

Esterification of an Unnatural Amino Acid Structurally Deviating from Canonical Amino Acids Promotes Its Uptake and Incorporation into Proteins in Mammalian Cells

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SUPPORTING INFORMATION

Experimental data for compounds

Compound 6. ^1H NMR (500 MHz, CDCl_3): δ_{H} = 8.56 (d, J = 8.5 Hz, 1 H), 8.25-8.22 (m, 2 H), 7.57-7.50 (m, 2 H), 7.18 (d, J = 7.0 Hz, 1 H), 5.45 (t, J = 6.5 Hz, 1 H), 5.39 (d, J = 6.5 Hz, 1 H), 4.29 (brs, 1 H), 3.59 (s, 3 H), 3.35-3.26 (m, 2 H), 2.89 (s, 6 H), 1.40 (s, 9 H) ppm; ^{13}C NMR (125 MHz, CDCl_3): δ_{C} = 170.7, 155.5, 152.0, 134.4, 130.8, 130.0, 129.9, 129.6, 128.8, 128.6, 128.4, 123.3, 118.8, 115.4, 80.5, 53.5, 52.8, 45.5, 44.9, 28.2 ppm.

Compound 7. ^1H NMR (500 MHz, CDCl_3): δ_{H} = 8.55 (d, J = 8.5 Hz, 1 H), 8.25-8.22 (m, 2 H), 7.56 (t, J = 8.0 Hz, 1 H), 7.52 (t, J = 8.0 Hz, 1 H), 7.19 (d, J = 7.5 Hz, 1 H), 5.39 (t, J = 6.5 Hz, 1 H), 4.26 (brs, 1 H), 4.14-4.05 (m, 1 H), 4.03-3.97 (m, 1 H), 3.35-3.30 (m, 1 H), 3.28-3.23 (m, 1 H), 2.89 (s, 6 H), 1.52 (s, 9 H), 1.17 (t, J = 7.2 Hz, 3 H) ppm; ^{13}C NMR (125 MHz, CDCl_3): δ_{C} = 170.2, 155.5, 152.0, 134.4, 130.8, 130.0, 129.9, 129.6, 128.7, 123.3, 118.9, 115.5, 80.5, 62.2, 53.6, 45.6, 45.0, 28.3, 14.1 ppm.

Compound 2. ^1H NMR (500 MHz, d_6 -DMSO): δ_{H} = 8.51 (dt, J = 8.5, 1.0 Hz, 1 H), 8.48 (brs, 3 H), 8.37 (t, J = 6.5 Hz, 1 H), 8.25 (d, J = 8.5 Hz, 1 H), 8.11 (dd, J = 7.2, 1.0 Hz, 1 H), 7.67 (dd, J = 8.8, 7.2 Hz, 1 H), 7.63 (dd, J = 8.5, 7.5 Hz, 1 H), 7.31 (dd, J = 8.0, 1.0 Hz, 1 H), 4.09 (m, 1 H), 3.60 (s, 3 H), 3.26 (ddd, J = 14.5, 7.0, 5.5 Hz, 1 H), 3.18 (dt, J = 14.5, 6.0 Hz, 1 H), 2.85 (s, 6 H) ppm; ^{13}C NMR (125 MHz, d_6 -DMSO): δ_{C} = 167.8, 158.4 (q, J = 36.5 Hz), 151.2, 134.9, 129.9, 129.1, 128.9, 128.7, 128.2, 123.7, 119.1, 116.8, 115.5, 114.5, 52.9, 52.3, 45.1, 42.3 ppm. HRMS (ESI-FT): calcd for $\text{C}_{16}\text{H}_{22}\text{N}_3\text{O}_4\text{S}$ (M+1) 352.1326, found 352.1328.

Compound 3. ^1H NMR (500 MHz, d_6 -DMSO): δ_{H} = 8.51 (dd, J = 8.0, 1.0 Hz, 1 H), 8.48 (brs, 3 H), 8.34 (t, J = 6.2 Hz, 1 H), 8.28 (d, J = 8.5 Hz, 1 H), 8.12 (dd, J = 7.0, 1.0 Hz, 1 H), 7.67 (dd, J = 8.8, 7.2 Hz, 1 H), 7.63 (dd, J = 8.5, 7.5 Hz, 1 H), 7.32 (d, J = 7.5 Hz, 1 H), 4.11 (dq, J = 11.0, 7.0 Hz, 1 H), 4.08 (m, 1 H),

4.00 (dq, $J = 11.0, 7.0$ Hz, 1 H), 3.27 (ddd, $J = 14.5, 6.5, 5.5$ Hz, 1 H), 3.19 (dt, $J = 14.5, 5.5$ Hz, 1 H), 2.86 (s, 6 H), 1.15 (t, $J = 7.0$ Hz, 3 H) ppm; ^{13}C NMR (125 MHz, d6-DMSO): $\delta_{\text{C}} = 167.4, 158.6$ (q, $J = 36.5$ Hz), 151.0, 135.0, 129.9, 129.1, 129.0, 128.8, 128.2, 123.8, 119.3, 116.8, 115.6, 62.1, 52.3, 45.2, 42.4, 13.8 ppm. HRMS (ESI-FT): calcd for $\text{C}_{17}\text{H}_{24}\text{N}_3\text{O}_4\text{S}$ (M+1) 366.1482, found 366.1481.

Boc-DanAla-OBn. ^1H NMR (500 MHz, CDCl_3): $\delta_{\text{H}} = 8.53$ (d, $J = 8.5$ Hz, 1 H), 8.22-8.20 (m, 2 H), 7.53 (t, $J = 8.0$ Hz, 1 H), 7.48 (d, $J = 8.0$ Hz, 1 H), 7.35-7.32 (m, 3 H), 7.27-7.26 (m, 2 H), 7.17 (d, $J = 7.5$ Hz, 1 H), 5.41 (brs, 2 H), 5.08 (d, $J = 12.0$ Hz, 1 H), 4.97 (d, $J = 12.0$ Hz, 1 H), 4.31 (brs, 1 H), 3.32-3.30 (m, 2 H), 2.86 (s, 6 H), 1.38 (s, 6 H) ppm; ^{13}C NMR (125 MHz, CDCl_3): $\delta_{\text{C}} = 170.0, 155.3, 152.0, 134.9, 134.2, 130.7, 129.9, 129.8, 129.5, 128.6, 128.5$ (2), 128.3, 123.1, 118.6, 115.3, 80.4, 77.2, 67.6, 53.6, 45.4, 28.2 ppm.

Compound 9. ^1H NMR (500 MHz, CDCl_3): $\delta_{\text{H}} = 8.57$ (d, $J = 8.5$ Hz, 1 H), 8.37 (d, $J = 7.5$ Hz, 1 H), 7.81 (d, $J = 9.0$ Hz, 1 H), 7.53 (t, $J = 8.0$ Hz, 2 H), 7.41-7.32 (m, 5 H), 7.16 (d, $J = 8.0$ Hz, 1 H), 5.51 (d, $J = 9.0$ Hz, 1 H), 5.26 (d, $J = 12.5$ Hz, 1 H), 5.17 (d, $J = 12.5$ Hz, 1 H), 4.85 (dd, $J = 13.2, 7.8$ Hz, 1 H), 4.45 (dd, $J = 14.0, 5.0$ Hz, 1 H), 4.38 (dd, $J = 14.2, 7.8$ Hz, 1 H), 2.87 (s, 6 H), 1.47 (s, 9 H), 1.13 (s, 9 H) ppm; ^{13}C NMR (125 MHz, CDCl_3): $\delta_{\text{C}} = 170.4, 155.2, 152.0, 150.6, 135.3, 134.8, 131.9, 131.1, 129.6, 129.3, 128.7, 128.5, 128.4, 128.3, 122.8, 117.7, 115.1, 84.8, 80.0, 77.2, 67.5, 52.8, 47.3, 45.3, 28.3, 27.6$ ppm;

Compound 10. ^1H NMR (500 MHz, CDCl_3): $\delta_{\text{H}} = 8.57$ (d, $J = 8.5$ Hz, 1 H), 8.37 (d, $J = 7.5$ Hz, 1 H), 7.83 (d, $J = 8.5$ Hz, 1 H), 7.55 (dd, $J = 16.8, 8.2$ Hz, 2 H), 7.17 (d, $J = 8.0$ Hz, 1 H), 5.59 (d, $J = 8.5$ Hz, 1 H), 4.84-4.83 (m, 1 H), 4.44-4.41 (m, 2 H), 2.88 (s, 6 H), 1.49 (s, 9 H), 1.17 (s, 9H) ppm; ^{13}C NMR (125 MHz, CDCl_3): $\delta_{\text{C}} = 156.0, 151.9, 150.8, 134.7, 132.0, 131.2, 129.6, 129.4, 128.8, 122.8, 117.8, 115.2, 85.1, 80.6, 77.2, 47.0, 45.4, 28.3, 27.6$ ppm;

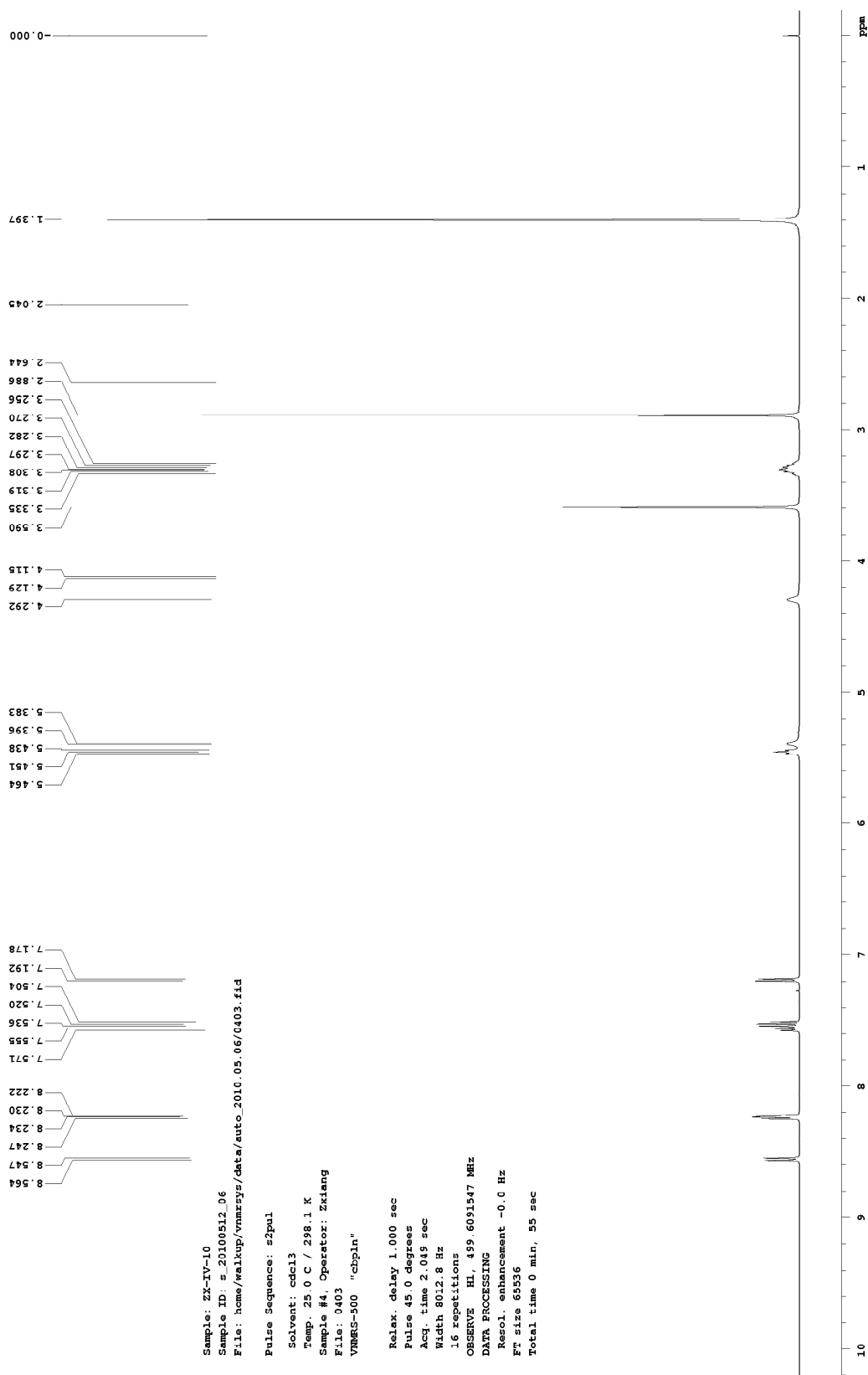
Compound 11. ^1H NMR (500 MHz, CDCl_3): $\delta_{\text{H}} = 8.58$ (d, $J = 8.0$ Hz, 1 H), 8.36 (d, $J = 7.0$ Hz, 1 H), 7.79 (d, $J = 9.0$ Hz, 1 H), 7.55 (dd, $J = 16.0, 8.5$ Hz, 2 H), 7.17 (d, 7.5 Hz, 1 H), 5.84-5.81 (m, 2 H), 5.51 (d, $J = 8.5$ Hz, 1 H), 4.82 (dd, $J = 12.8, 7.2$ Hz, 1 H), 4.46 (dd, $J = 14.5, 4.8$ Hz, 1 H), 4.39 (dd, $J = 14.5, 7.5$ Hz, 1 H), 2.88 (s, 6 H), 2.13 (s, 3 H), 1.48 (s, 9 H), 1.14 (s, 9 H) ppm; ^{13}C NMR (125 MHz, CDCl_3): $\delta_{\text{C}} = 169.5, 169.4, 155.1, 152.0, 150.6, 134.7, 132.0, 131.2, 129.6, 129.3, 128.8, 122.8, 117.7, 115.2, 85.0, 80.2, 79.9, 77.2, 52.7, 47.0, 45.3, 28.3, 27.5, 20.7$ ppm.

Compound 4. ^1H NMR (500 MHz, d6-DMSO): $\delta_{\text{H}} = 8.51$ (d, $J = 8.5$ Hz, 1 H), 8.37 (t, $J = 6.5$ Hz, 1 H), 8.26 (d, $J = 8.5$ Hz, 1 H), 8.11 (dd, $J = 7.2, 1.2$ Hz, 1 H), 7.67 (dd, $J = 8.2, 7.8$ Hz, 1 H), 7.63 (dd, $J = 8.5, 7.5$ Hz, 1 H), 7.32 (d, $J = 8.0$ Hz, 1 H), 5.73 (d, $J = 6.0$ Hz, 1 H), 5.59 (d, $J = 5.5$ Hz, 1 H), 4.20 (t, $J = 4.8$ Hz, 1 H), 3.27 (ddd, $J = 14.5, 7.0, 5.5$ Hz, 1 H), 3.17 (dt, $J = 14.5, 5.5$ Hz, 1 H), 2.85 (s, 6 H), 2.07 (s, 3 H) ppm; ^{13}C NMR (125 MHz, d6-DMSO): $\delta_{\text{C}} = 169.1, 166.7, 158.5$ (q, $J = 36.1$ Hz), 151.1, 134.8, 129.9, 129.1, 128.9, 128.8, 128.2, 123.8, 119.1, 115.6, 79.8, 52.3, 45.2, 42.2, 20.5 ppm. LC-MS (ESI): (m/z) 410.2 $[\text{M}+\text{H}]^+$. HRMS (ESI-FT): calcd for $\text{C}_{18}\text{H}_{24}\text{N}_3\text{O}_6\text{S}$ (M+1) 410.1380, found 410.1382.

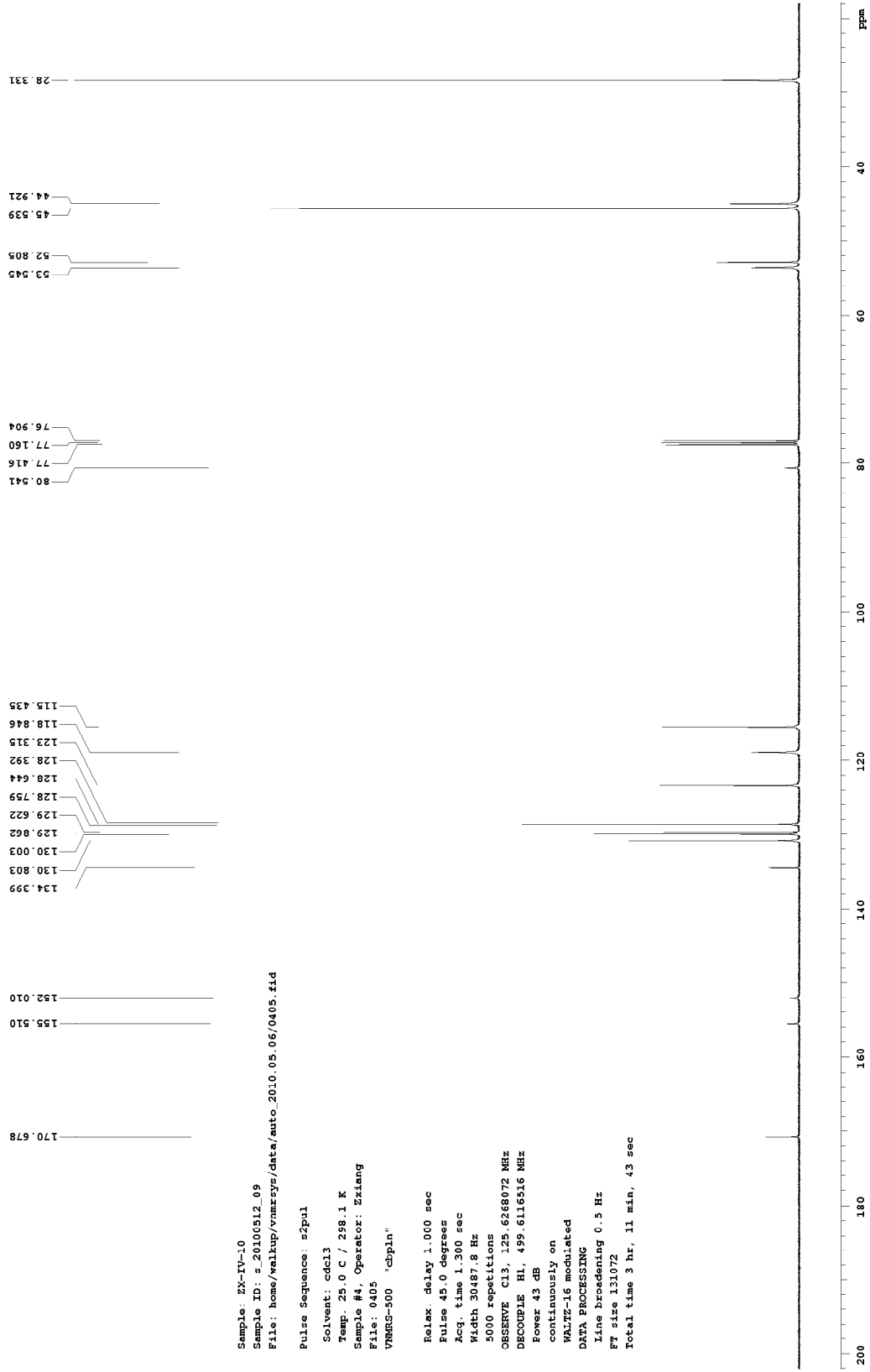
NMR Spectra

¹ H NMR (500 MHz, CDCl ₃) of Compound 6	4
¹³ C NMR (125 MHz, CDCl ₃) of Compound 6	5
¹ H NMR (500 MHz, CDCl ₃) of Compound 7	6
¹³ C NMR (125 MHz, CDCl ₃) of Compound 7	7
¹ H NMR (500 MHz, d6-DMSO) of Compound 2	8
¹³ C NMR (125 MHz, d6-DMSO) of Compound 2	9
¹ H NMR (500 MHz, d6-DMSO) of Compound 3	10
¹³ C NMR (125 MHz, d6-DMSO) of Compound 3	11
¹ H NMR (500 MHz, CDCl ₃) of Compound 8	12
¹³ C NMR (125 MHz, CDCl ₃) of Compound 8	13
¹ H NMR (500 MHz, CDCl ₃) of Boc-DanAla-OBn.....	14
¹³ C NMR (125 MHz, CDCl ₃) of Boc-DanAla-OBn.....	15
¹ H NMR (500 MHz, CDCl ₃) of Compound 9	16
¹³ C NMR (125 MHz, CDCl ₃) of Compound 9	17
¹ H NMR (500 MHz, CDCl ₃) of Compound 10	18
¹³ C NMR (125 MHz, CDCl ₃) of Compound 10	19
¹ H NMR (500 MHz, CDCl ₃) of Compound 11	20
¹³ C NMR (125 MHz, CDCl ₃) of Compound 11	21
¹ H NMR (500 MHz, d6-DMSO) of Compound 4	22
¹³ C NMR (125 MHz, d6-DMSO) of Compound 4	23

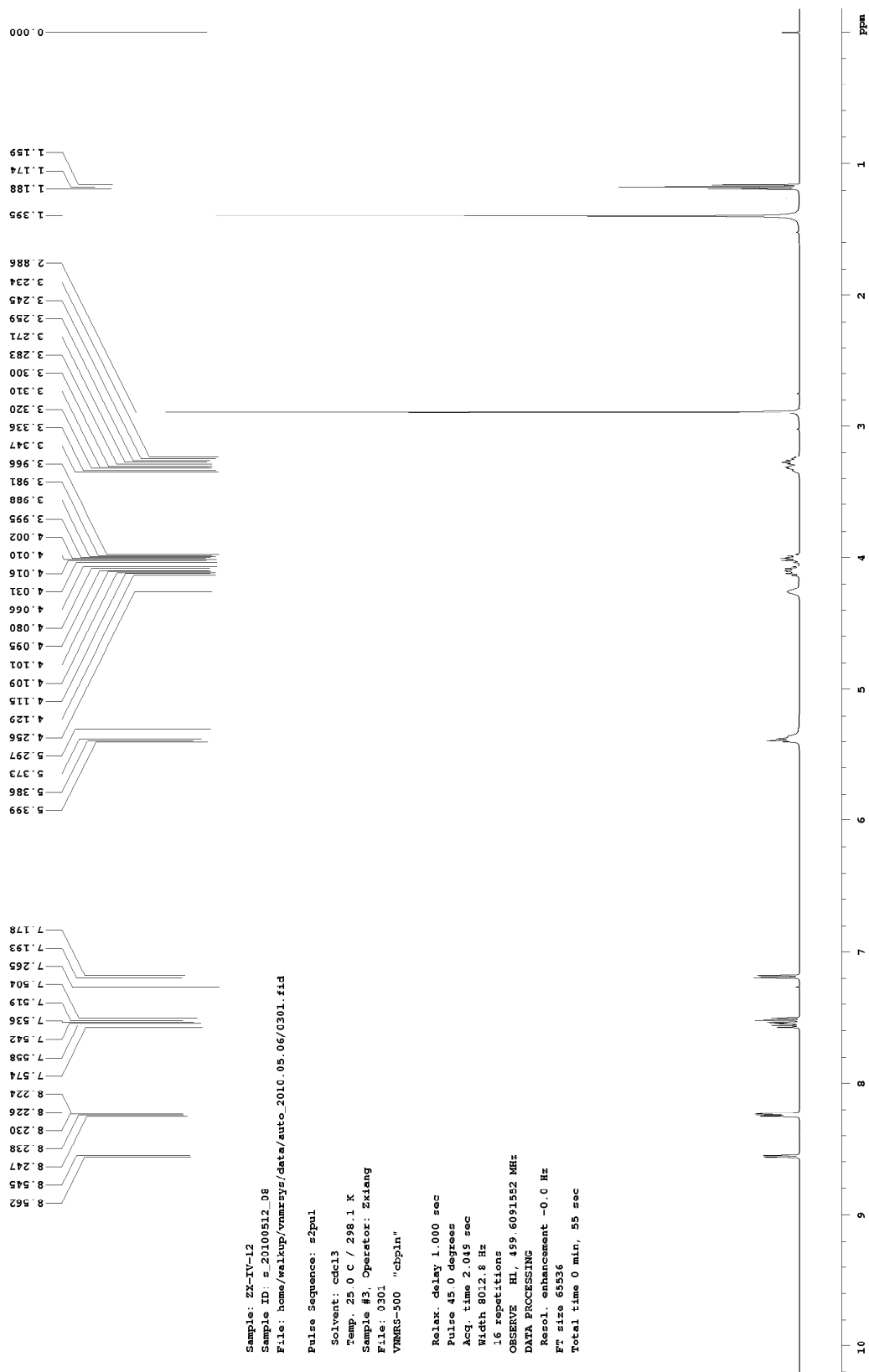
¹H NMR (500 MHz, CDCl₃) of Compound 6



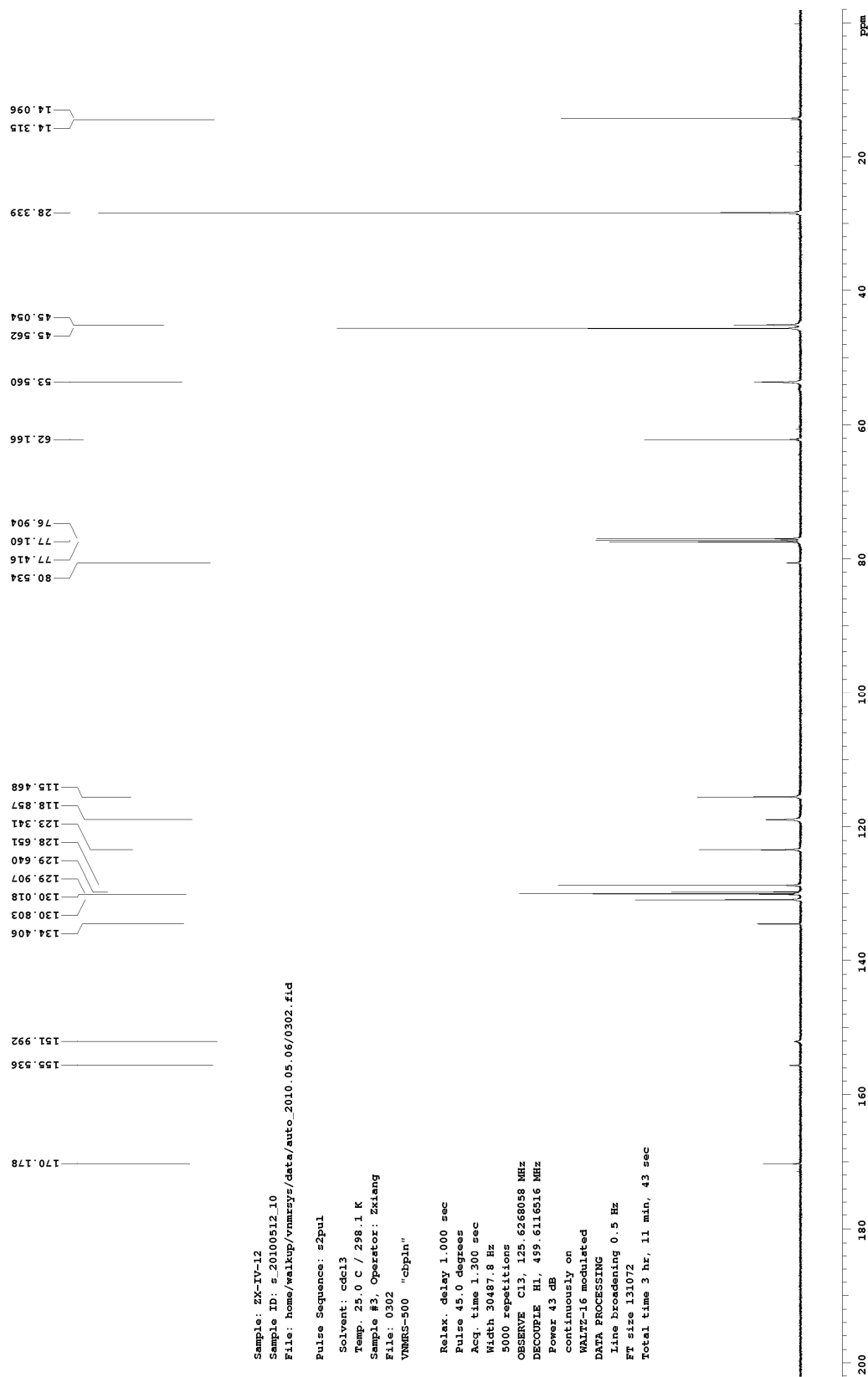
¹³C NMR (125 MHz, CDCl₃) of Compound 6



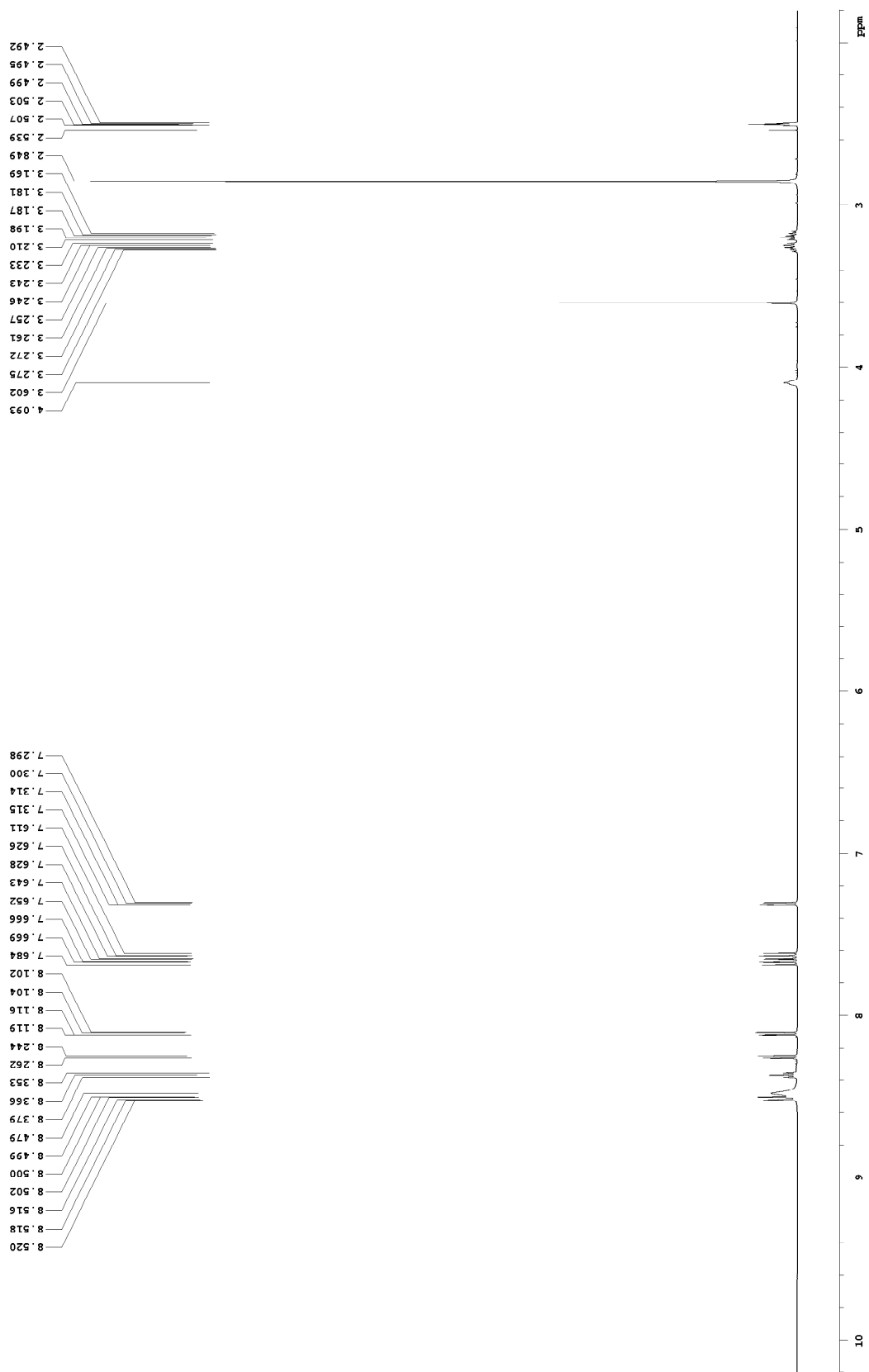
¹H NMR (500 MHz, CDCl₃) of Compound 7



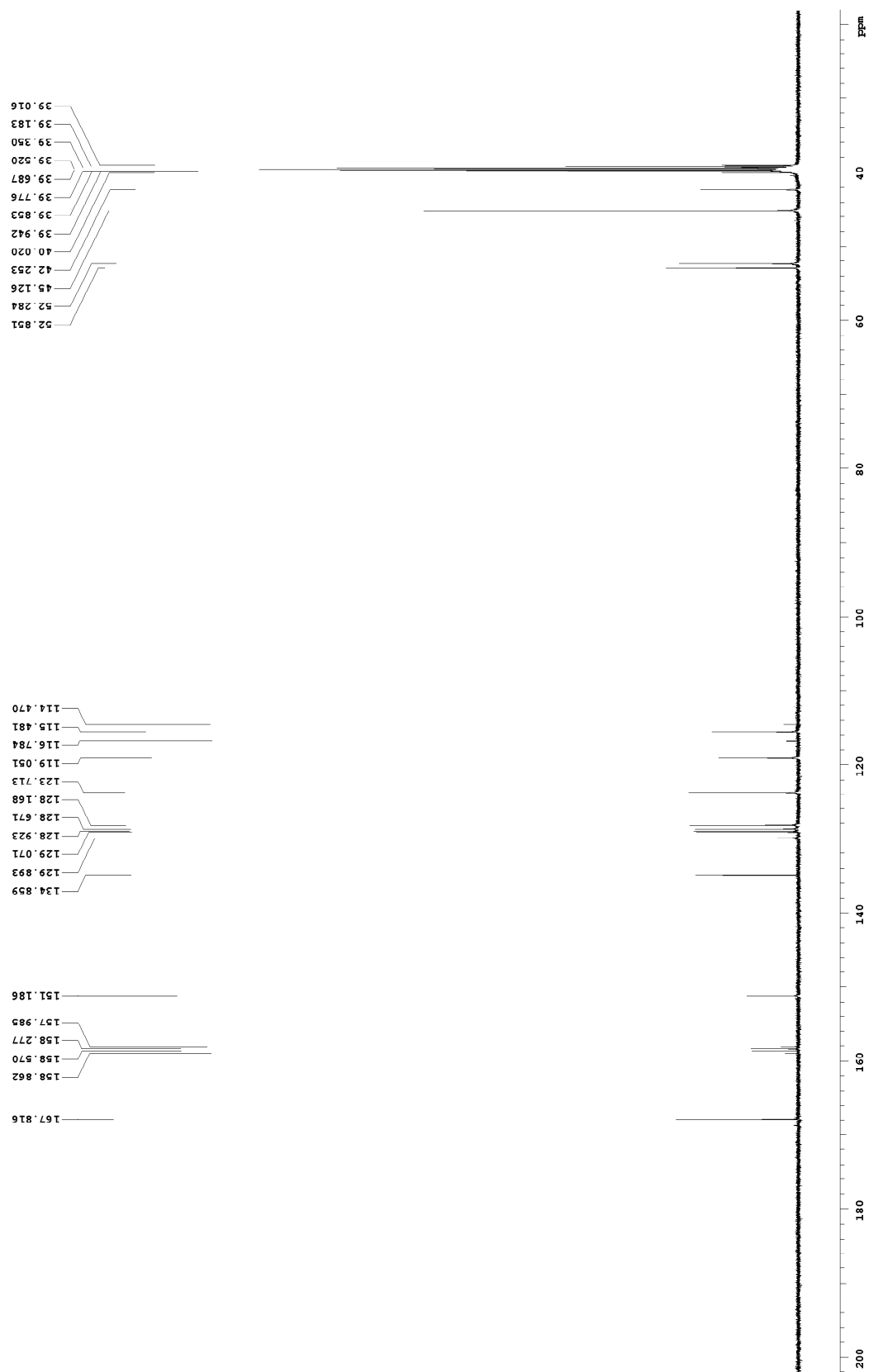
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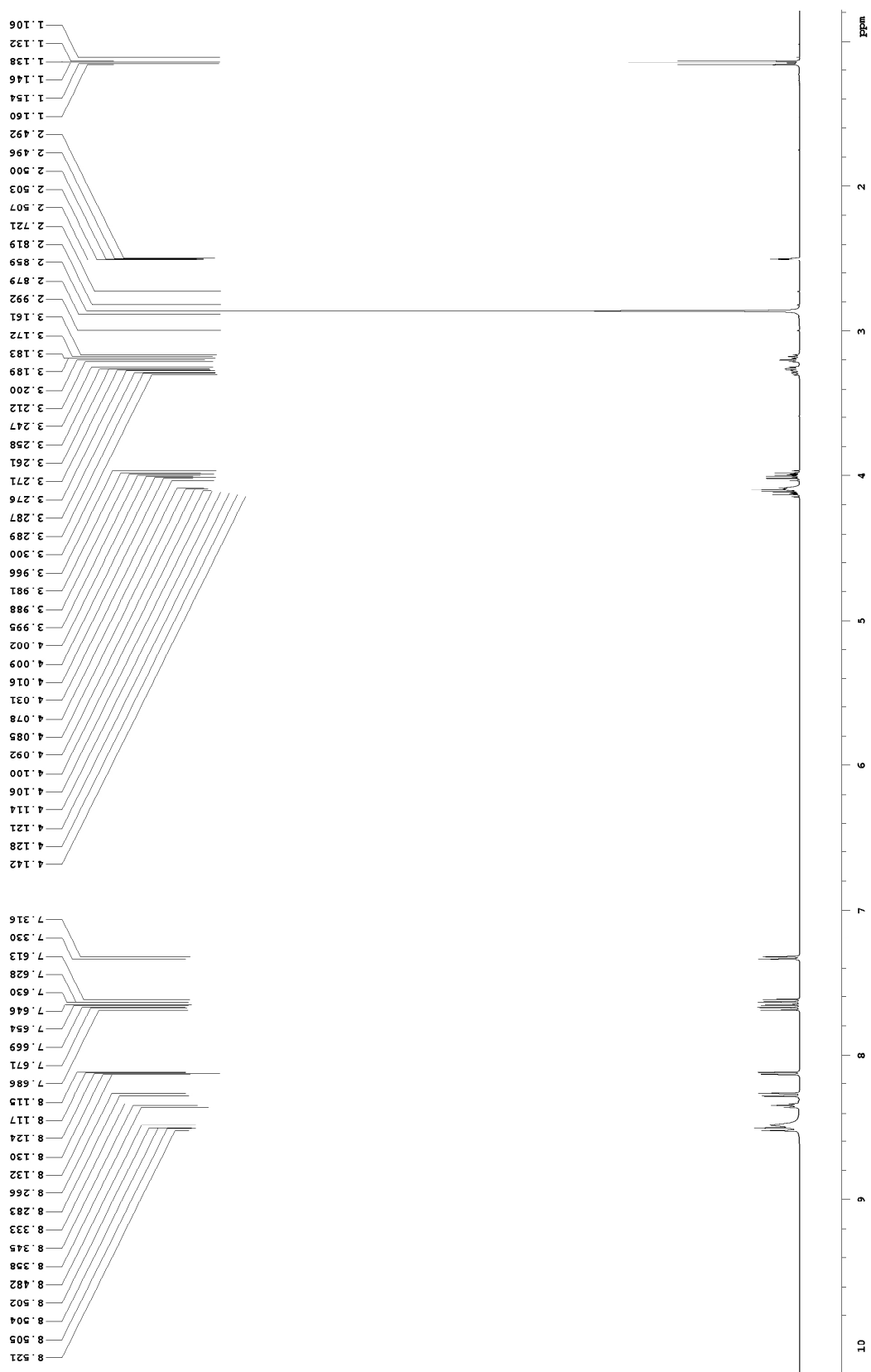
¹H NMR (500 MHz, d6-DMSO) of Compound 2



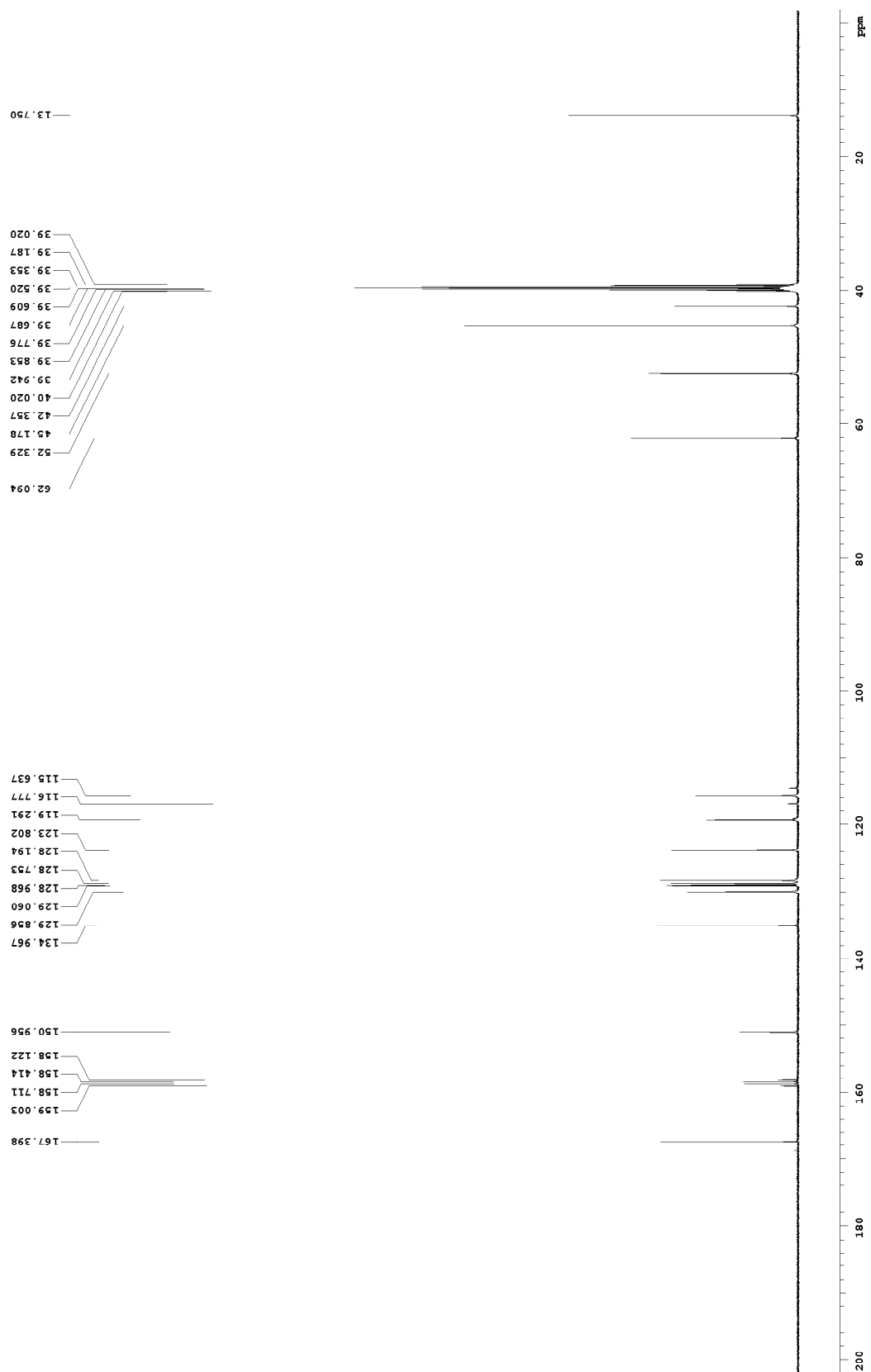
¹³C NMR (125 MHz, d6-DMSO) of Compound 2



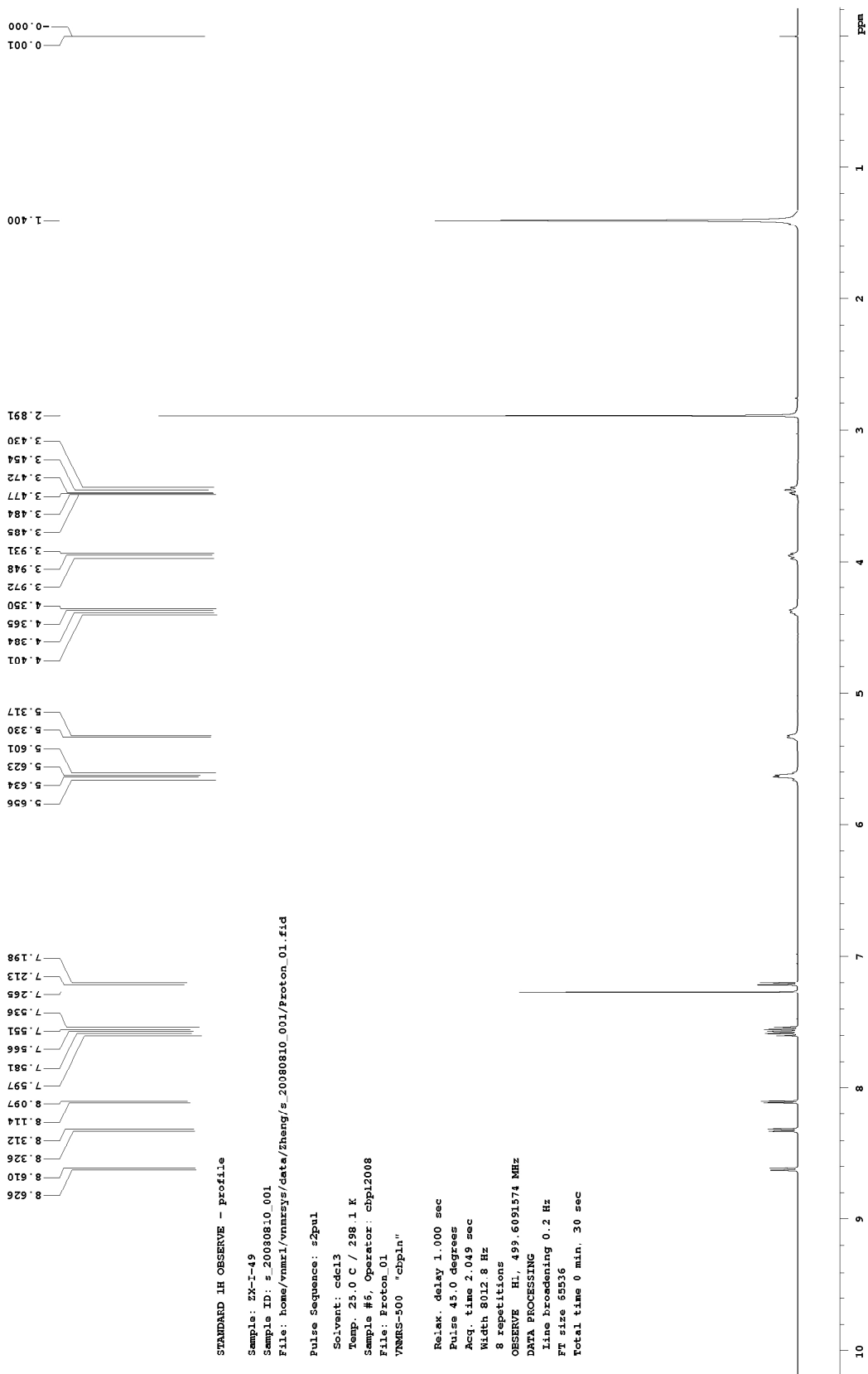
¹H NMR (500 MHz, d6-DMSO) of Compound 3



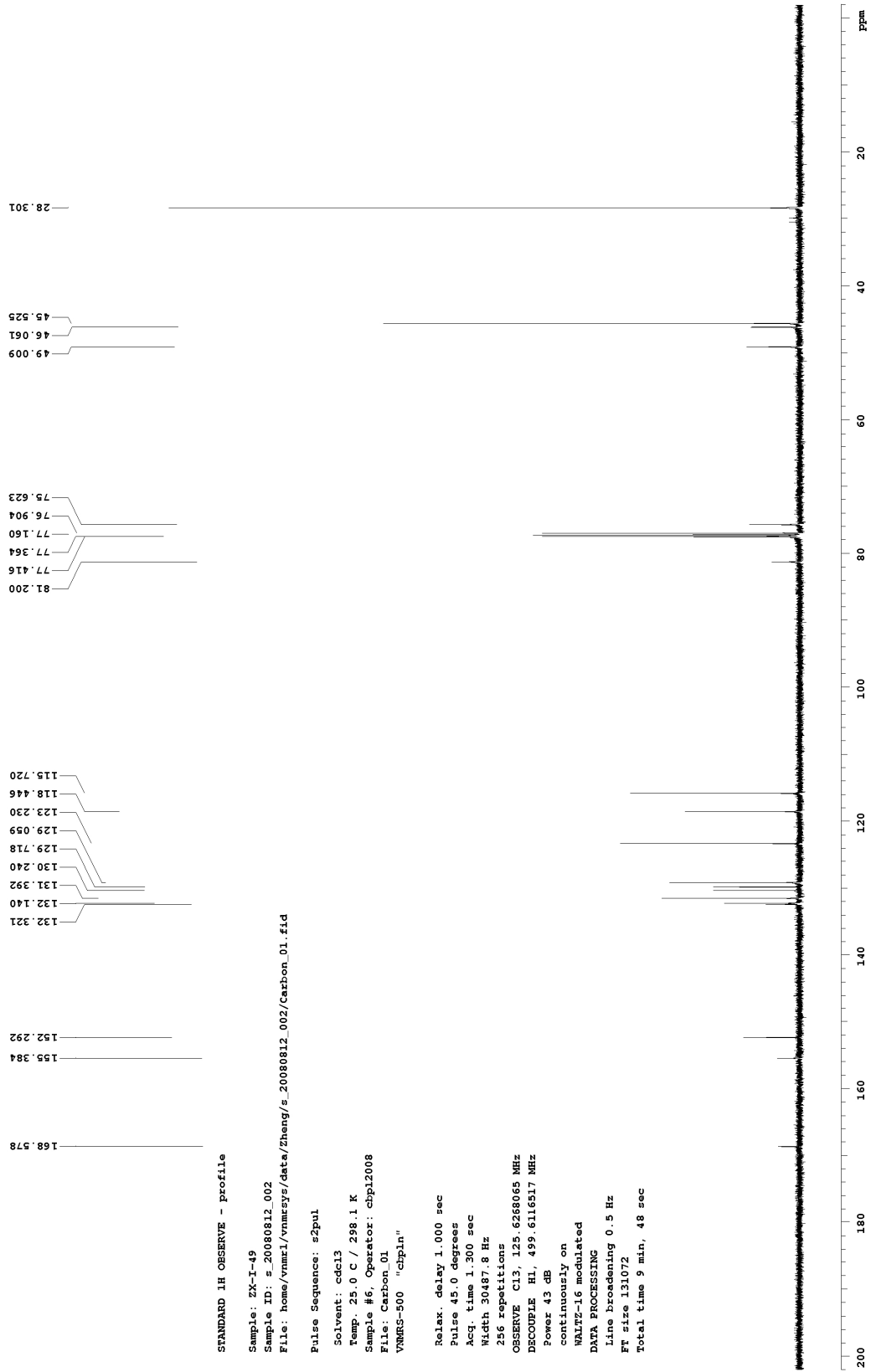
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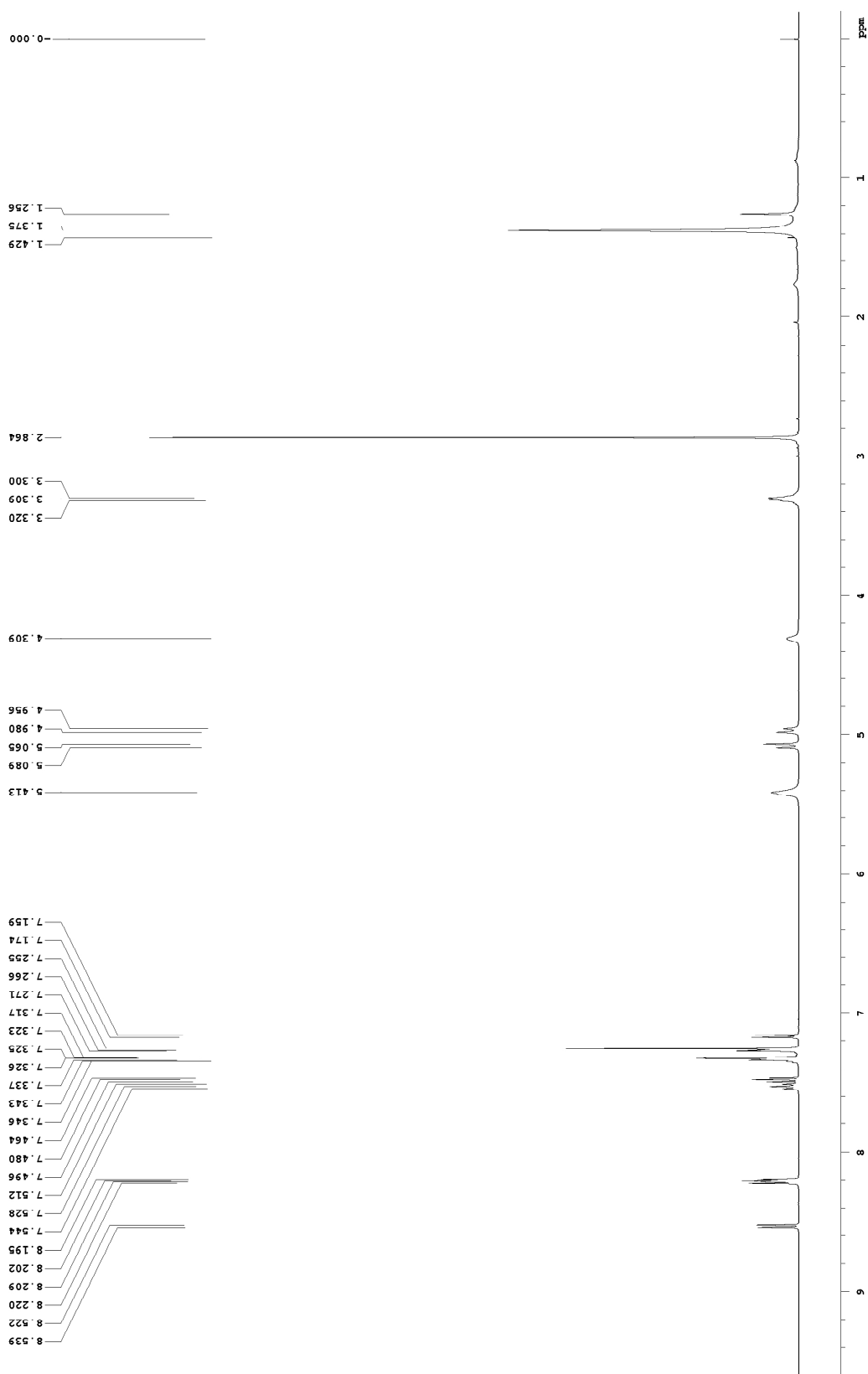
¹H NMR (500 MHz, CDCl₃) of Compound 8



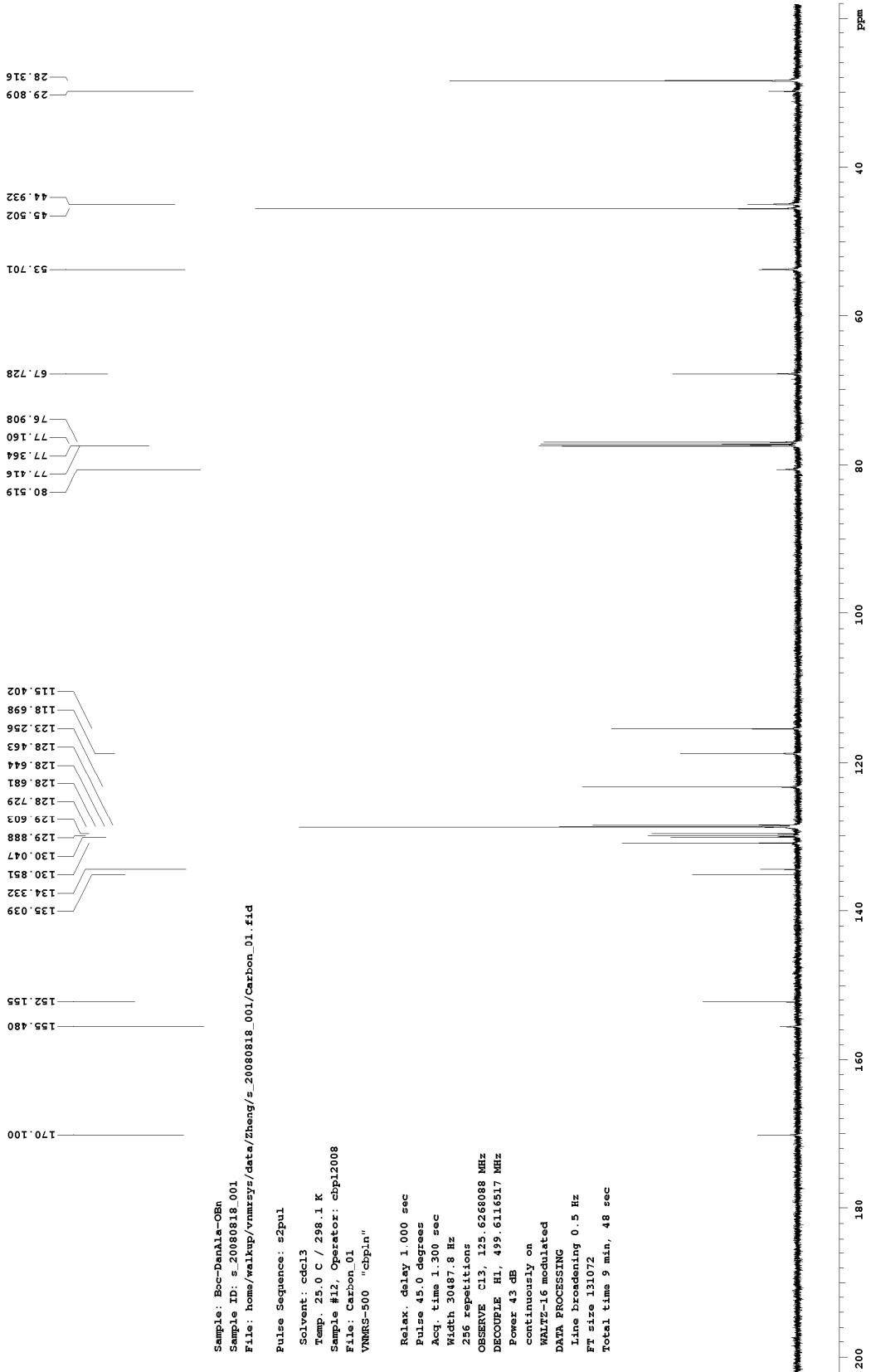
¹³C NMR (125 MHz, CDCl₃) of Compound 8



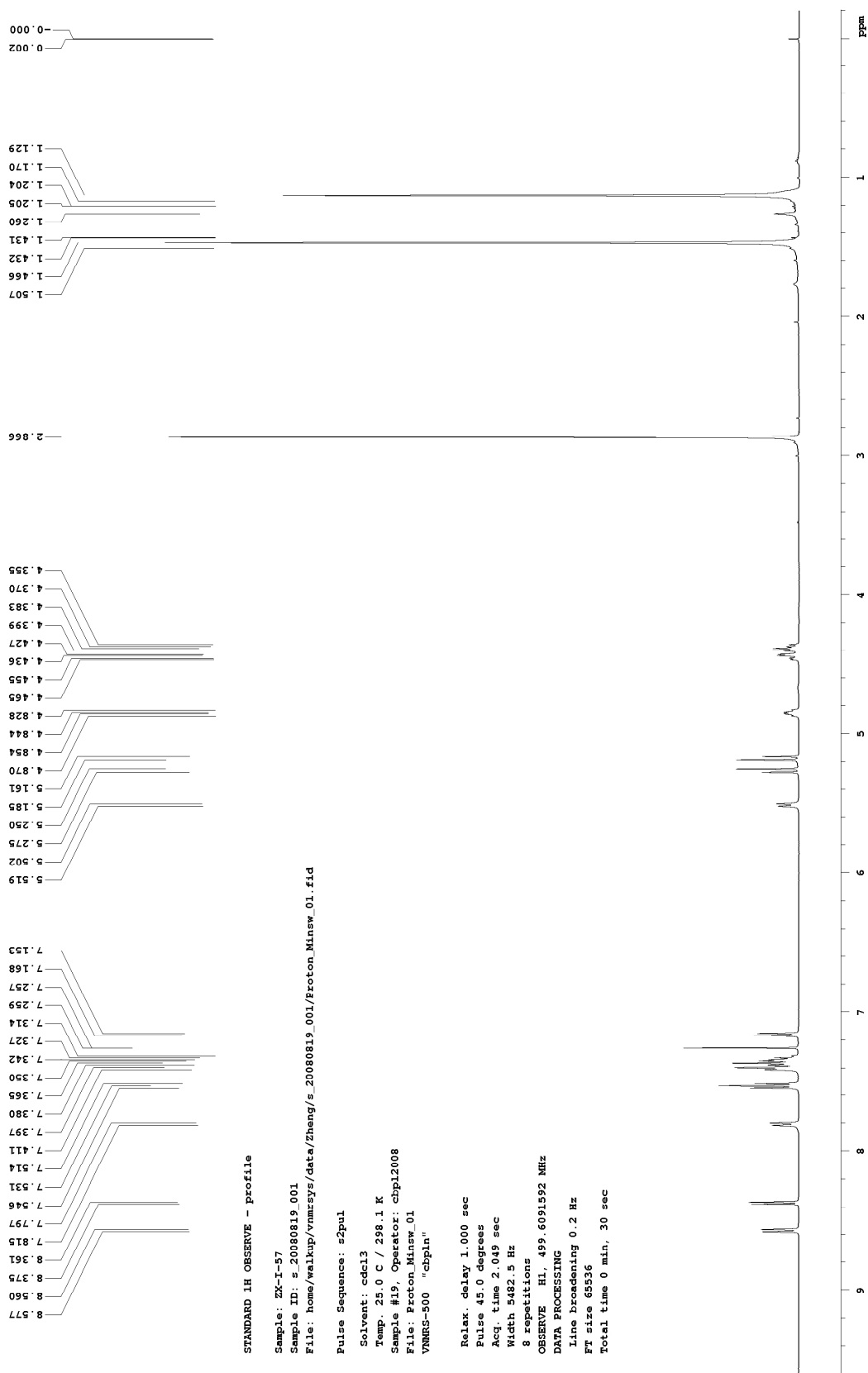
^1H NMR (500 MHz, CDCl_3) of Boc-DanAla-OBn



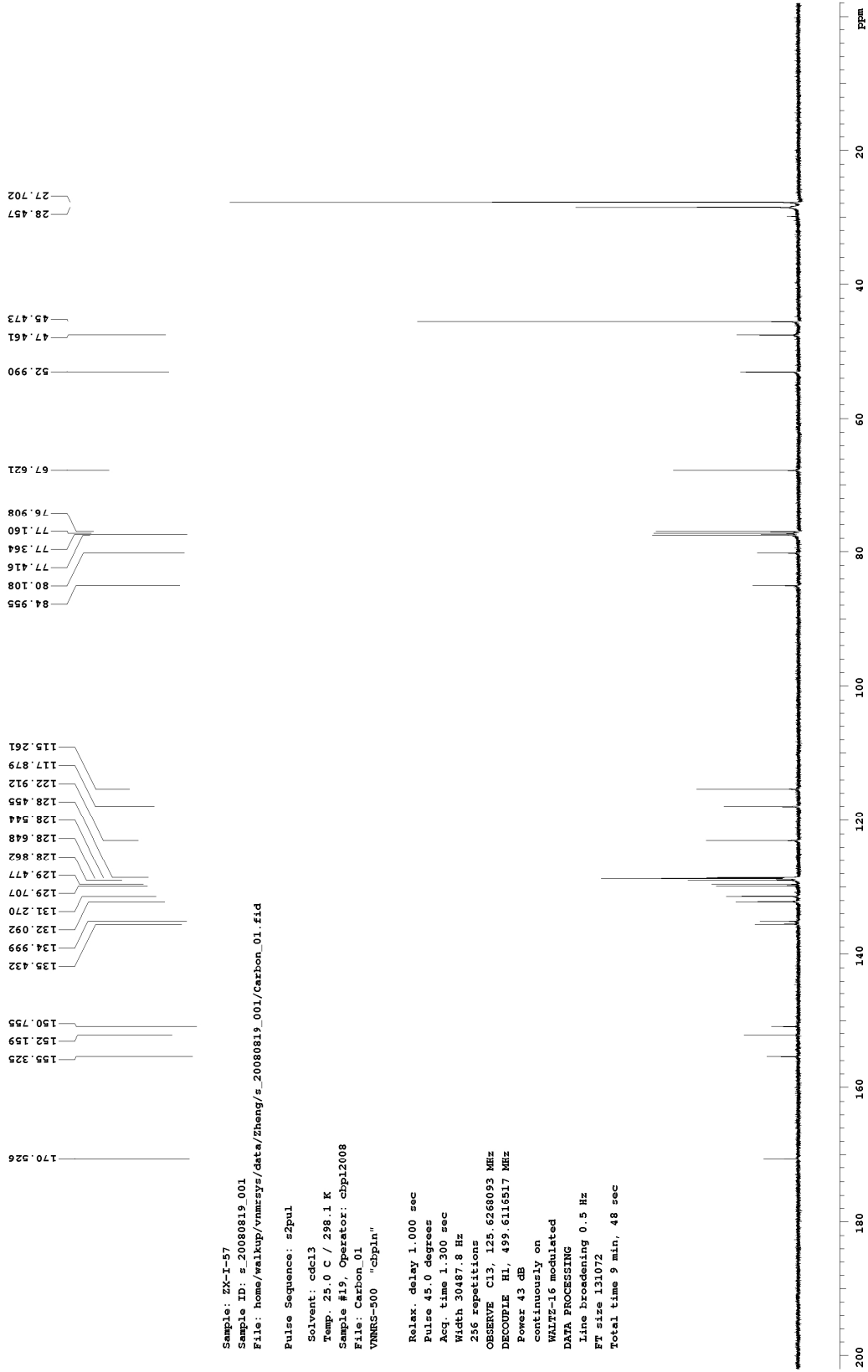
¹³C NMR (125 MHz, CDCl₃) of Boc-DanAla-OBn



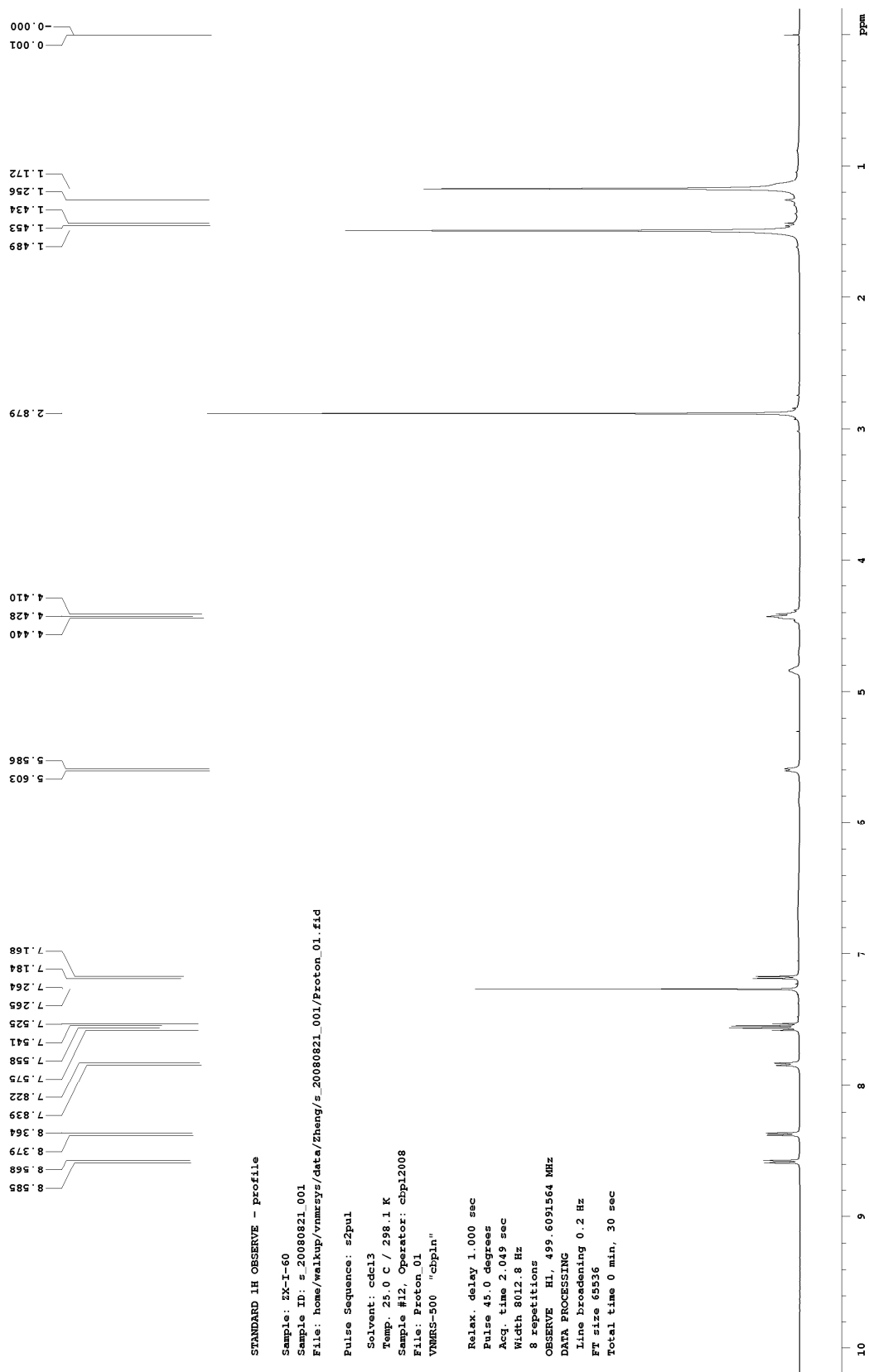
¹H NMR (500 MHz, CDCl₃) of Compound 9



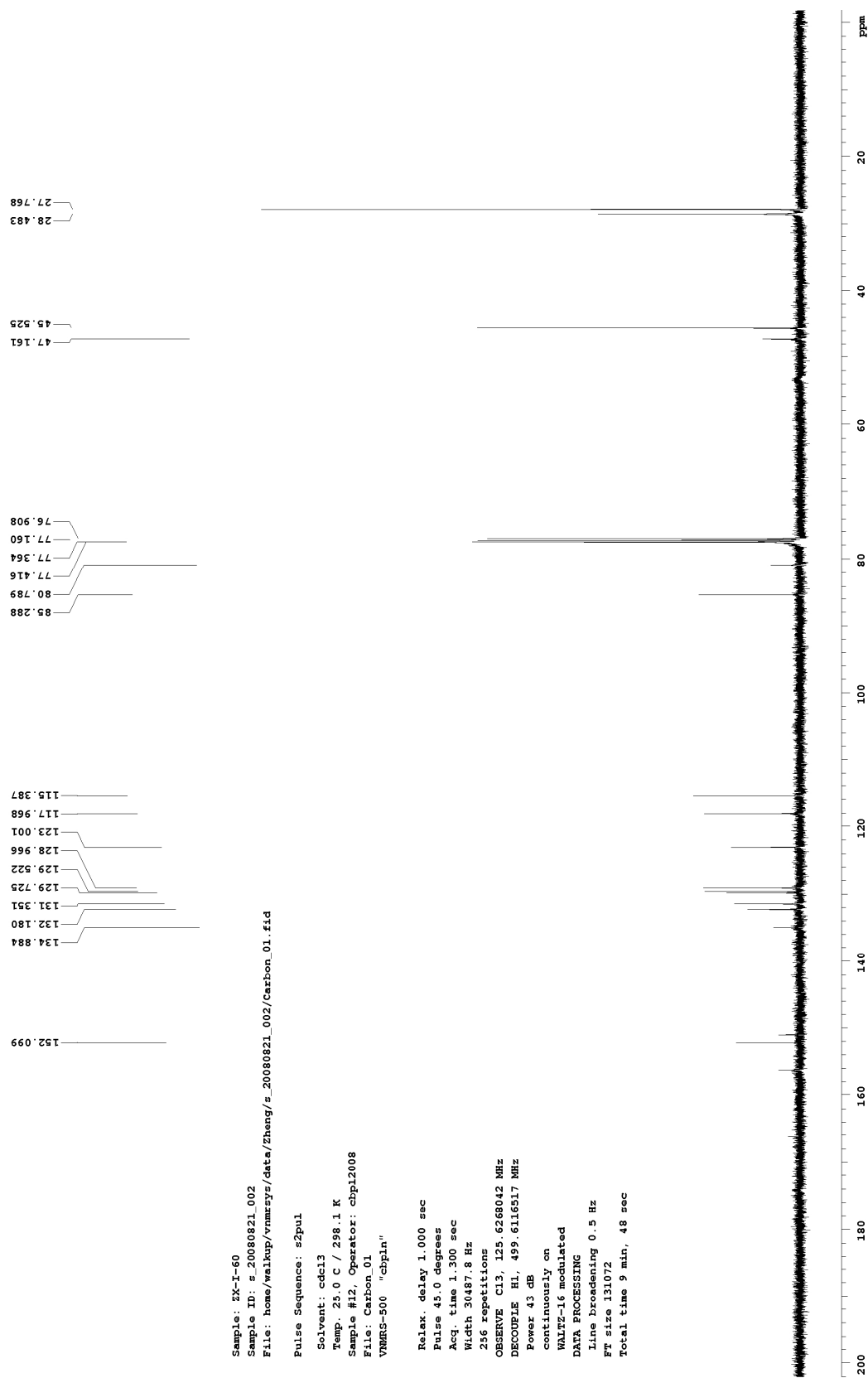
¹³C NMR (125 MHz, CDCl₃) of Compound 9



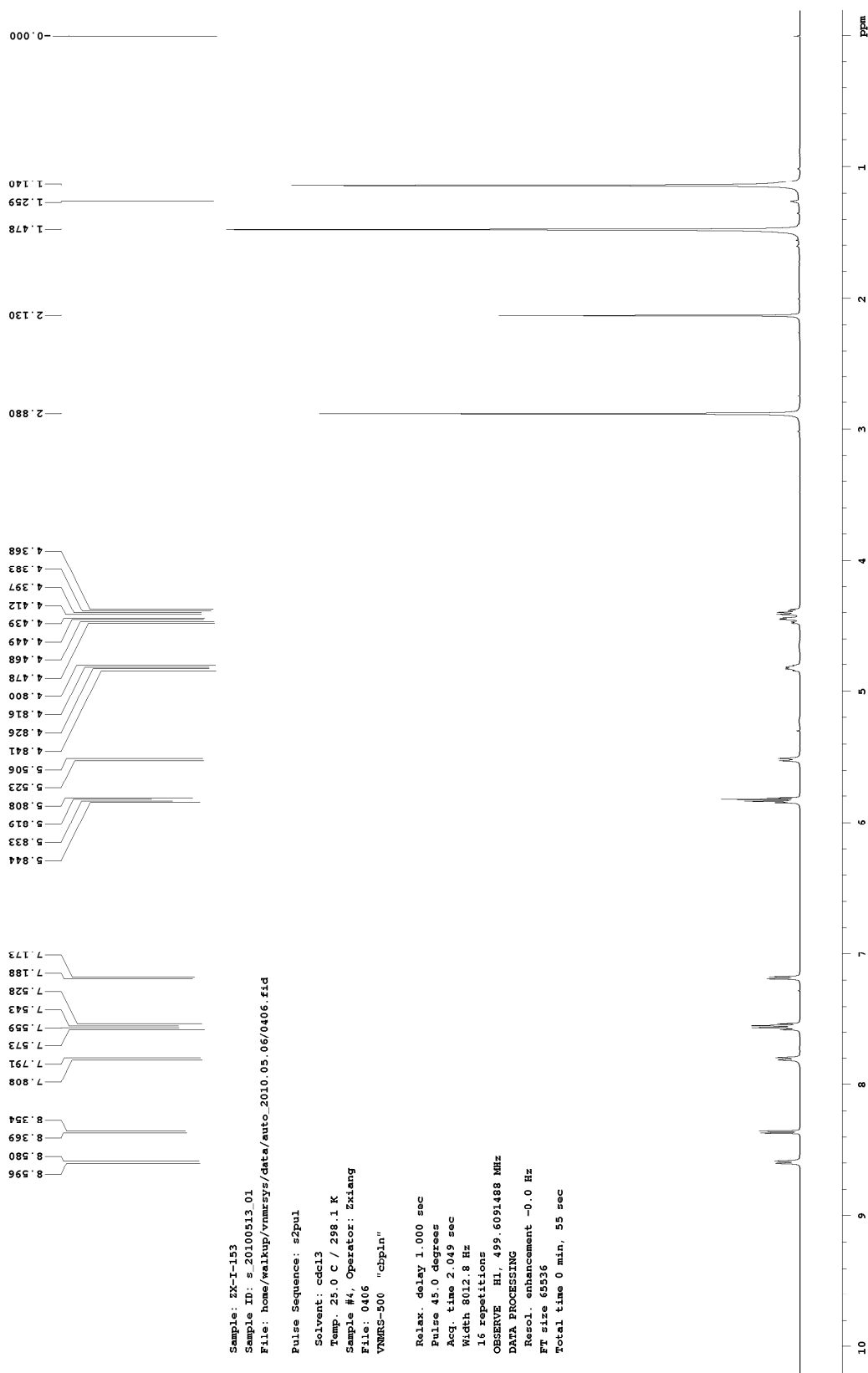
¹H NMR (500 MHz, CDCl₃) of Compound 10



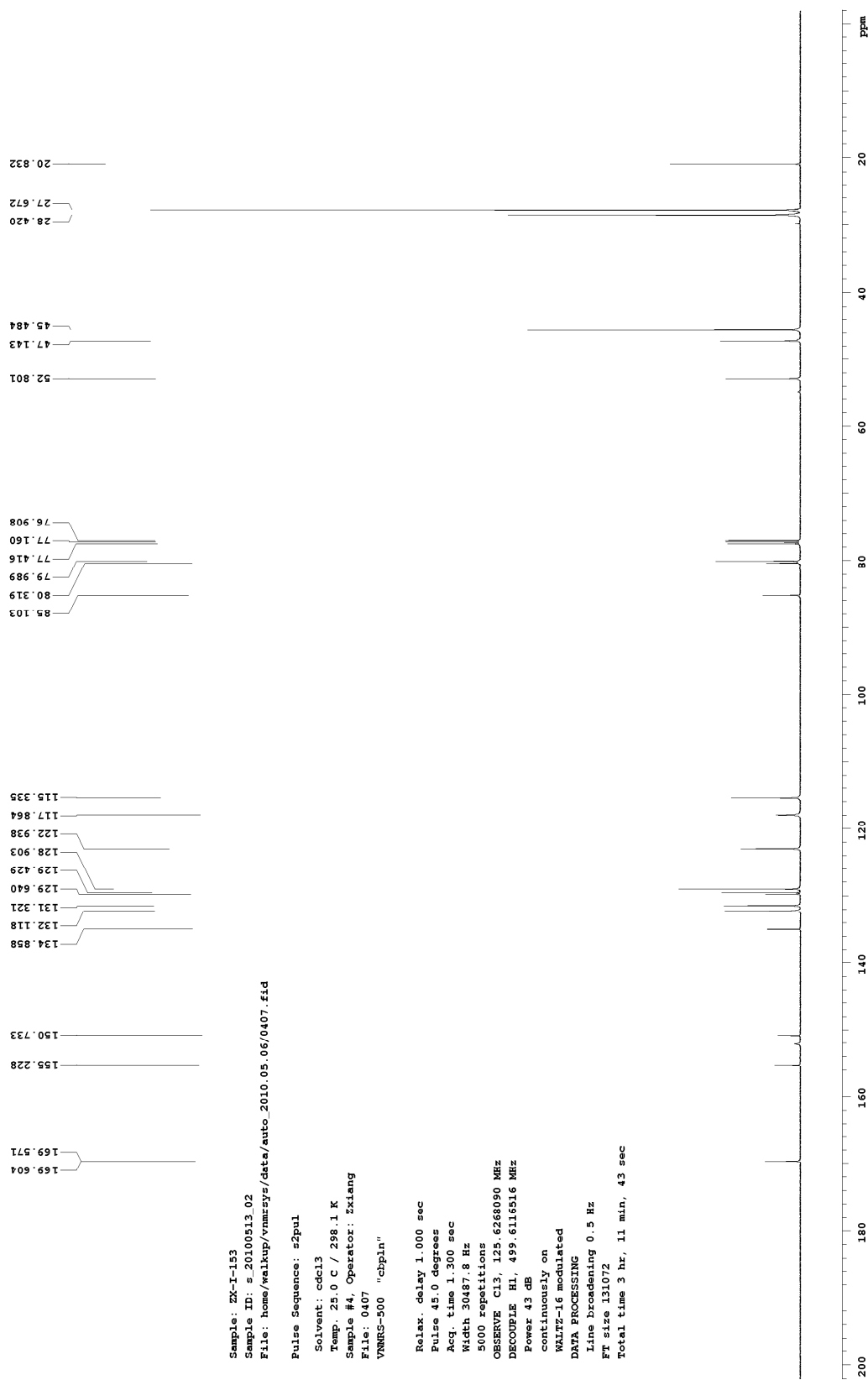
¹³C NMR (125 MHz, CDCl₃) of Compound 10



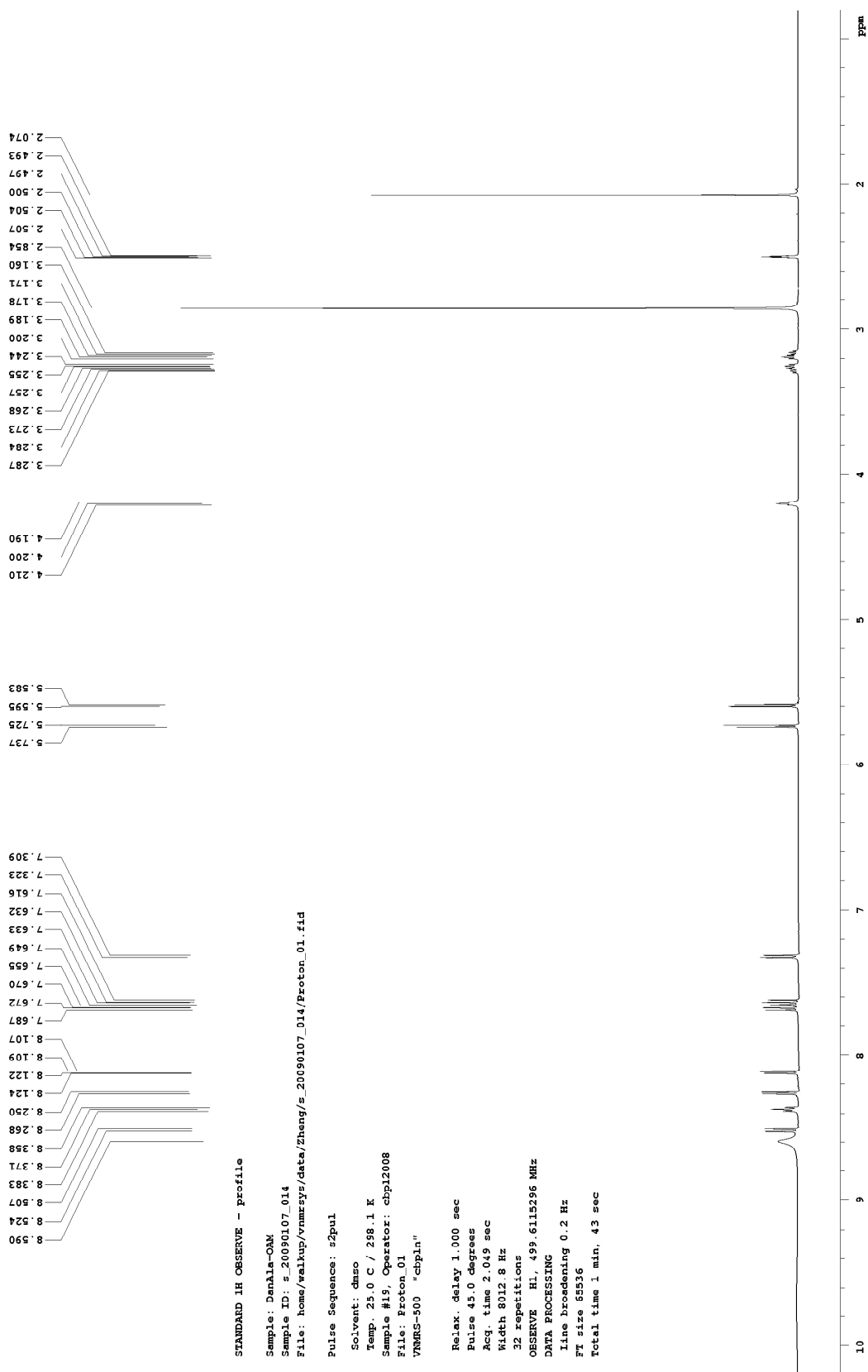
¹H NMR (500 MHz, CDCl₃) of Compound 11



¹³C NMR (125 MHz, CDCl₃) of Compound 11



¹H NMR (500 MHz, d6-DMSO) of Compound 4



¹³C NMR (125 MHz, d6-DMSO) of Compound 4

