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Supplementary Material

The Synthesis of a Two-photon Fluorescence Labelling Probe and Its Immunochromatographic Strip for Rapid Diagnosis of COVID-19

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Materials

NMR spectra were recorded on a VARIAN INOVA 400 MHz NMR spectrometer. Mass spectral determinations were made on a Q-TOF mass spectrometry (Micromass, England). High resolution mass spectra measurements were performed at a GC-TOF mass spectrometry (Micromass, US) (Electron Ionization Source). Fluorescence mea-surements were performed on a PTI-C-700 Felix and Time-Master system. Fluoresce-nce quantum yields were measured using standard methods [S1] on air-equilibrated samples at room temperature. Quinine bisulfate in 0.05M H_2SO_4 (Φ = 0.546) was used as a reference [S1].

TPEF (two-photon-excited fluorescence) action cross-section spectra were measured according to the experimental protocol established by Xu and Webb [S2], using a mode-locked Ti/sapphire laser that delivers ~ 80 fs pulses at 80 MHz. Fluorescein (10^{-4} M in 0.1M NaOH), whose TPEF action cross-sections are well-known [S2], served as the reference. The quadratic dependence of the fluorescence intensity on the excitation intensity was verified for each data point, indicating that the measurements were carried out in intensity regimes in which saturation or photodegradation do not occur. The measurements were performed at room temperature on air-equilibrated solutions (10^{-5} M). The experimental uncertainty on the absolute action cross-sections determined by this method has been estimated to be ± 20% [S2]. Absorption spectra were measured on a HP-8453 spectrophotometer. Solvents were generally dried and distilled prior to use. Reactions were monitored by thin-layer chromatography on Merck silica gel 60 F_{254} precoated aluminum sheets. Column chromatography: Merck silica gel Si 60 (40-63 μm, 230-400 mesh).

Negative-positive discrimination

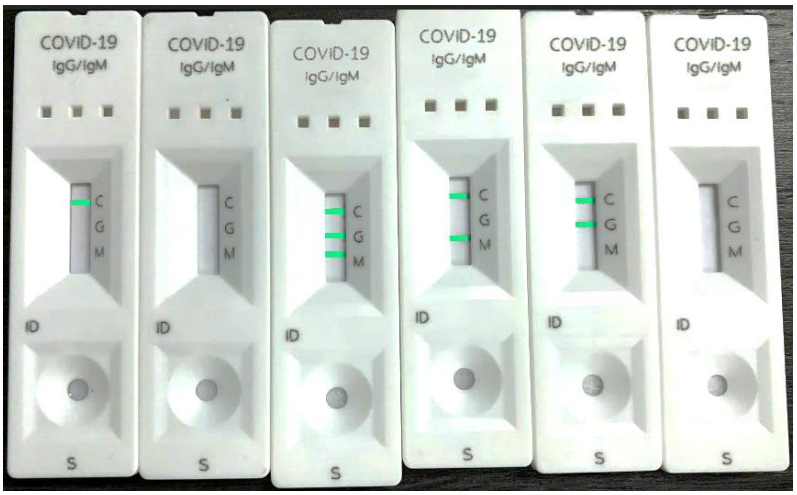


Figure S1 The diagram of negative-positive identification

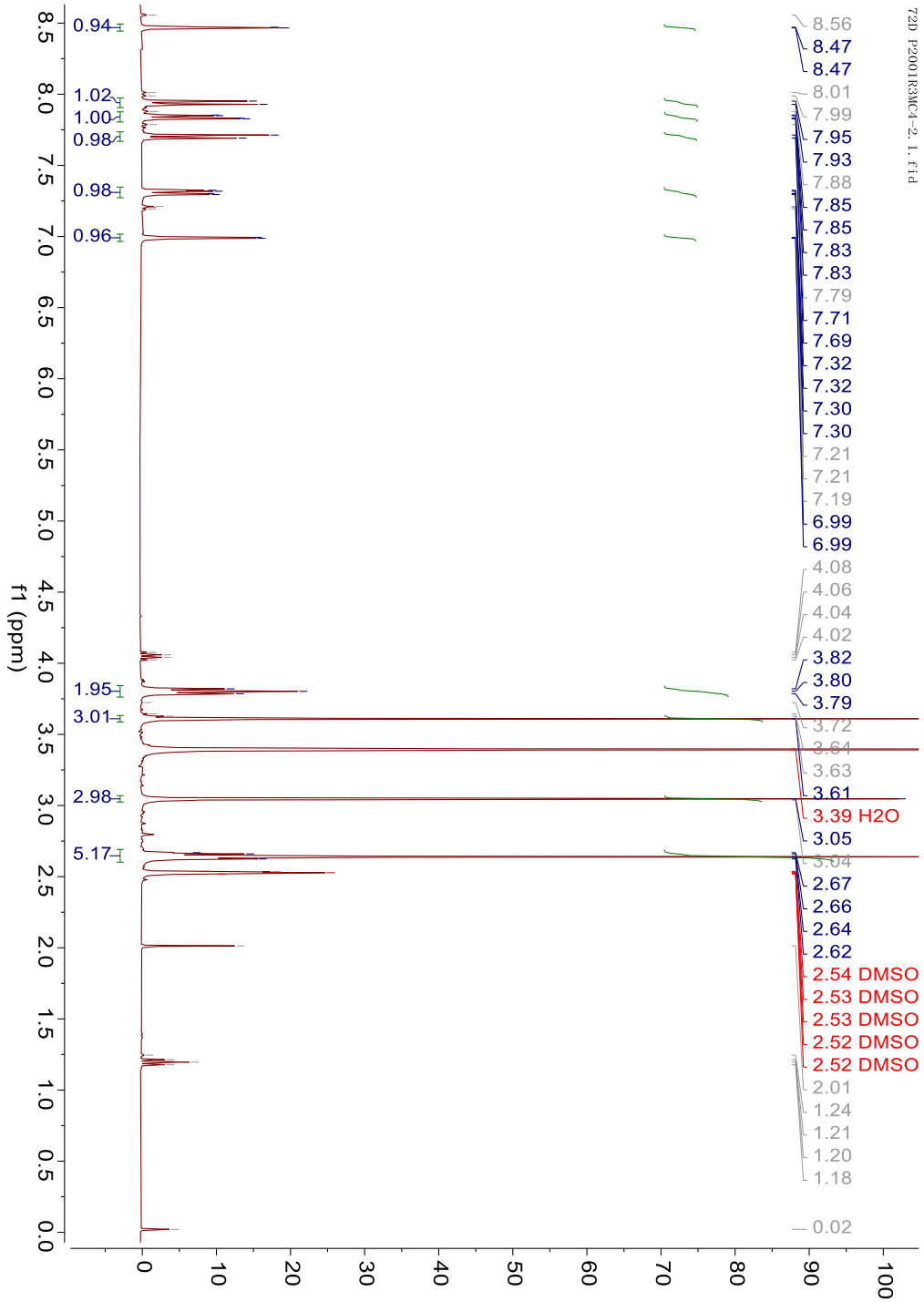
References

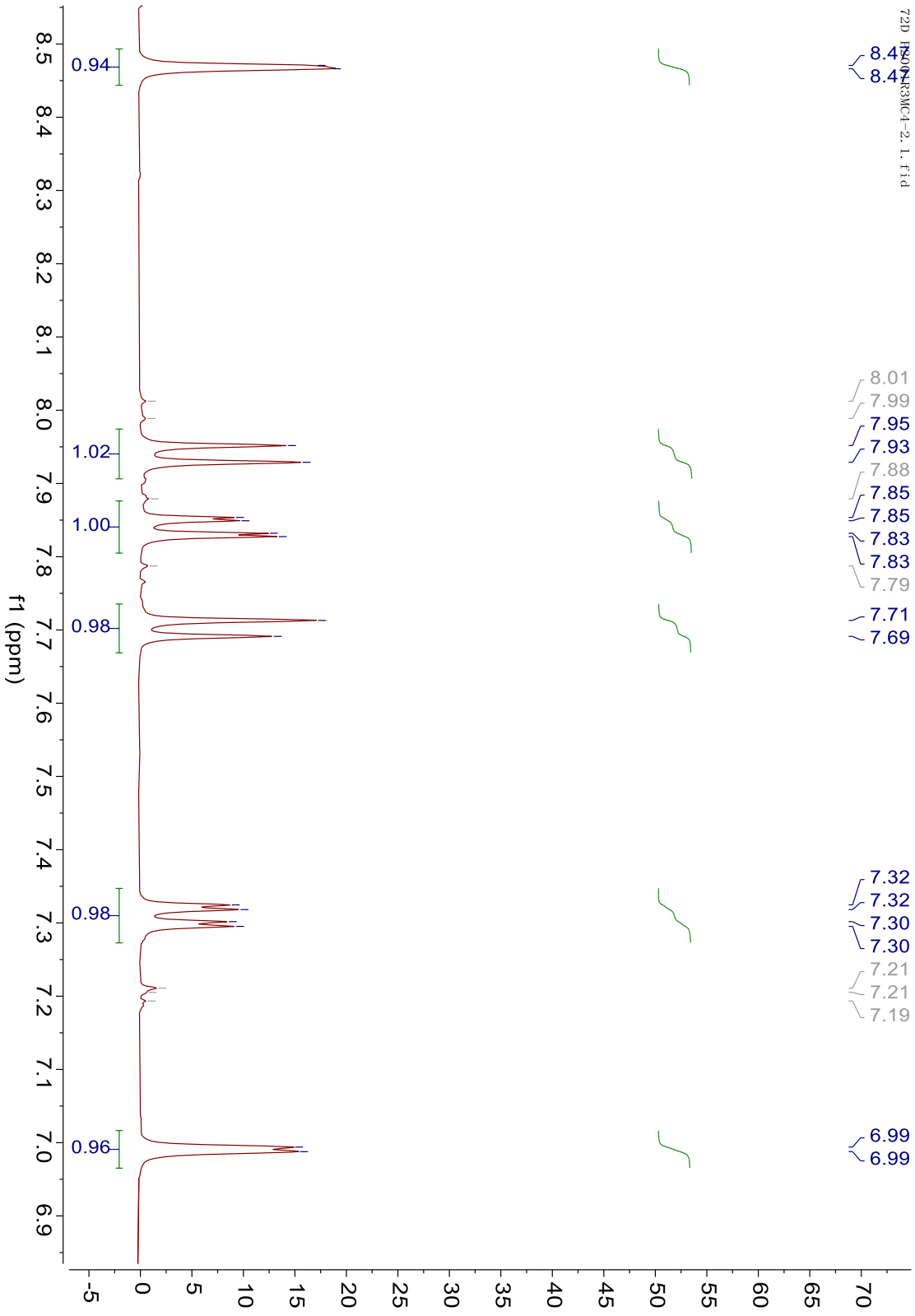
[S1] Eaton, D. F. *J. Photochem. Photobiol. B*, **1988**, *2*, 523.

[S2] Xu, C.; Webb, W. W. *J. Opt. Soc. Am. B*, **1996**, *13*, 481.

NMR Spectra

Figure S2. ^{1}H NMR spectra for compound 4 in DMSO-d_6




72D H2O+R3MC4-2. 1. fid
8.47
8.47
8.01
7.99
7.95
7.93
7.88
7.85
7.85
7.83
7.83
7.79
7.71
7.69
7.32
7.32
7.30
7.30
7.21
7.21
7.19
6.99
6.99
0.94
1.02
1.00
0.98
0.98
0.96
f1 (ppm)

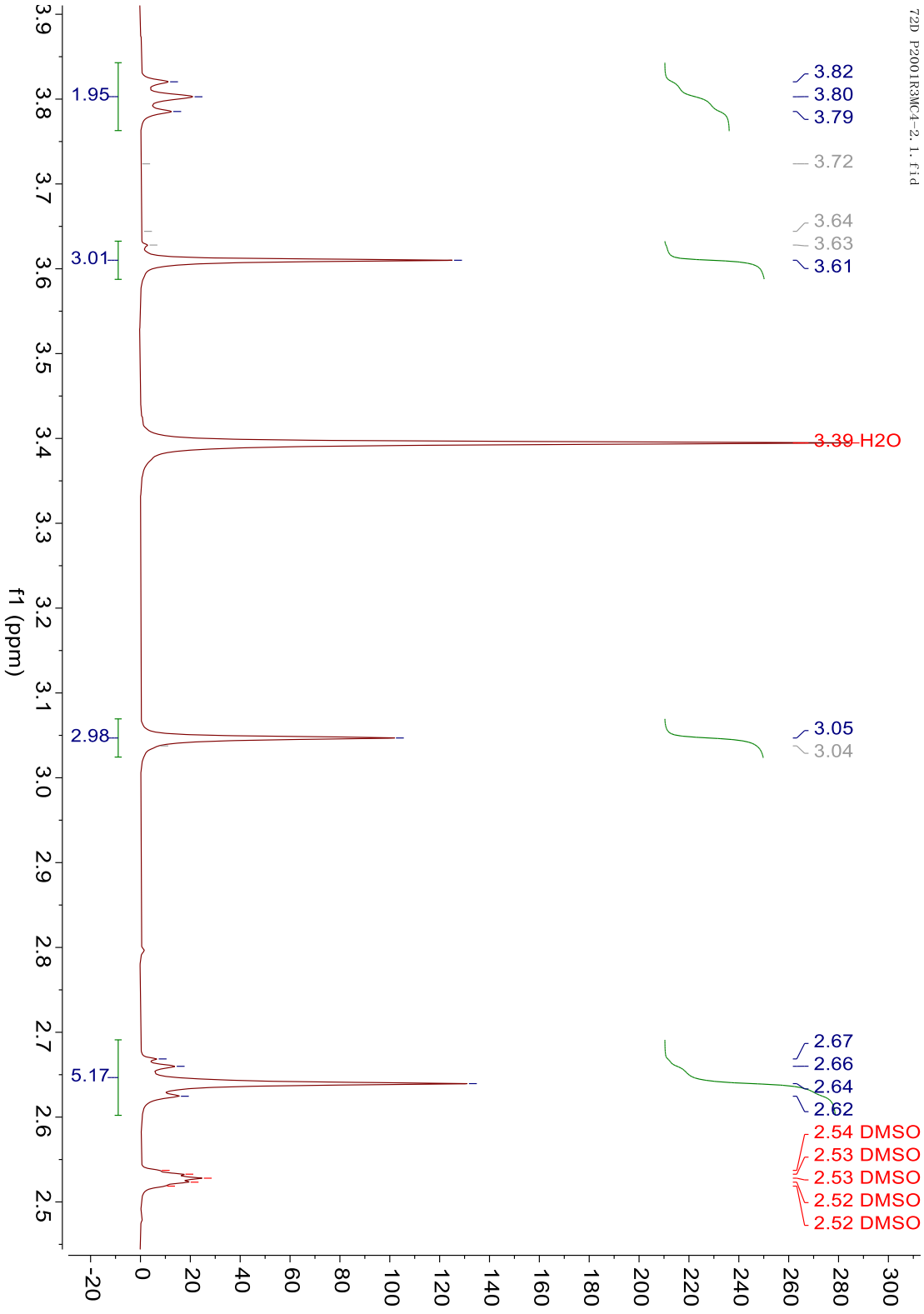

72D P2001R3MC4-2, 1, fid
3.82
3.80
3.79
3.72
3.64
3.63
3.61
3.39 H2O
3.05
3.04
2.67
2.66
2.64
2.62
2.54 DMSO
2.53 DMSO
2.53 DMSO
2.52 DMSO
2.52 DMSO
1.95
3.01
2.98
5.17
f1 (ppm)

Figure S3. ^{13}C NMR spectrum for compound 4 in $CHCl_3$-*d*

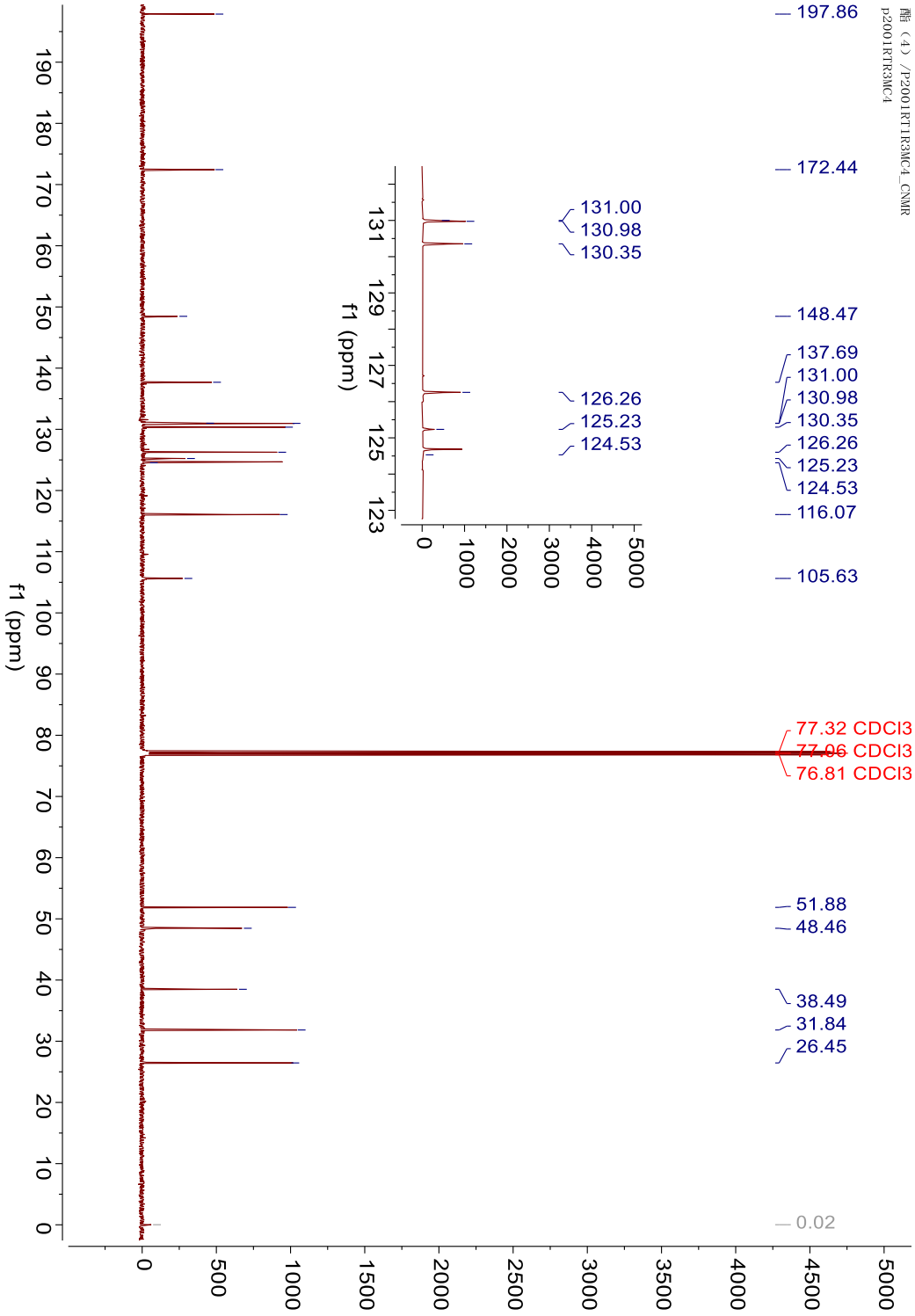
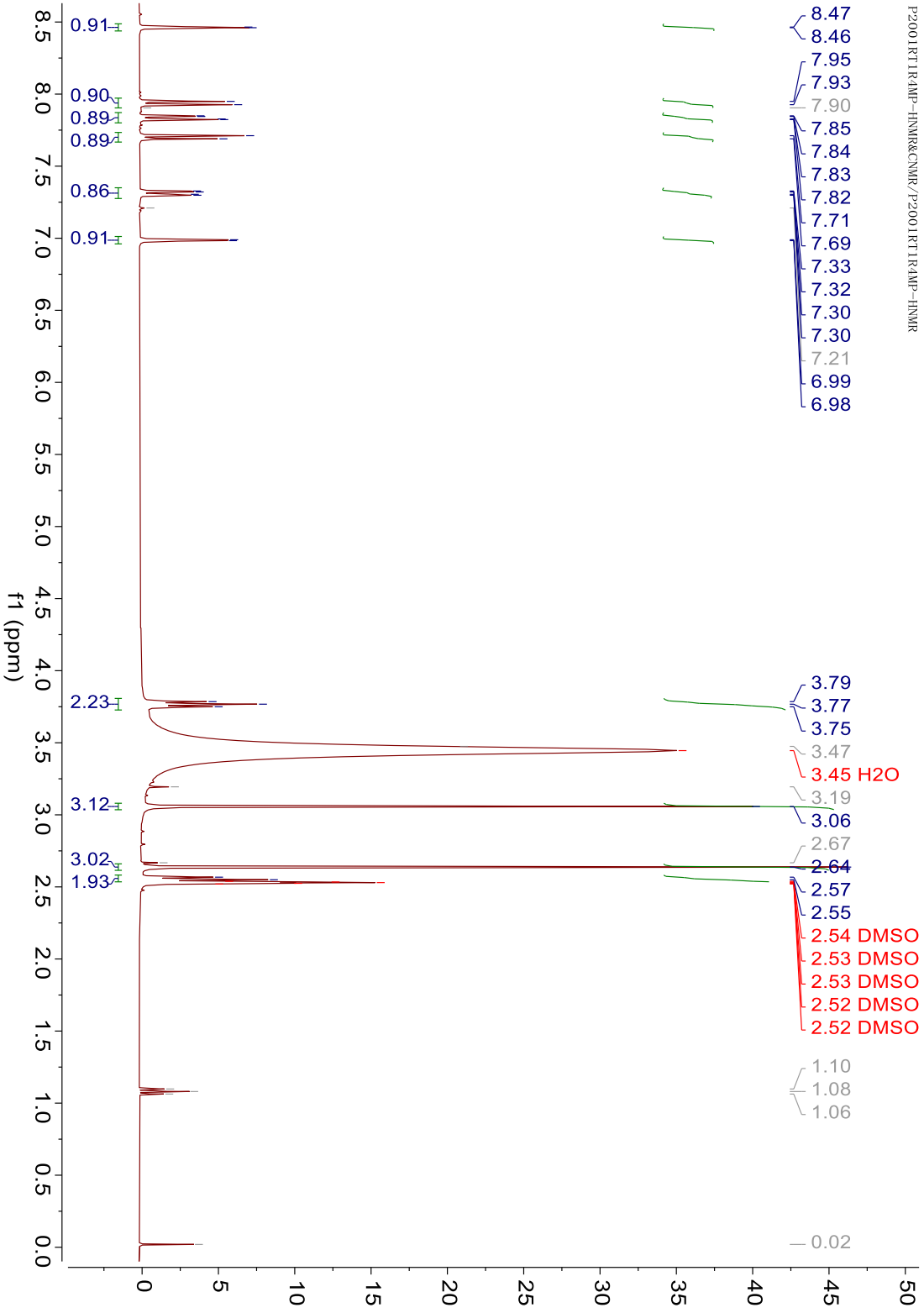
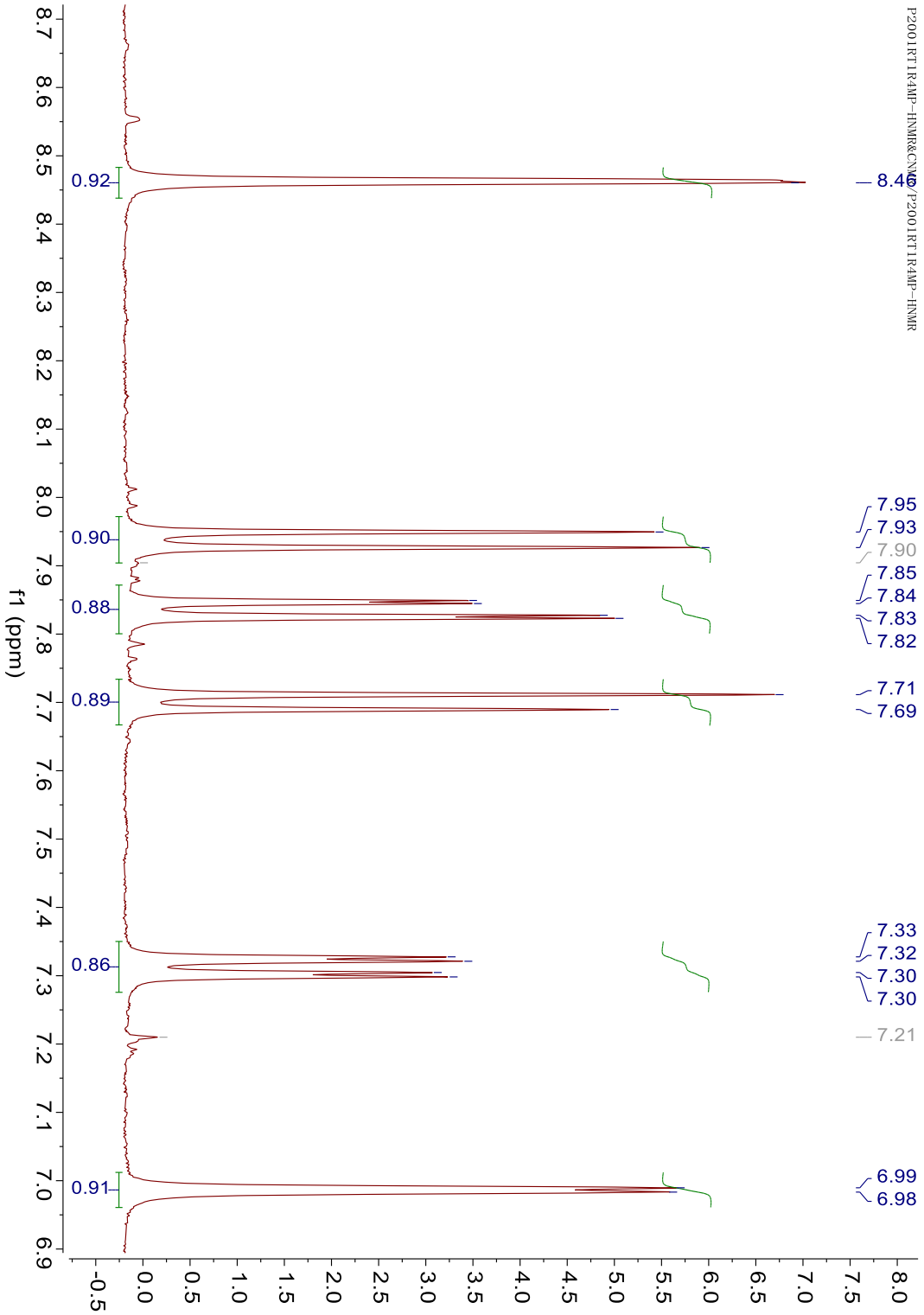


Figure S4. ^{1}H NMR spectra for compound 5 in DMSO-d_6




P2001RT1R4MP-HNMR&CNMR/P2001RT1R4MP-HNMR
f1 (ppm)
0.92
0.90
0.88
0.89
0.86
0.91
8.46
7.95
7.93
7.90
7.85
7.84
7.83
7.82
7.71
7.69
7.33
7.32
7.30
7.30
7.21
6.99
6.98

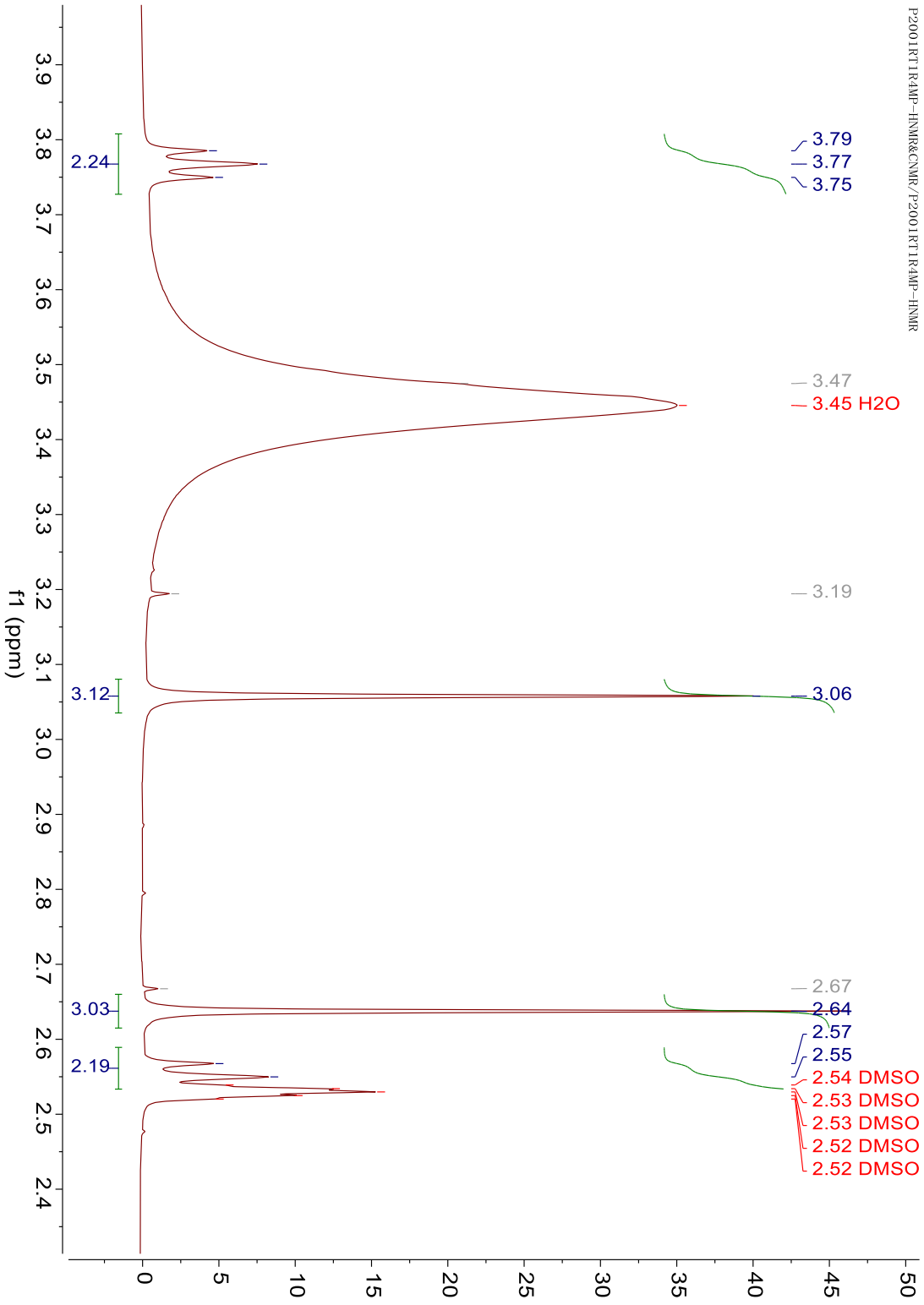

P2001RT1R4MP-HNMR&CNMR/P2001RT1R4MP-HNMR
f1 (ppm)
2.24
3.12
3.03
2.19
3.79
3.77
3.75
3.47
3.45 H2O
3.19
3.06
2.67
2.64
2.57
2.55
2.54 DMSO
2.53 DMSO
2.53 DMSO
2.52 DMSO
2.52 DMSO

Figure S5. ^{13}C NMR spectrum for compound 5 in DMSO-d_6

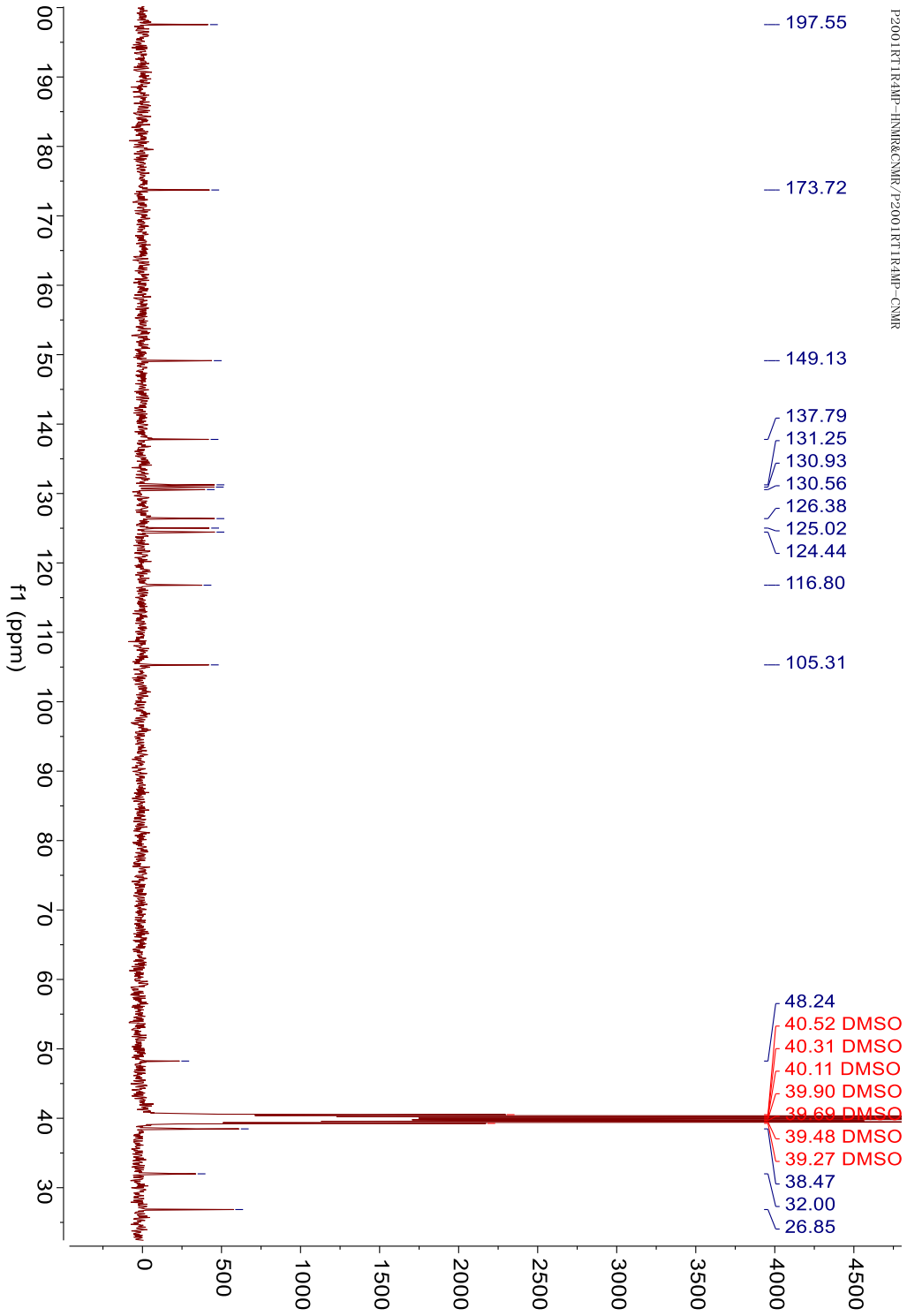
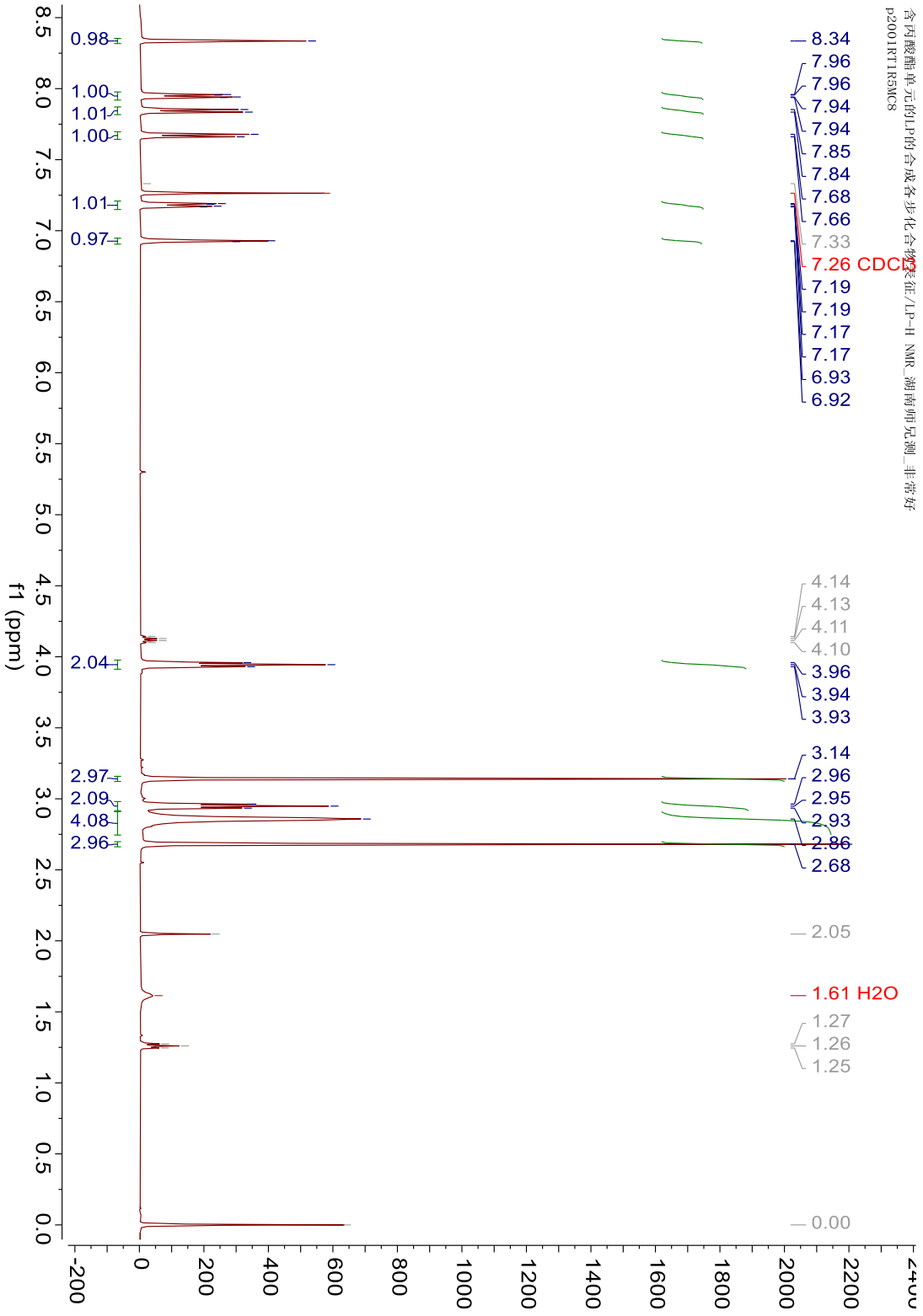
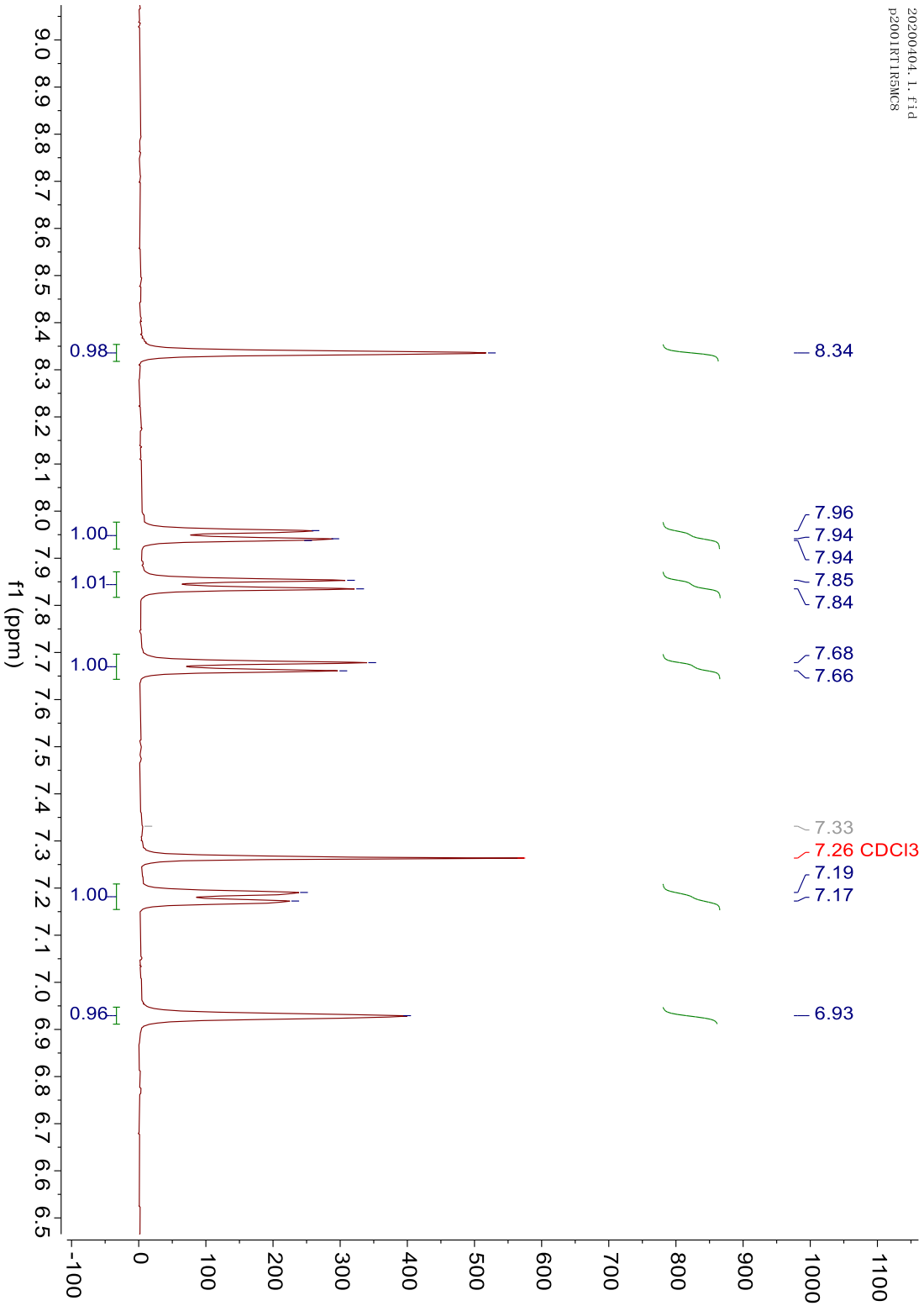


Figure S6. ^{1}H NMR spectra for compound LP in $CHCl_3$-*d*




20200404_1.fid
p2001RT1R5MC8
f1 (ppm)
0.98
1.00
1.01
1.00
1.00
0.96
8.34
7.96
7.94
7.94
7.85
7.84
7.68
7.66
7.33
7.26 CDCl3
7.19
7.17
6.93

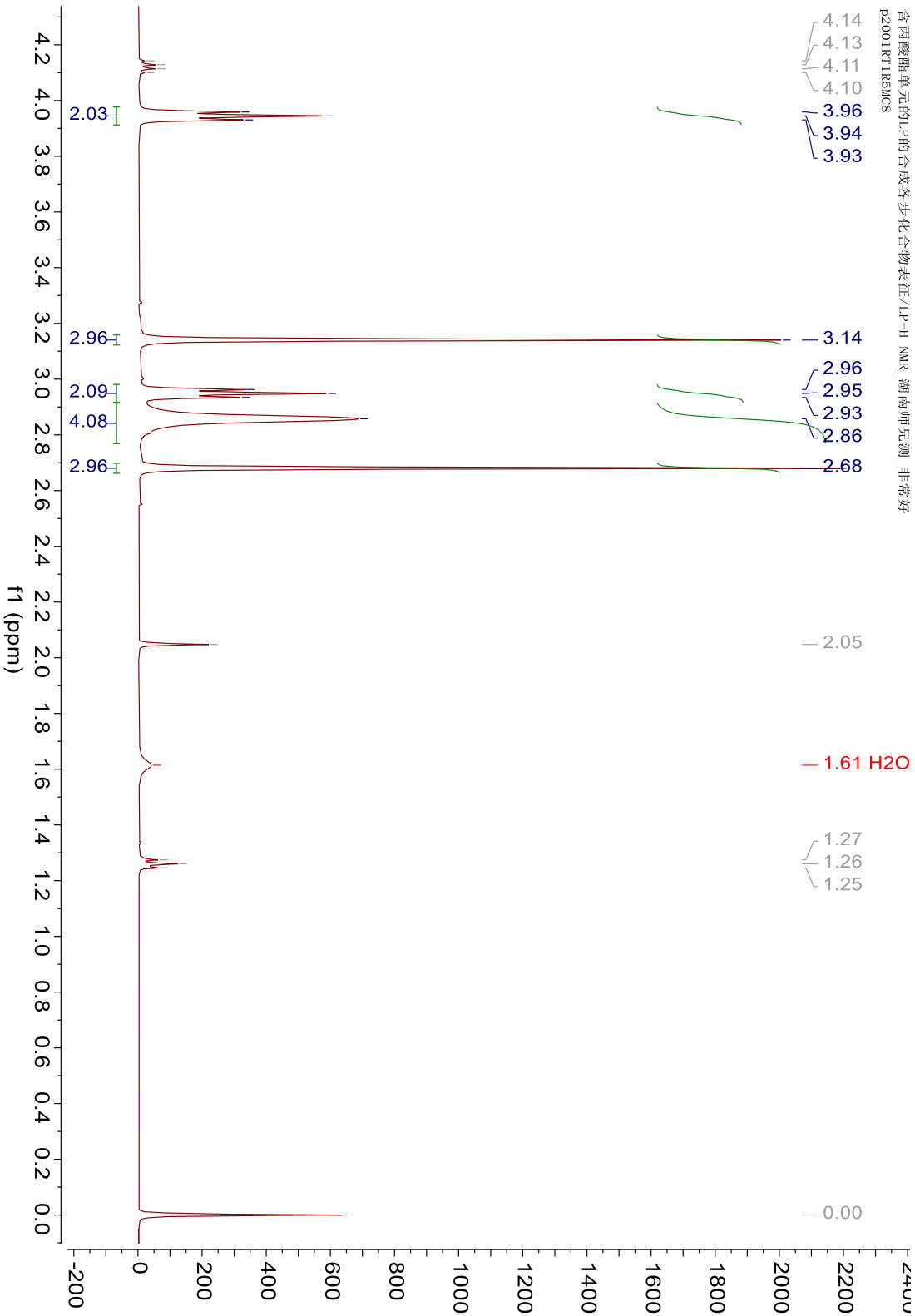

含丙酸酯单元的LP的合成各步化合物表征/LP-1 NMR_湖南师兄测_非常好
p2001RT1R5MC8
4.14
4.13
4.11
4.10
3.96
3.94
3.93
3.14
2.96
2.95
2.93
2.86
2.68
2.05
1.61 H2O
1.27
1.26
1.25
0.00
2.03
2.96
2.09
4.08
2.96
f1 (ppm)

Figure S7. ^{13}C NMR spectrum for compound LP in $CHCl_3$-*d*