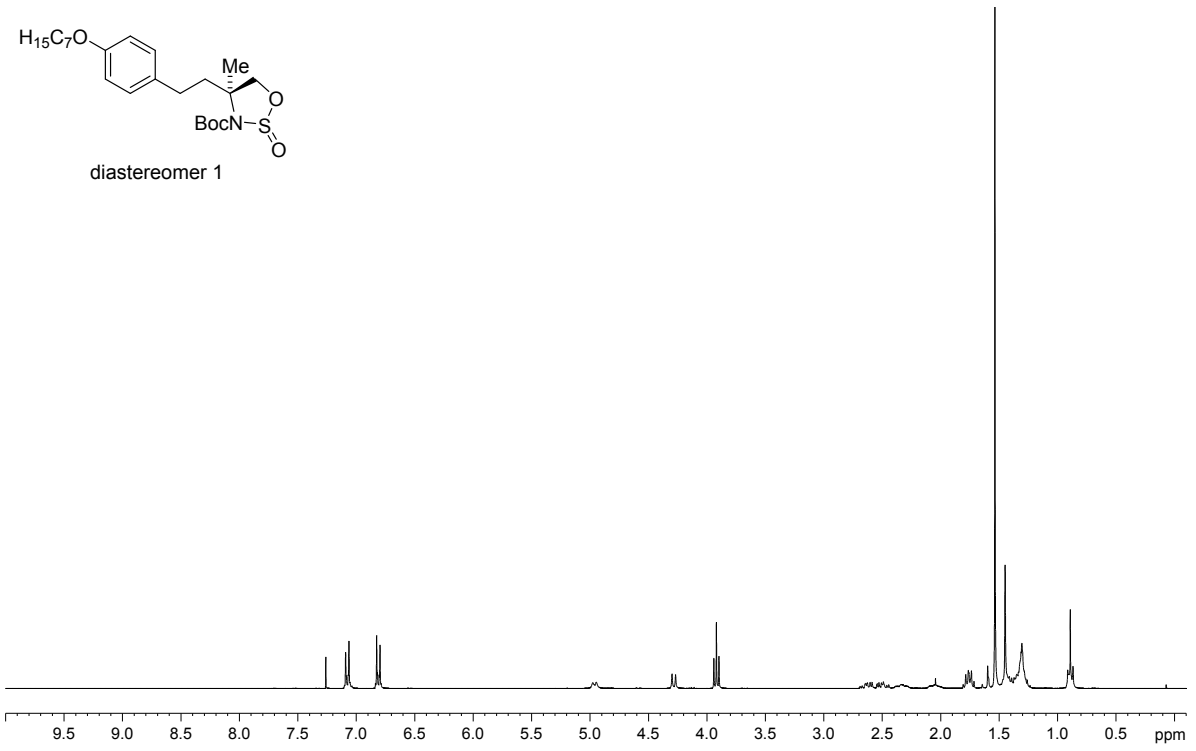
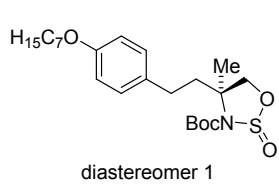
(d) The crude material (**S9**) was dissolved in acetone (8 mL) and sodium iodide (2.59 g, 17.19 mmol) was added as a solid in one portion at room temperature. The solution was stirred at room temperature, protected from light, for 19 h. The acetone was removed under reduced pressure. The residue was taken up in brine and extracted with dichloromethane ($\times 3$). The organic extracts were combined and washed with water and brine, then dried (Na_2SO_4). The solvent was removed under reduced pressure and the crude material was purified by flash chromatography on silica gel, eluting with 1 % ethyl acetate/*n*-hexane, to afford the title compound **30** as a clear colourless oil (0.44 g, 74 %). With all analytical data matching that reported in the literature.⁸ ^1H NMR (300 MHz; CDCl_3) δ 0.88 (t, $J = 6.9$ Hz, 3H), 1.20 – 1.30 (m, 10H), 1.55 – 1.62 (m, 2H), 2.55 – 2.63 (m, 2H), 3.12 – 3.18 (m, 2H), 3.31 – 3.36 (m, 2H), 7.08 – 7.15 (m, 4H).

3. Scheme Summarising the Intermediates and NMR Data for the Synthesis of (2S)-1-Fluoro-4-(4'-heptyloxyphenyl)-2-methylbutan-2-amine (13)

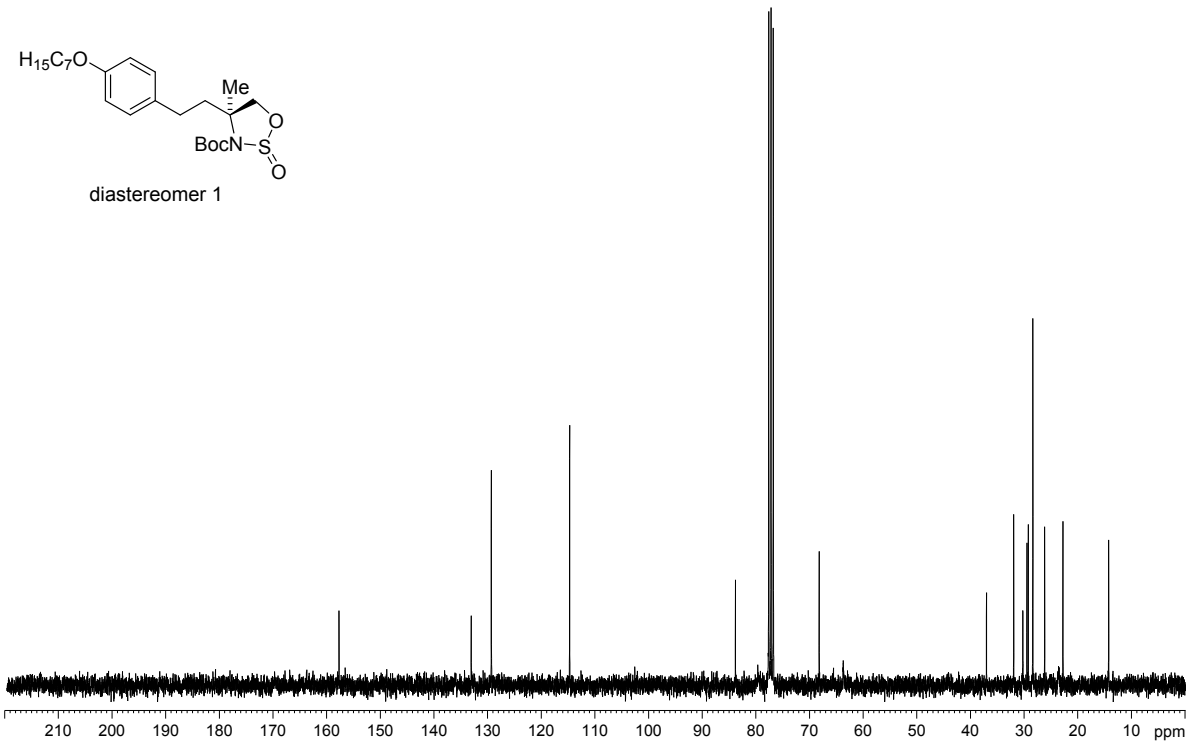
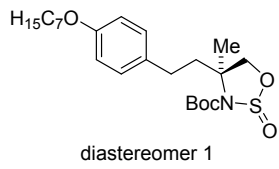
Scheme used to synthesise (2S)-1-Fluoro-4-(4'-heptyloxyphenyl)-2-methylbutan-2-amine (13).



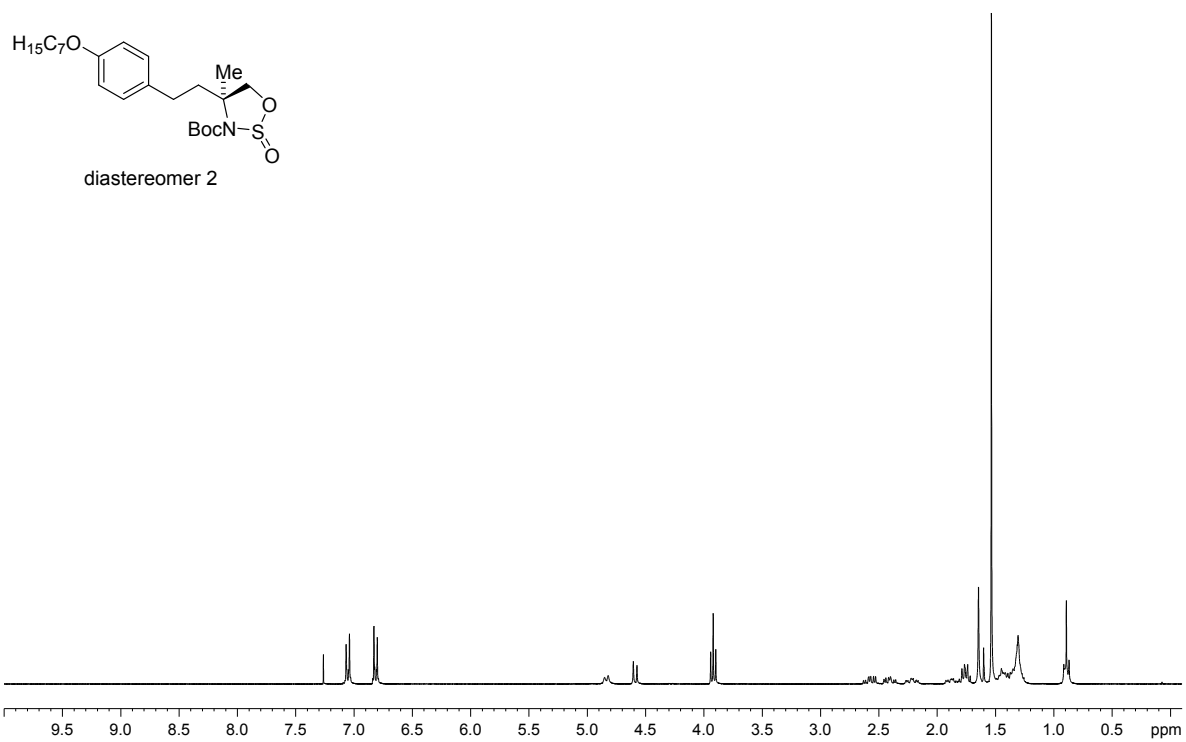
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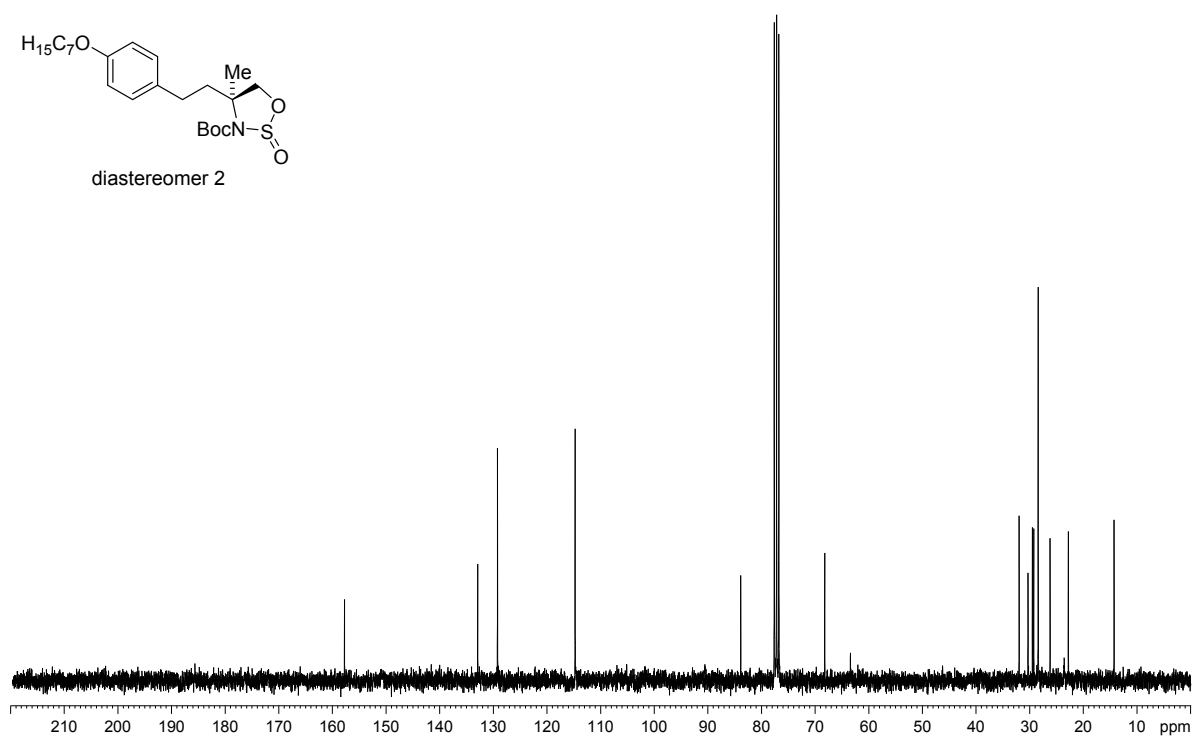
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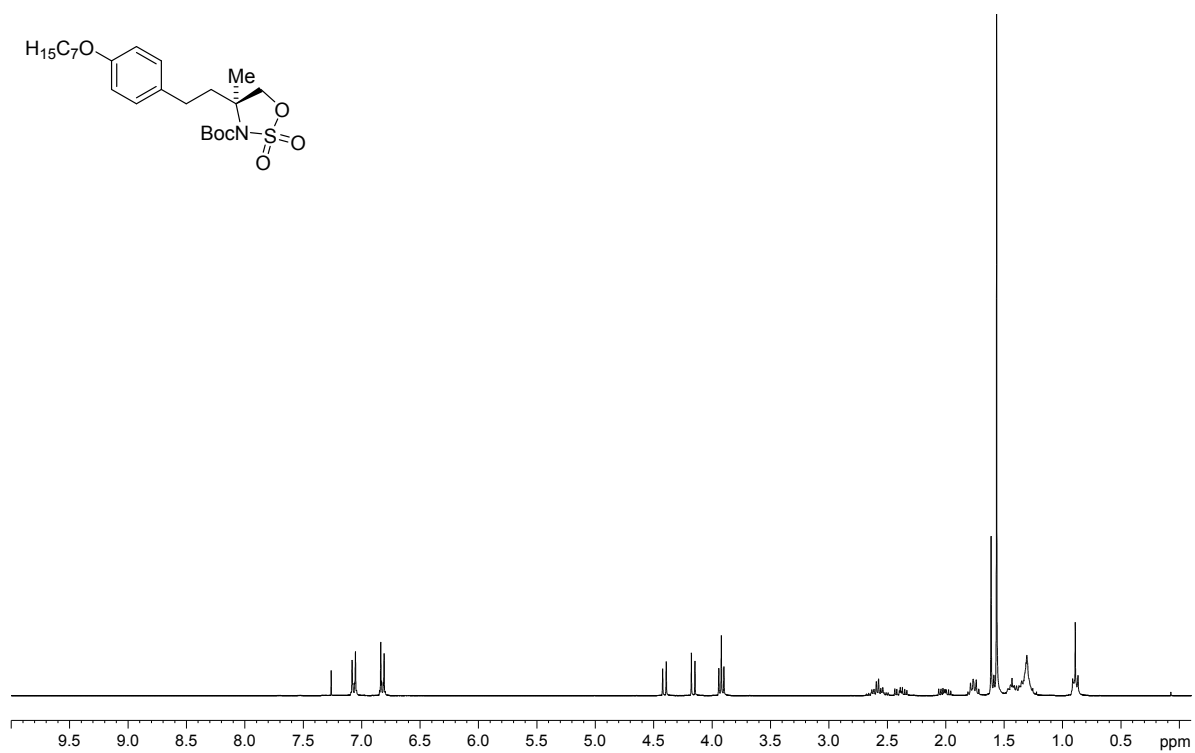
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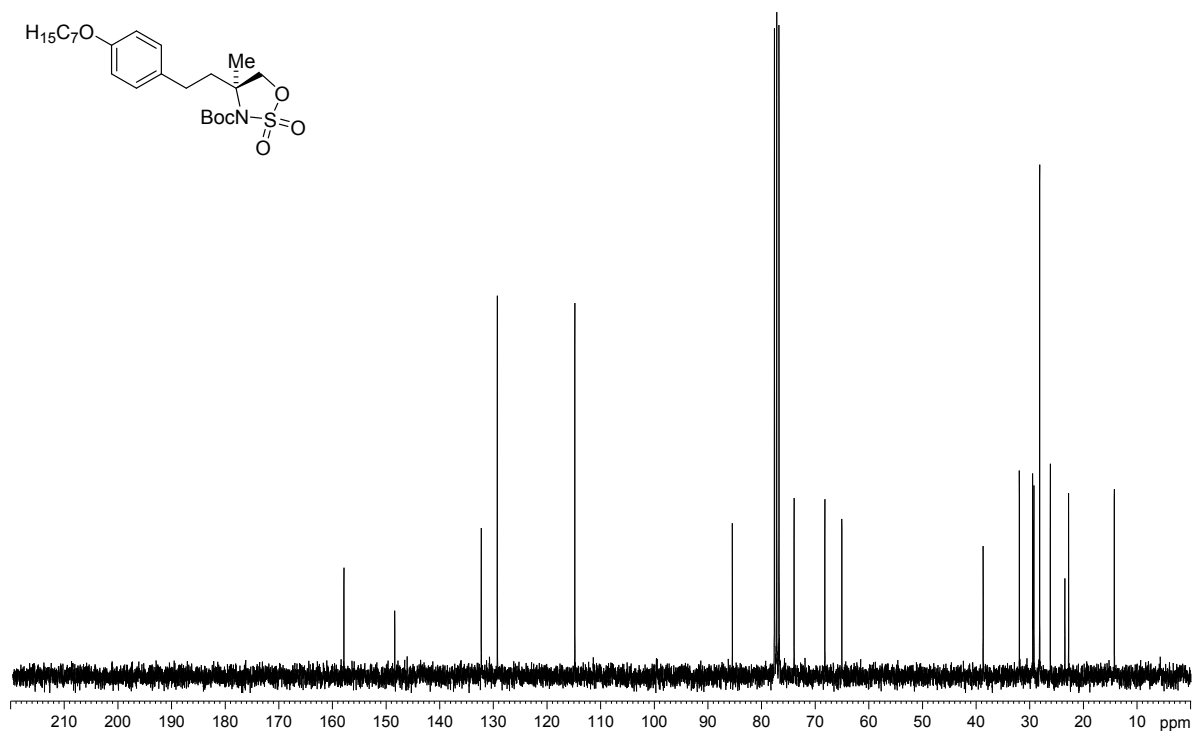
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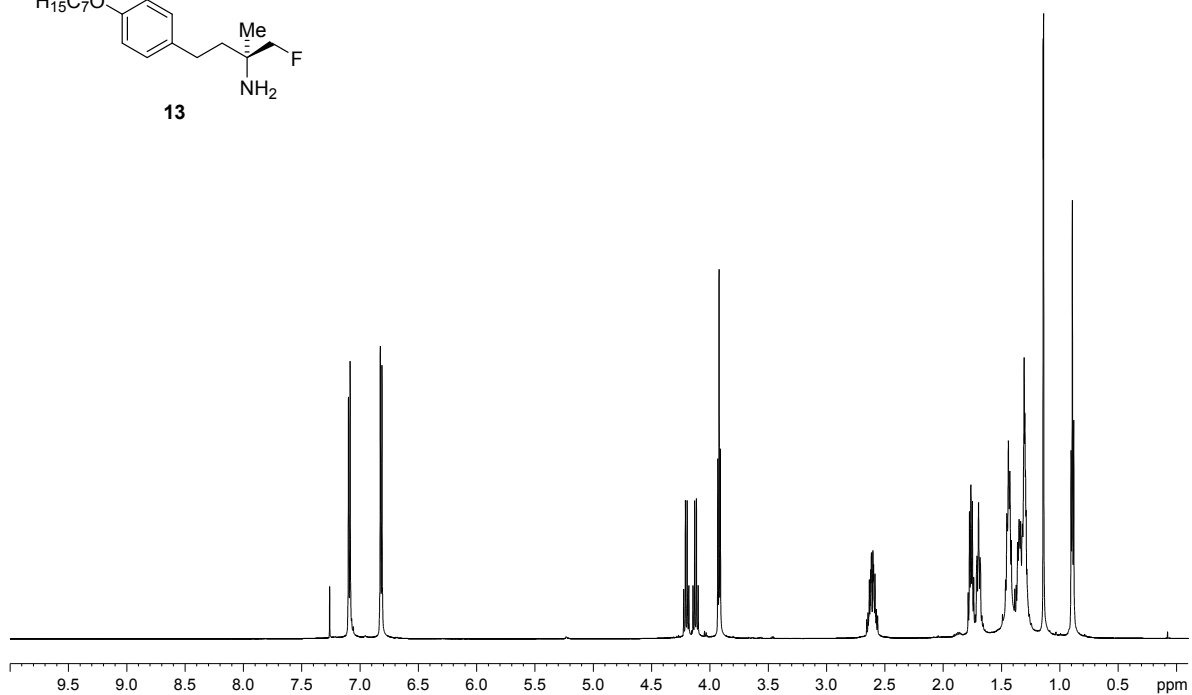
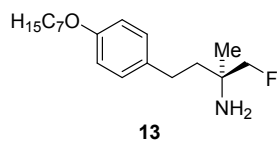
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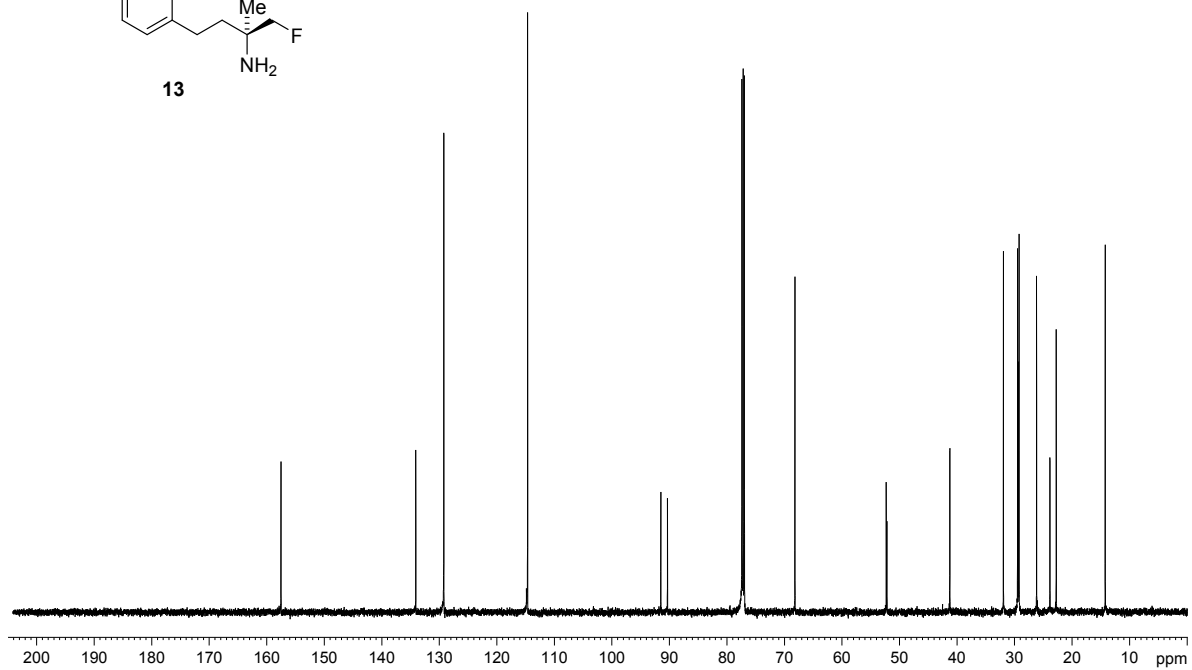
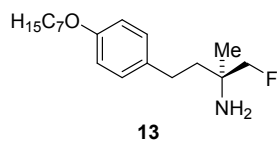
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01/02/2012 HDTI014_2; 600 MHz; CDCl3

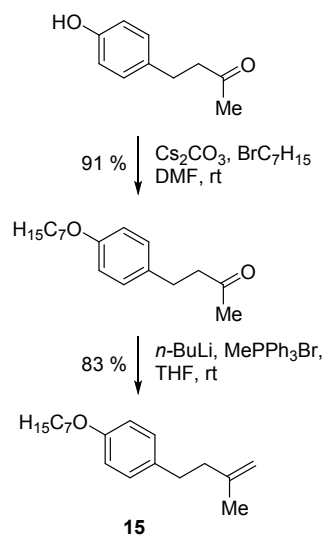


01/02/2012 HDTI014_2; 600 MHz; CDCl3

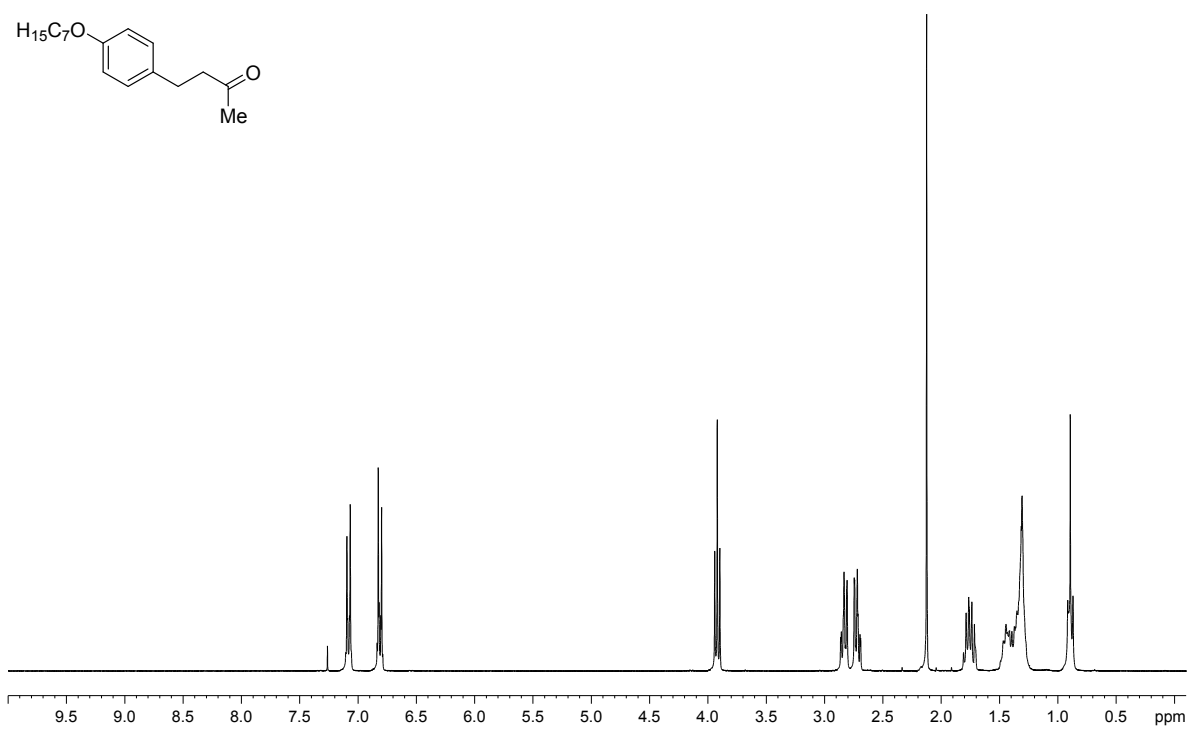
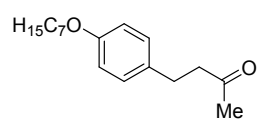


4. Scheme Summarising the Intermediates and NMR Data for the Synthesis of 1-(Heptyloxy)-4-(3'-methylbut-3'-enyl)benzene (**15**)

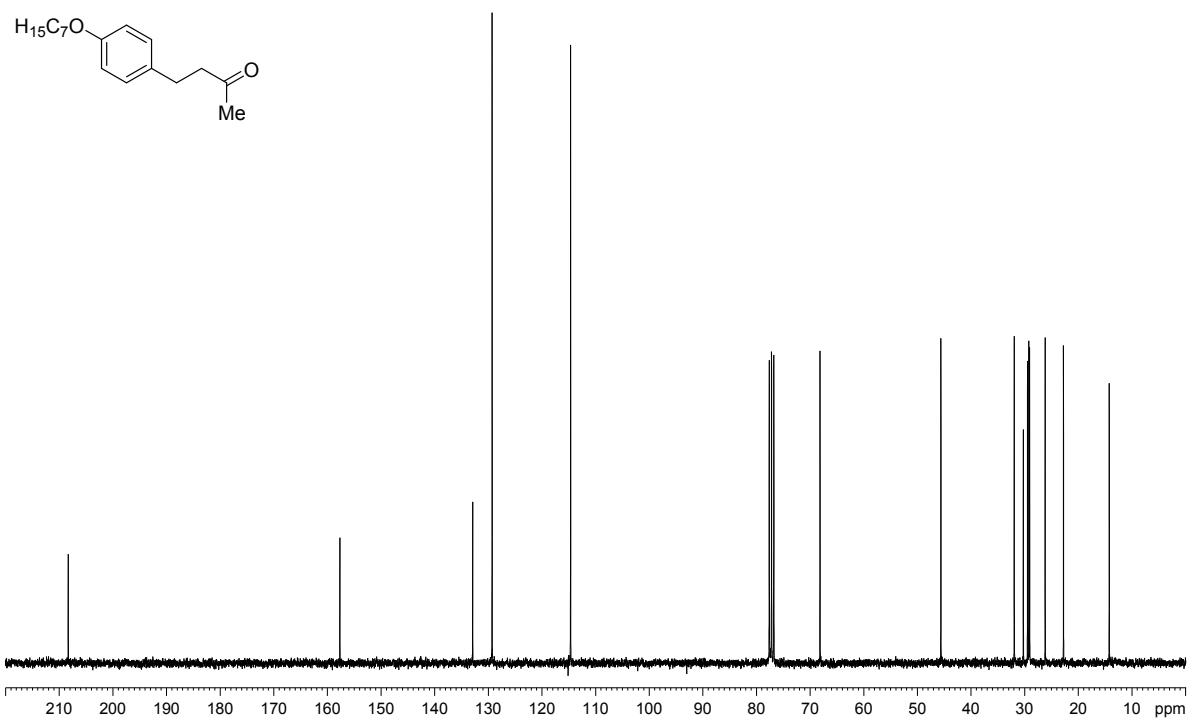
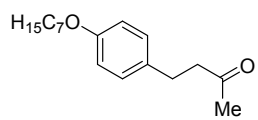
Scheme used to synthesise 1-(Heptyloxy)-4-(3'-methylbut-3'-enyl)benzene (**15**).



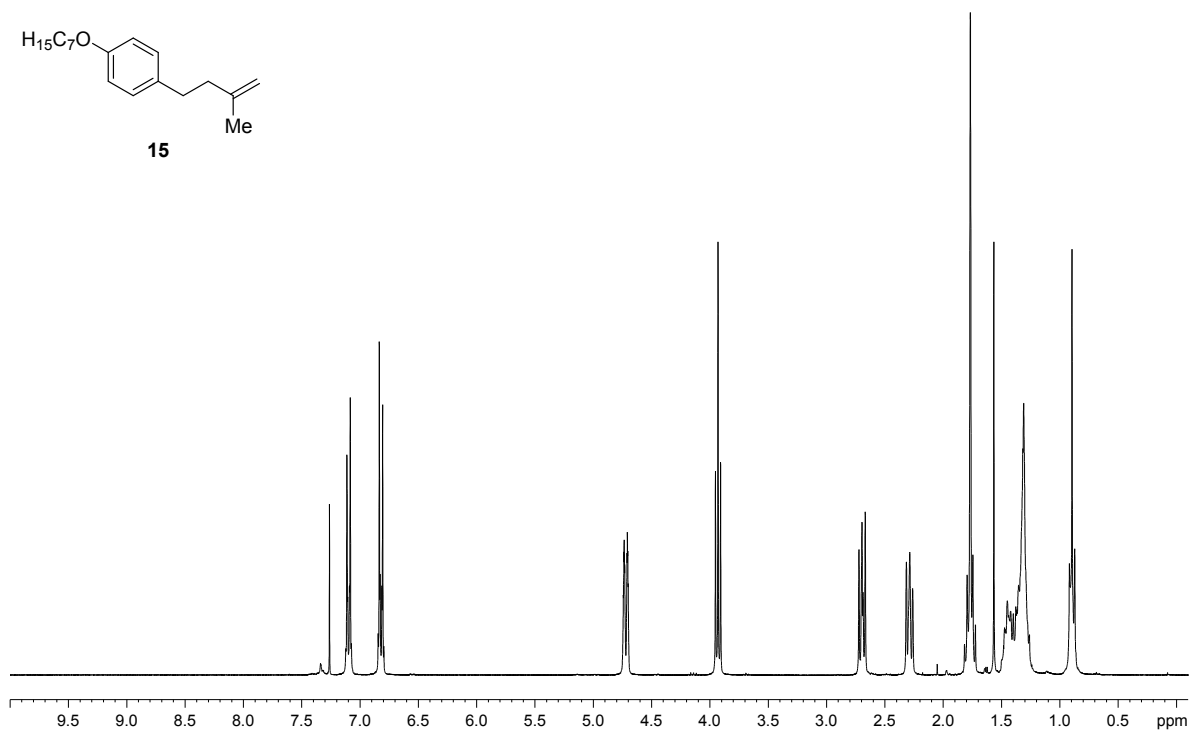
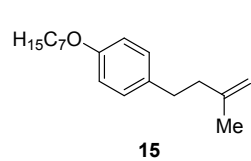
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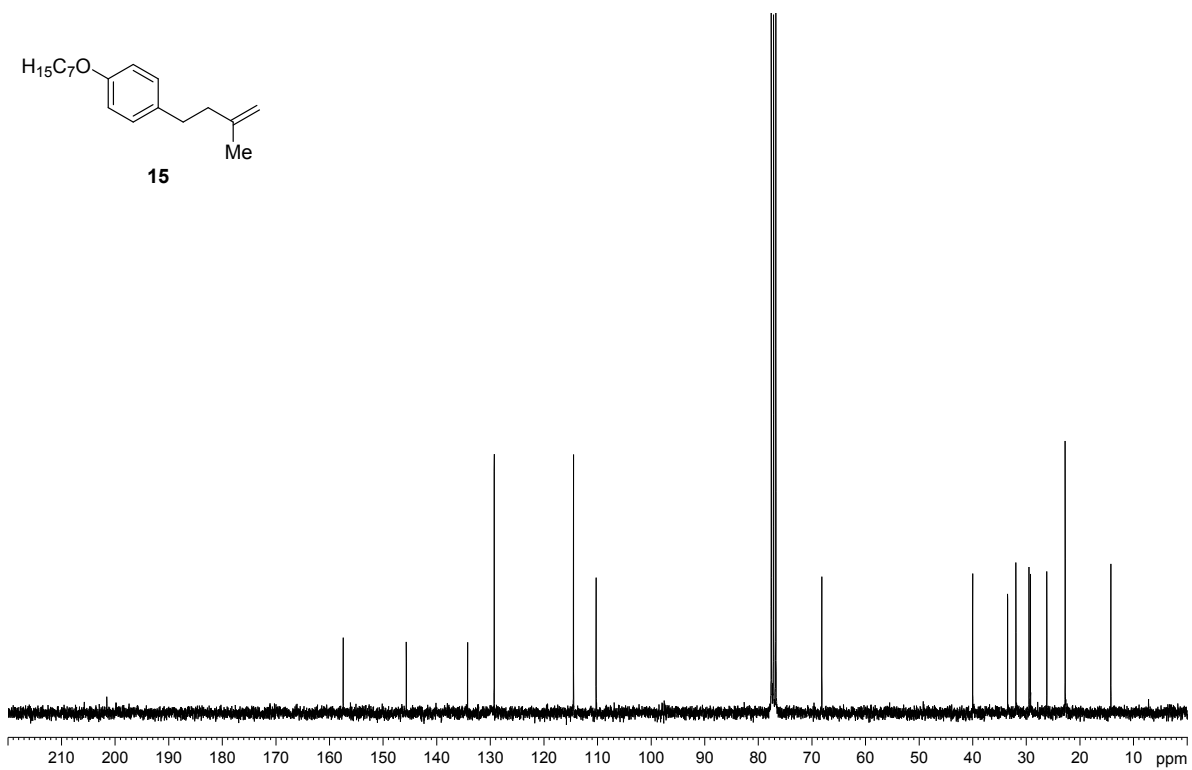
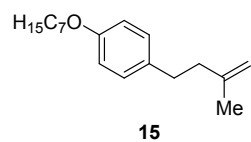
26/04/2011 HDTF005_2; 300 MHz; CDCl₃



01/05/2011 HDTF009_2; 300 MHz; CDCl3

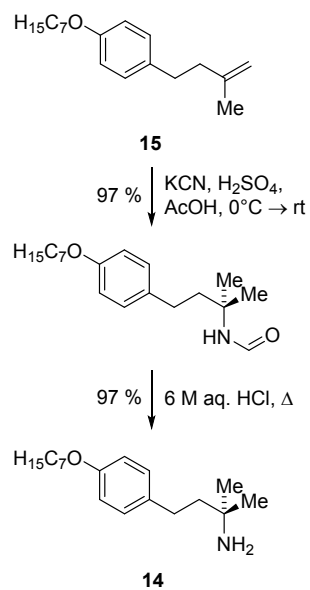


01/05/2011 HDTF009_2; 300 MHz; CDCl3

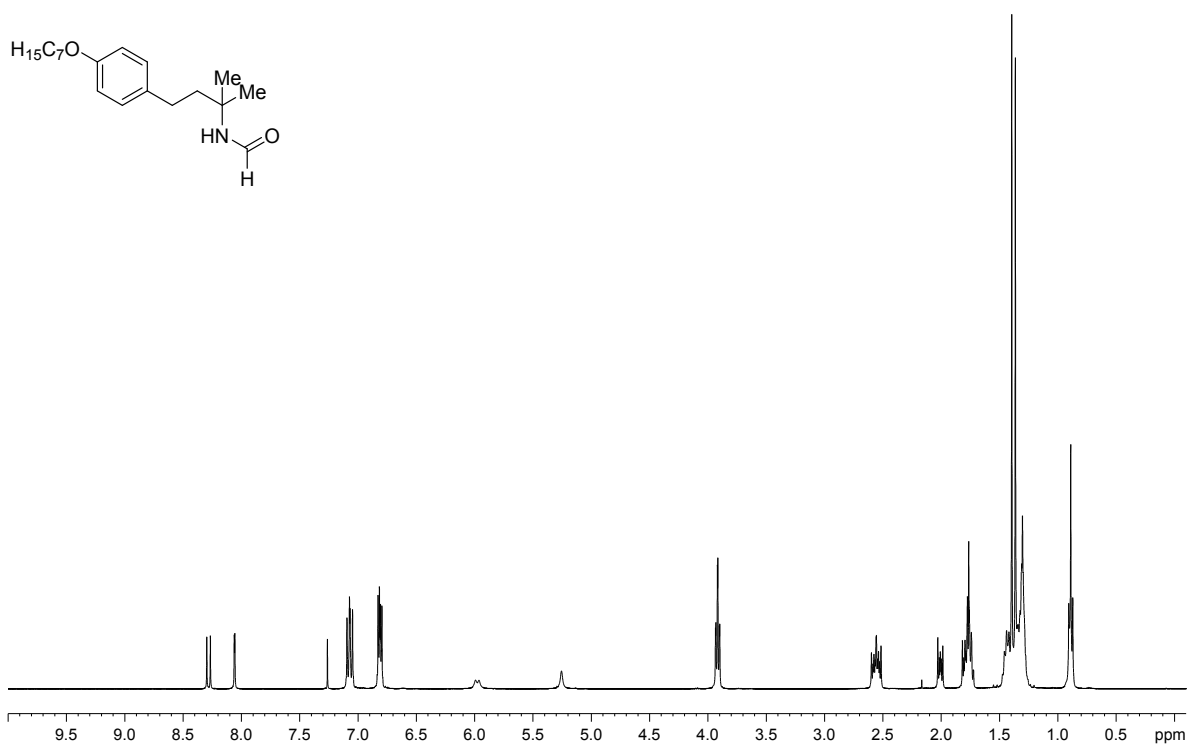


5. Scheme Summarising the Intermediates and NMR Data for the Synthesis of 4-(4'-Heptyloxyphenyl)-2-methylbutan-2-amine (14)

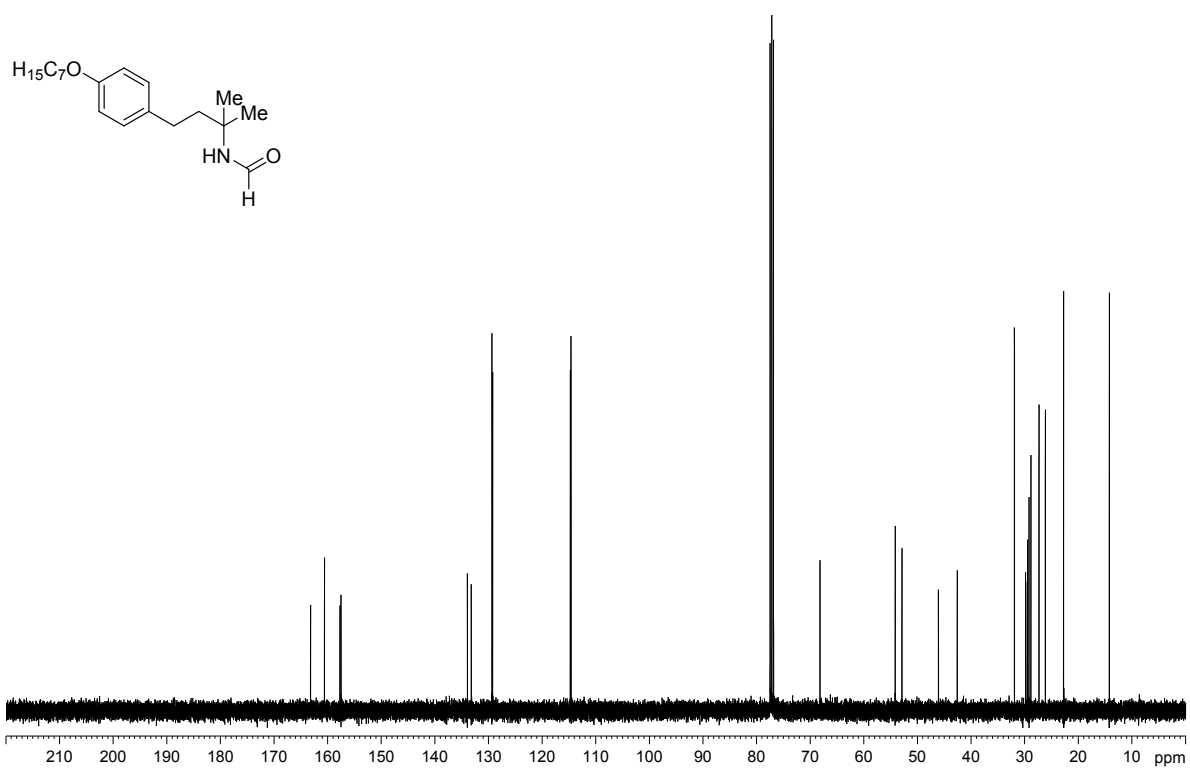
Scheme used to synthesise 4-(4'-Heptyloxyphenyl)-2-methylbutan-2-amine (14).



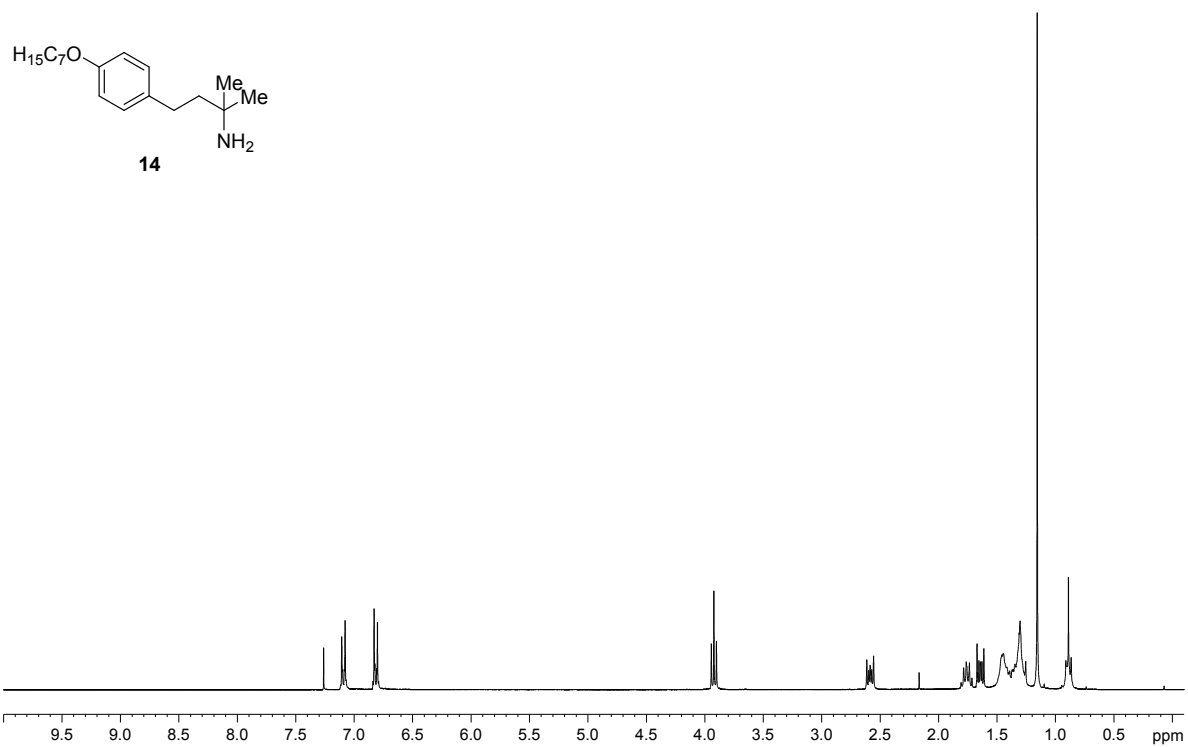
26/07/2011 HDTF019_#, 400 MHz; CDCl3



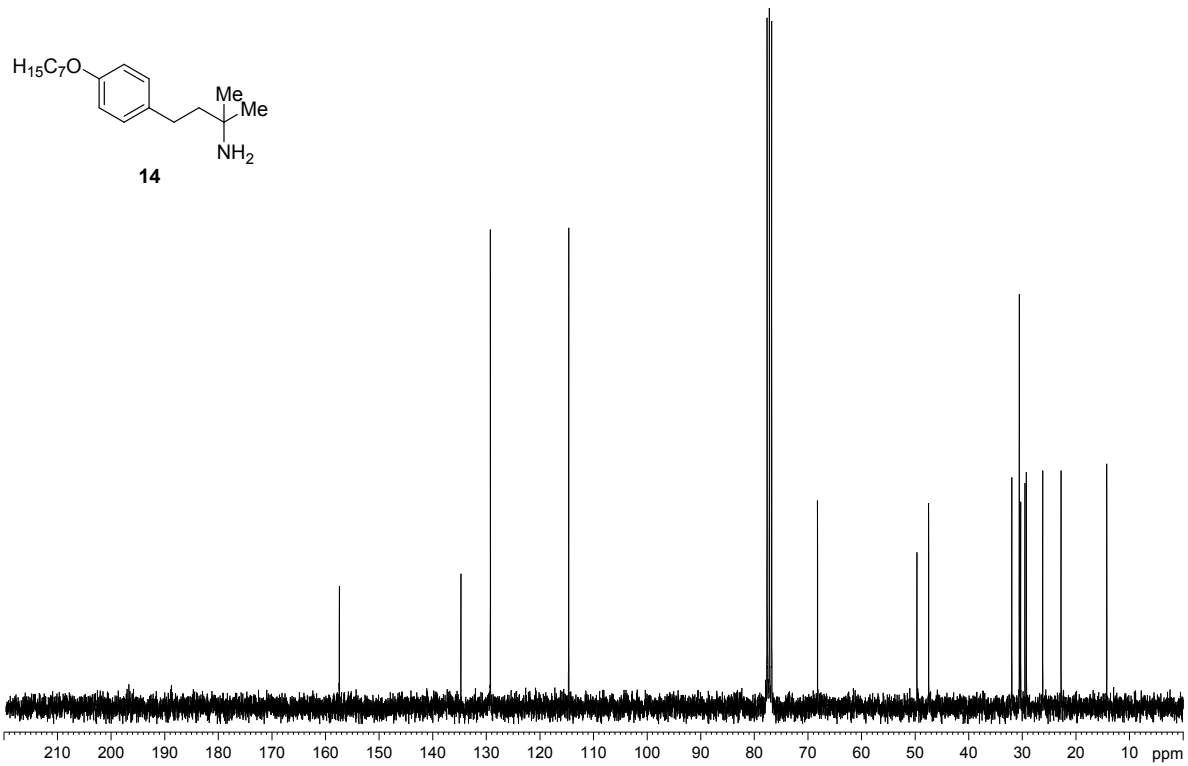
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29/08/2011 HDTG052_1; 300 MHz; CDCl3

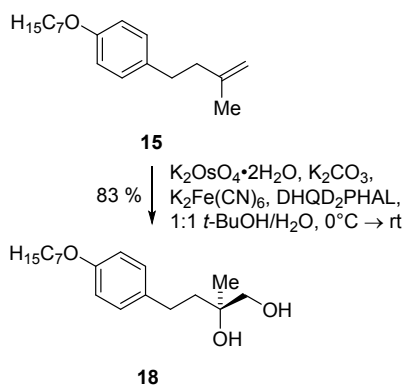


29/08/2011 HDTG052_1; 300 MHz; CDCl3

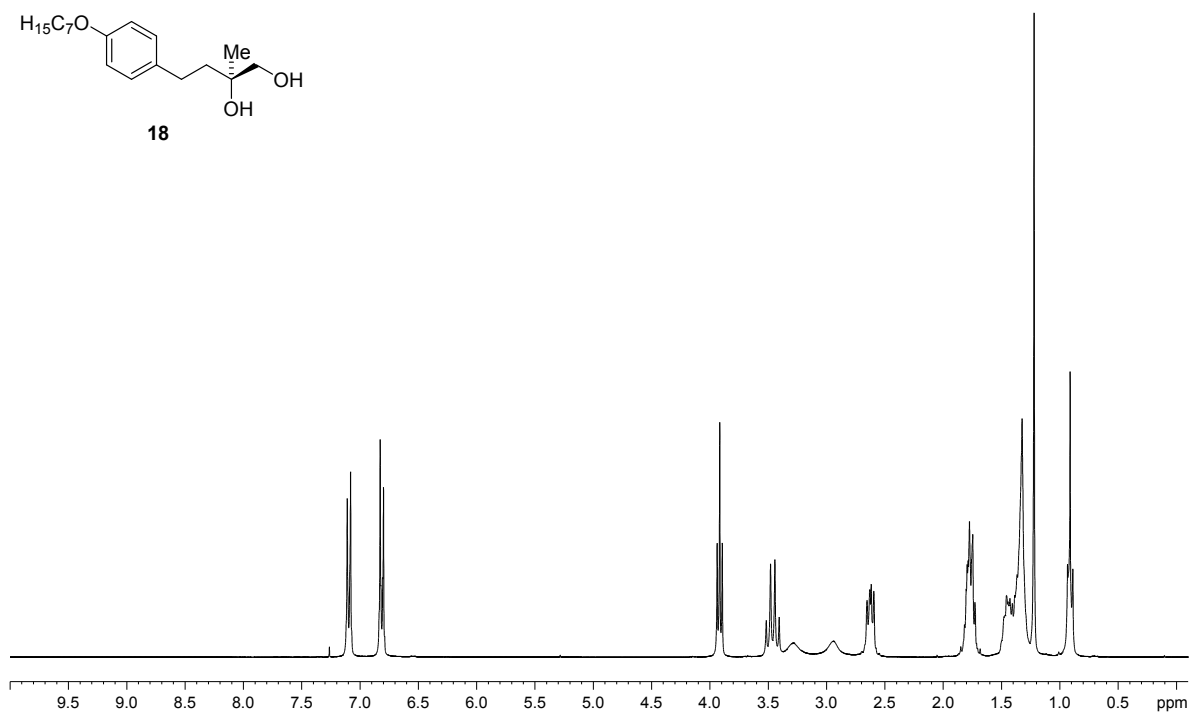
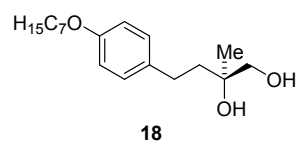


6. Scheme Summarising the Intermediates and NMR Data for the Synthesis of (2S)-4-(4'-Heptyloxyphenyl)-2-methylbutane-1,2-diol (18)

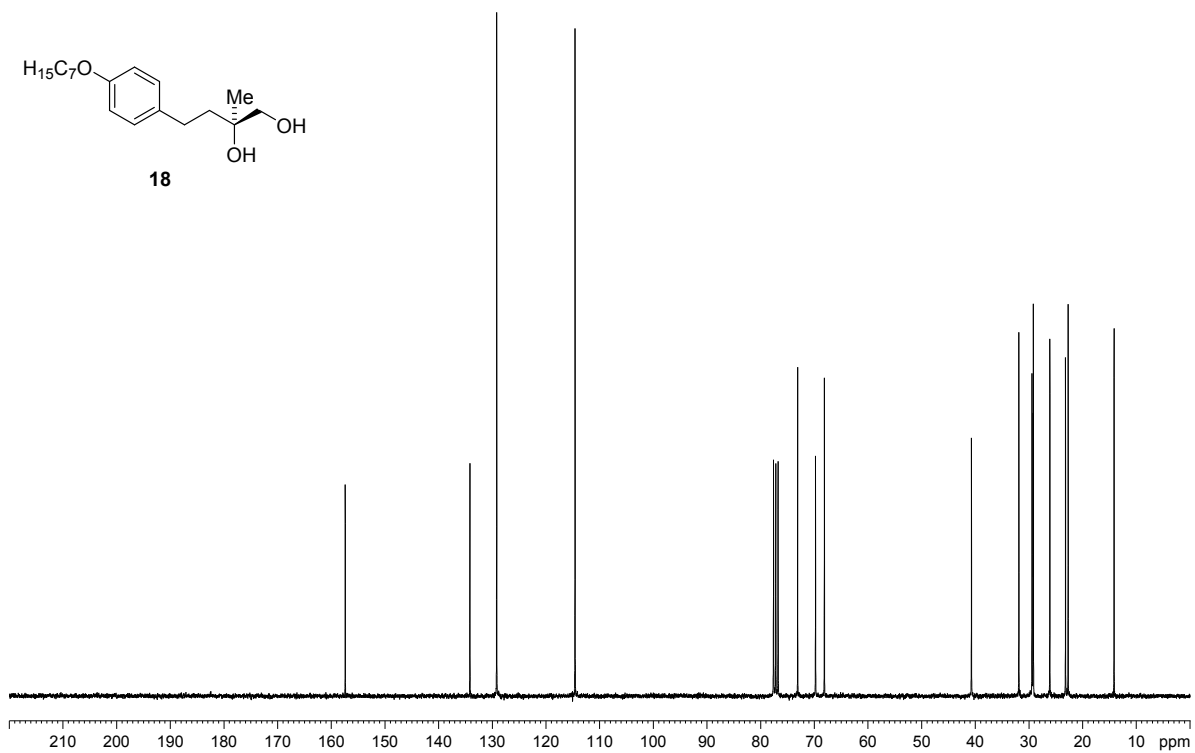
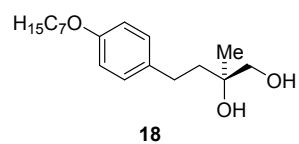
Scheme used to synthesise (2S)-4-(4'-Heptyloxyphenyl)-2-methylbutane-1,2-diol (**18**)



24/05/2011 HDTF020_2; 300 MHz; CDCl3

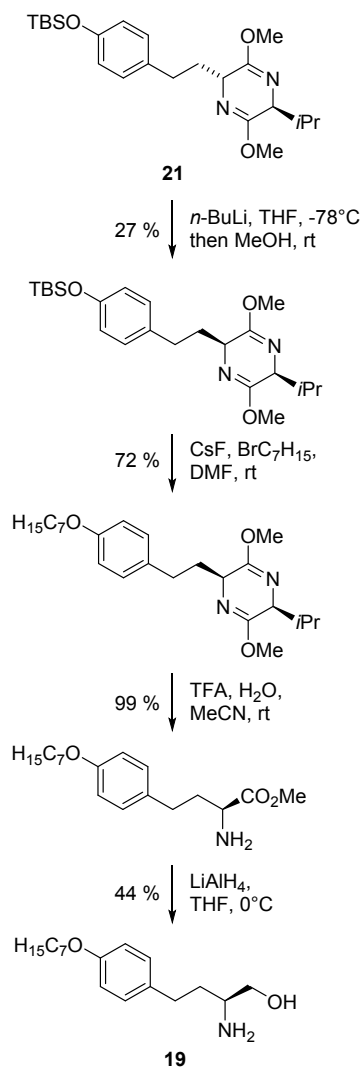


24/05/2011 HDTF020_2; 300 MHz; CDCl3

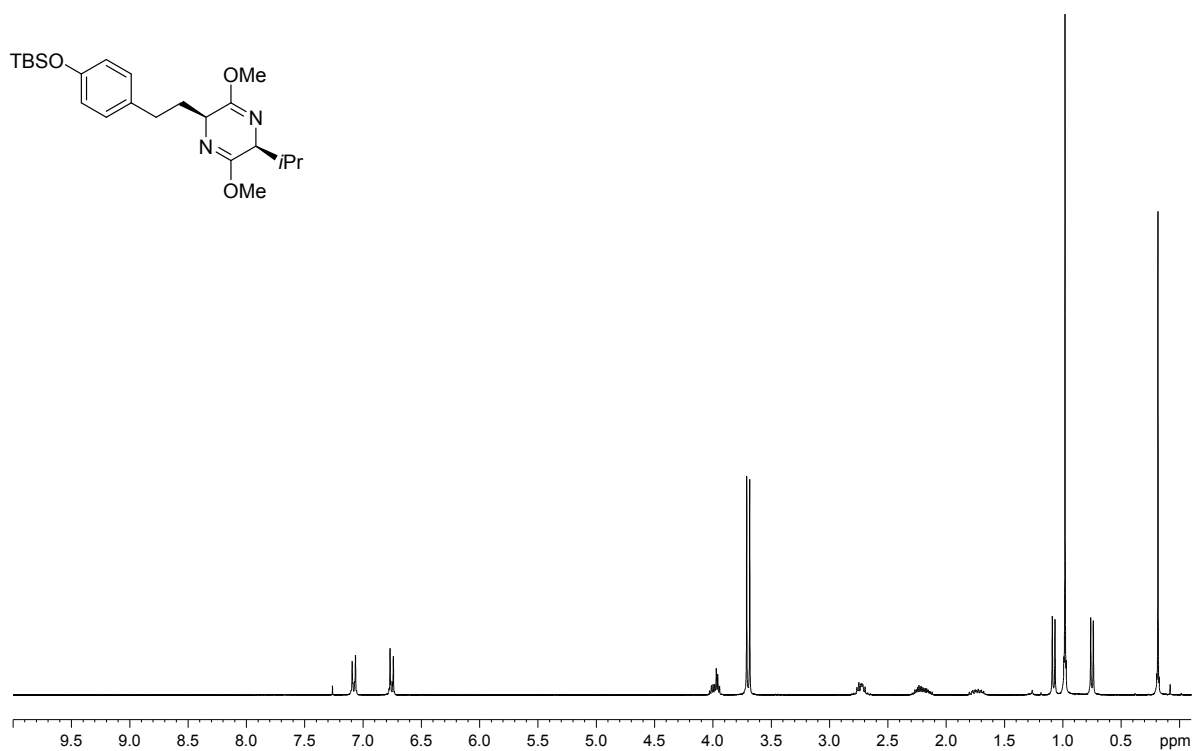


7. Scheme Summarising the Intermediates and NMR Data for the Synthesis of (2S)-2-Amino-4-(4'-heptyloxyphenyl)butan-1-ol (19)

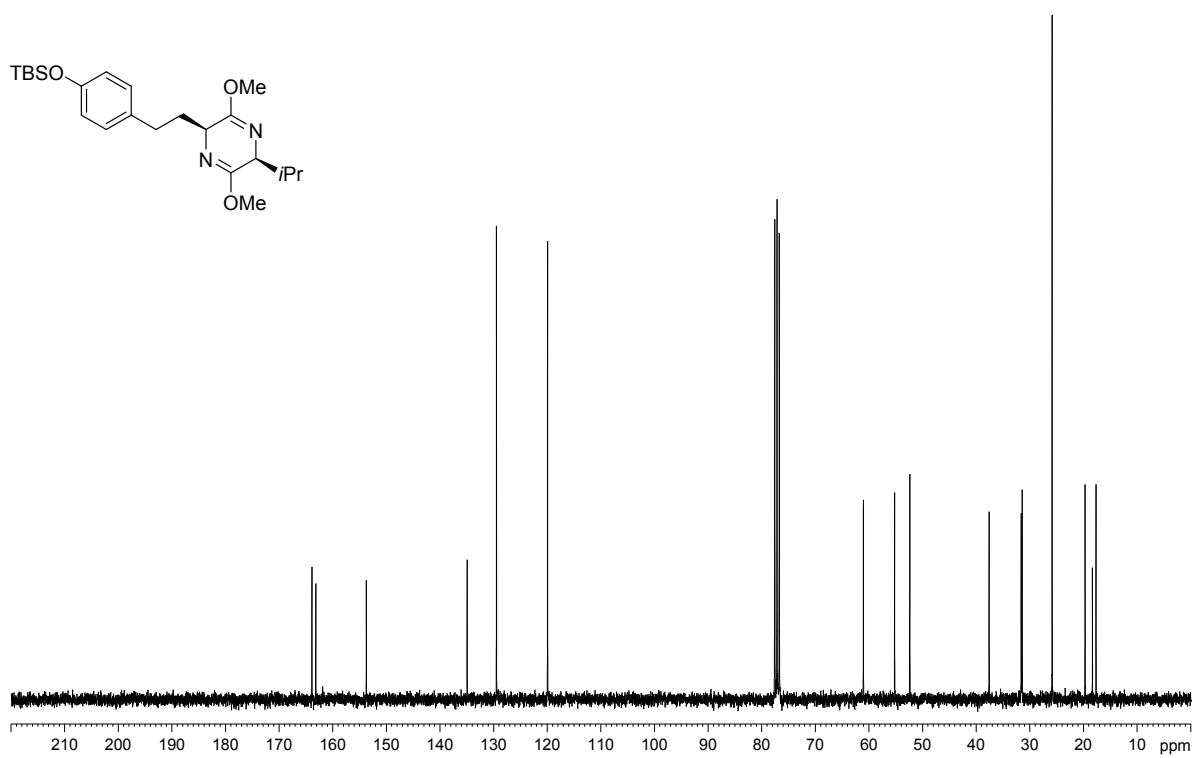
Scheme used to synthesise (2S)-2-Amino-4-(4'-heptyloxyphenyl)butan-1-ol (19)



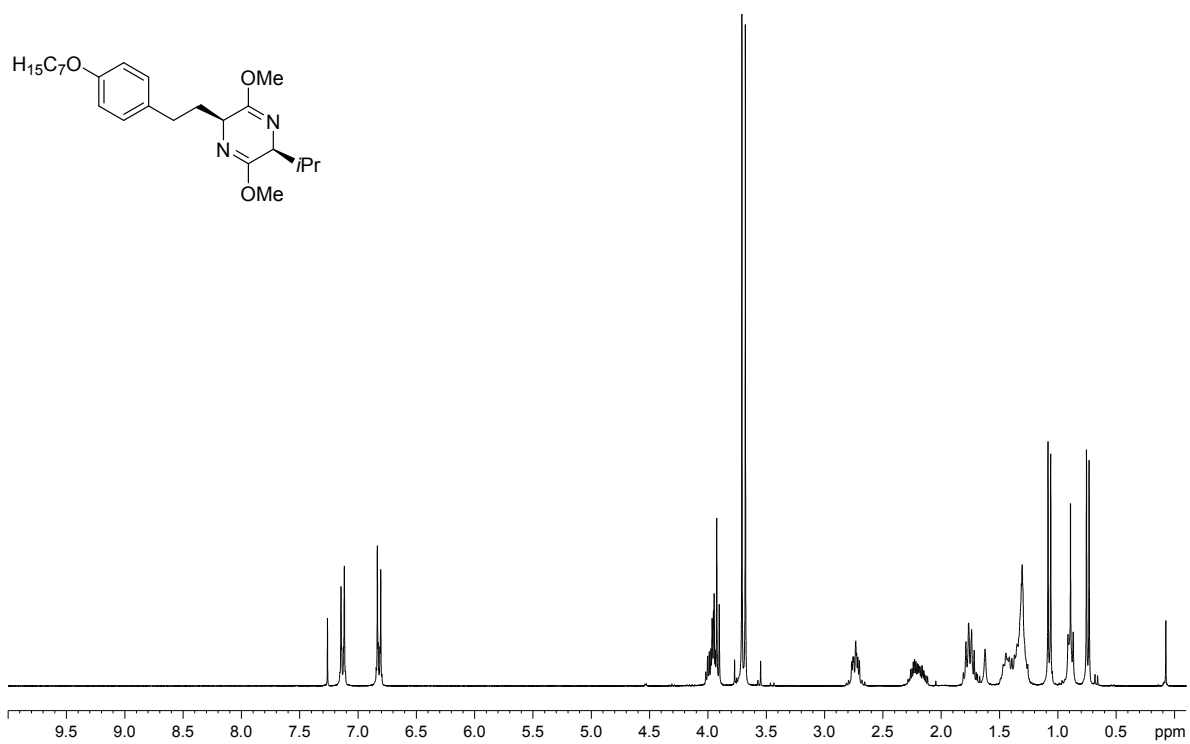
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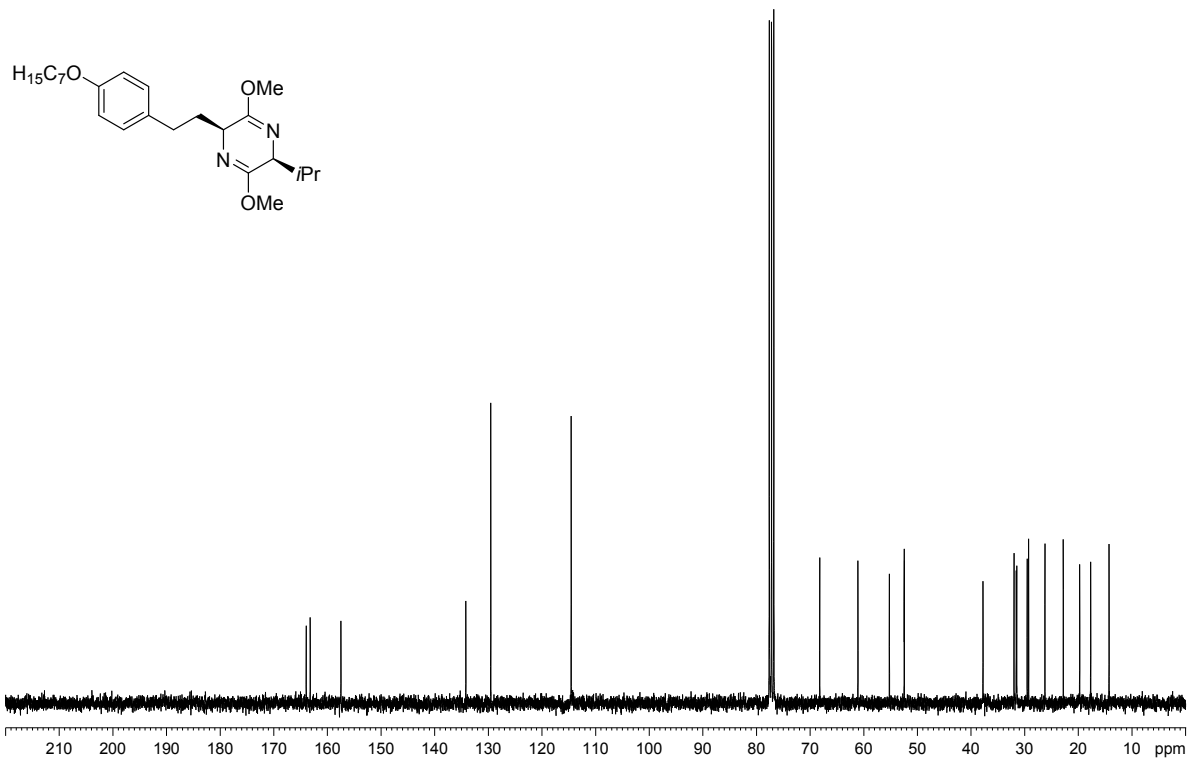
13/05/2011 HDTF008_4; 300 MHz; CDCl₃



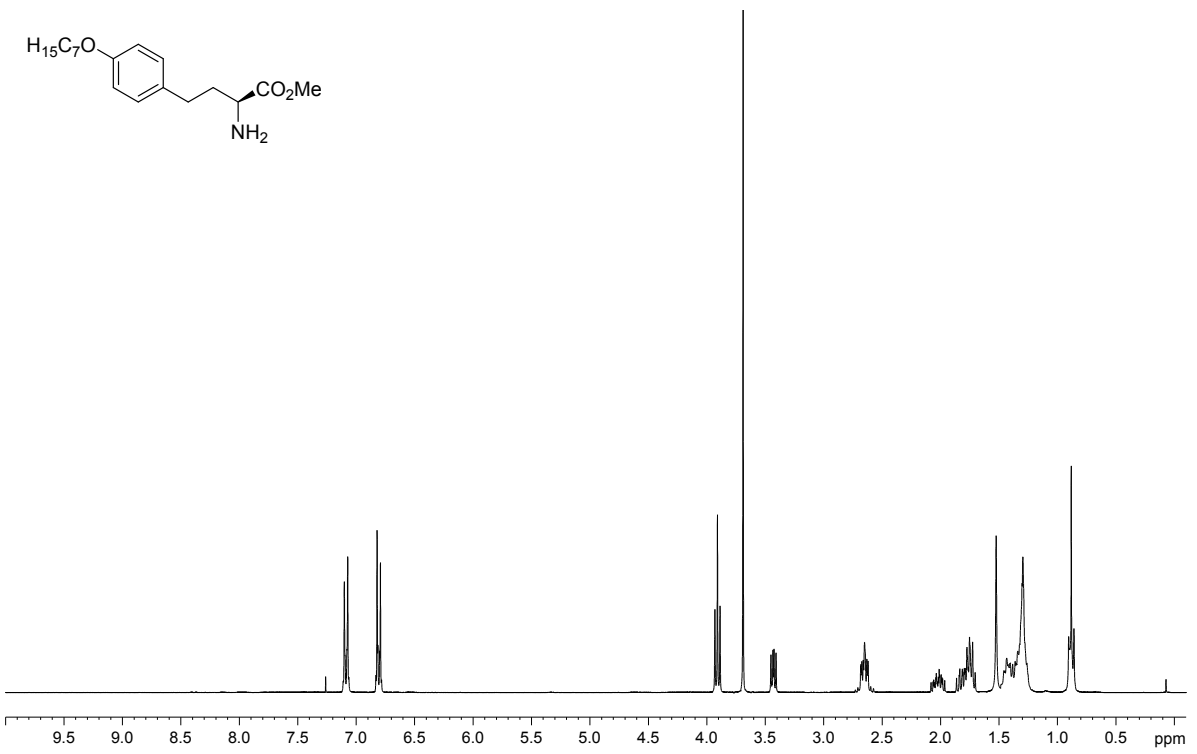
21/05/2011 HDTF018_3; 300 MHz; CDCl₃



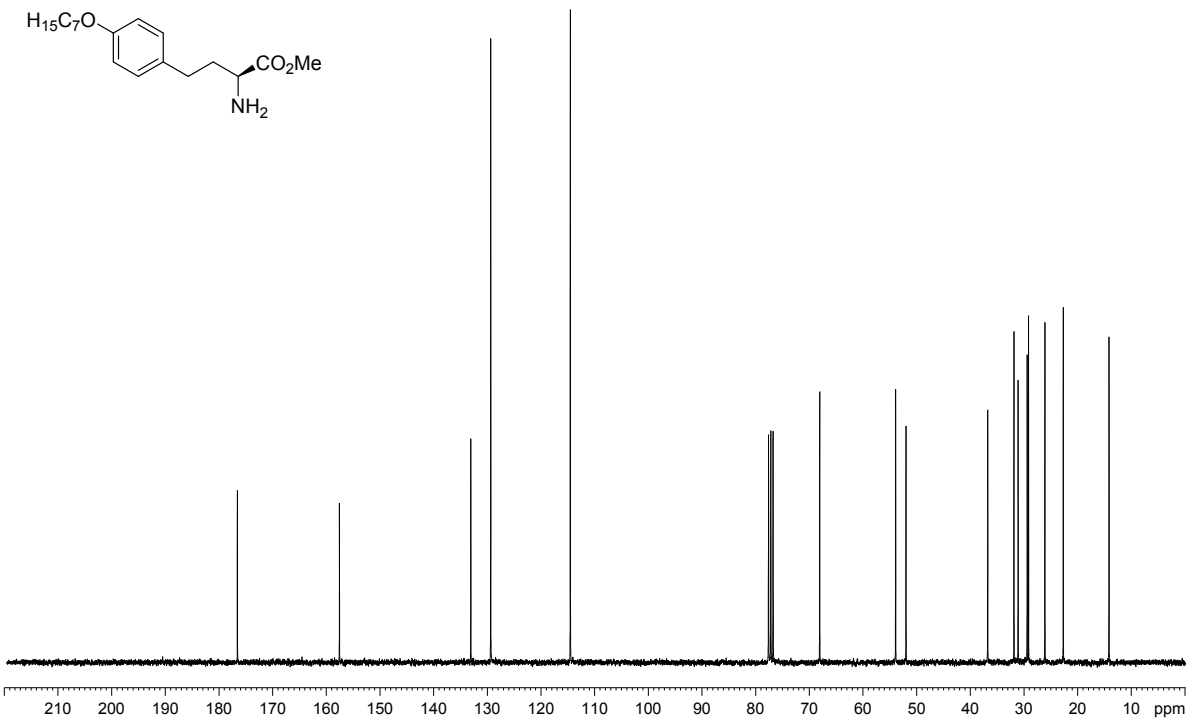
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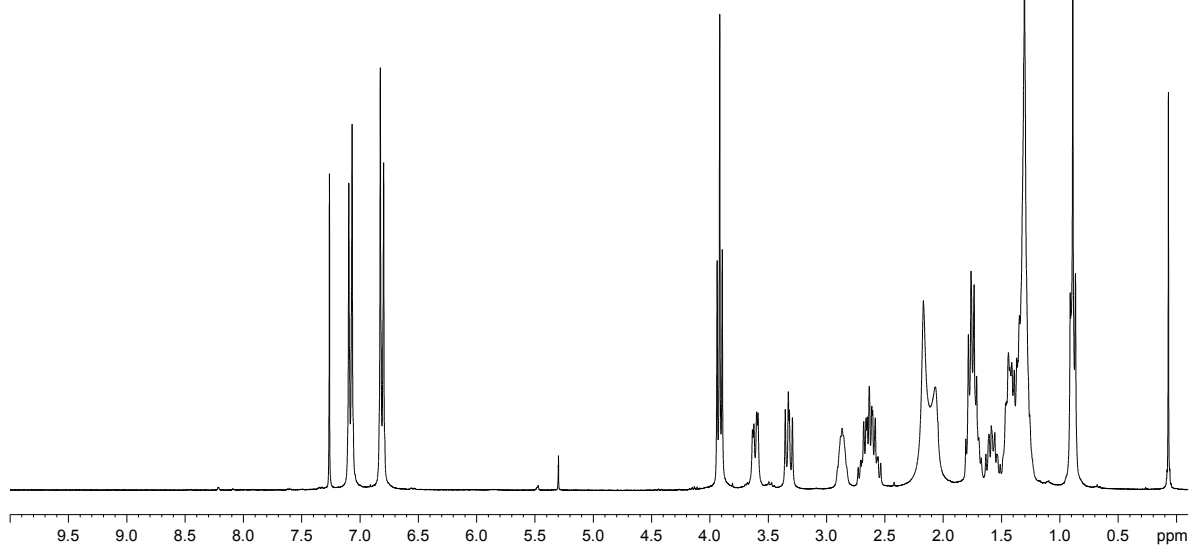
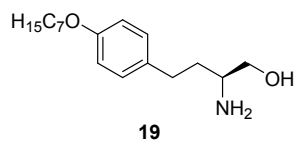
07/09/2011 HDTH001_1; 300 MHz; CDCl3



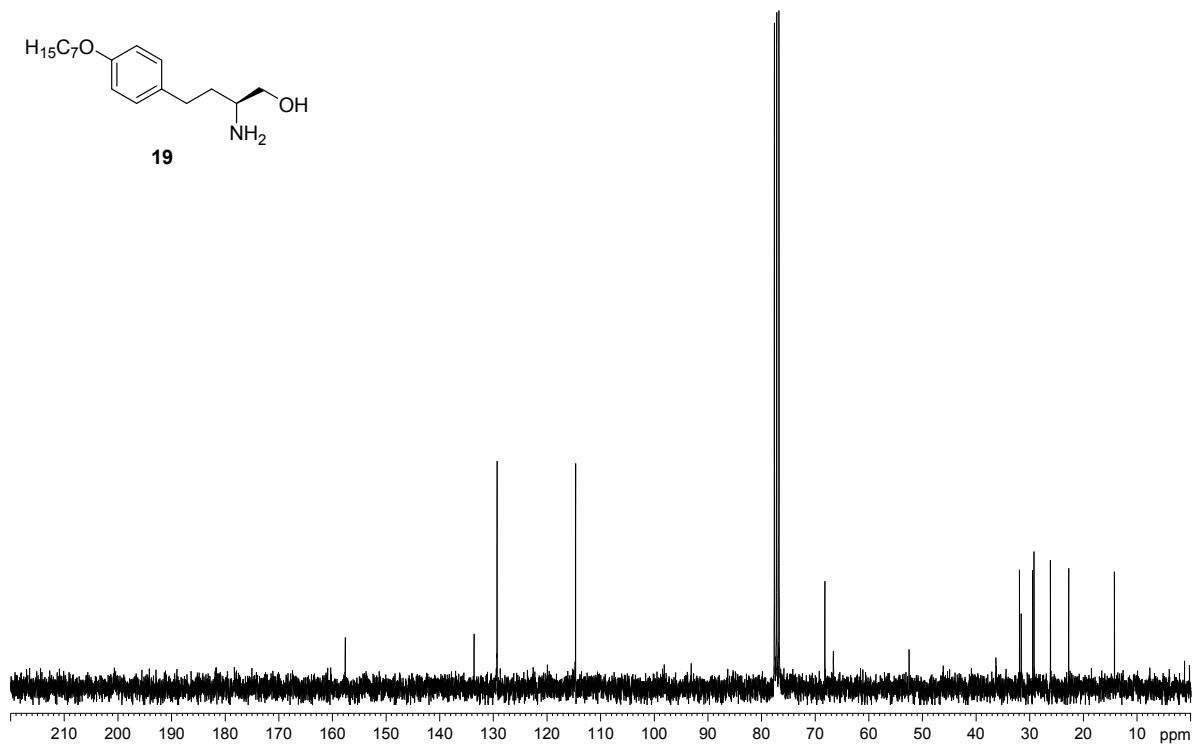
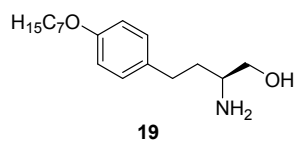
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13/07/2012 HDTK014_X; 300 MHz; CDCl3

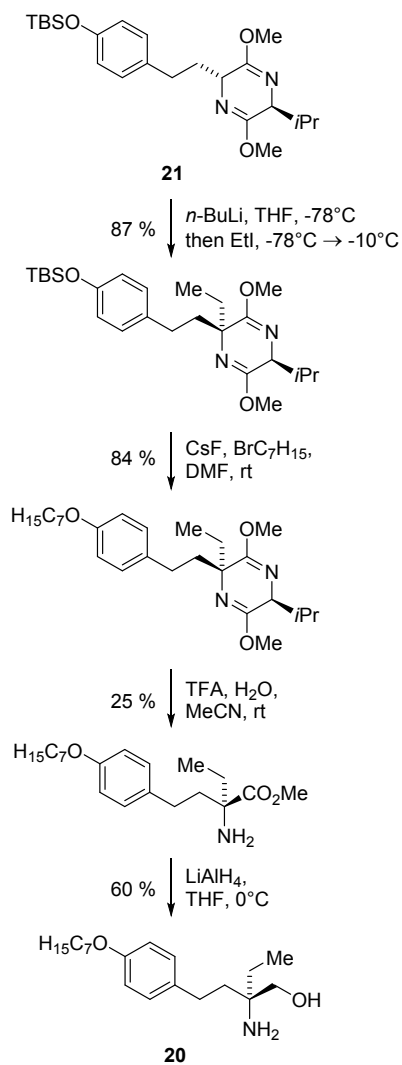


13/07/2012 HDTK014_X; 300 MHz; CDCl3

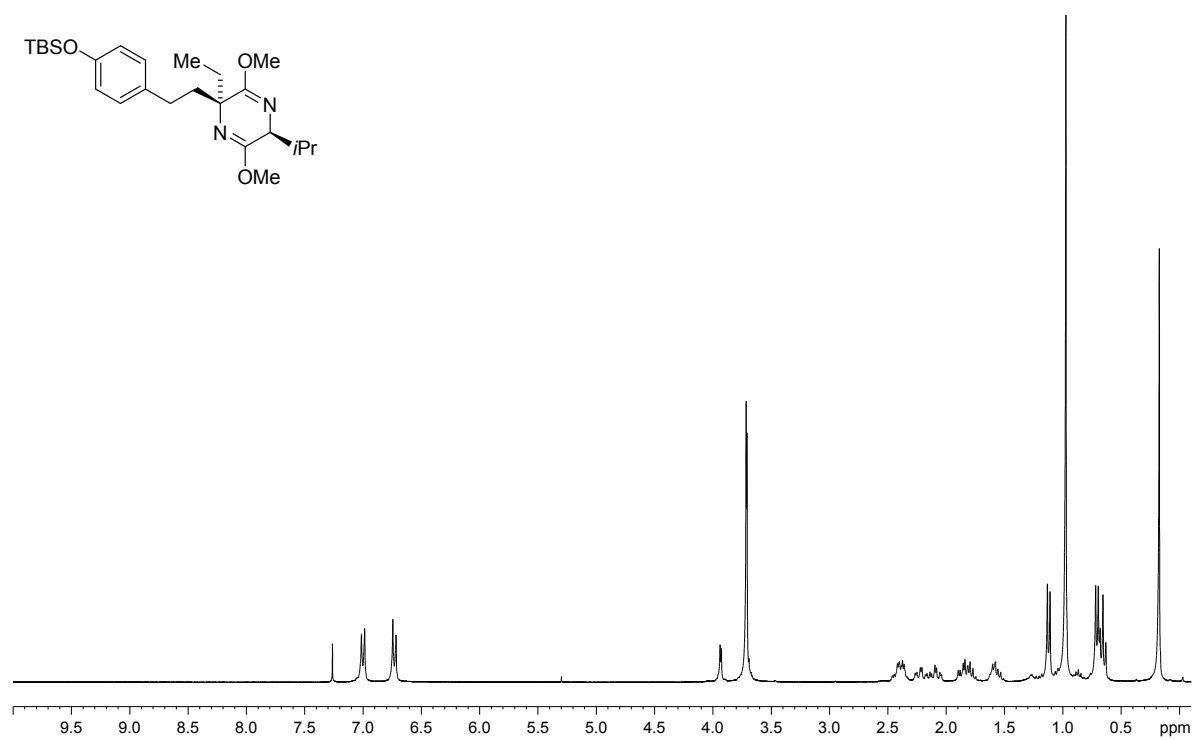


8. Scheme Summarising the Intermediates and NMR Data for the Synthesis of (2S)-2-Amino-2-ethyl-4-(4'-heptyloxyphenyl)butan-1-ol (20)

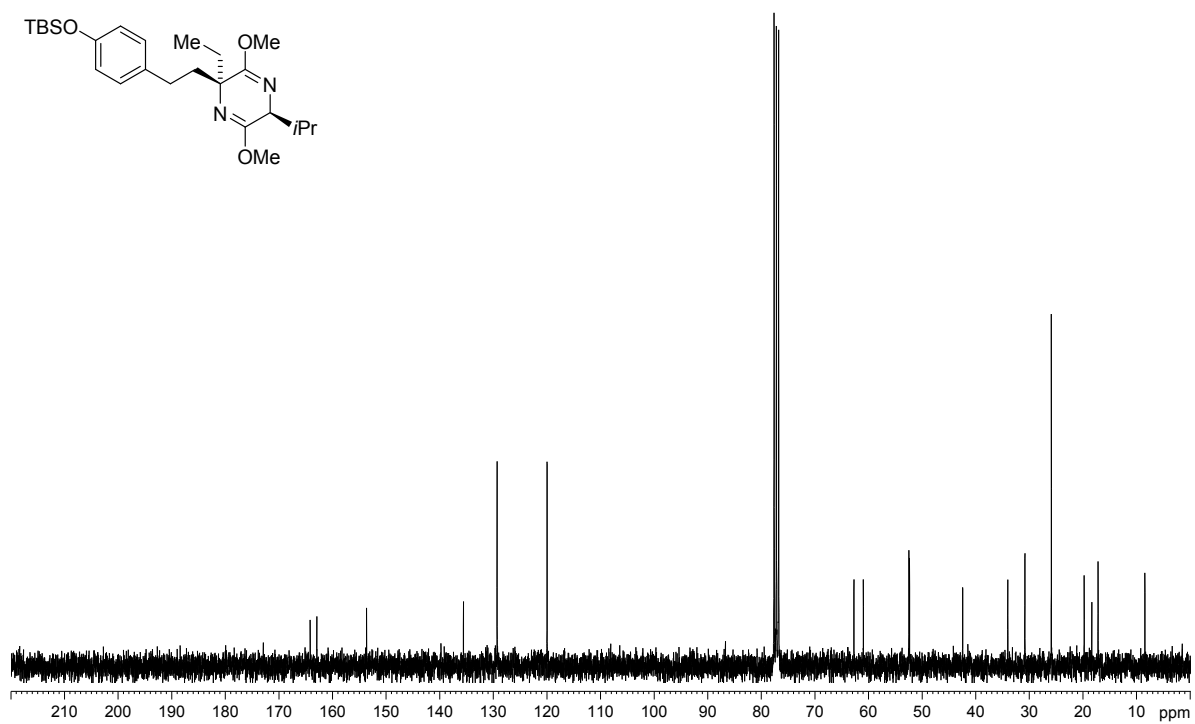
Scheme used to synthesise (2S)-2-Amino-2-ethyl-4-(4'-heptyloxyphenyl)butan-1-ol (20)



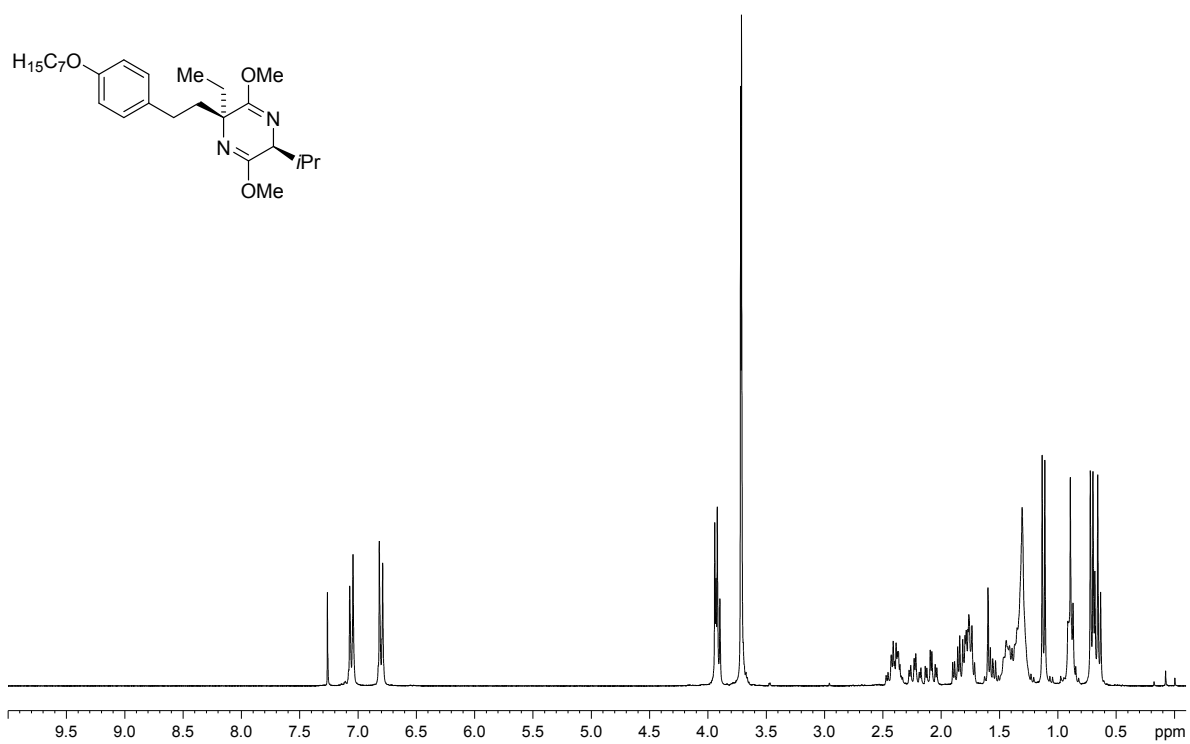
11/07/2012 HDTK001_2; 300 MHz; CDCl₃



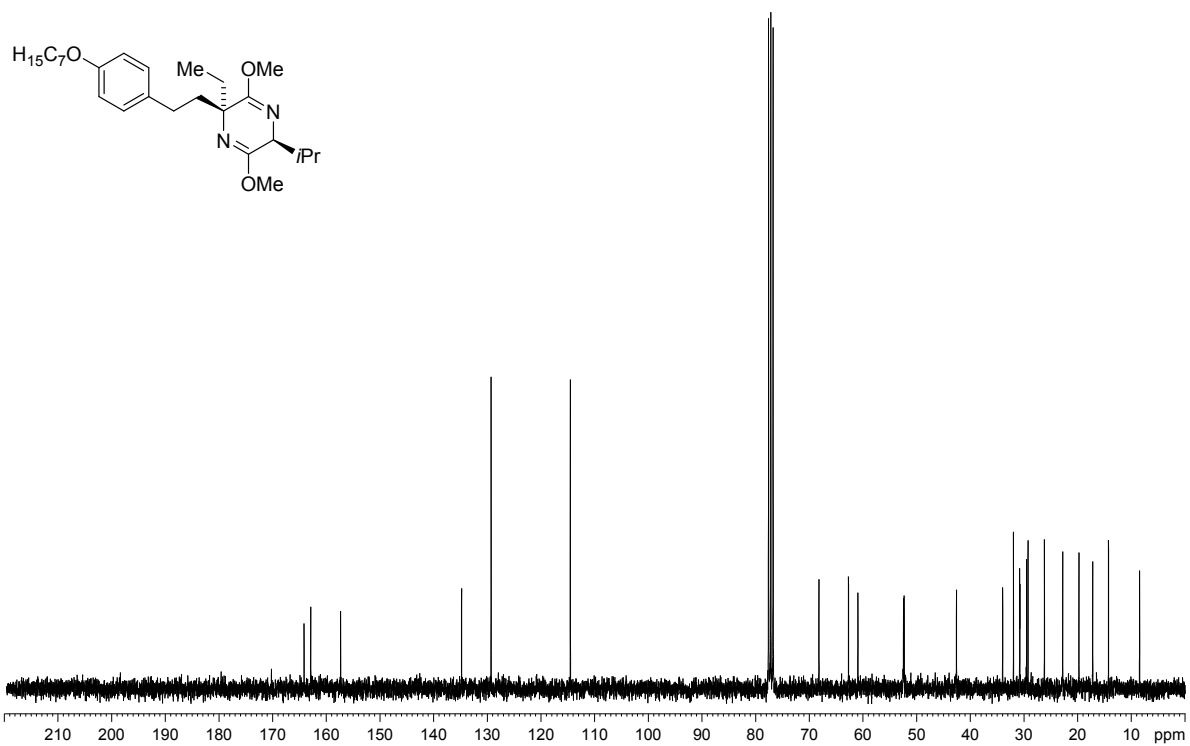
11/07/2012 HDTK001_2; 300 MHz; CDCl₃



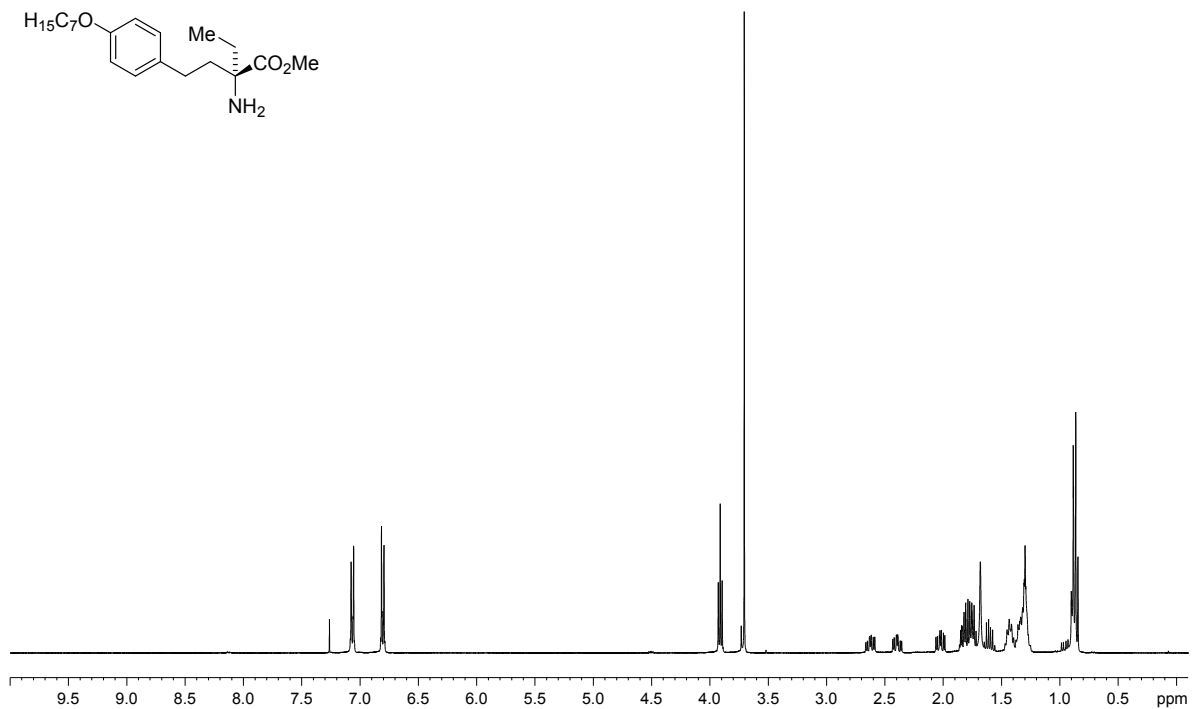
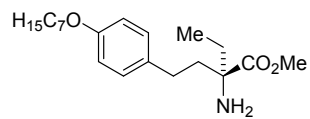
16/07/2012 HDTK017_2; 300 MHz; CDCl₃



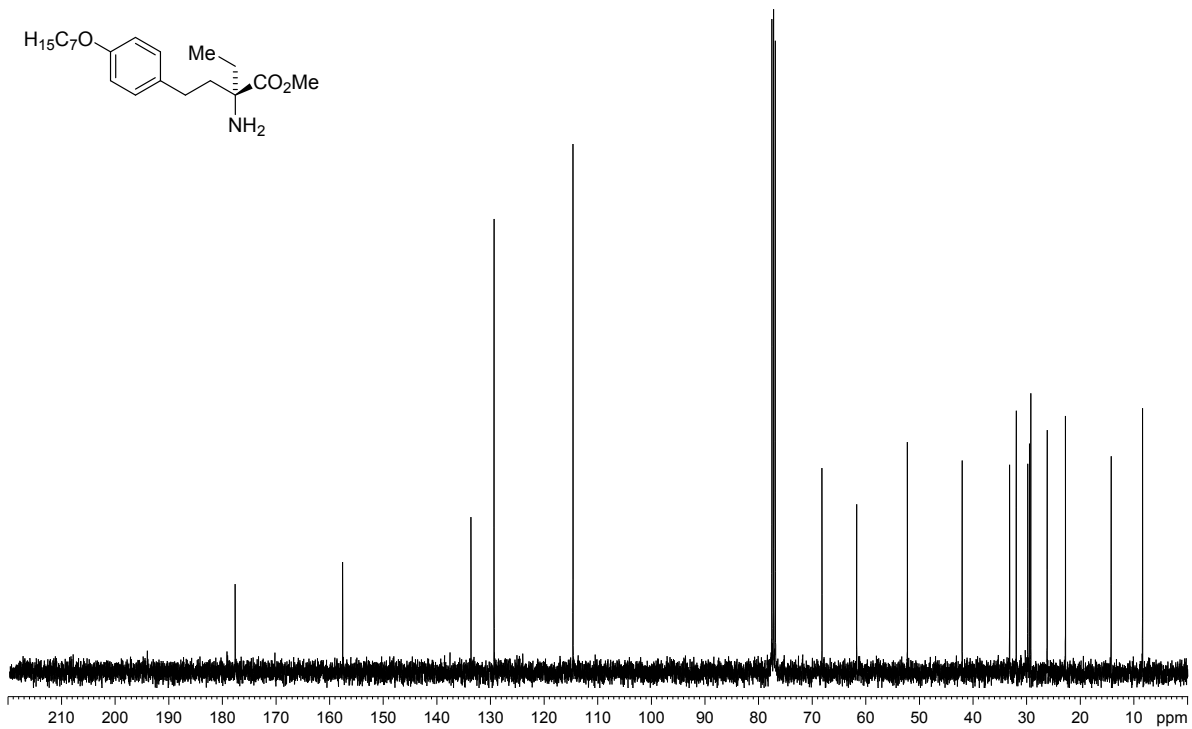
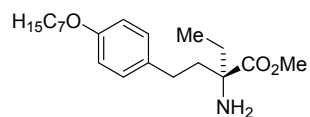
16/07/2012 HDTK017_2; 300 MHz; CDCl₃



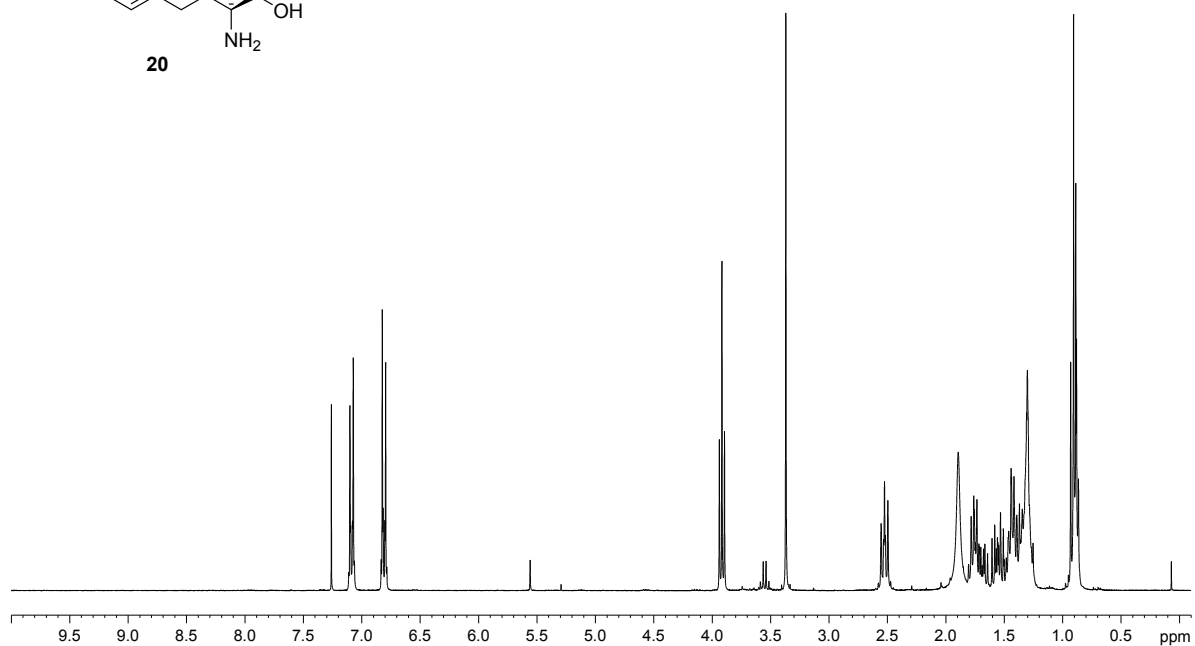
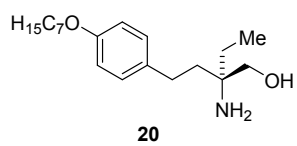
18/11/2012 HDTK025_X; 400 MHz; CDCl3



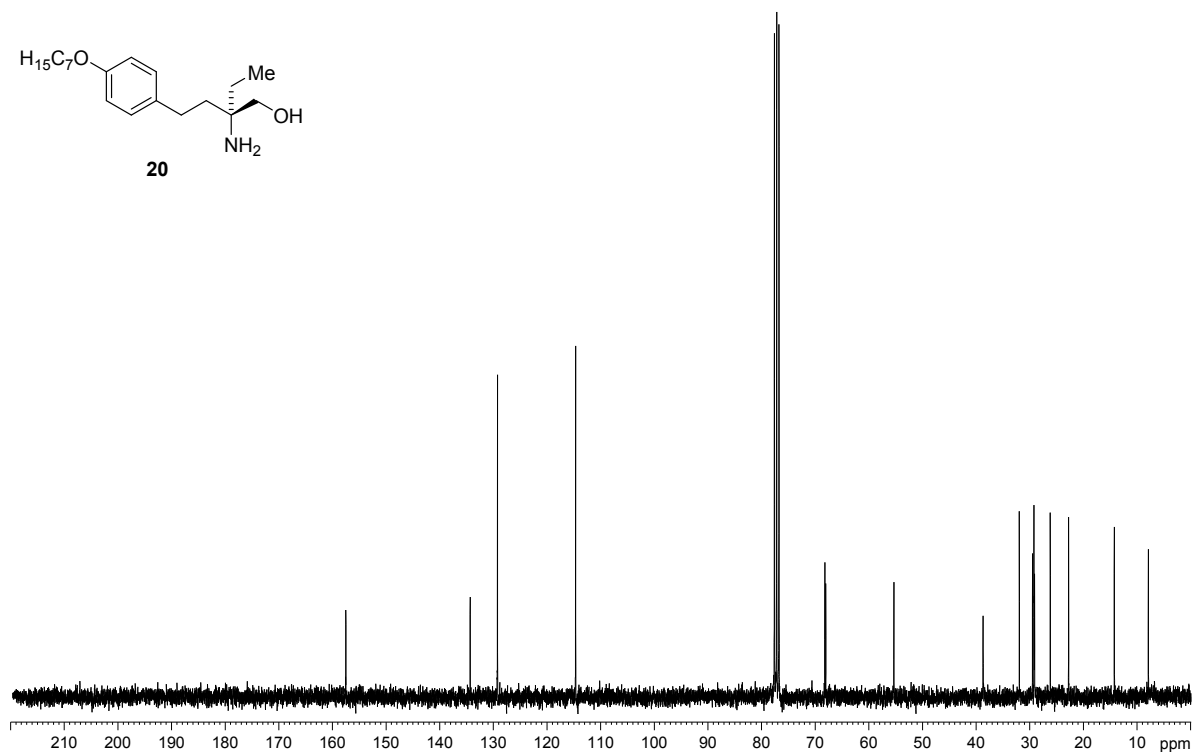
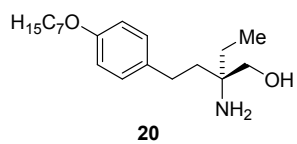
18/11/2012 HDTK025_X; 400 MHz; CDCl3



27/11/2012 HDTM008_2; 300 MHz; CDCl3

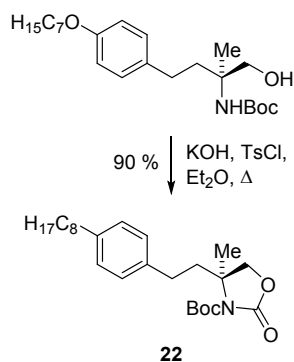


27/11/2012 HDTM008_2; 300 MHz; CDCl3

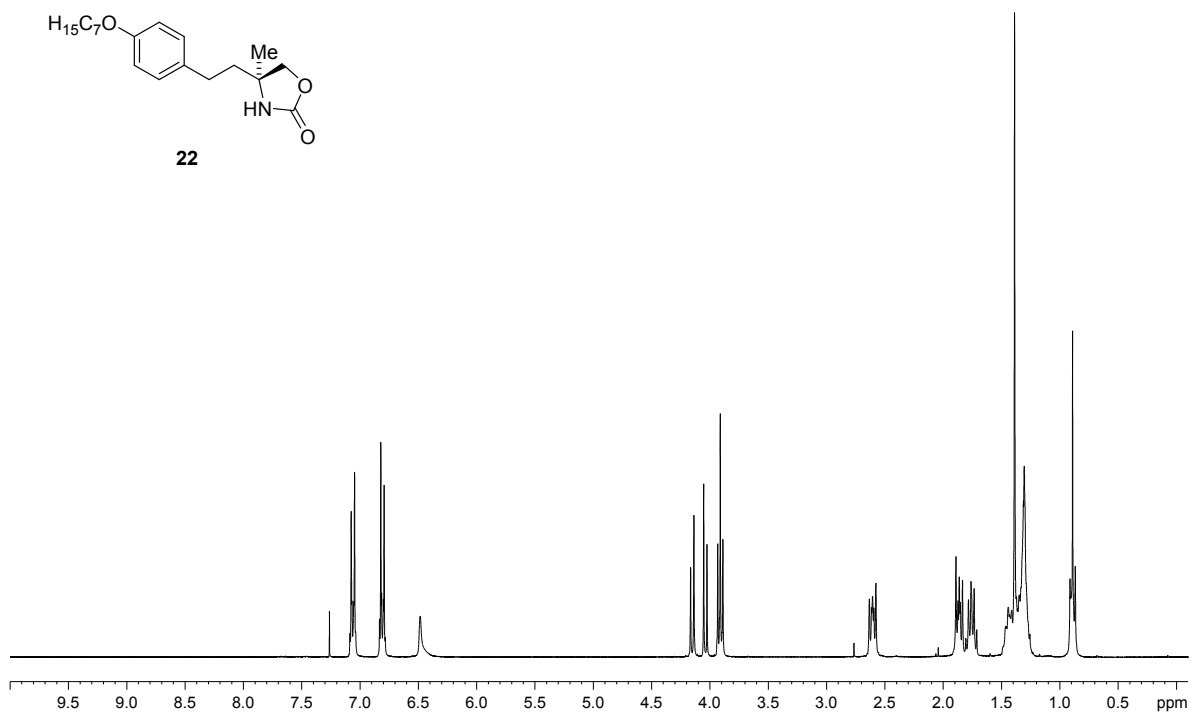
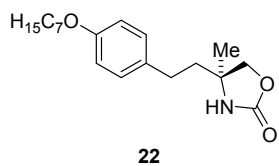


9. Scheme Summarising the Intermediates and NMR Data for the Synthesis of (4S)-4-(4'-Heptyloxyphenethyl)-4-methyloxazolidin-2-one (**22**)

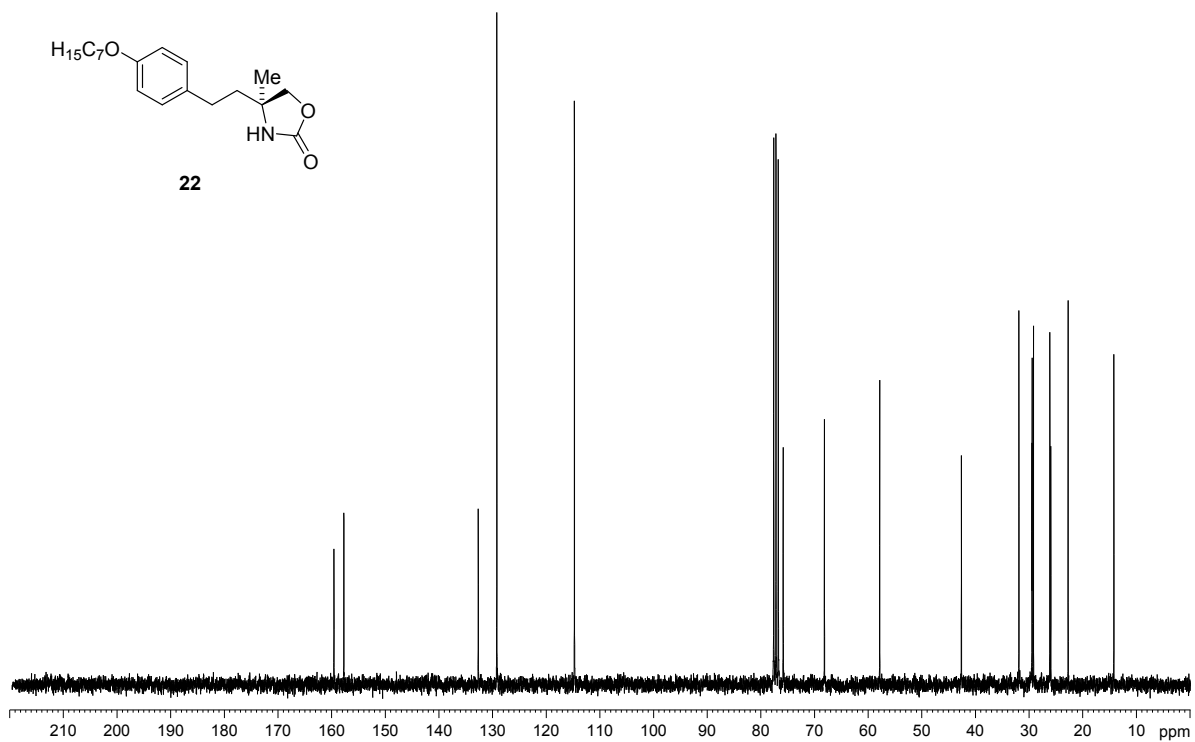
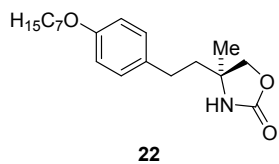
Scheme used to synthesise (4S)-4-(4'-Heptyloxyphenethyl)-4-methyloxazolidin-2-one (**22**)



21/11/2011 HDT_oxazolidinone; 300 MHz; CDCl3

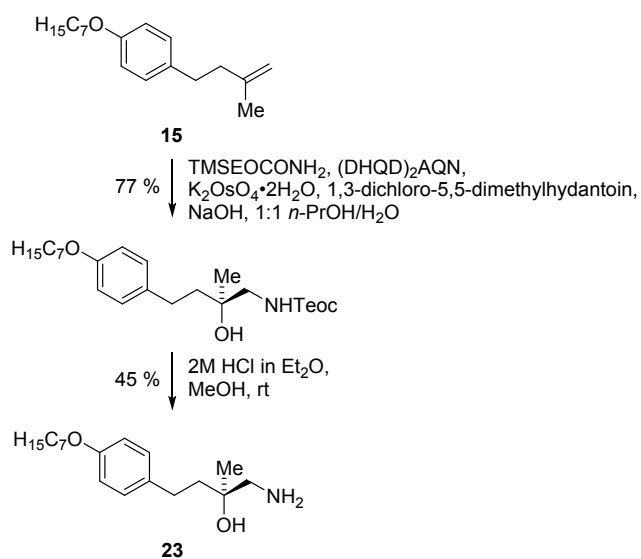


21/11/2011 HDTH_oxazolidinone; 300 MHz; CDCl3

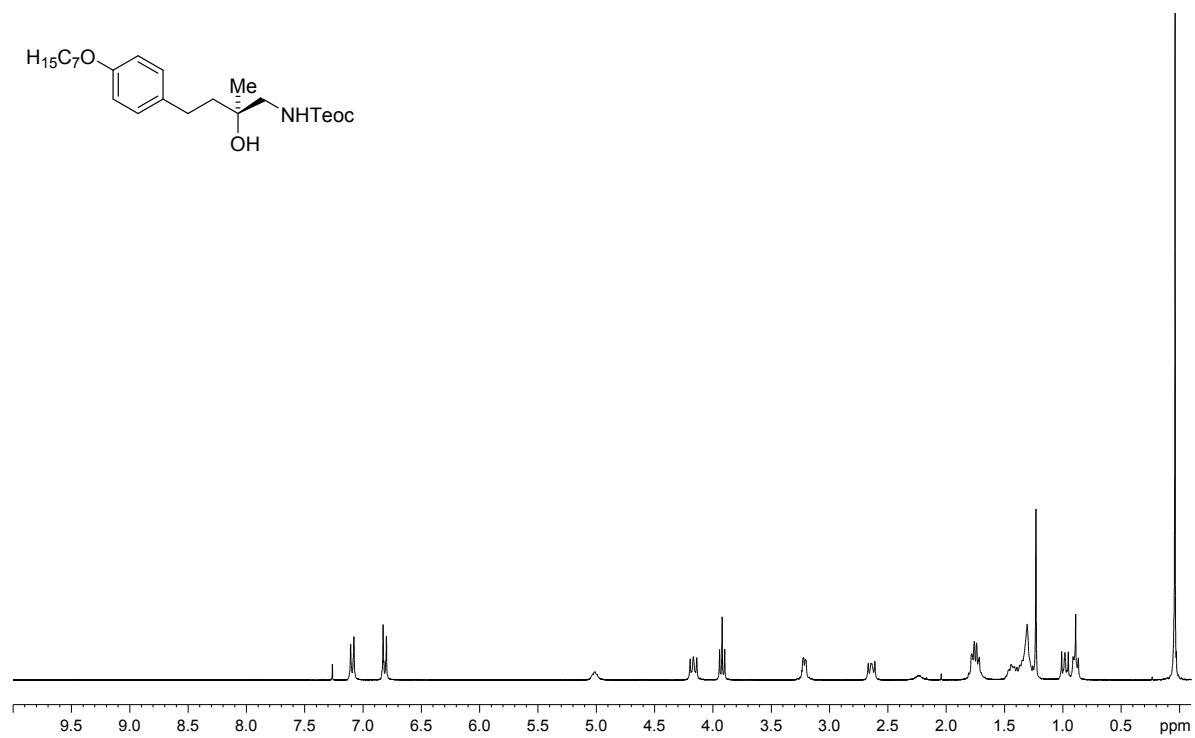


10. Scheme Summarising the Intermediates and NMR Data for the Synthesis of (2S)-1-Amino-4-(4'-heptyloxyphenyl)-2-methylbutan-2-ol (23)

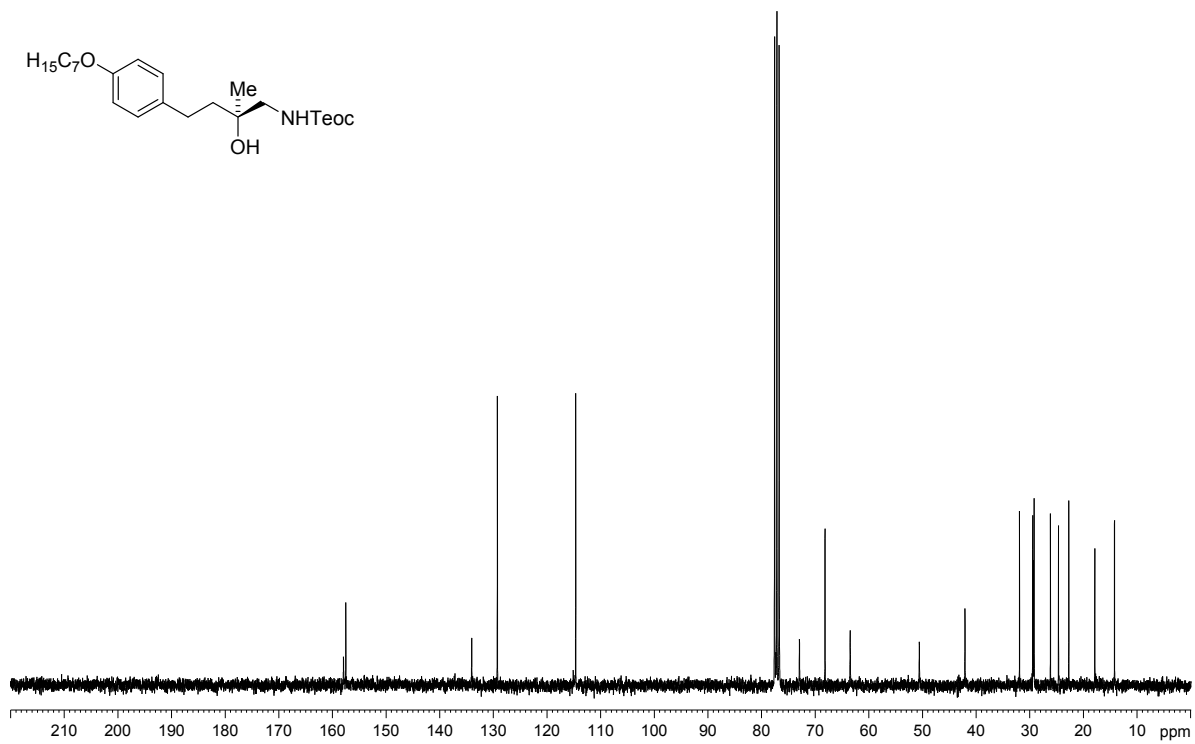
Scheme used to synthesise (2S)-1-Amino-4-(4'-heptyloxyphenyl)-2-methylbutan-2-ol (**23**)



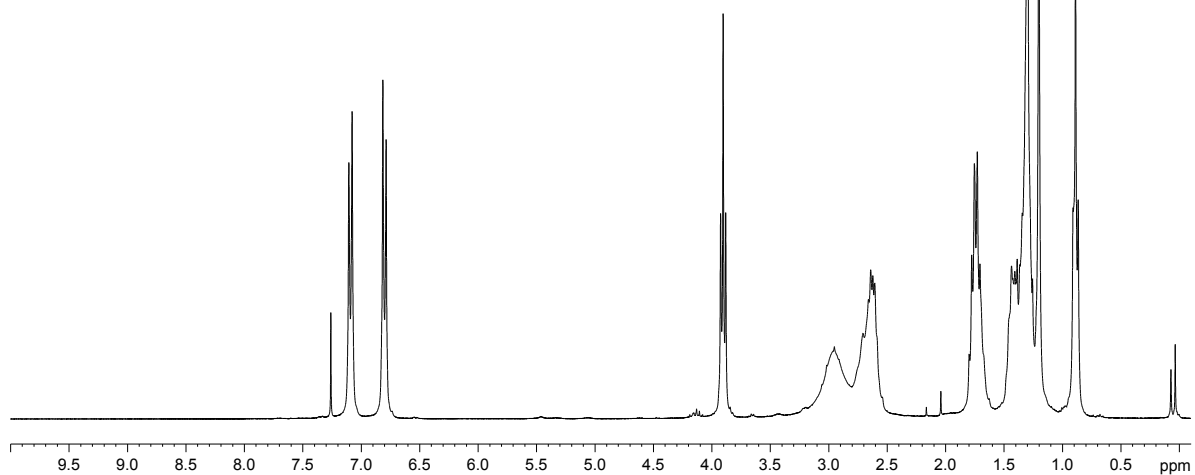
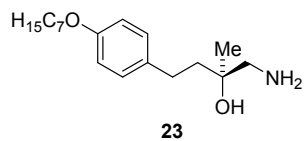
27/05/2011 HDTF021_2; 300 MHz; CDCl3



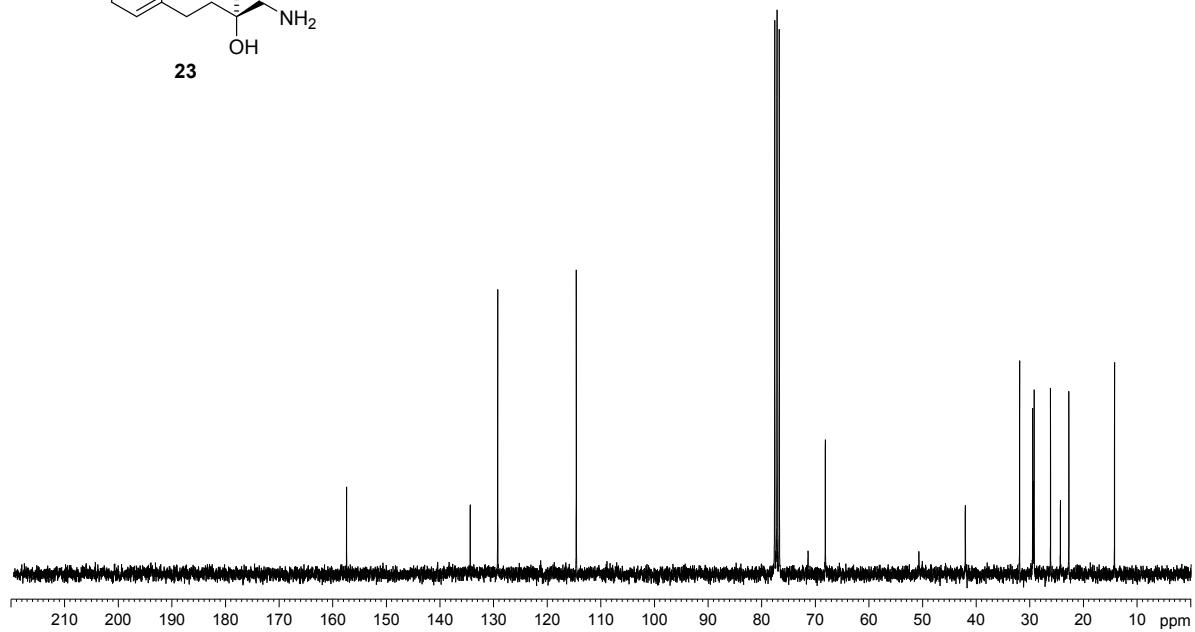
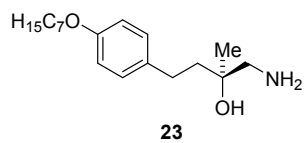
27/05/2011 HDTF021_2; 300 MHz; CDCl3



10/07/2011 HDTG006_1; 300 MHz; CDCl3

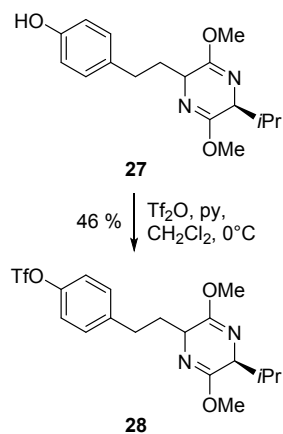


10/07/2011 HDTG006_1; 300 MHz; CDCl3

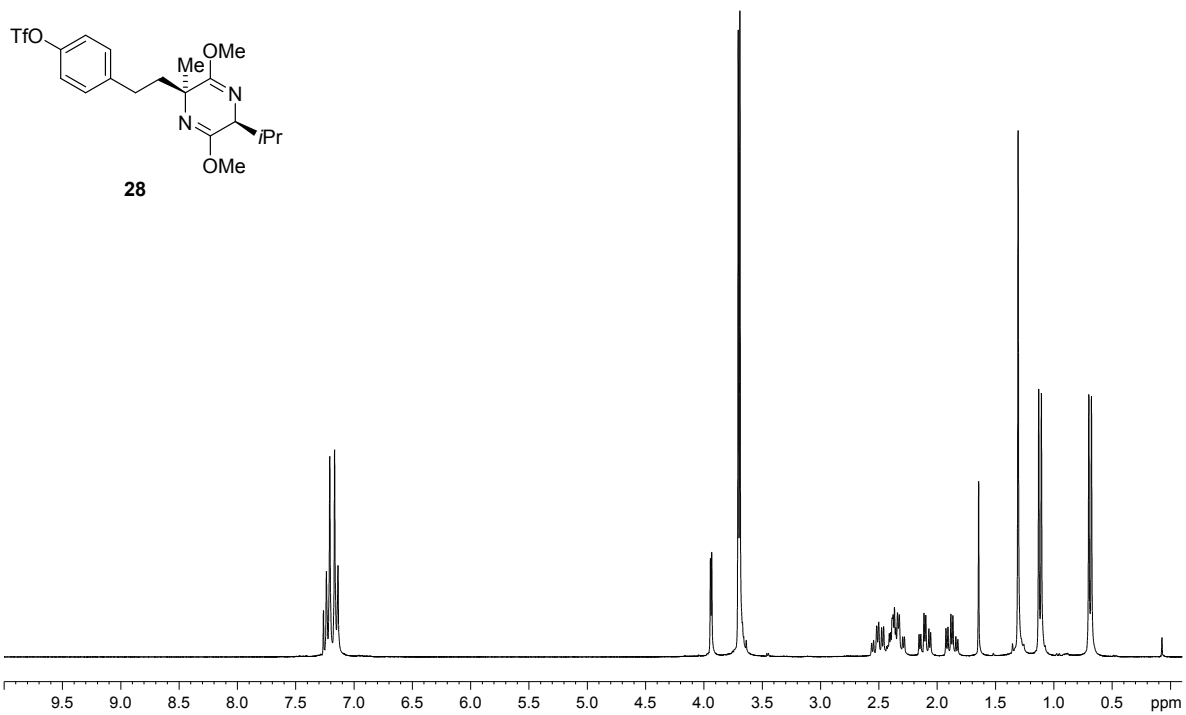


11. Scheme Summarising the Intermediates and NMR Data for the Synthesis of 4-(2-((2S,5S)-5-Isopropyl-3,6-dimethoxy-2-methyl-2,5-dihydropyrazin-2-yl)ethyl)phenyl trifluoromethanesulfonate (28)

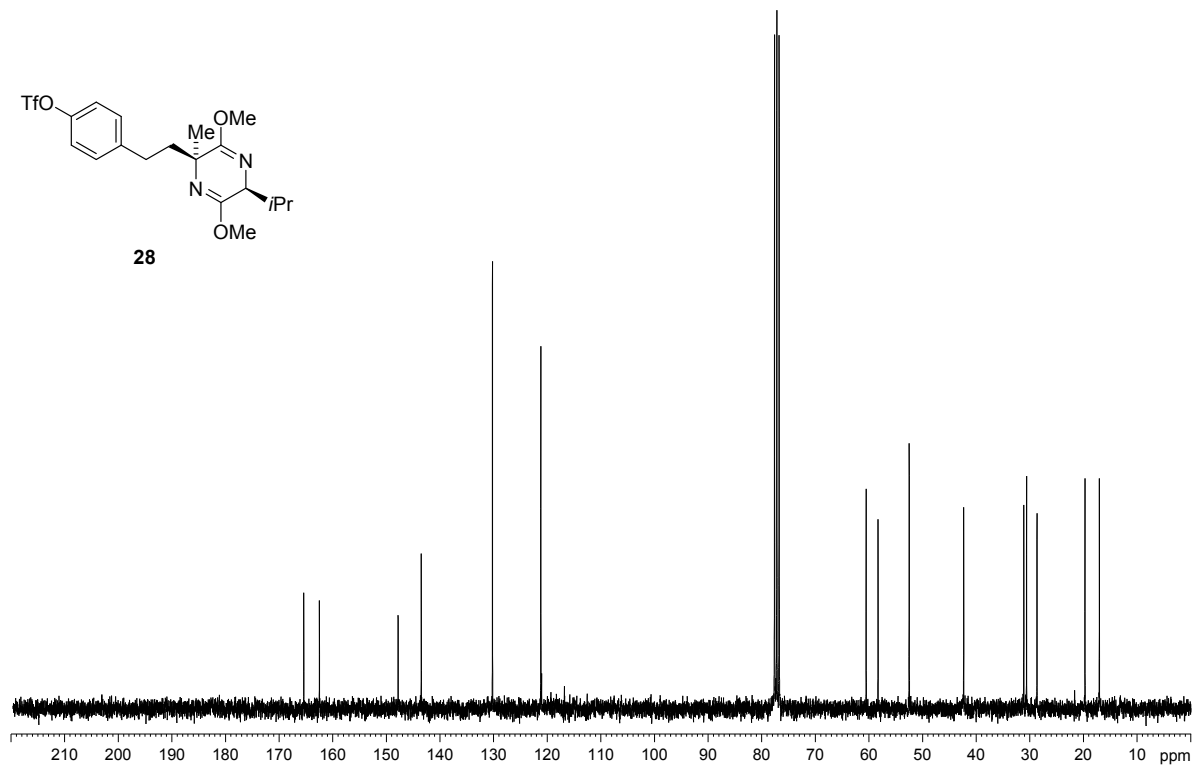
Scheme used to synthesise 4-(2-((2S,5S)-5-Isopropyl-3,6-dimethoxy-2-methyl-2,5-dihydropyrazin-2-yl)ethyl)phenyl trifluoromethanesulfonate (**28**)



04/02/2014 HDTQ054_1; 300 MHz; CDCl3

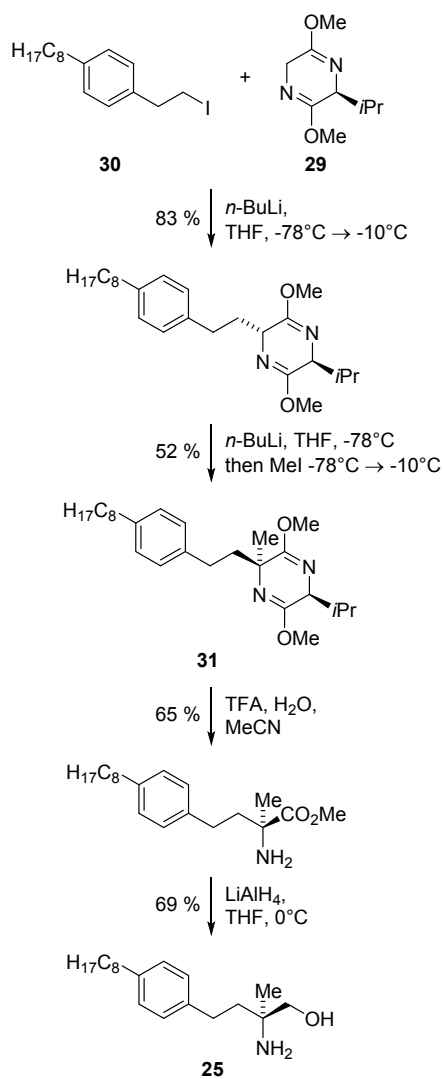


04/02/2014 HDTQ054_1; 300 MHz; CDCl3

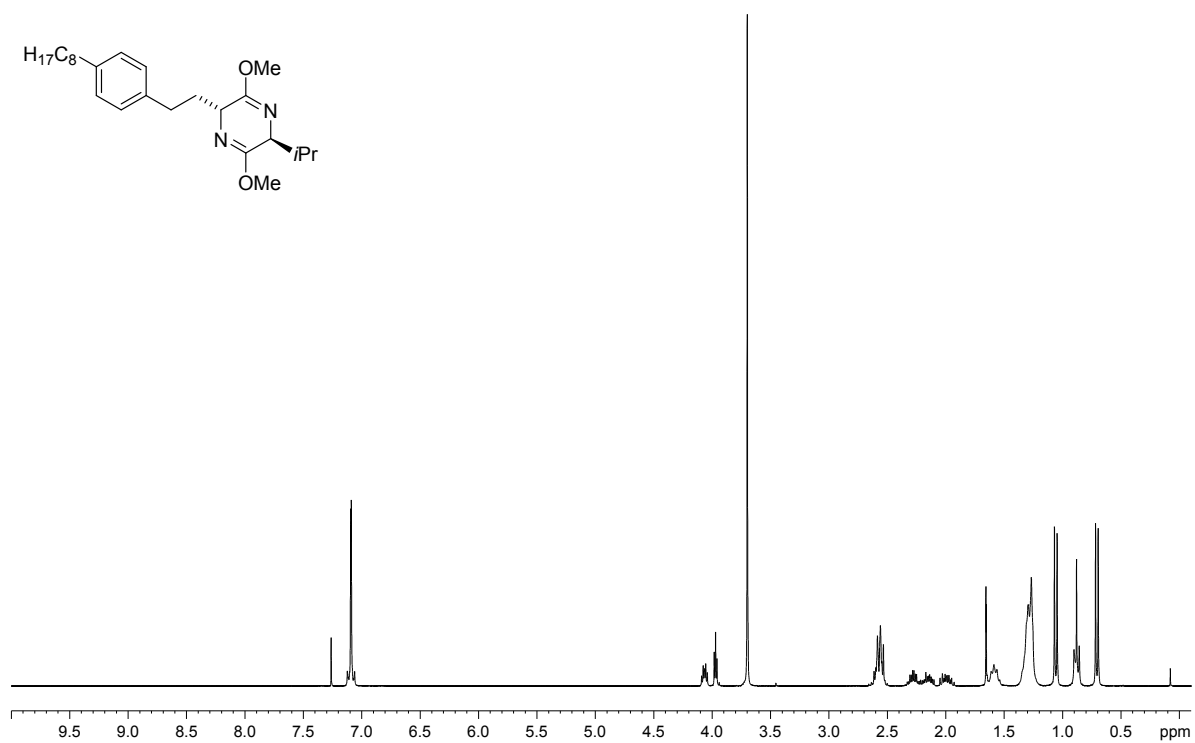


12. Scheme Summarising the Intermediates and NMR Data for the Synthesis of (2S)-2-Amino-2-methyl-4-(4'-octylphenyl)butan-1-ol (25)

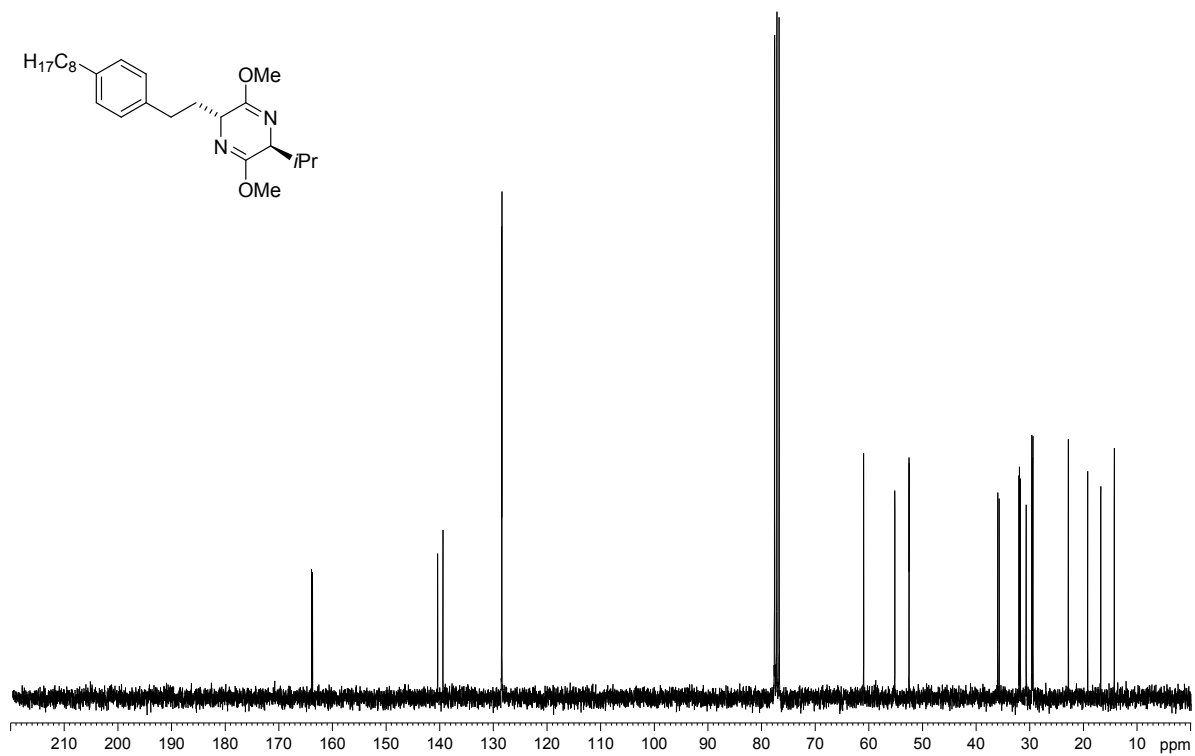
Scheme used to synthesise (2S)-2-Amino-2-methyl-4-(4'-octylphenyl)butan-1-ol (25)



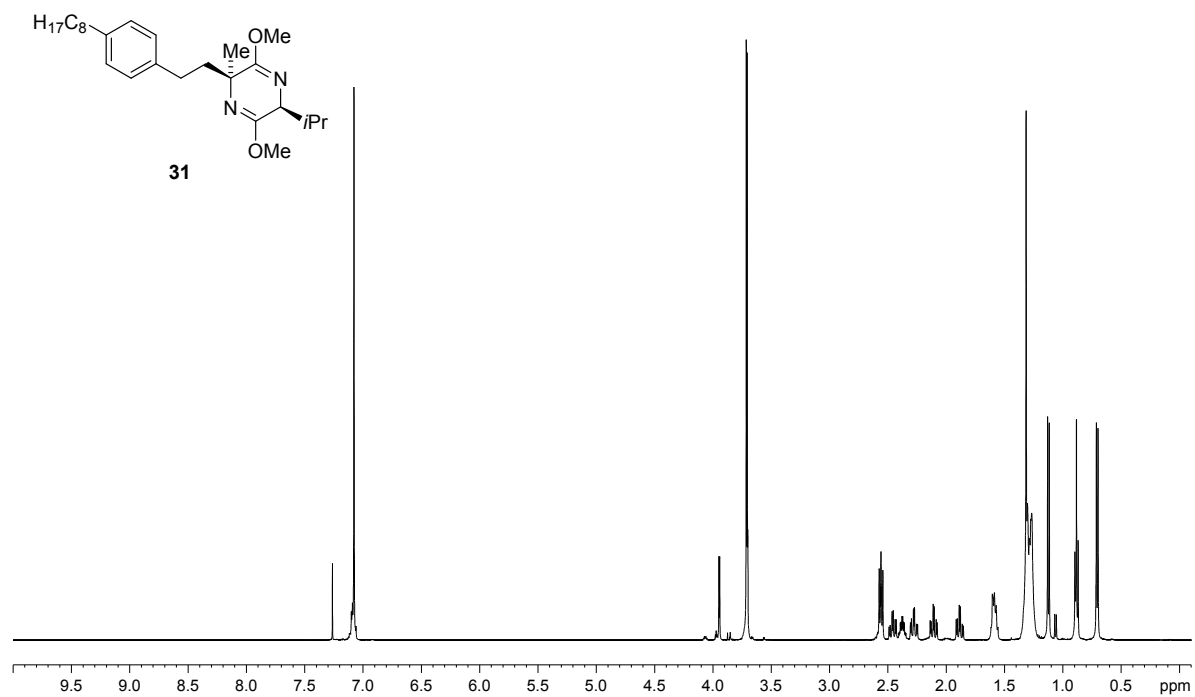
19/03/2014 HDTR034_c1f13; 300 MHz; CDCl3



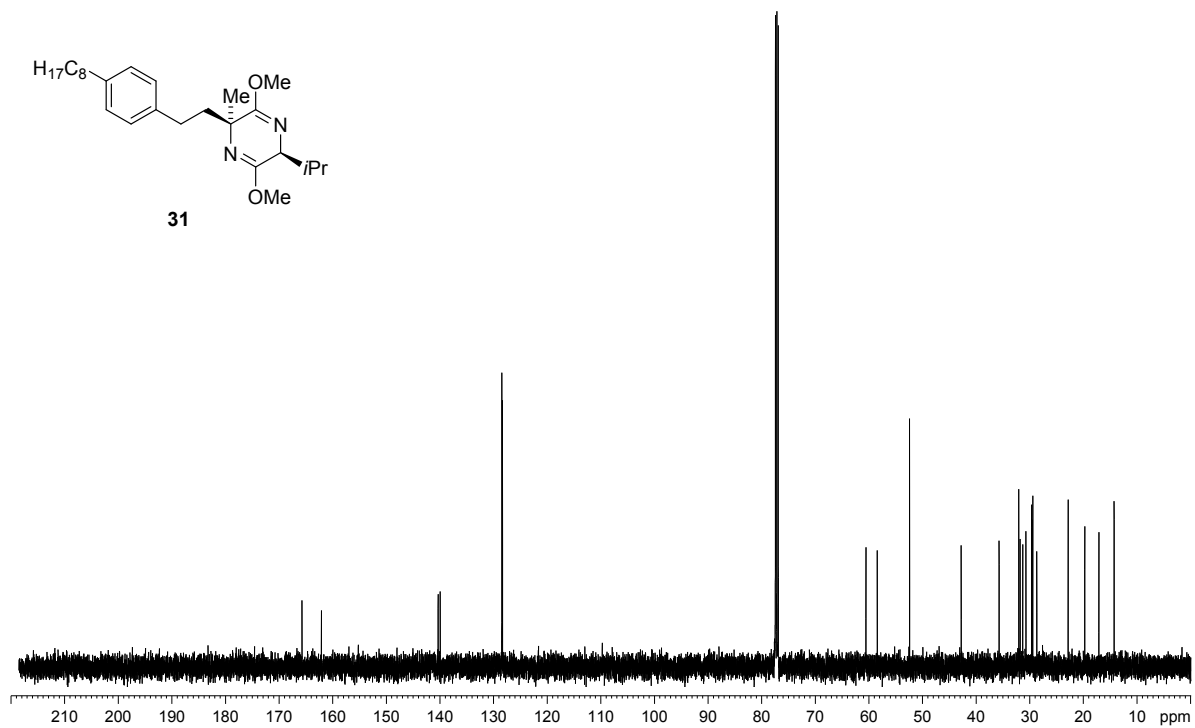
19/03/2014 HDTR034_c1f13; 300 MHz; CDCl3



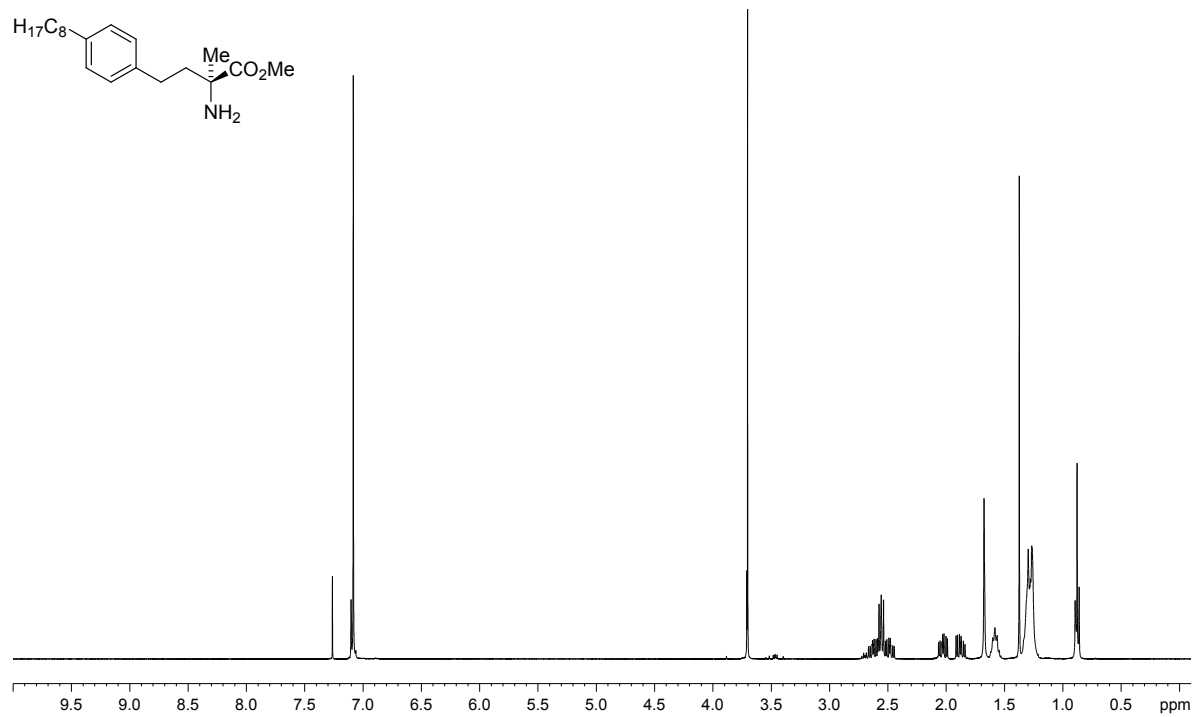
14/07/2014 HDTR037_2; 500 MHz; CDCl3



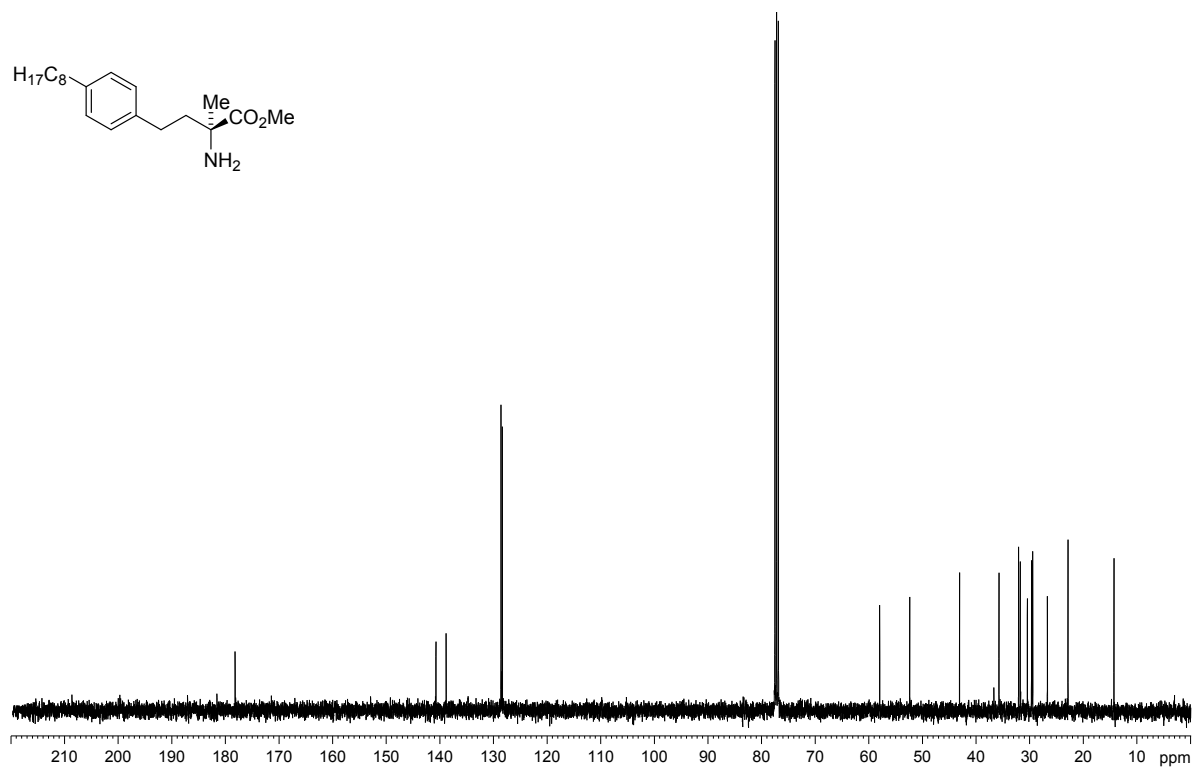
14/07/2014 HDTR037_2; 500 MHz; CDCl3



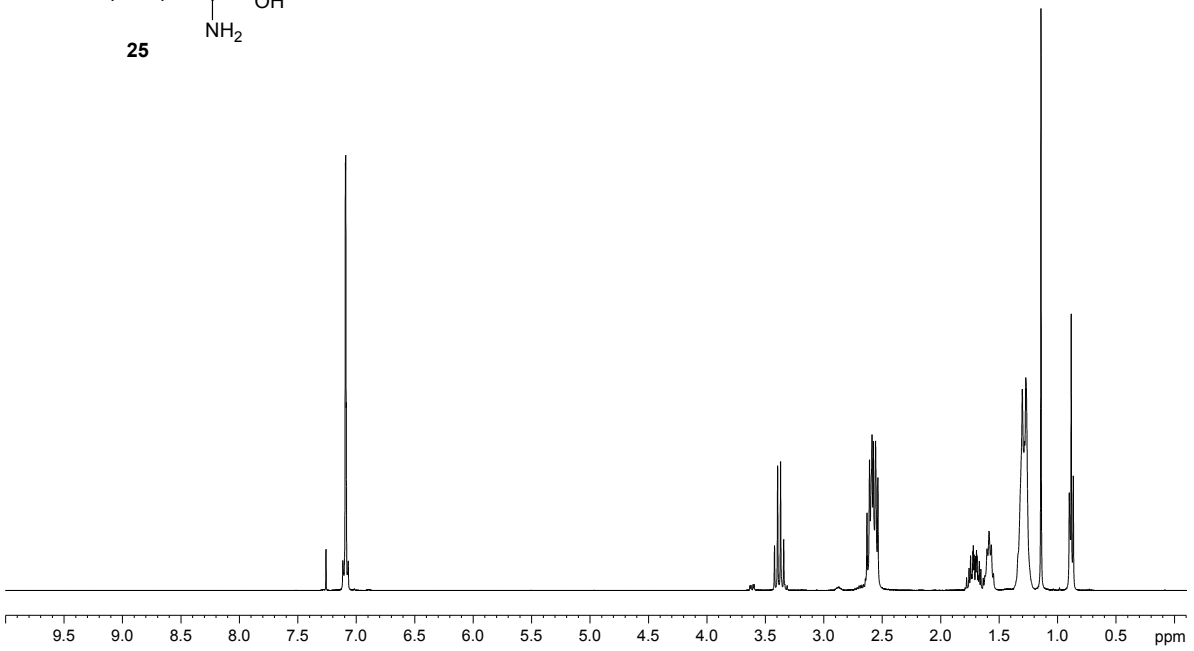
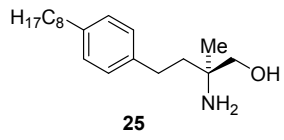
16/07/2014 HDTR054_1; 400 MHz; CDCl3



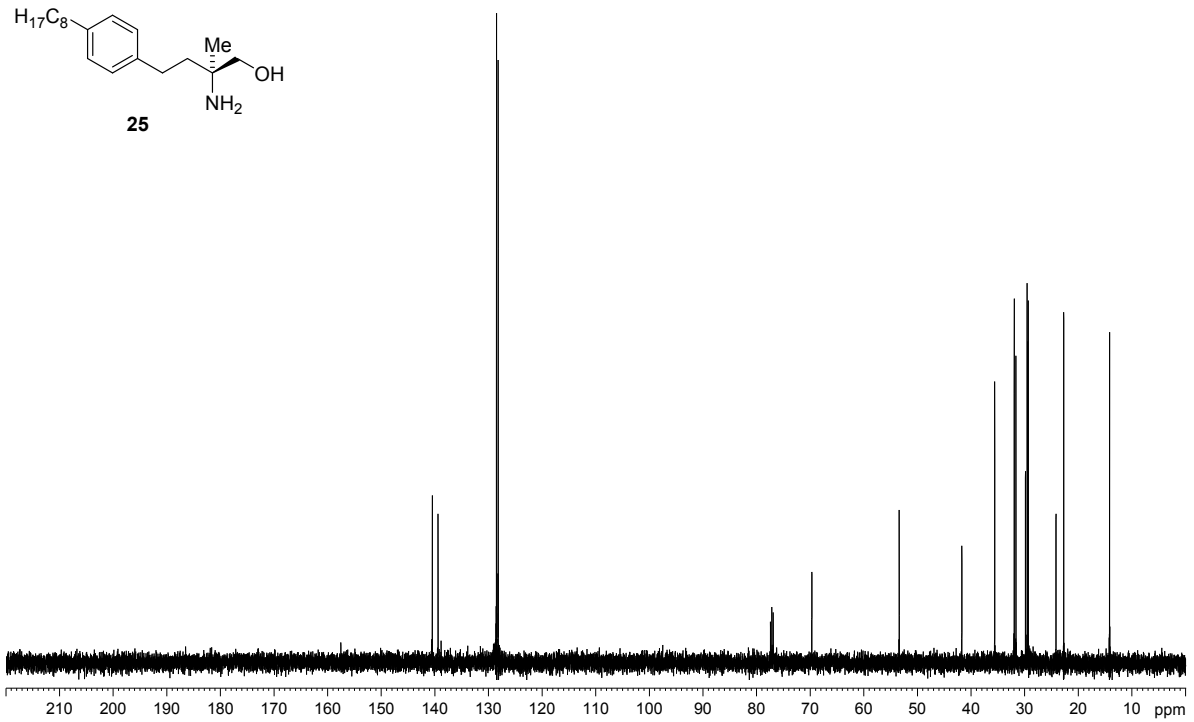
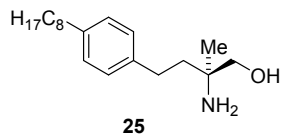
16/07/2014 HDTR054_1; 400 MHz; CDCl3



23/07/2014 HDTR057; 400 MHz; CDCl3



17/07/2014 HDTR057_1; 500 MHz; CDCl3



13. References

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- 5) H. D. Toop, A. S. Don, J. C. Morris, *Org. Biomol. Chem.* 2015, **13**, 11593.
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