Supporting Information

Synthesis and preliminary studies of ^{11}C-labeled tetrahydro-1,7-naphthyridine-2-carboxamides for PET imaging of metabotropic glutamate receptor 2

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[†]These two authors contributed equally to this work.

Contents

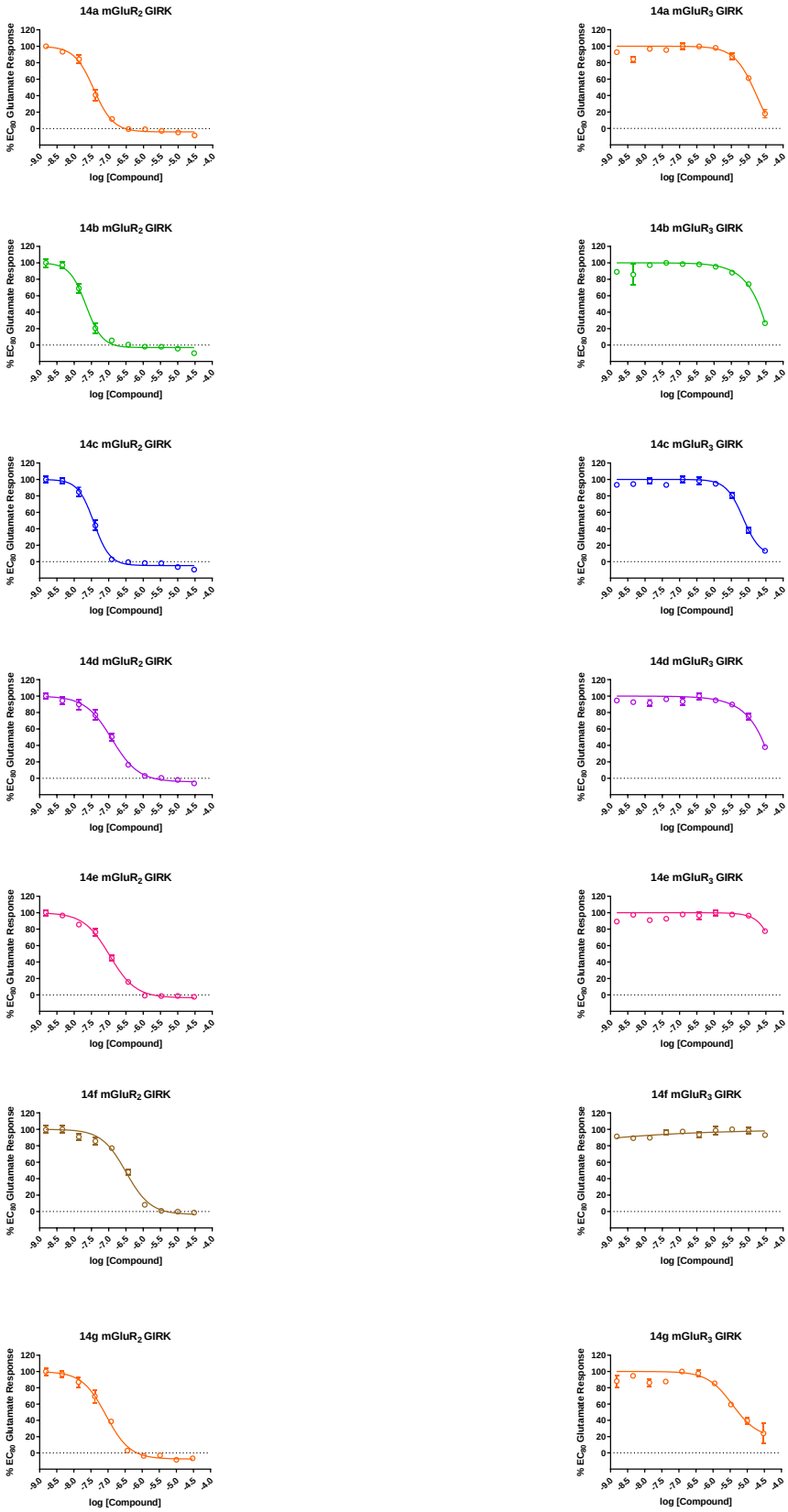
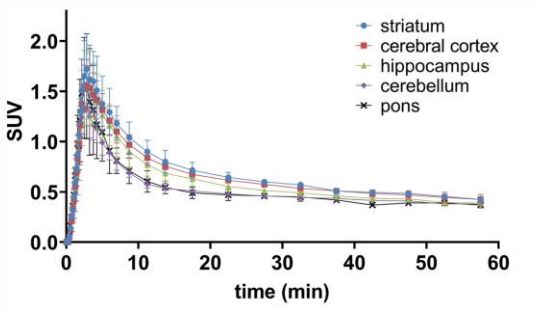


Figure S1. Concentration−response curves of $mGlu_2$ NAMs compounds **14a-14g** in $mGlu_2$ GIRK or $mGlu_3$ GIRK functional assays.

A. Baseline studies of [^{11}C]14a



B. Baseline studies with pretreatment of 14a

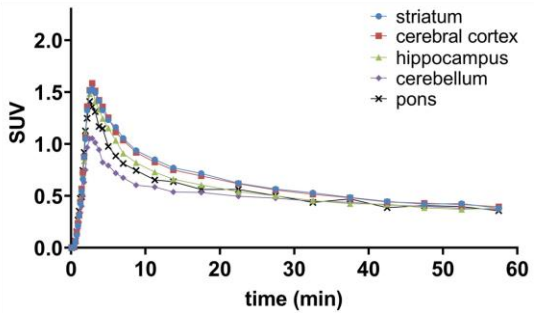


Figure S2. Time-activity curves of [^{11}C]**14a** in rat brains. [a]Blocking conditions: **14a** (1 mg/kg), 30 min *i.v.* before radioligand injection.

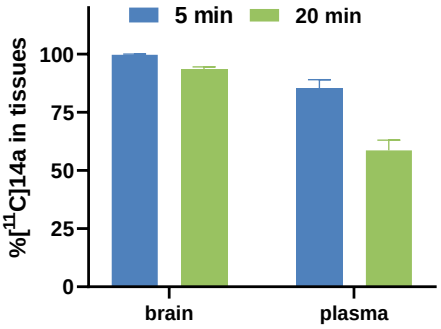


Figure S3. Radiometabolite analysis of [^{11}C]**14a** in rat brain (average two runs)

Table S1. Radiometabolite and parent (unchanged) fraction of [^{11}C]**14a** in rat brain and plasma

	Metabolite (%)	Unchanged (%)
Plasma 5 mins−1	11.96	88.04
Plasma 5 mins−2	17.06	82.94
Plasma 20 mins−1	44.51	55.49
Plasma 20 mins−2	38.26	61.74
Brain 5 mins−1	0.06	99.94
Brain 5 mins−2	0.49	99.51
Brain 20 mins−1	7.3	92.7
Brain 20 mins−2	5.76	94.24

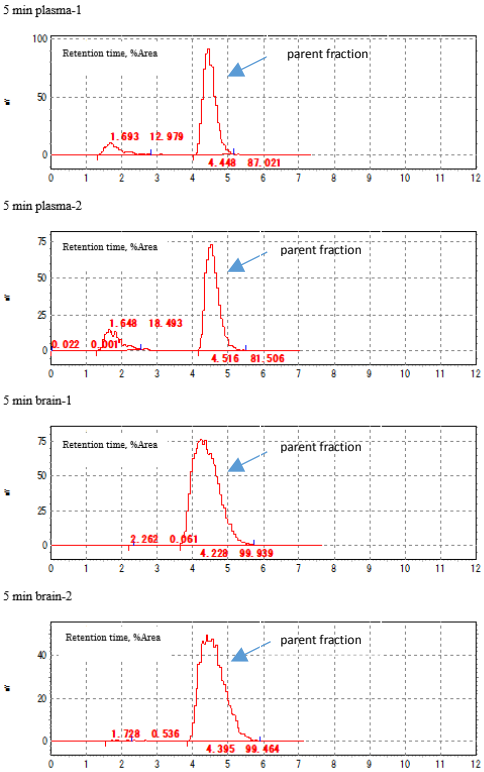


Figure S4. RadioHPLC chromatogram in the brain and plasma 5 min post injection of [^{11}C]**14a**

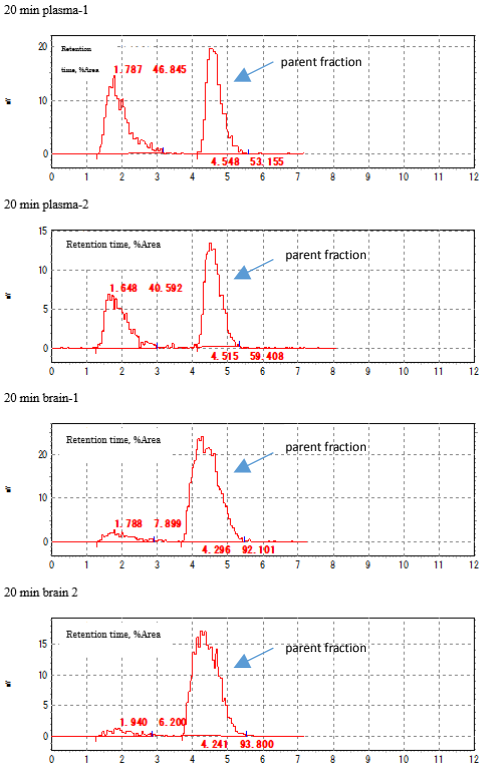


Figure S5. RadioHPLC chromatogram in the brain and plasma 20 min post injection of [^{11}C]**14a**

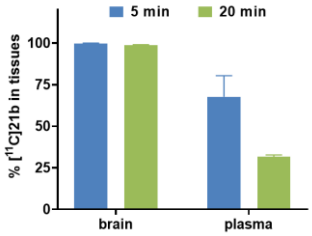
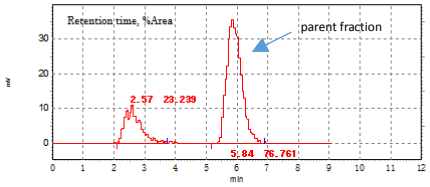


Figure S6. Radiometabolite analysis of [^{11}C]**14b** in rat brain (average two runs)

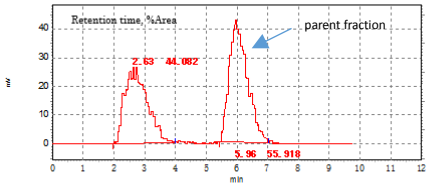
Table S2. Radiometabolite and parent (unchanged) fraction of [^{11}C]**14b** in rat brain and plasma

	Metabolite(%)	Unchanged (%)
Plasma 5 mins−1	21.32	76.68
Plasma 5 mins−2	41.31	58.69
Plasma 20 mins−1	69.22	30.78
Plasma 20 mins−2	67.60	32.40
Brain 5 mins−1	0.28	99.72
Brain 5 mins−2	0.04	99.96
Brain 20 mins−1	0.97	99.03
Brain 20 mins−2	1.22	98.78

5 min plasma-1



5 min plasma-2



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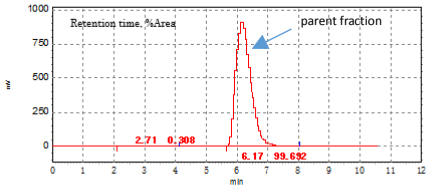

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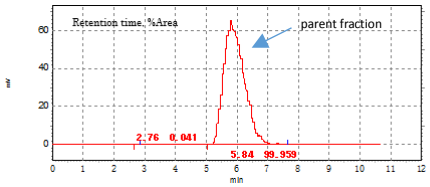


Figure S7. RadioHPLC chromatogram in the brain and plasma 5 min post injection of [^{11}C]**14b**

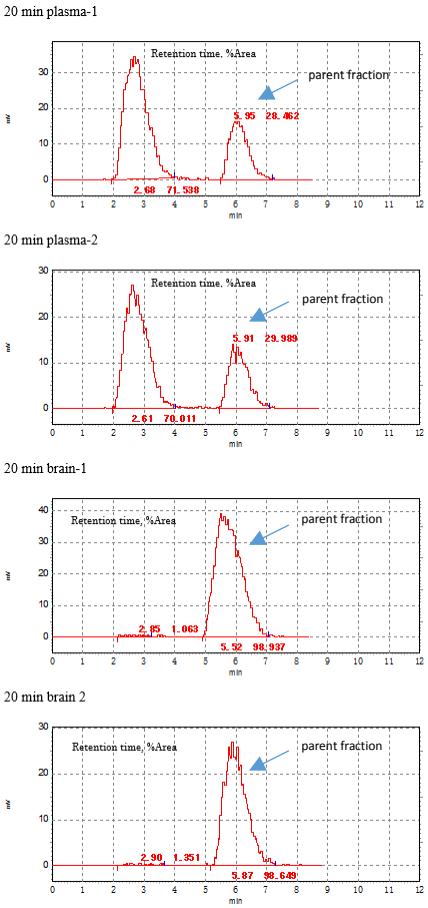


Figure S8. RadioHPLC chromatogram in the brain and plasma 20 min post injection of [^{11}C]**14b**

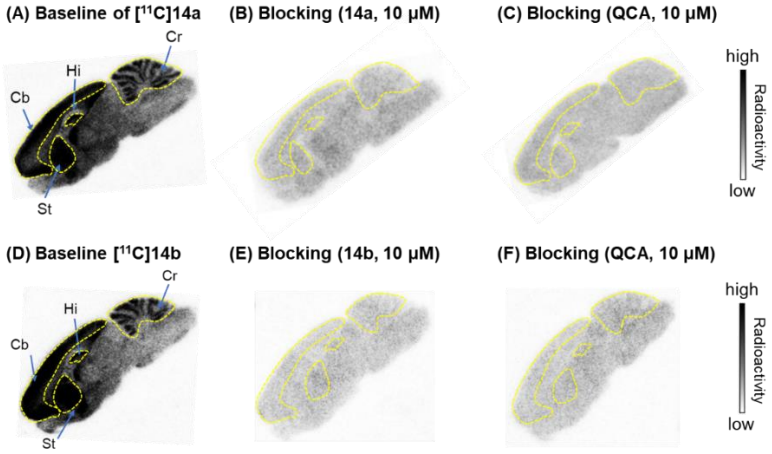


Figure S9. Image of the ROIs used for quantification of in vitro autoradiography. (A) Brain sections were treated with [^{11}C]**14a**; (B) Brain sections were pre-treated with **14a** (10 μM), followed by [^{11}C]**14a**; (C) Brain sections were pre-treated with **QCA** (10 μM), followed by [^{11}C]**14a**; (A) Brain sections were treated with [^{11}C]**14b**; (B) Brain sections were pre-treated with **14b** (10 μM), followed by [^{11}C]**14b**; (C) Brain sections were pre-treated with **QCA** (10 μM), followed by [^{11}C]**14b**.

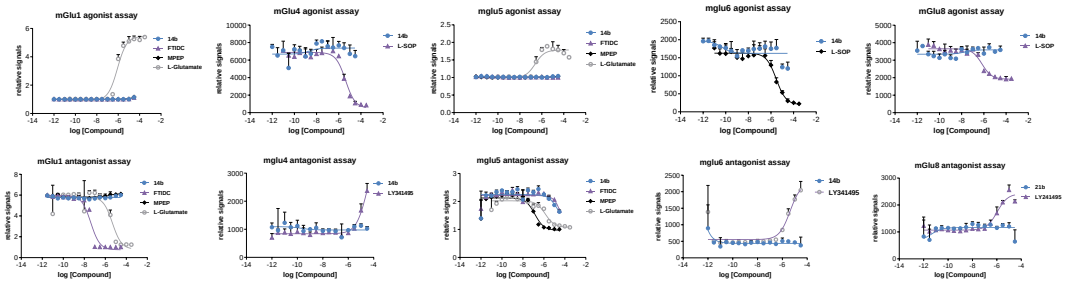


Figure S10. Functional (agonist and antagonist) assays of compound **14b** towards mGlu receptors, including mGlu$_1$, mGlu$_4$, mGlu$_5$, mGlu$_6$ and mGlu$_8$.

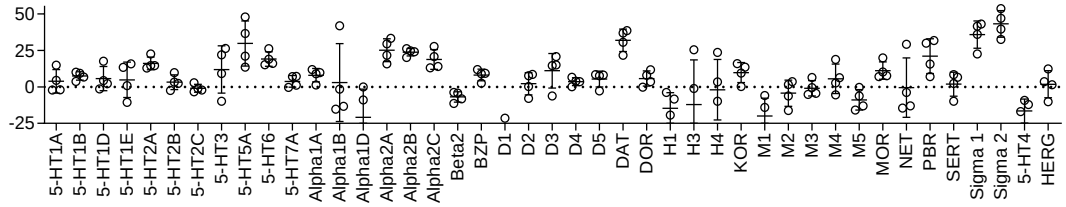


Figure S11. Off-target pharmacological evaluation of compound **14b** at a concentration of 10 μM against major CNS targets, including common GPCRs, enzymes, ion channels and transporters: Initial screening at a concentration of 10 μM. All data are mean ± SD (n = 4). No significant off-target binding (>50%) was observed at 10 μM compound testing concentration.

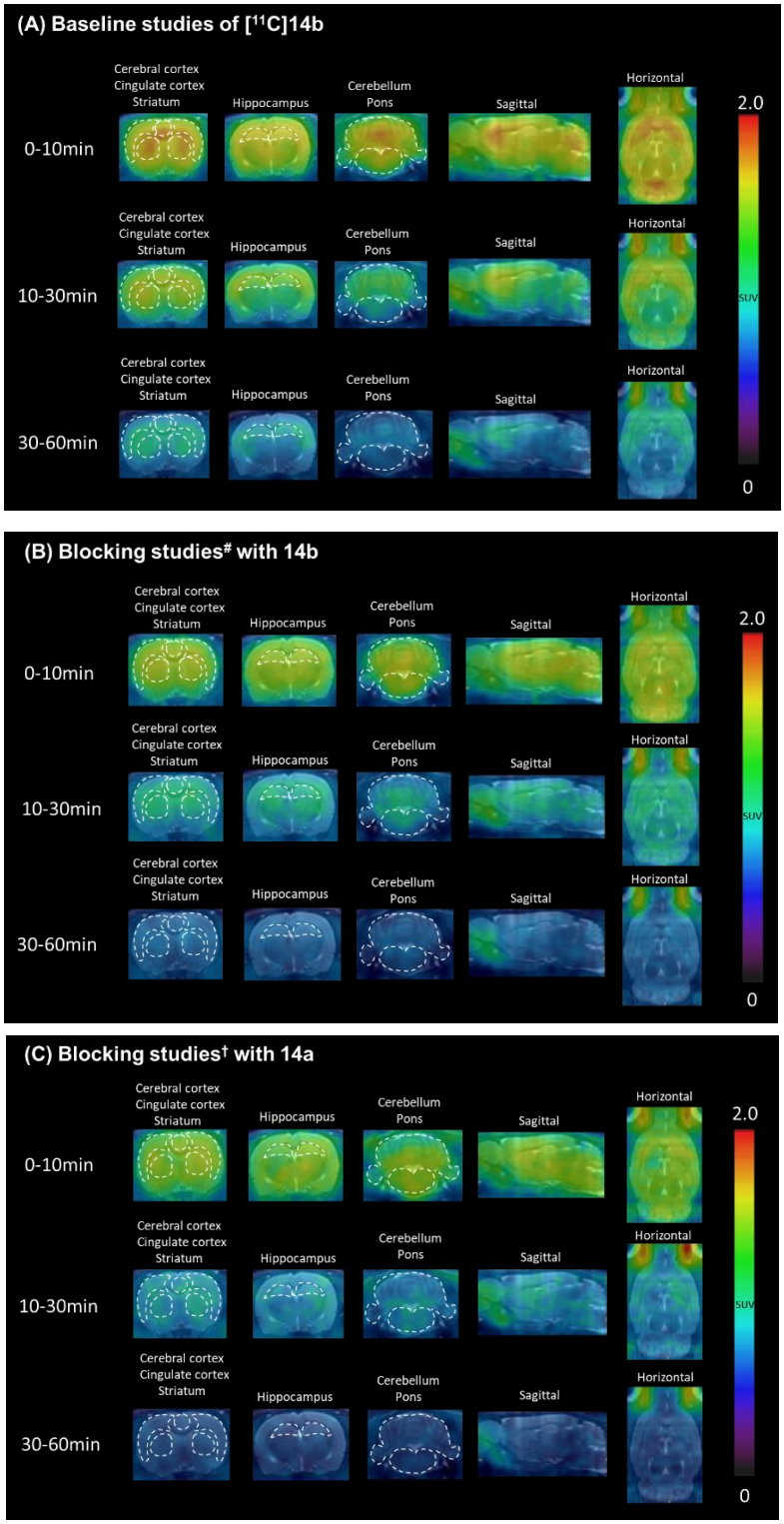
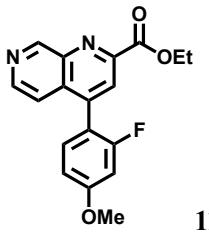


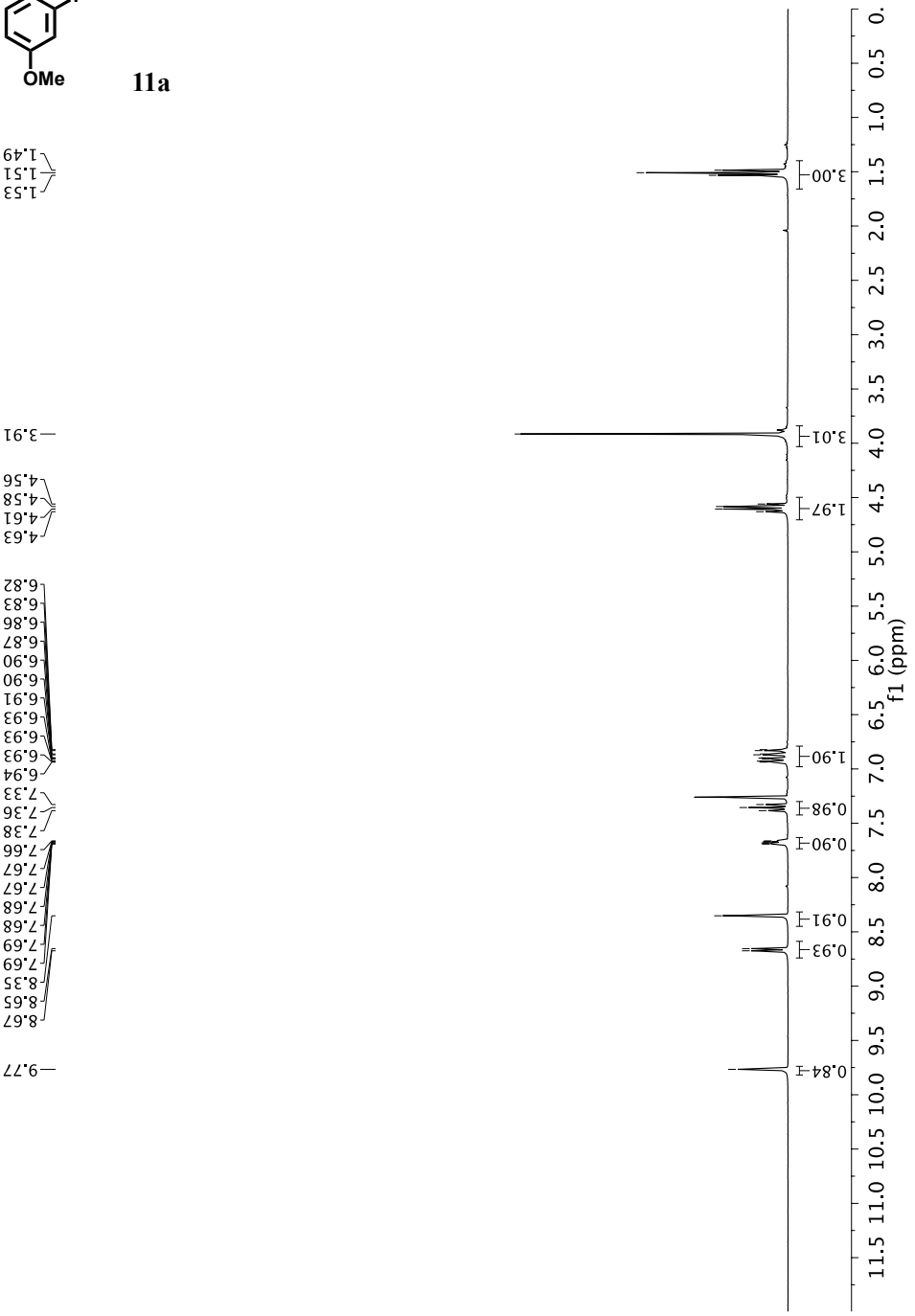
Figure S12. Representative PET/MRI fused images (summed at 0-10 min, 10-30 min and 30-60 min) and time-activity curves of [^{11}C]**14b** under baseline and blocking conditions in SD rat brain. #Blocking conditions: **14b** (1 mg/kg), 30 min *i.v.* before radioligand injection; †Blocking conditions: **14a** (3 mg/kg), 30 min *i.v.* before radioligand injection. Data are presented as mean ± SEM (n = 3).

NMR spectra of synthesized compounds

^{1}H spectrum of 11a



11a



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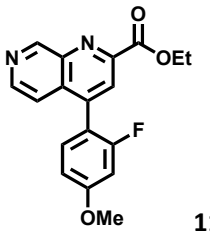
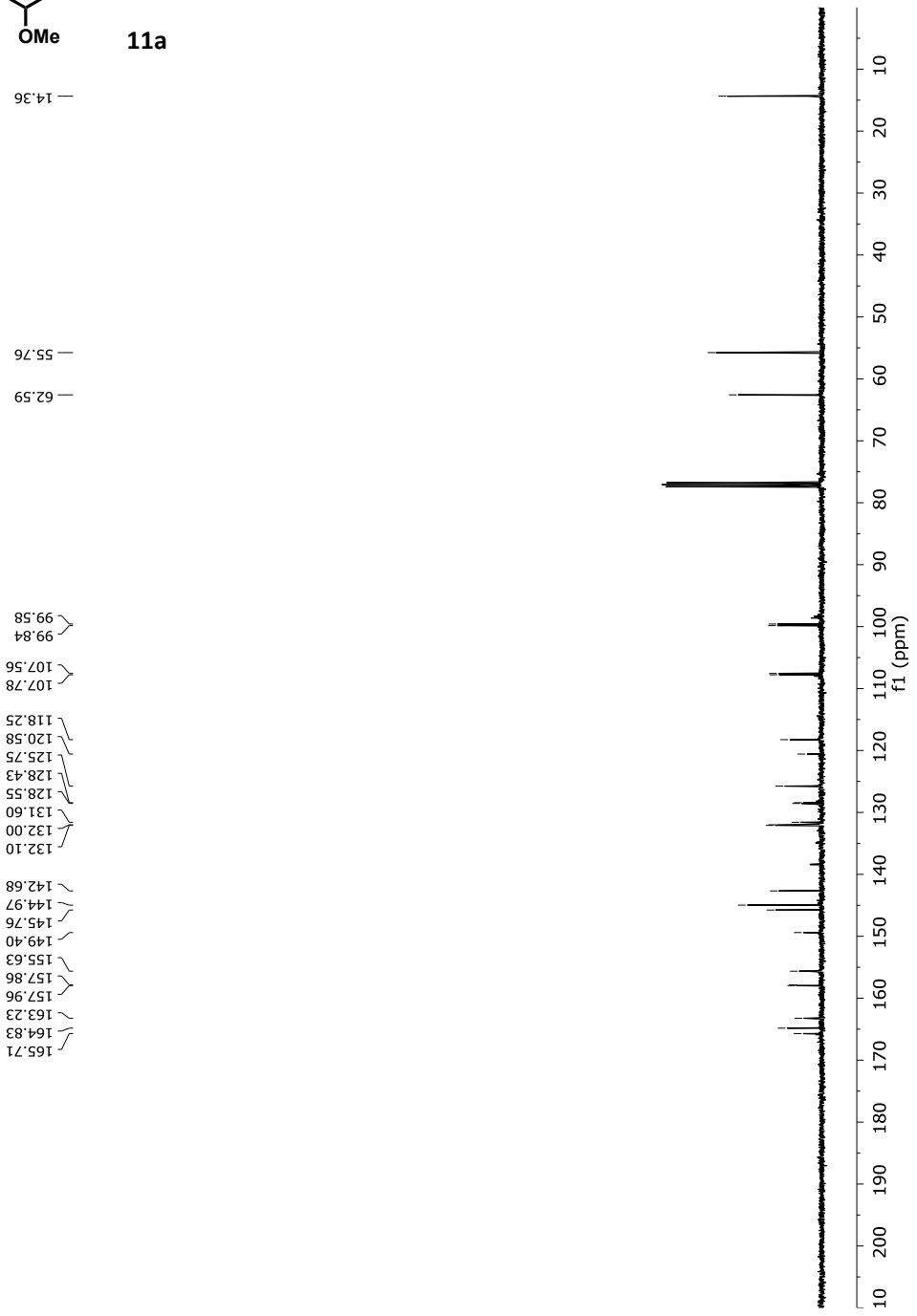


11a



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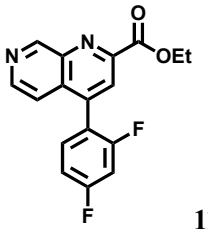

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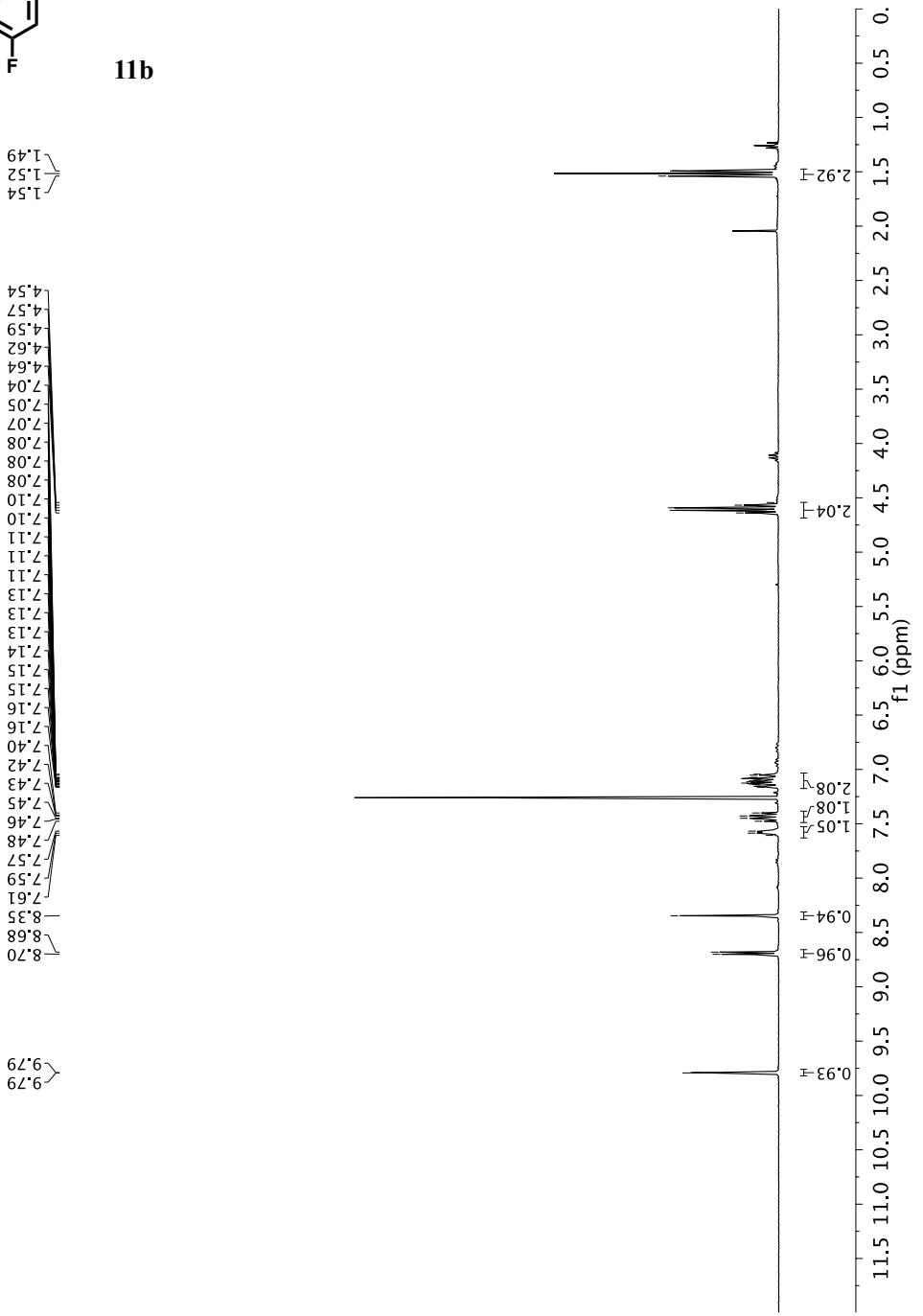

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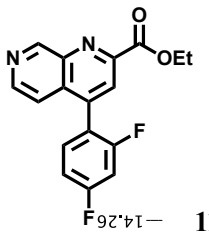

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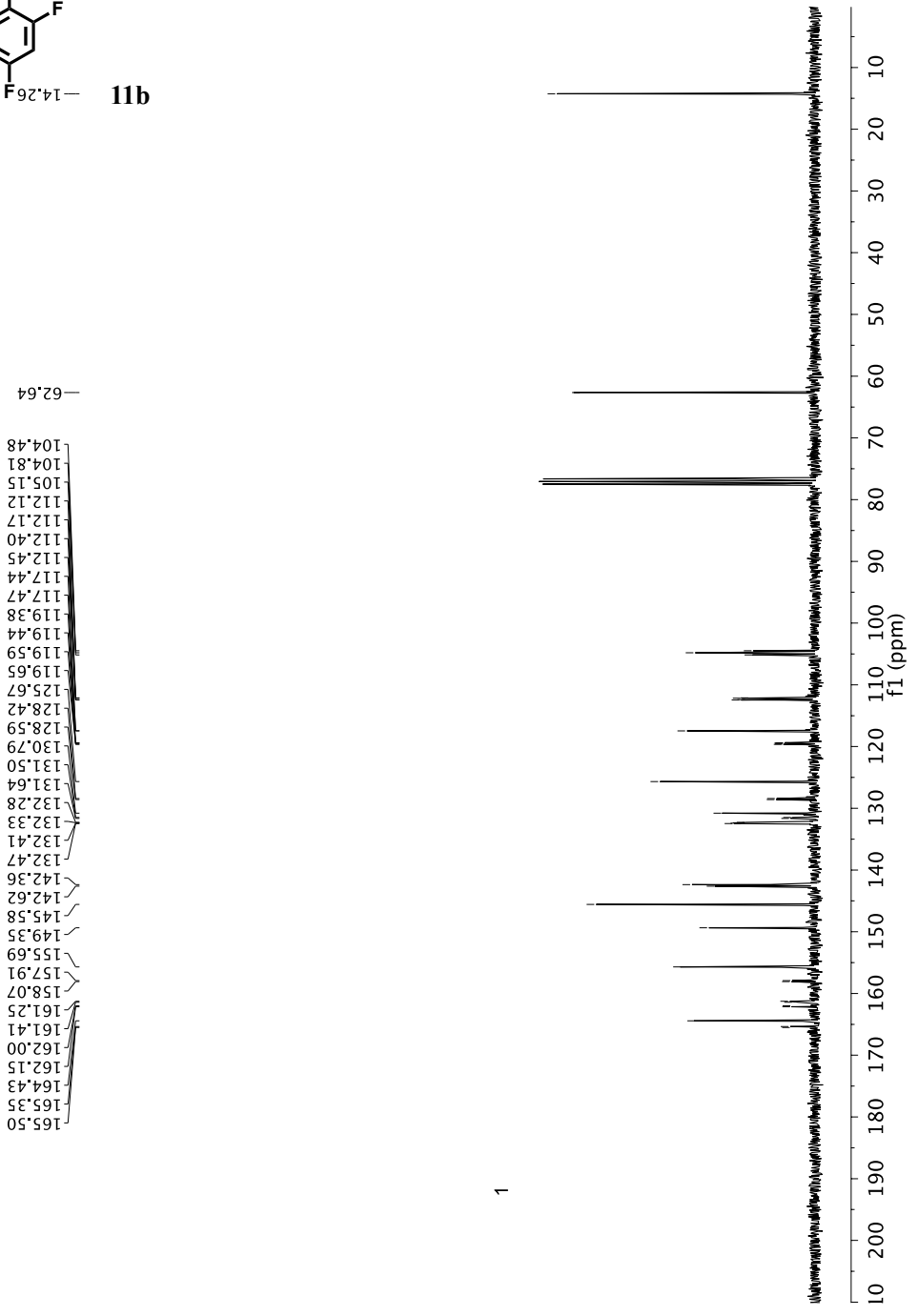

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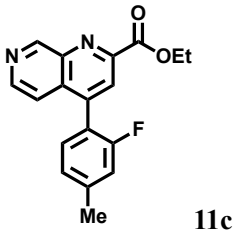

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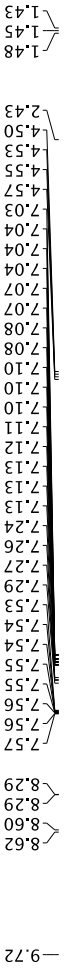

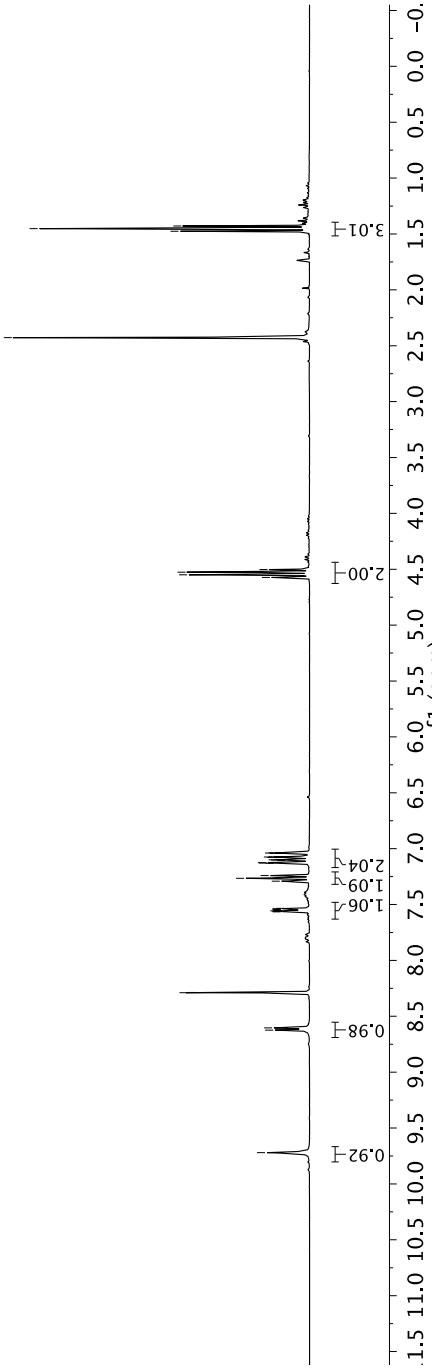

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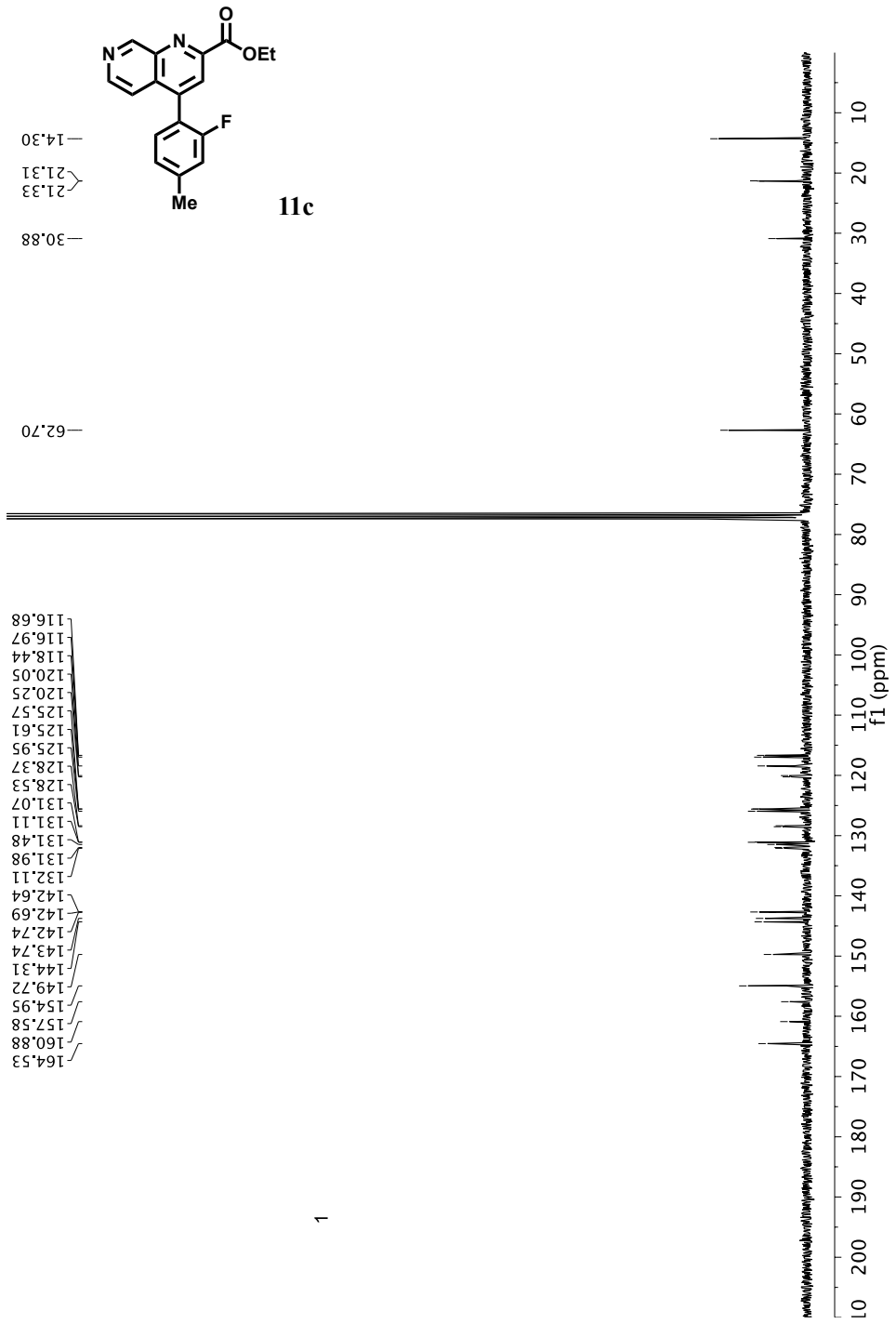

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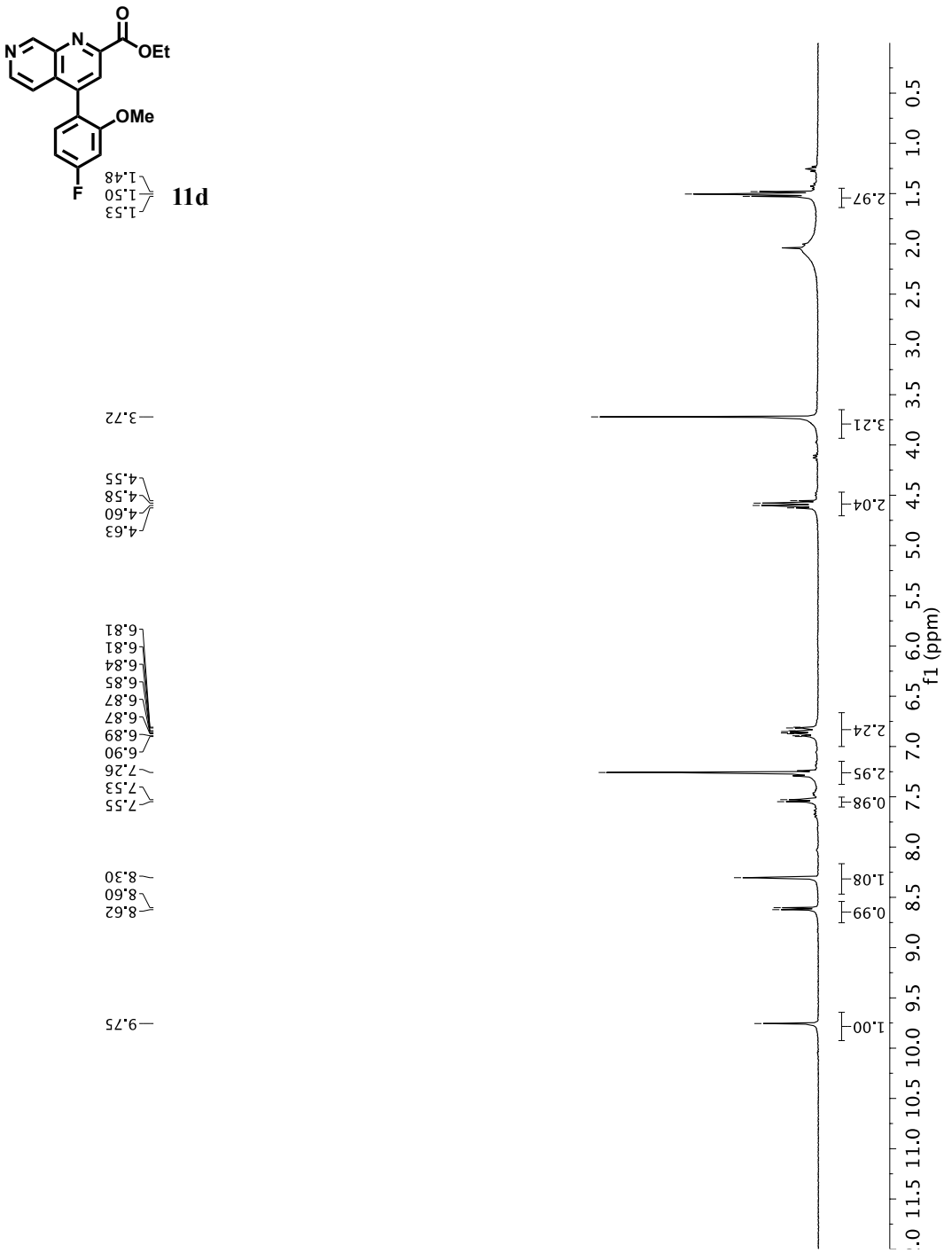

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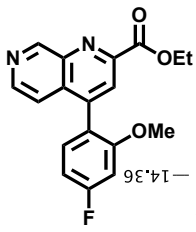

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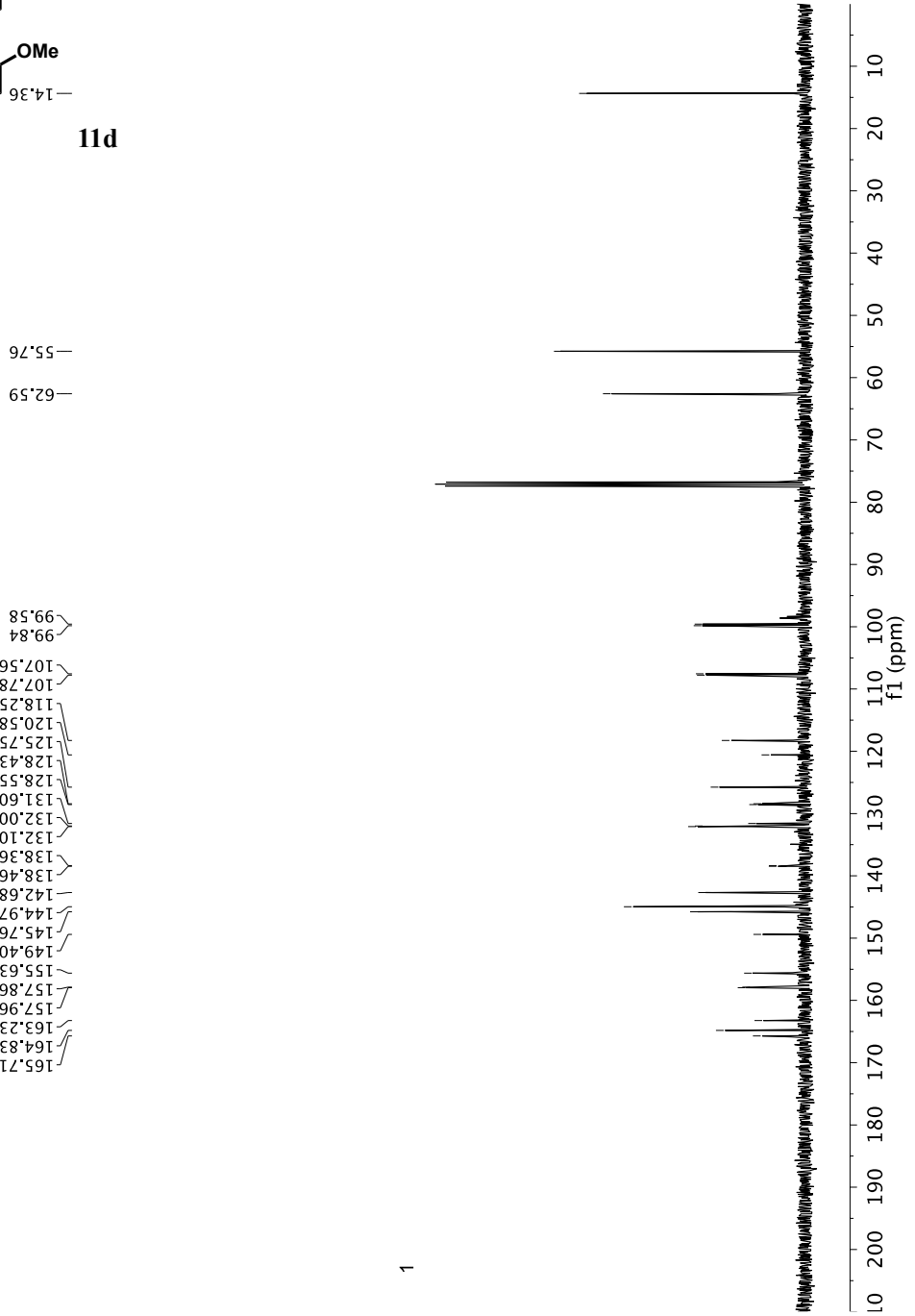

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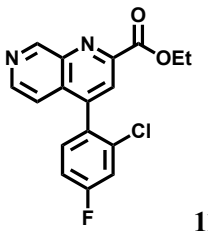

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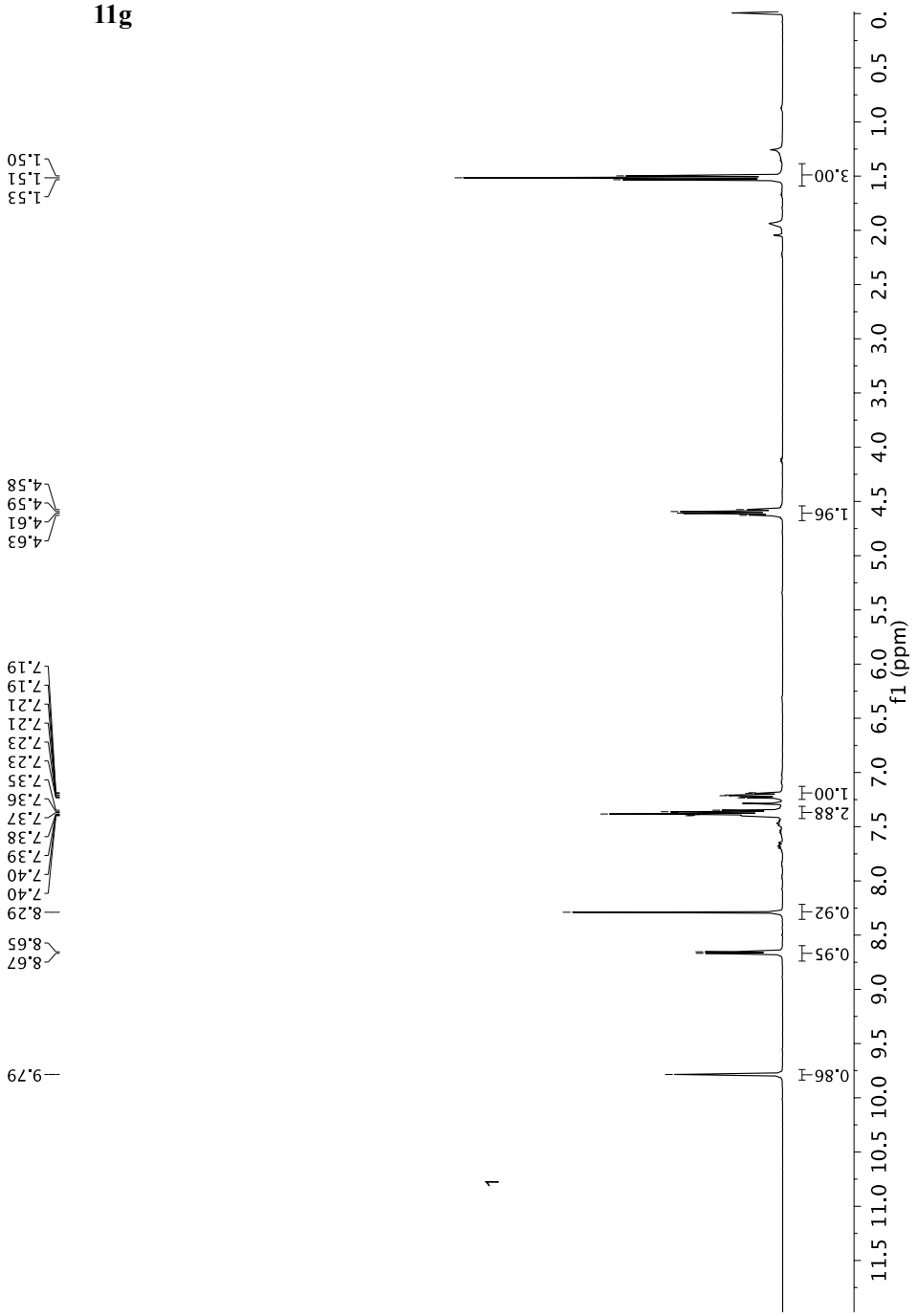

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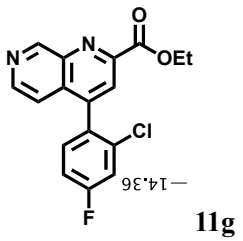

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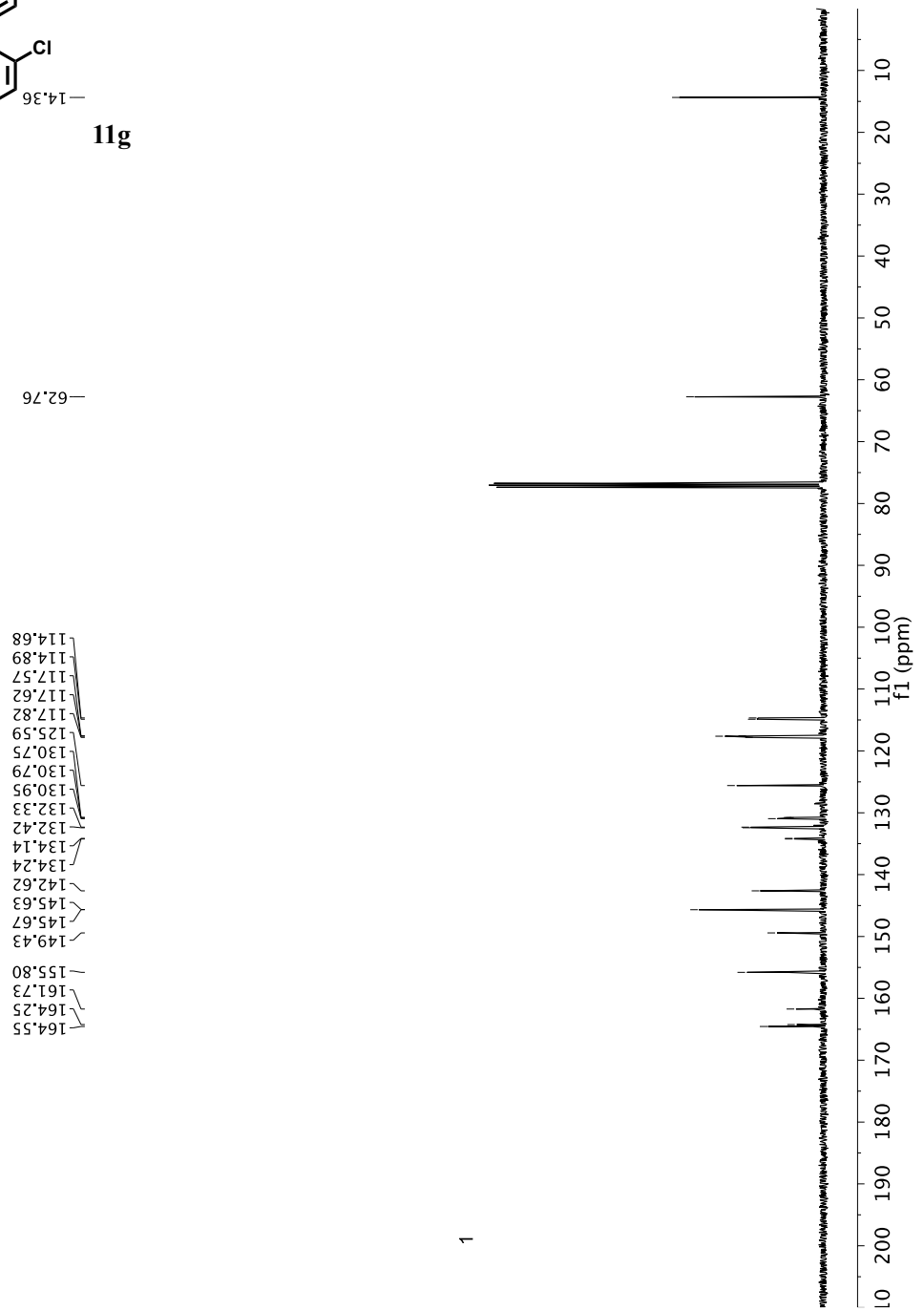

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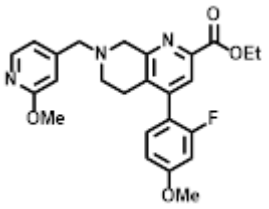

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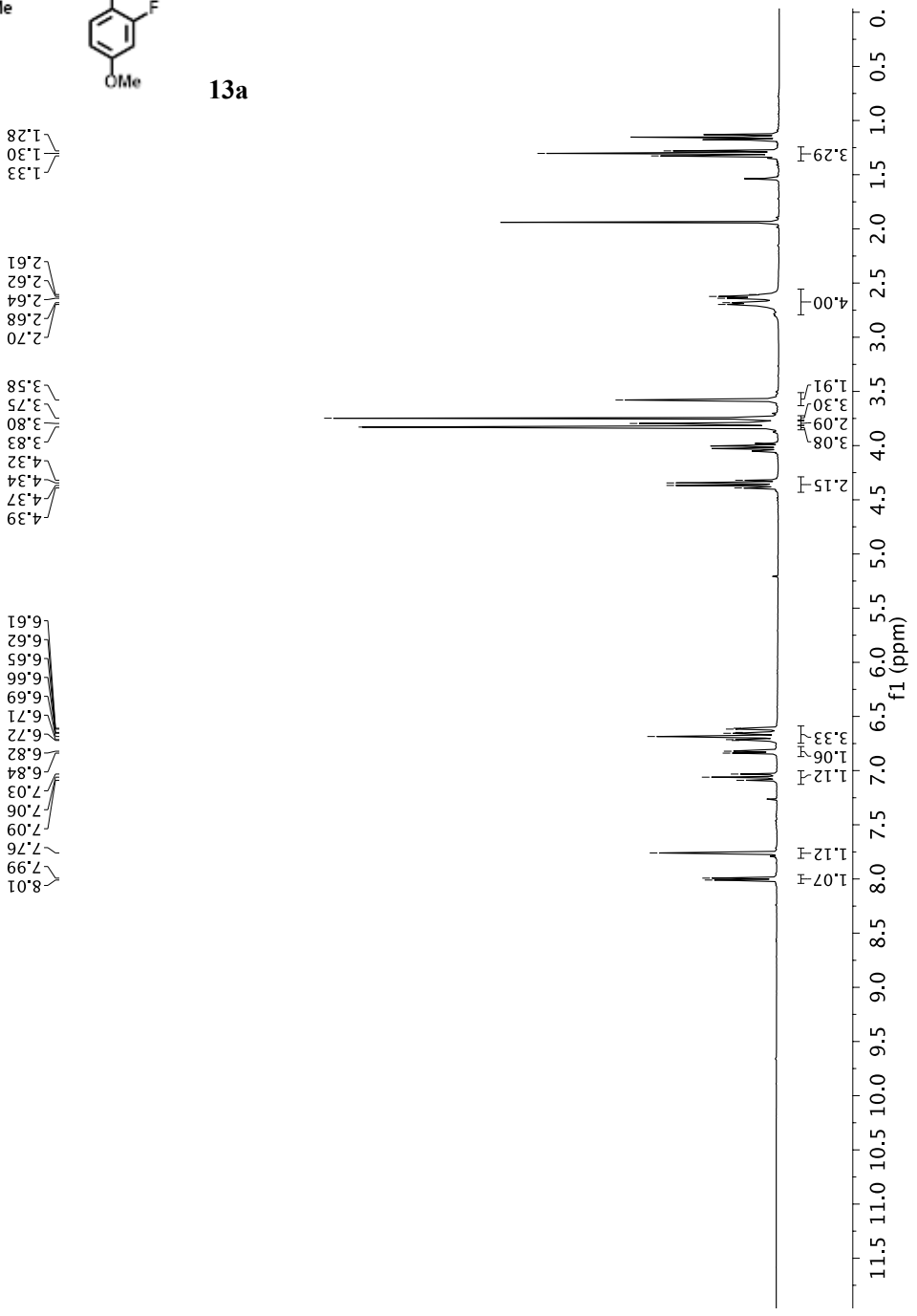

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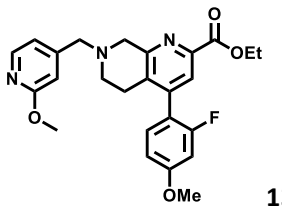

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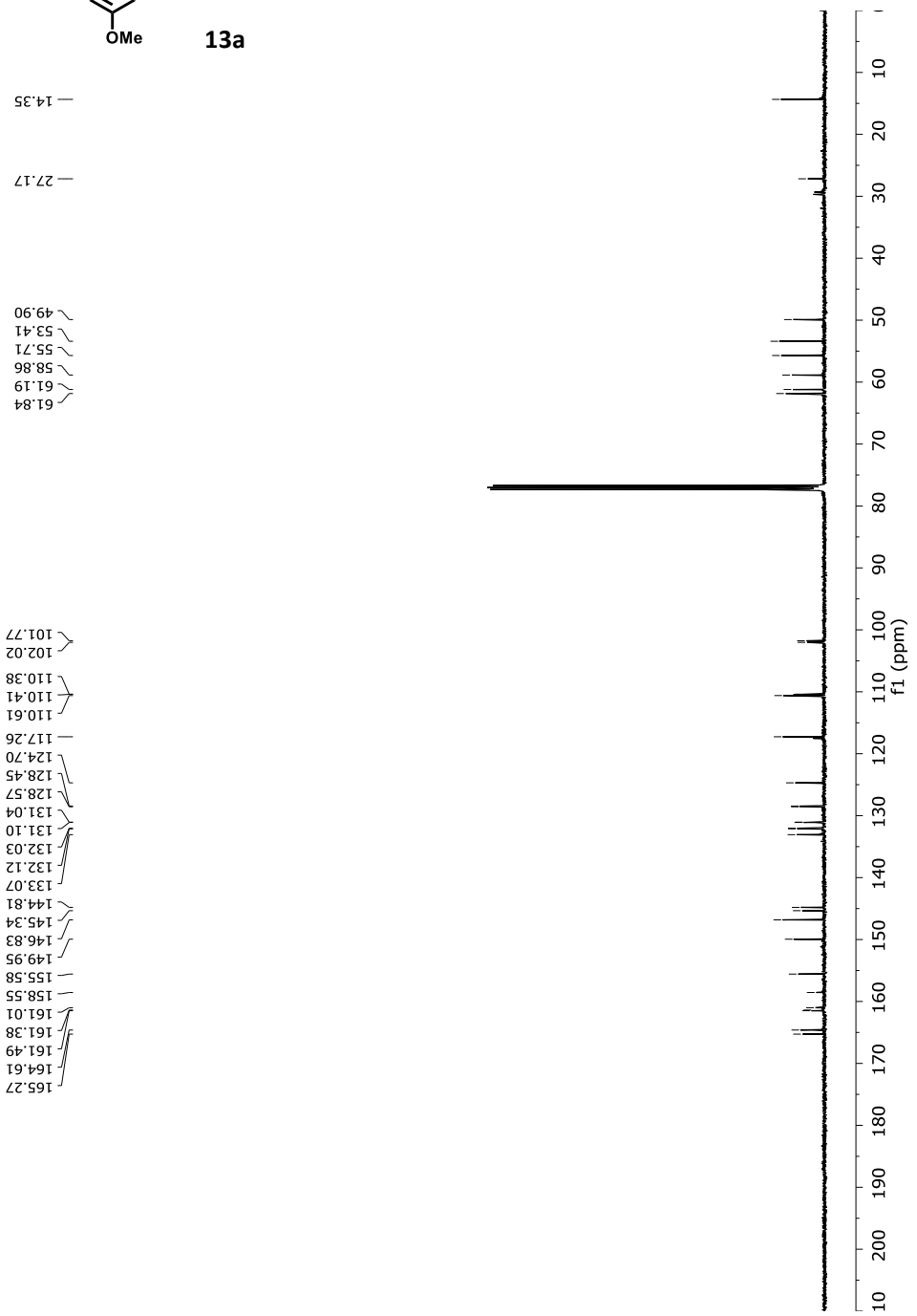

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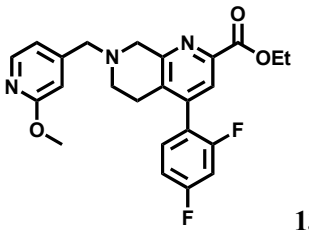

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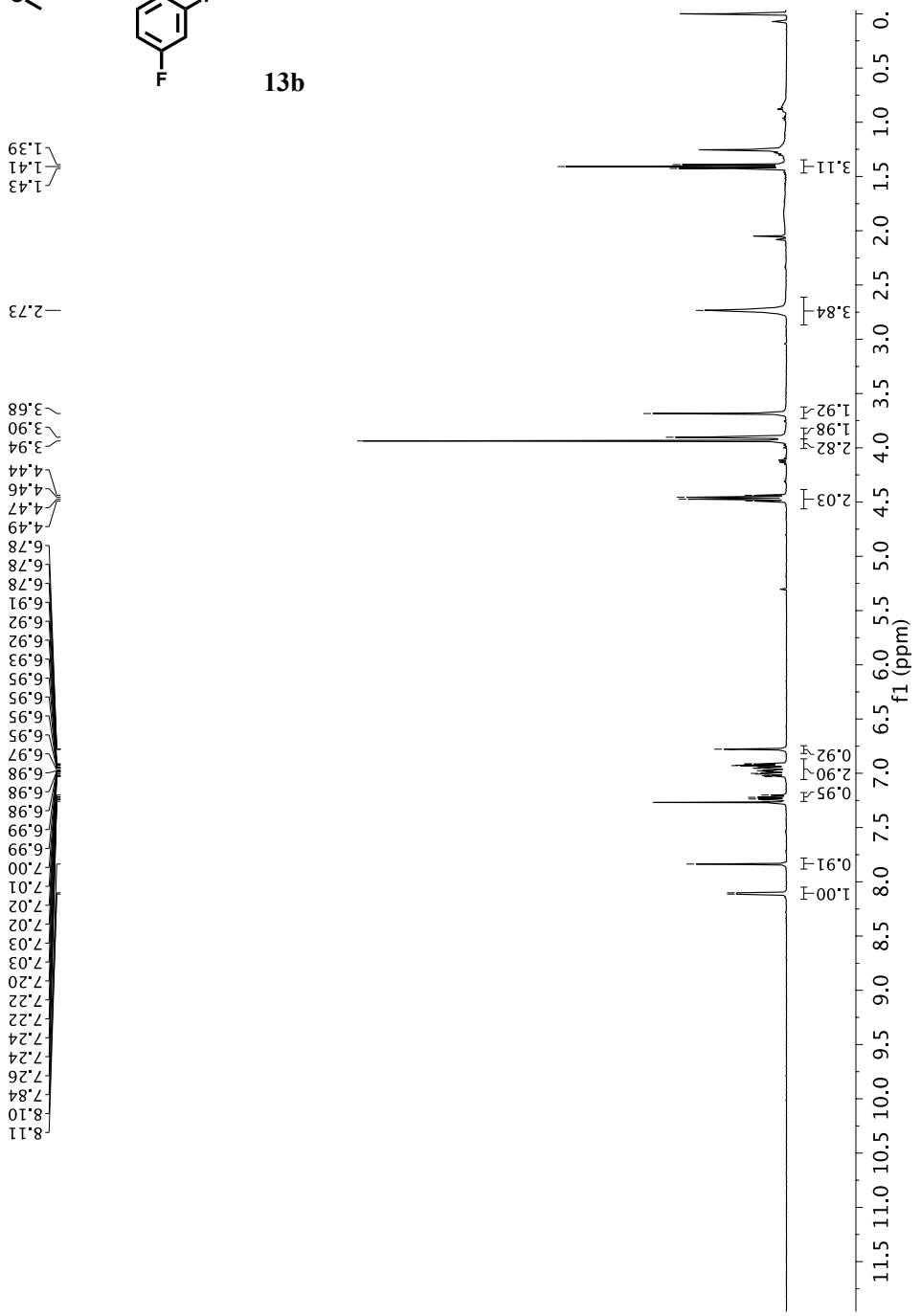

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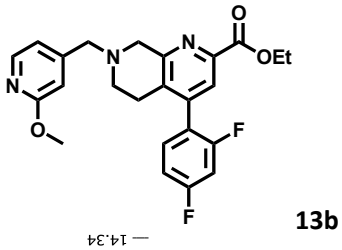

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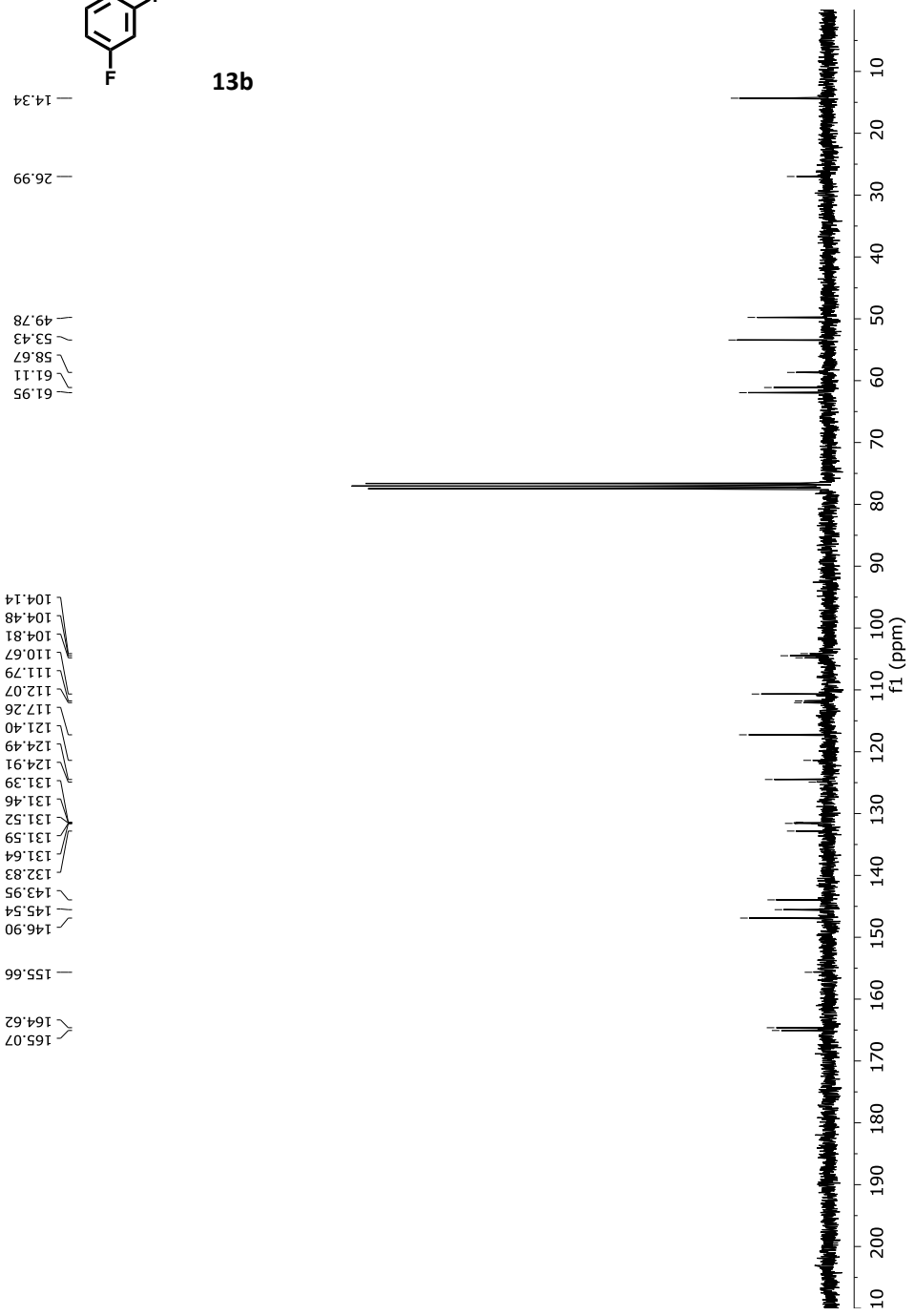

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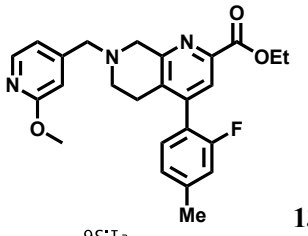

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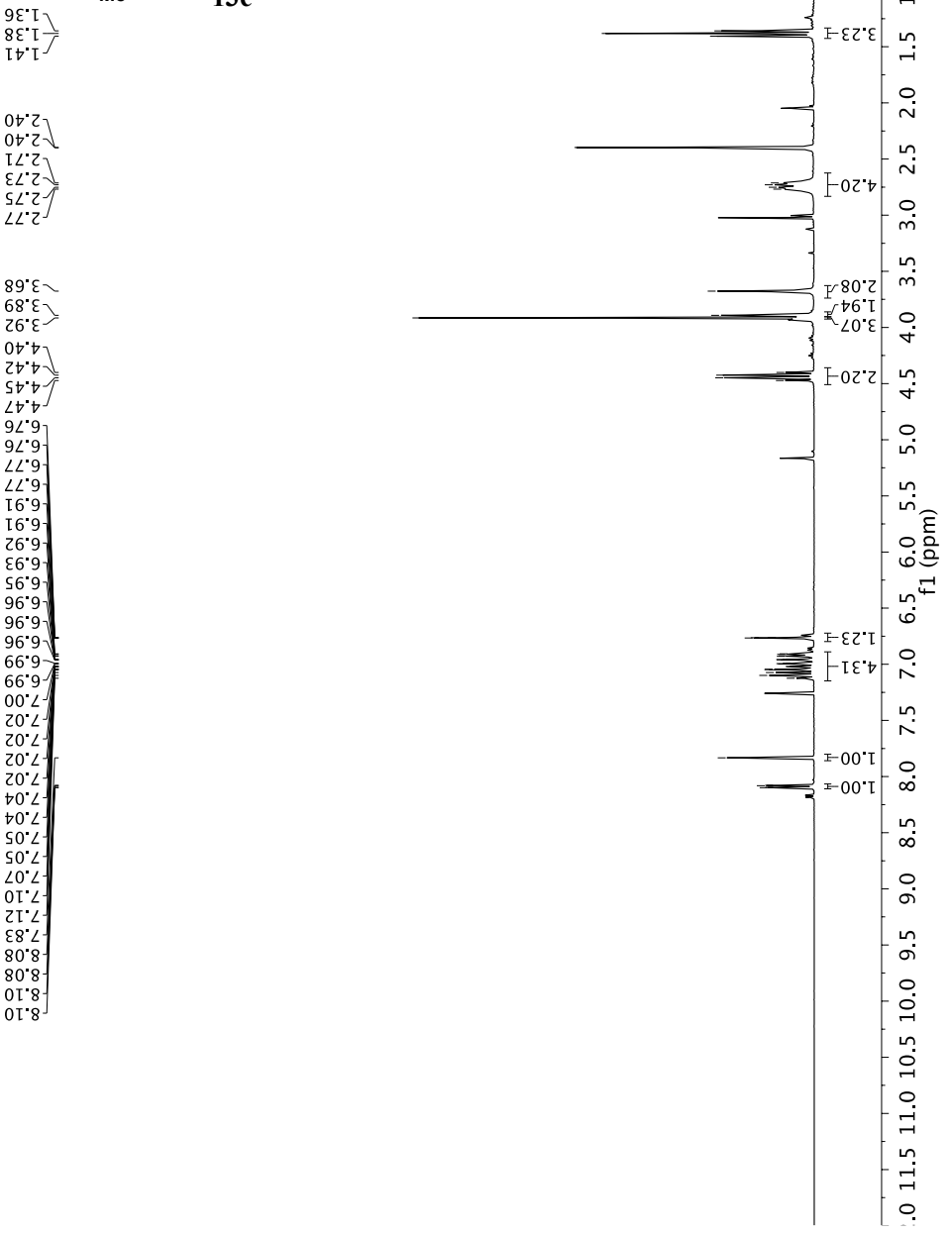

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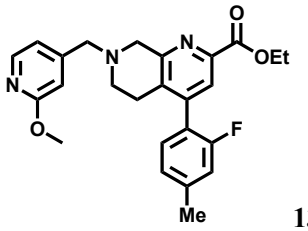

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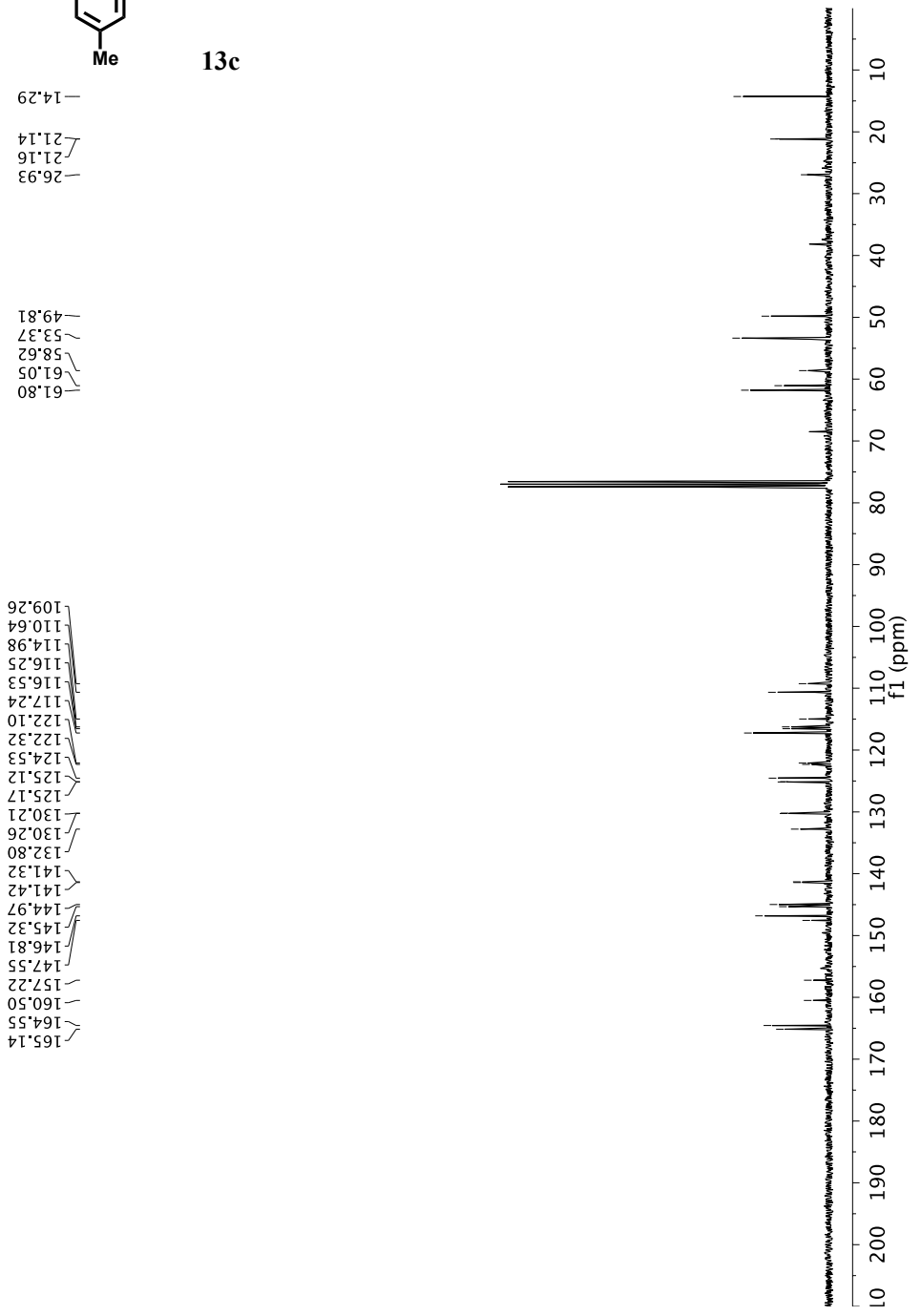

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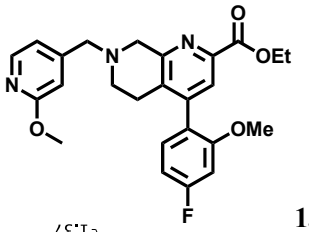
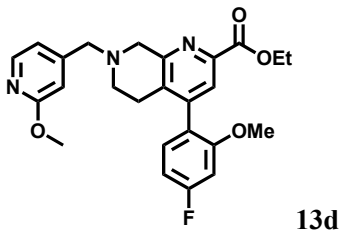


13d

^{13}C spectrum of 13d



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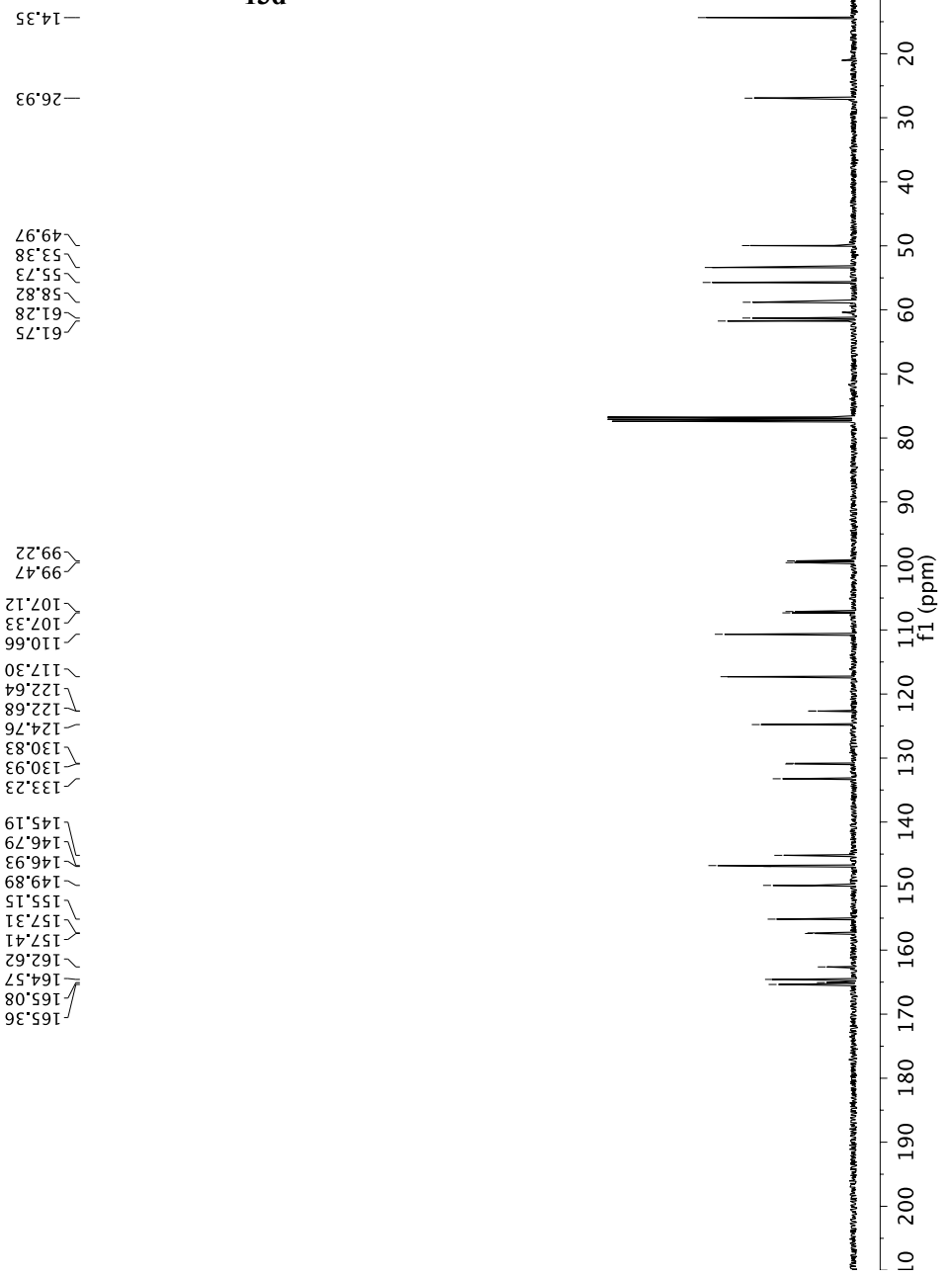
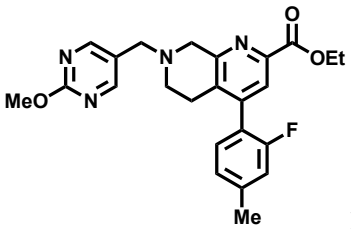


^{1}H spectrum of 13e



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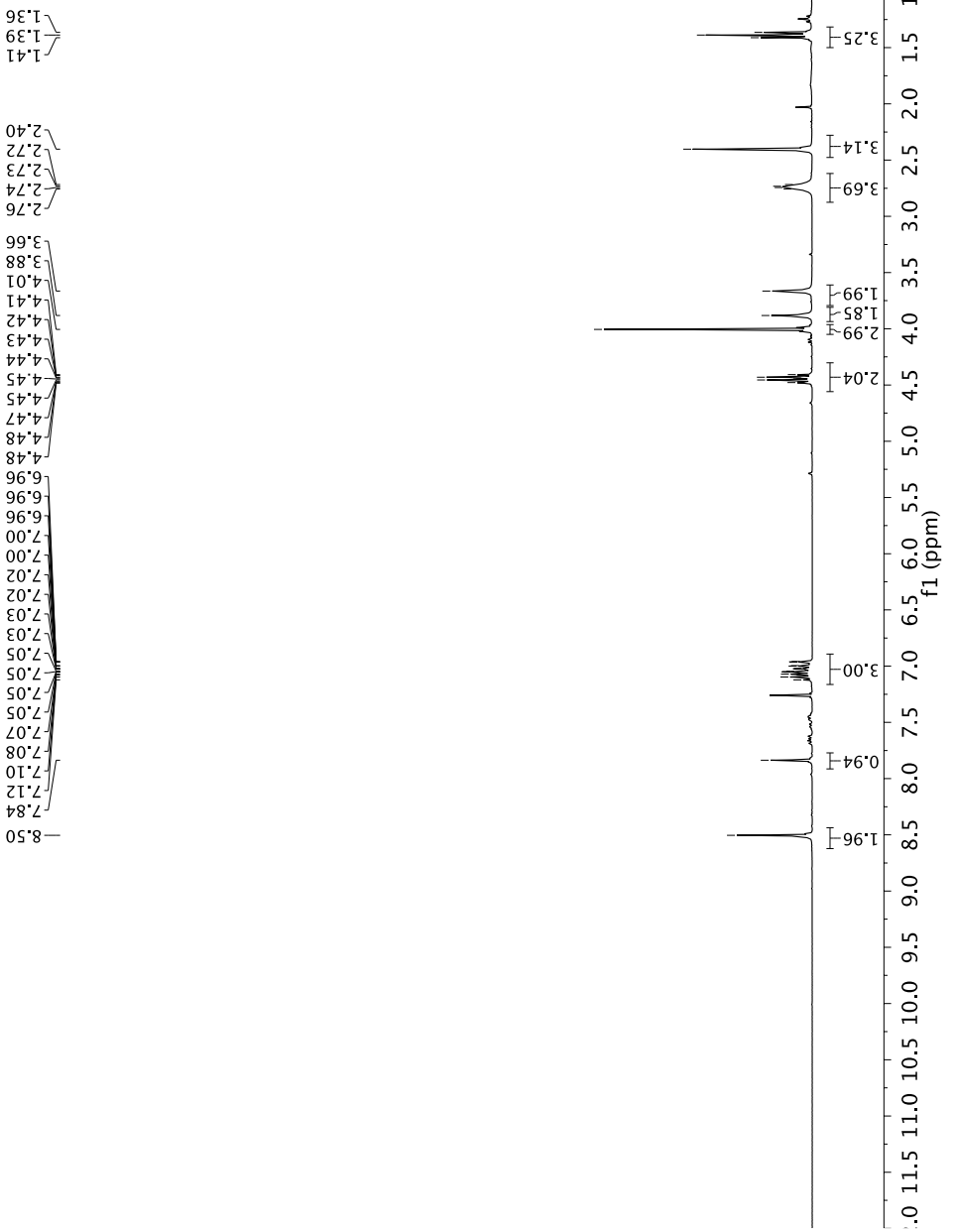
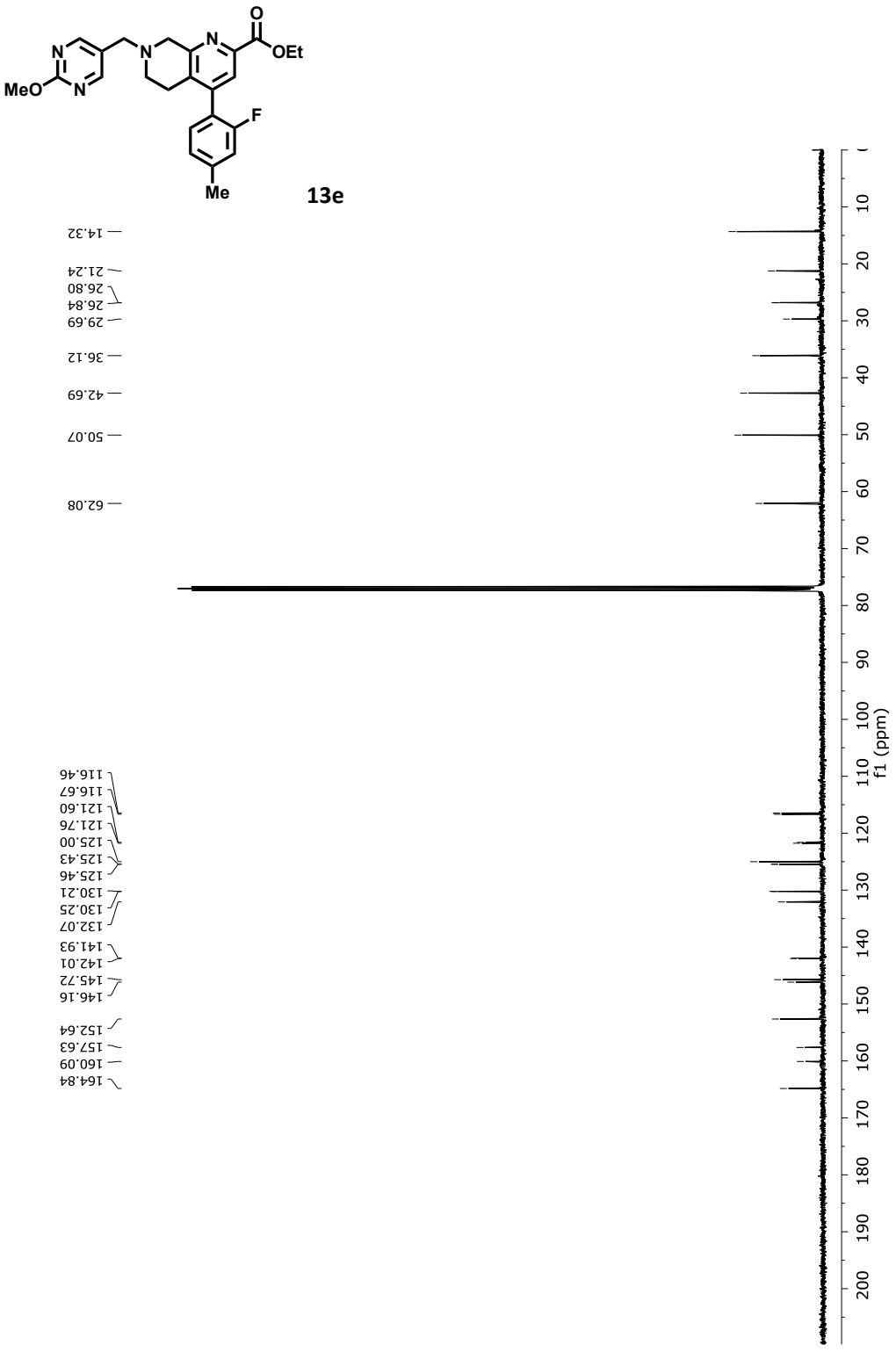
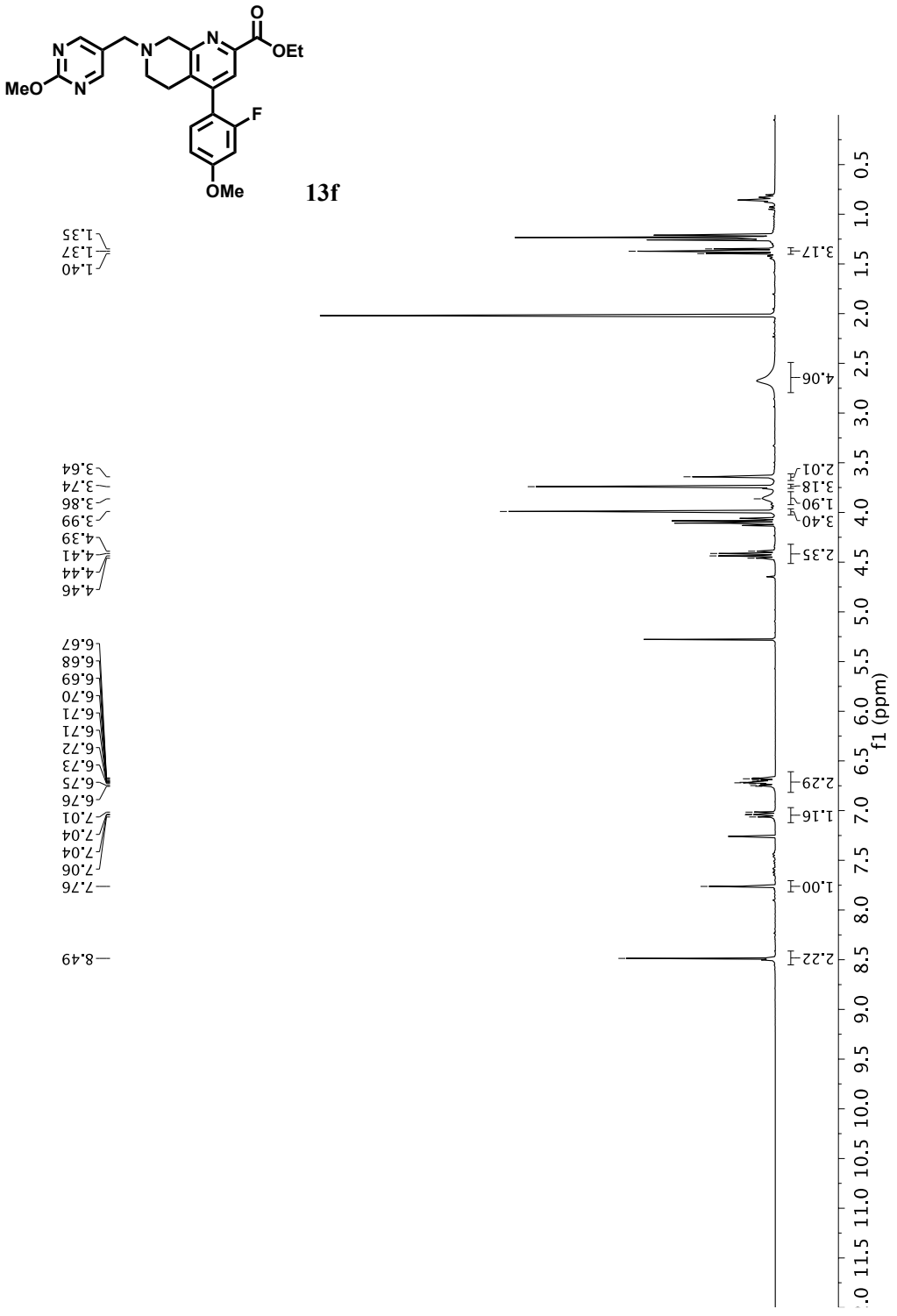


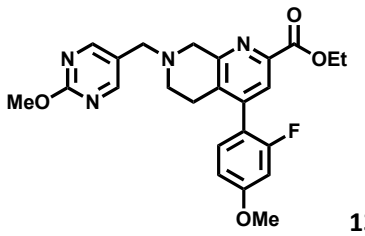
^{13}C spectrum of 13e



^{1}H spectrum of 13f



^{13}C spectrum of 13f



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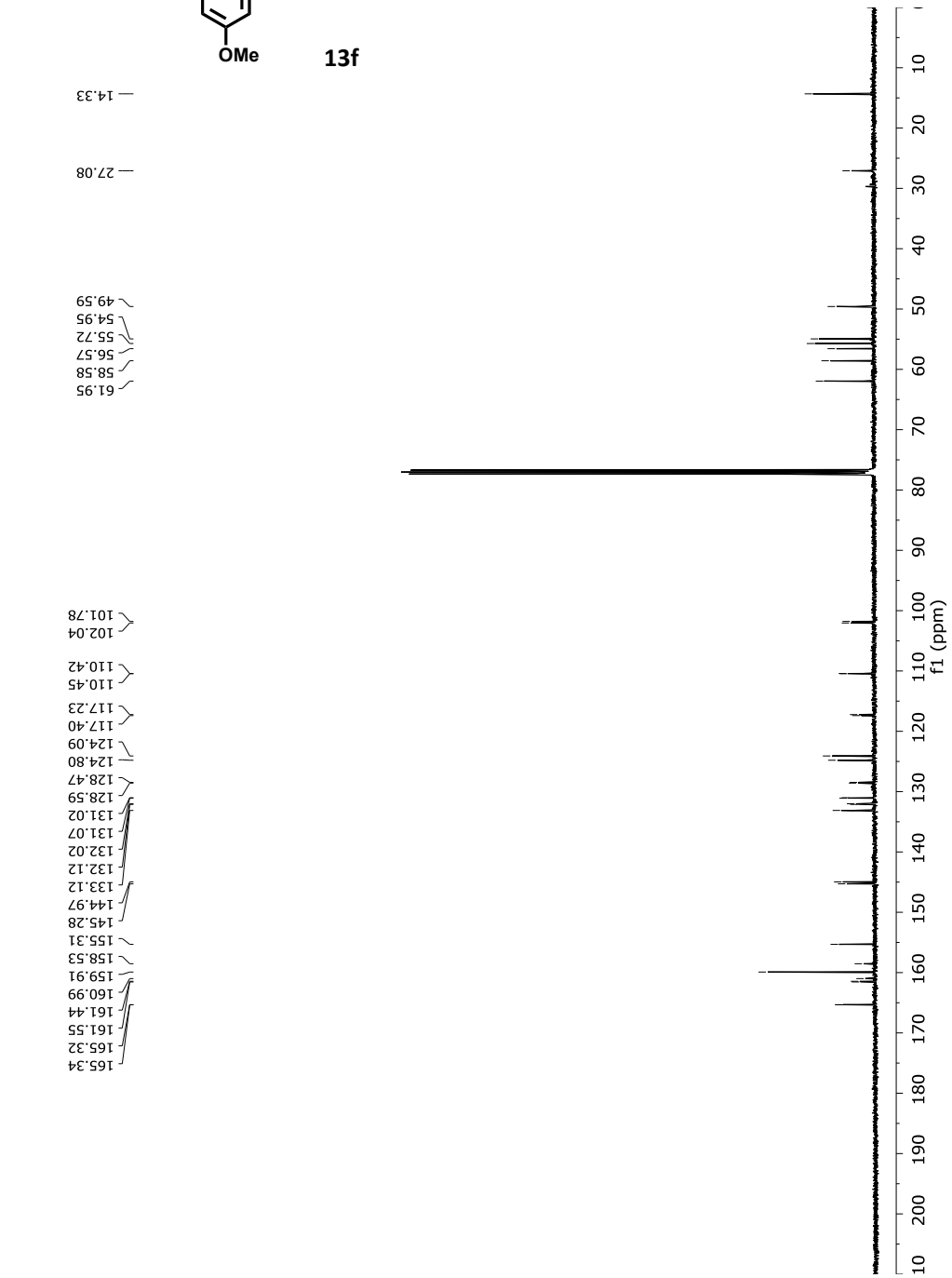
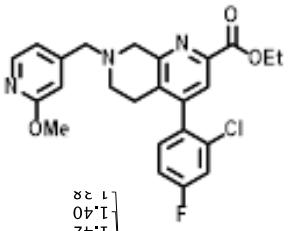


^{1}H spectrum of 13g



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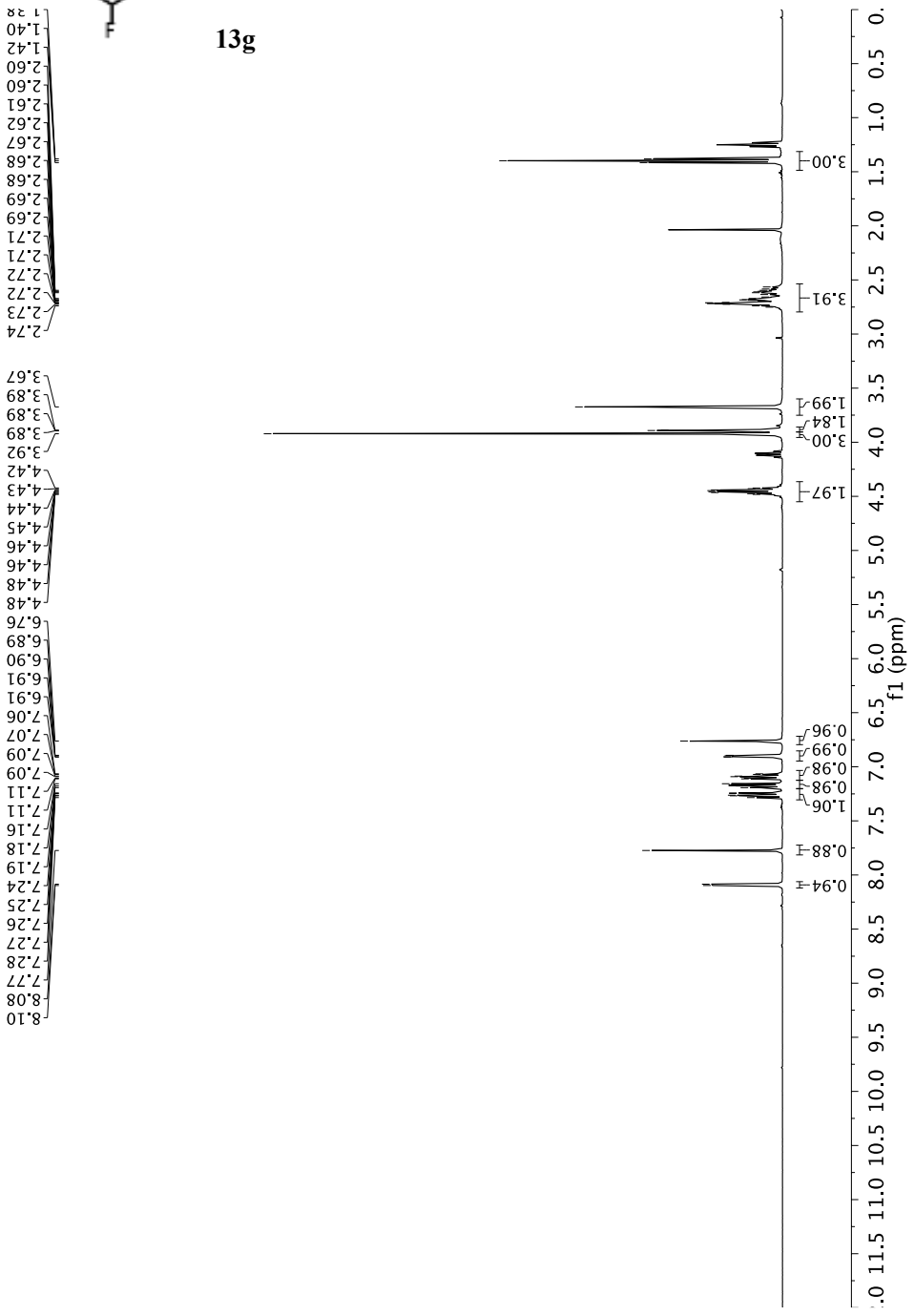
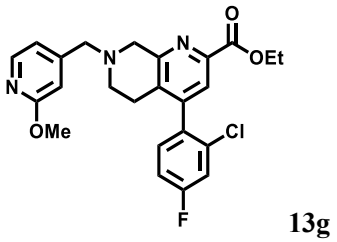
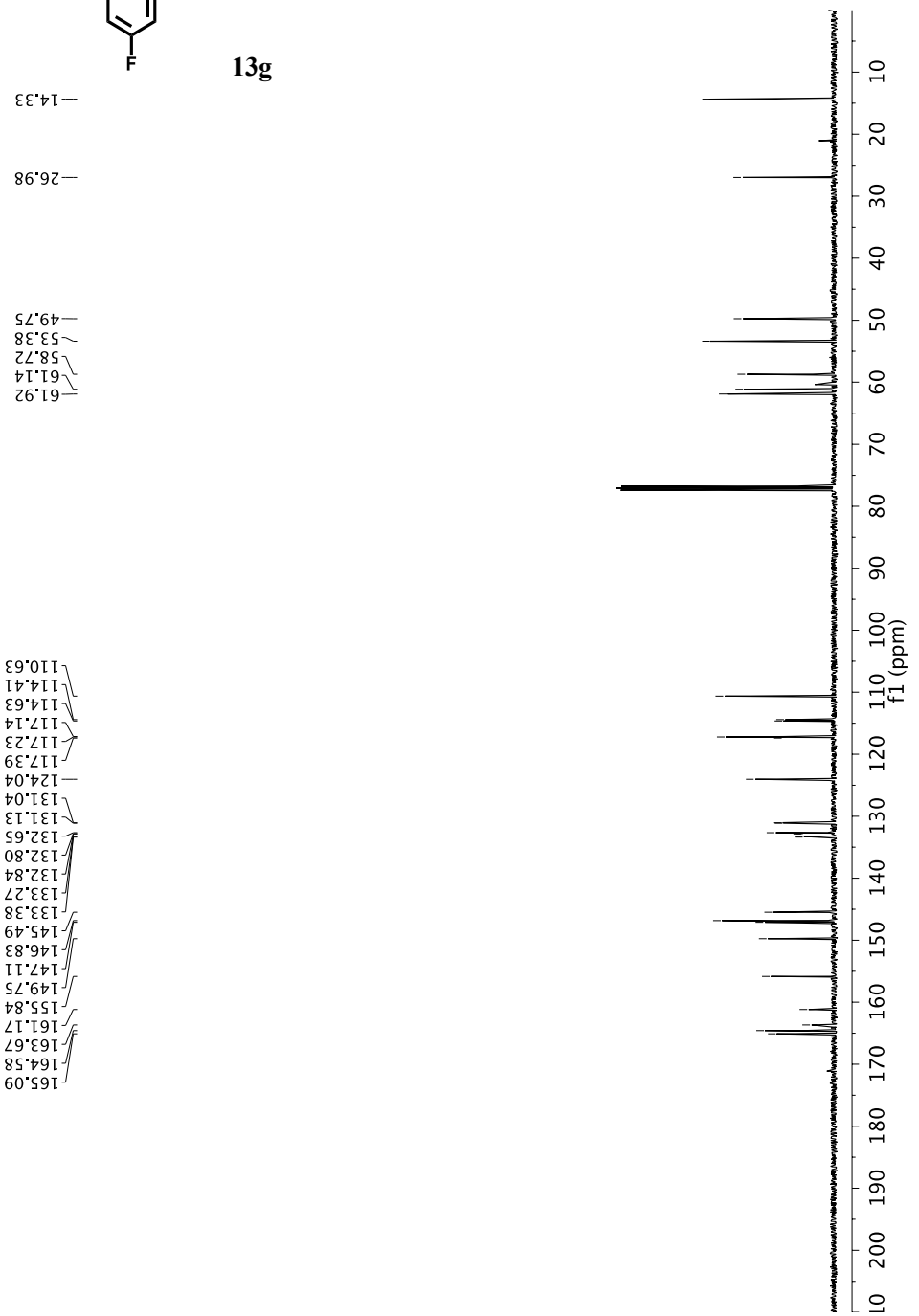


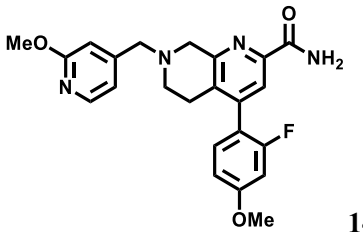
^{13}C spectrum of 13g



13g



^{1}H spectrum of 14a



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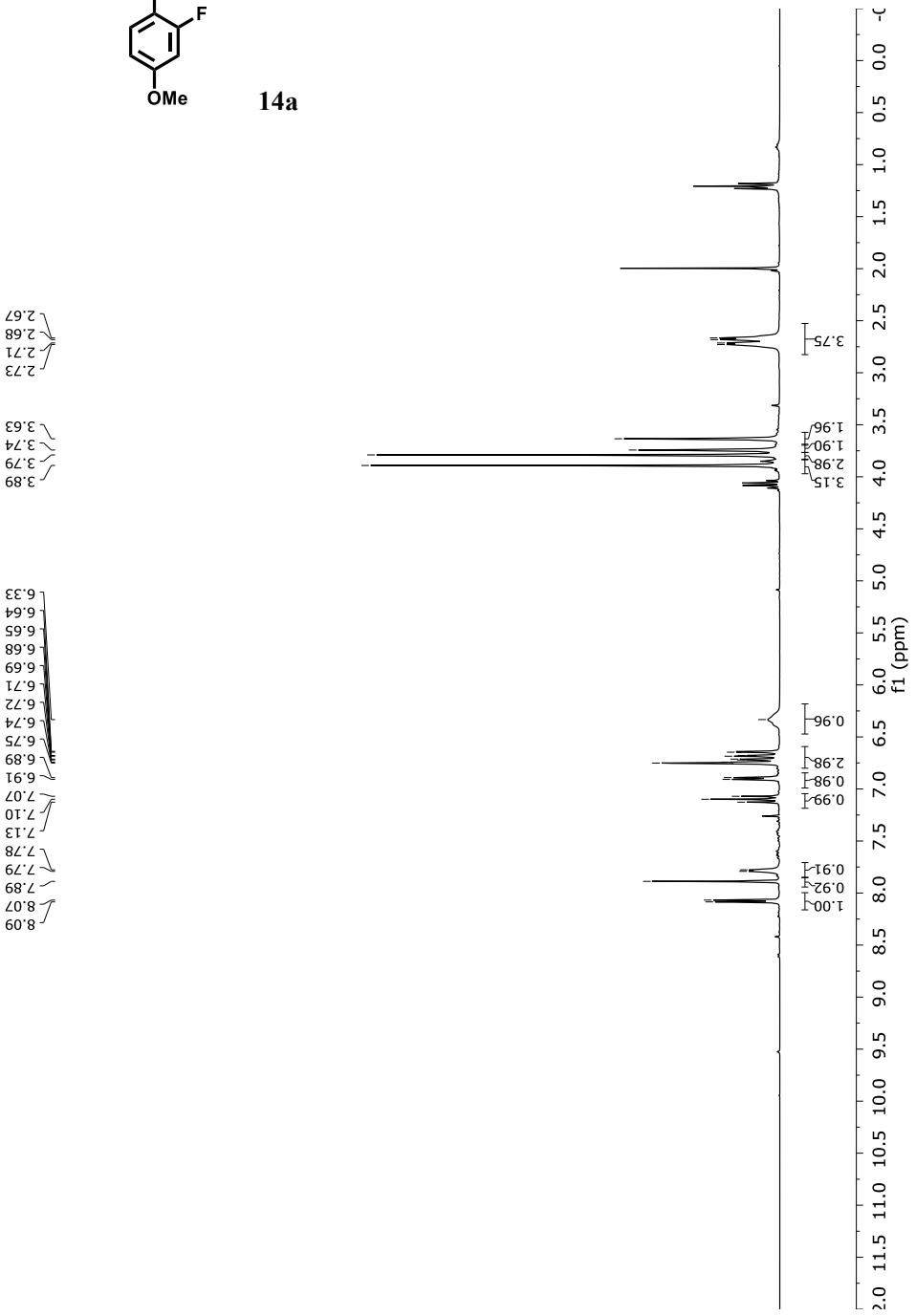
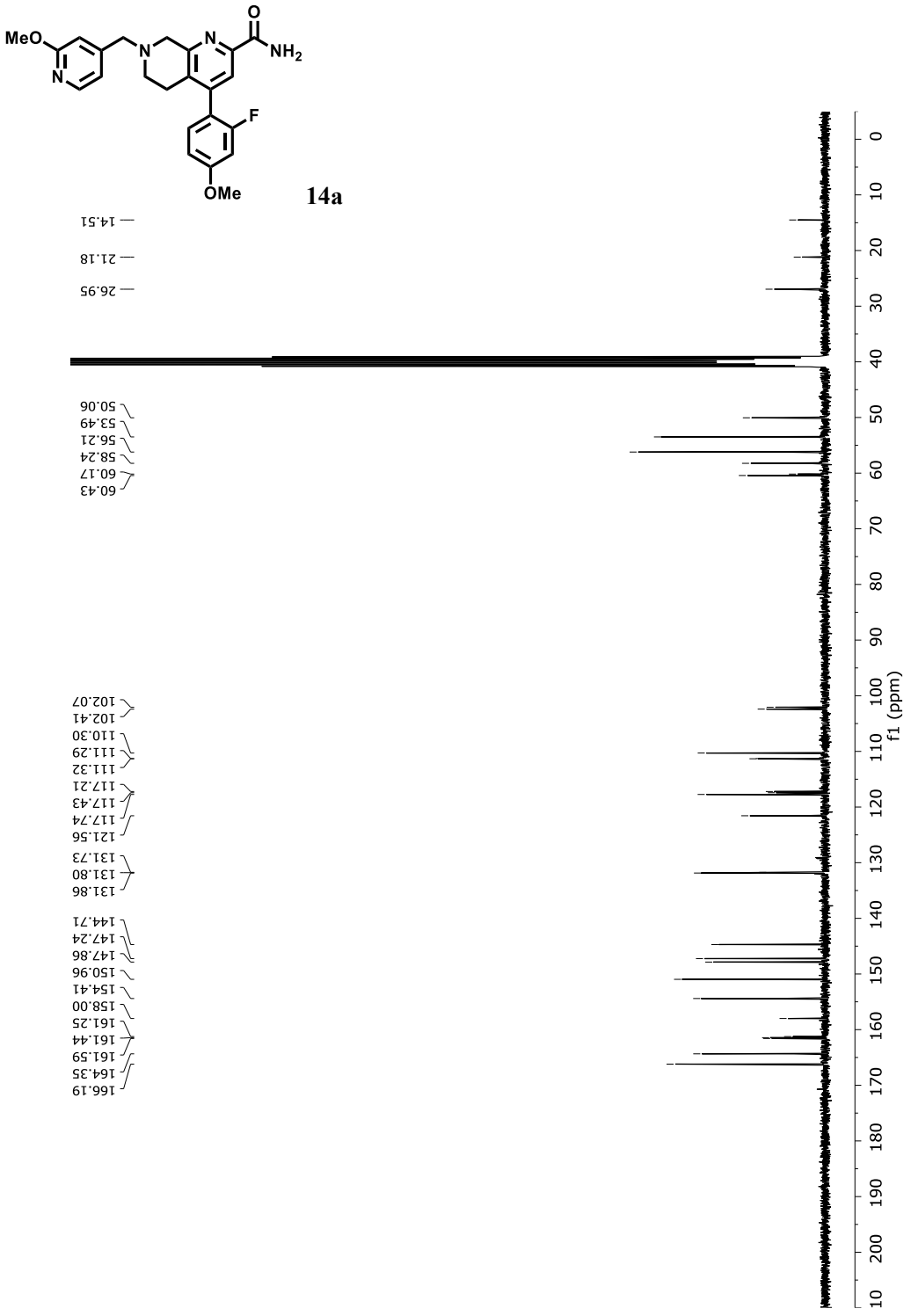
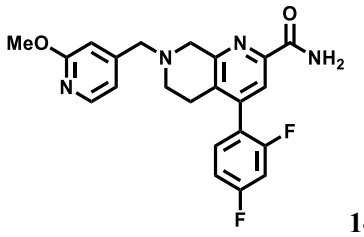


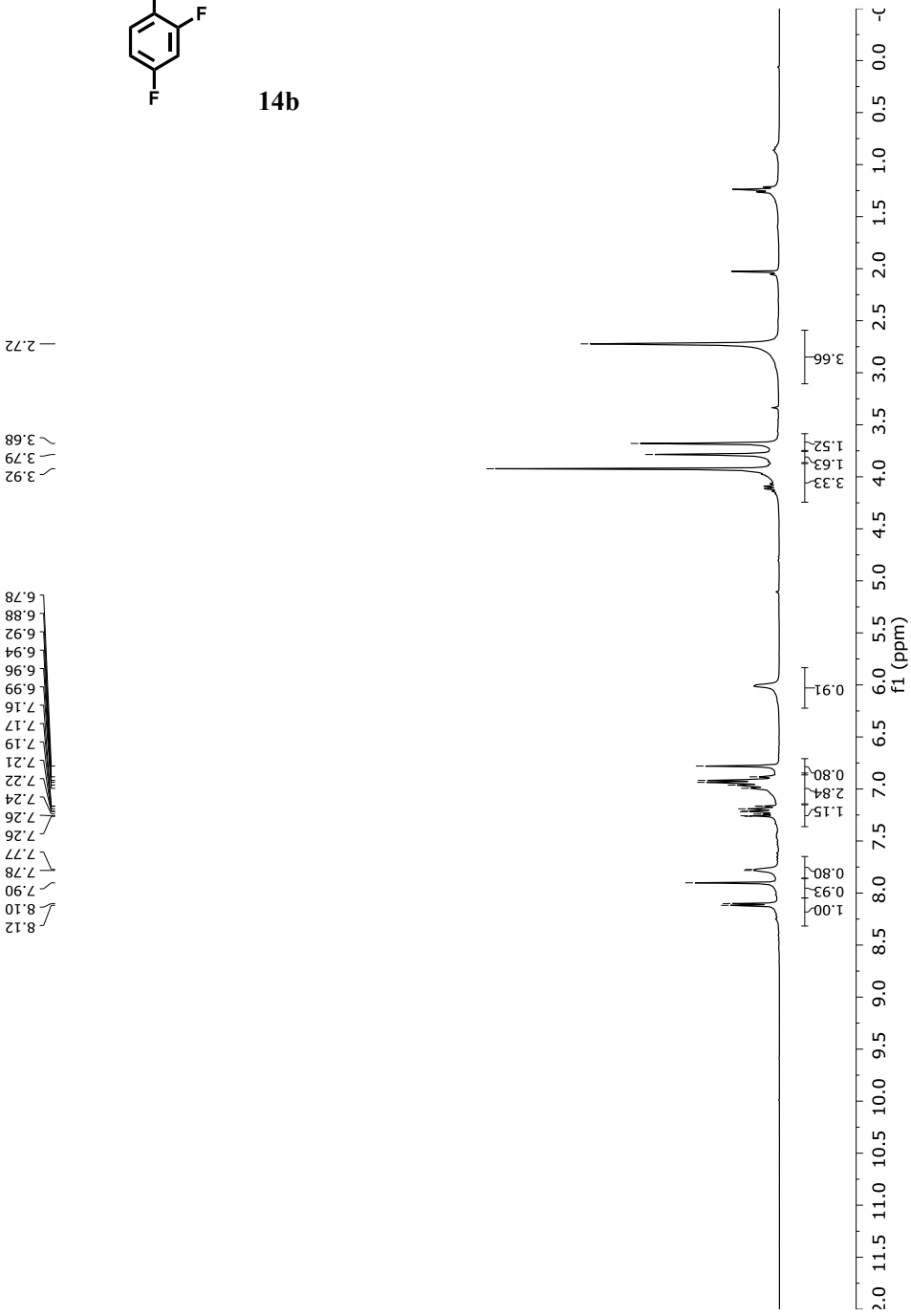
^{13}C spectrum of 14a



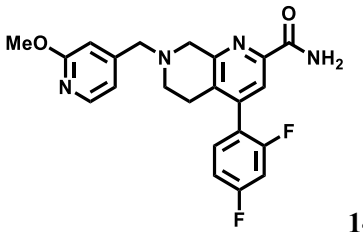
^{1}H spectrum of 14b



14b



^{13}C spectrum of 14b



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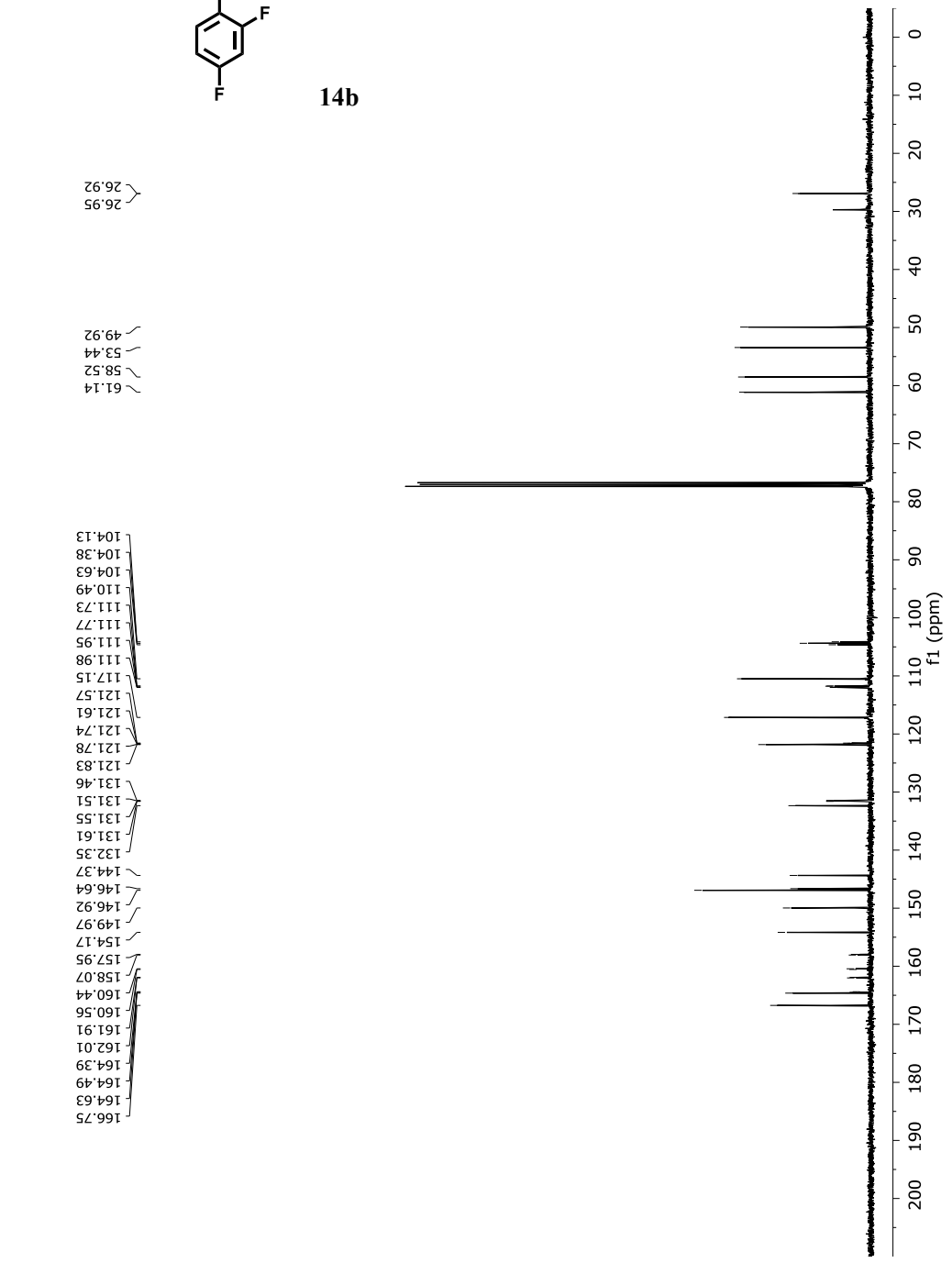
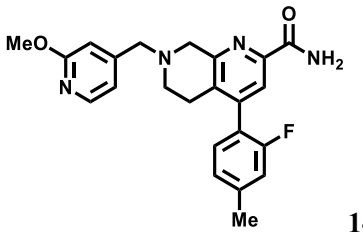


^{1}H spectrum of 14c



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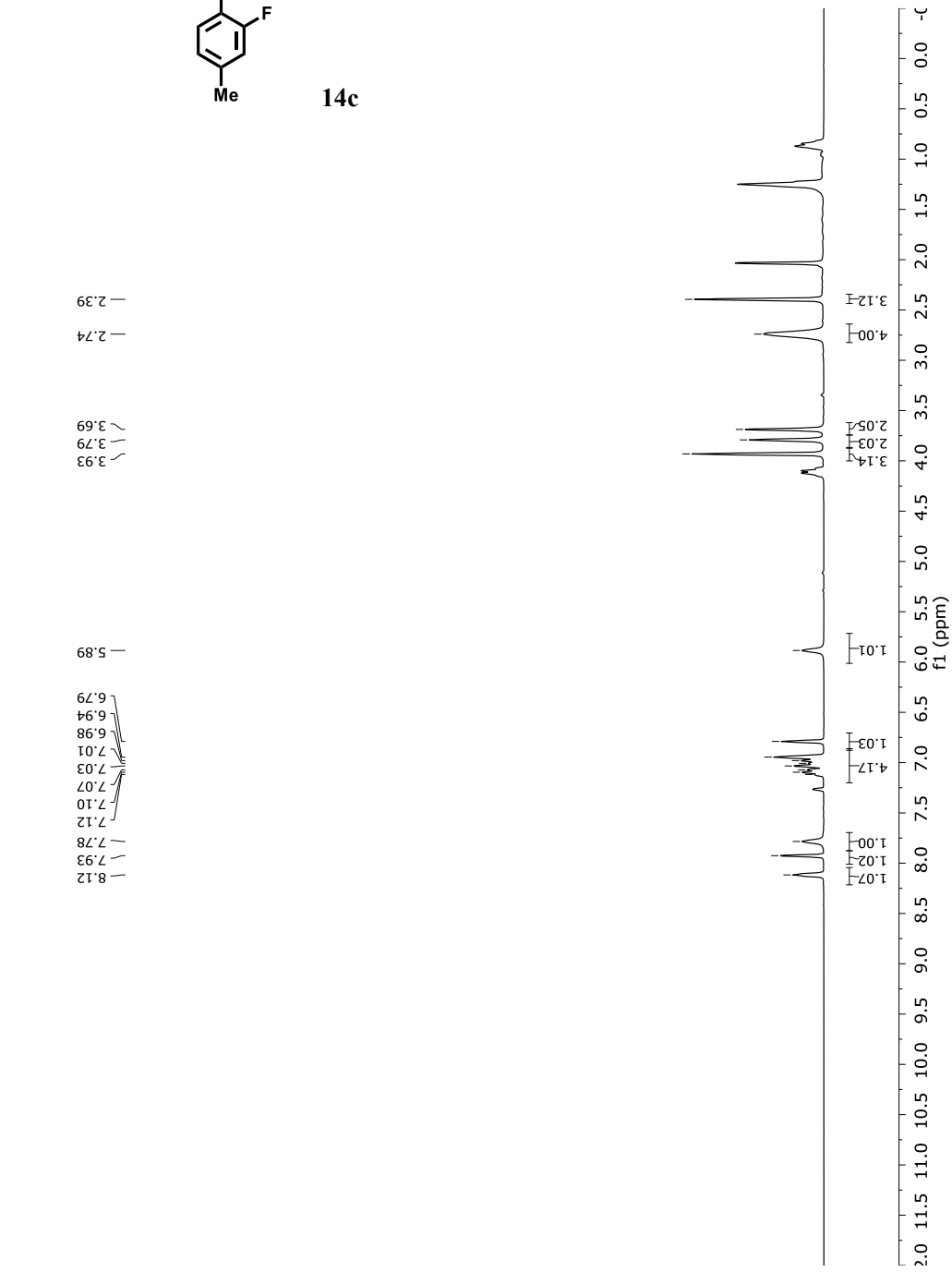
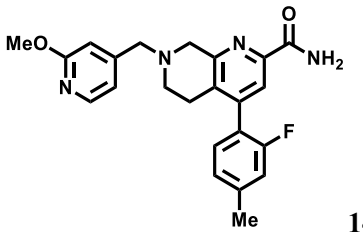
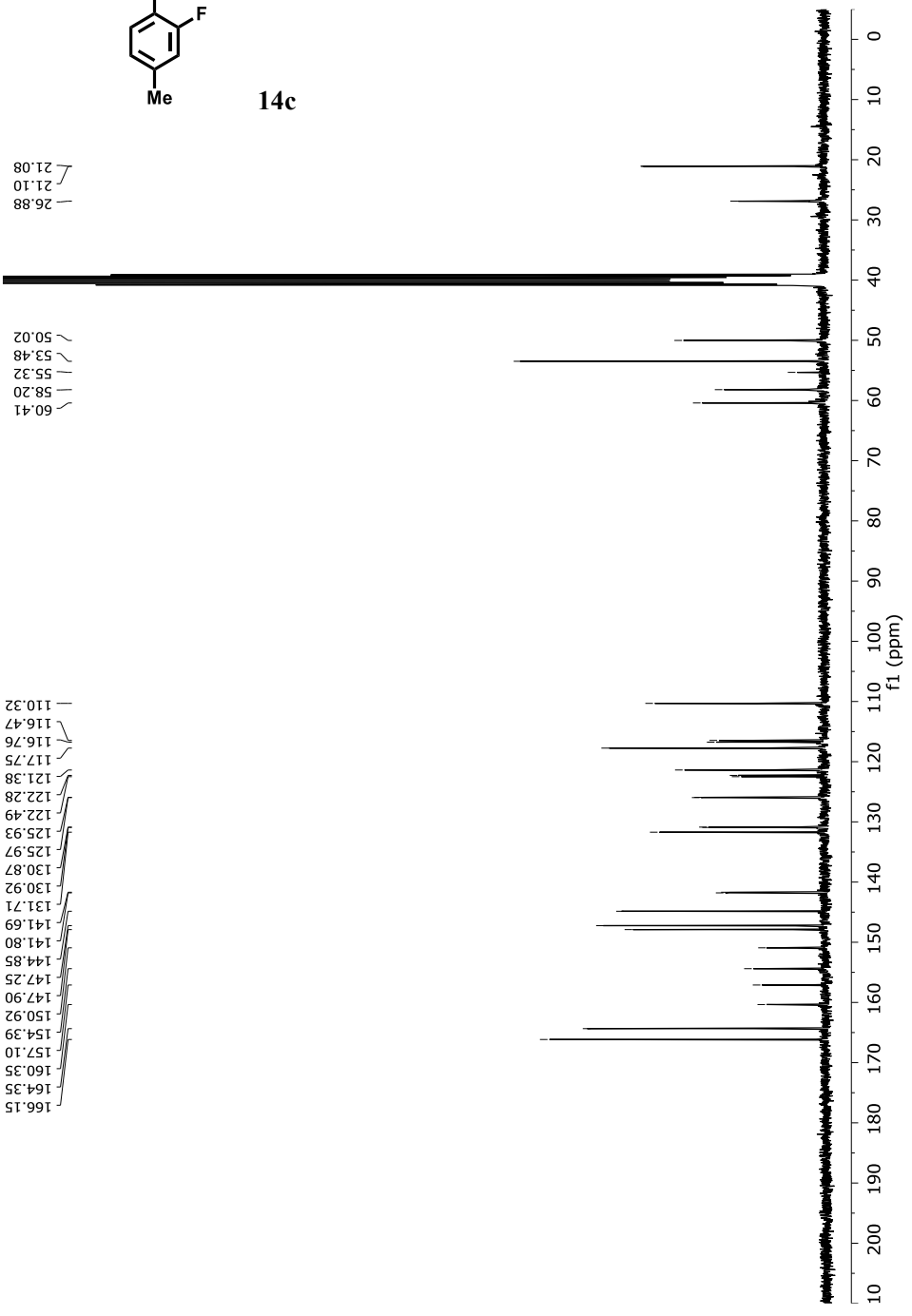


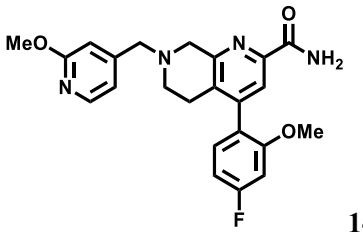
^{13}C spectrum of 14c



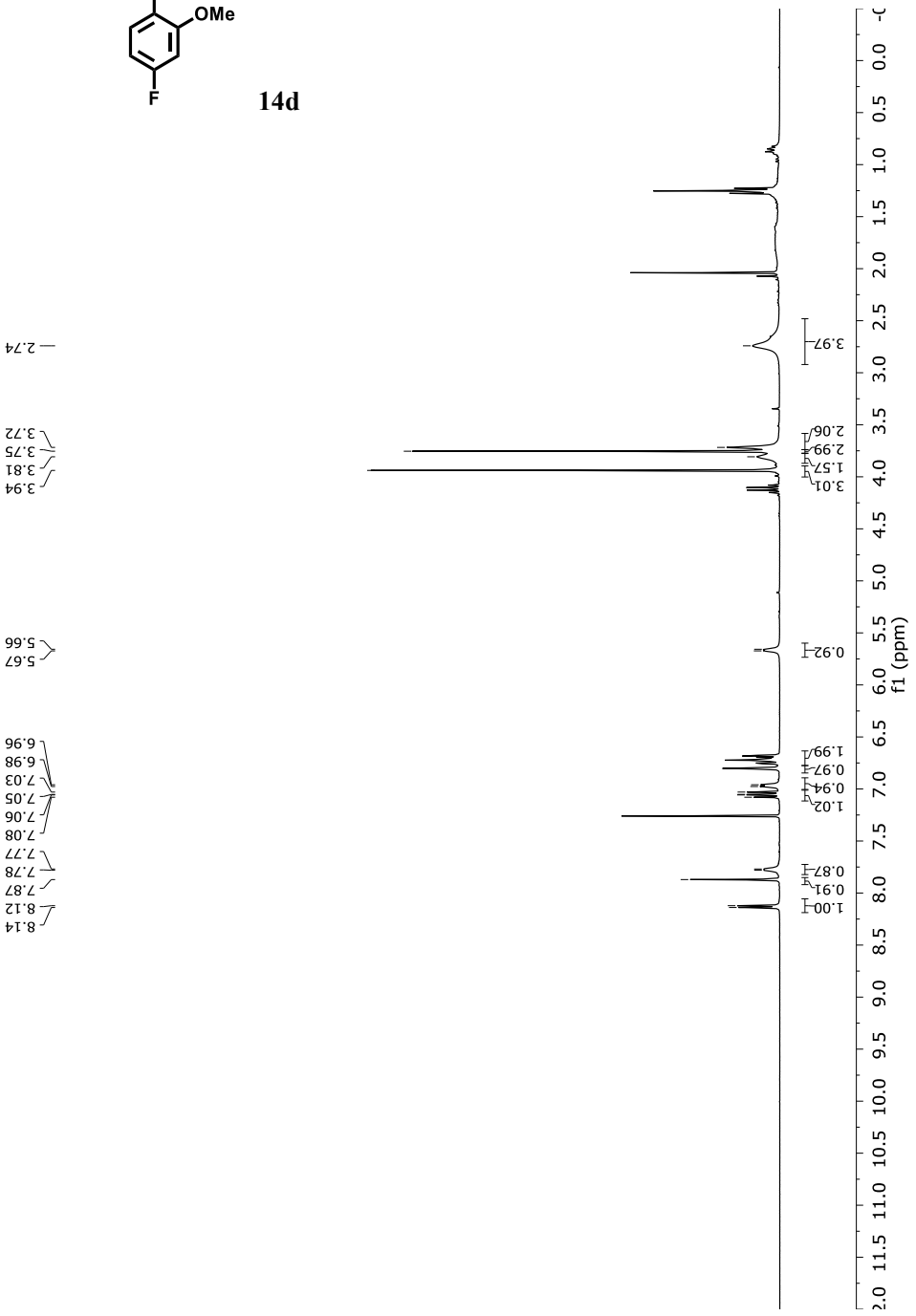
14c



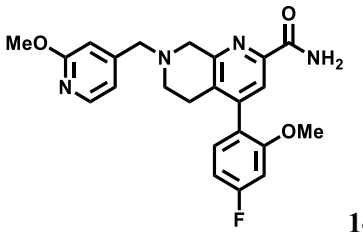
^{1}H spectrum of 14d



14d



^{13}C spectrum of 14d



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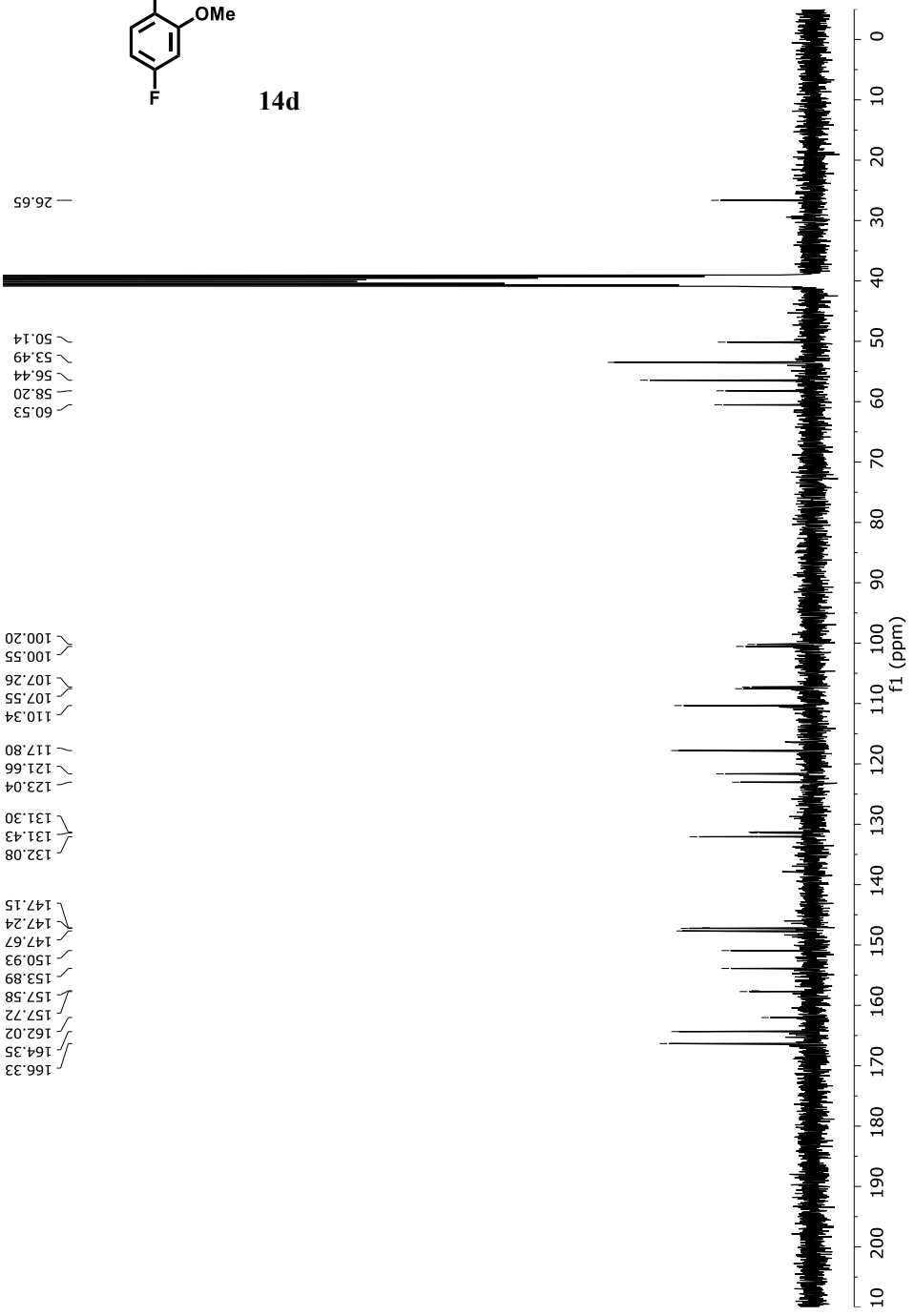

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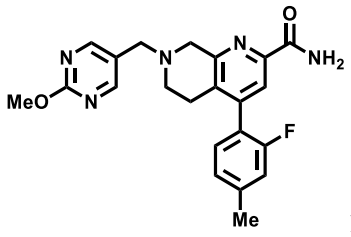

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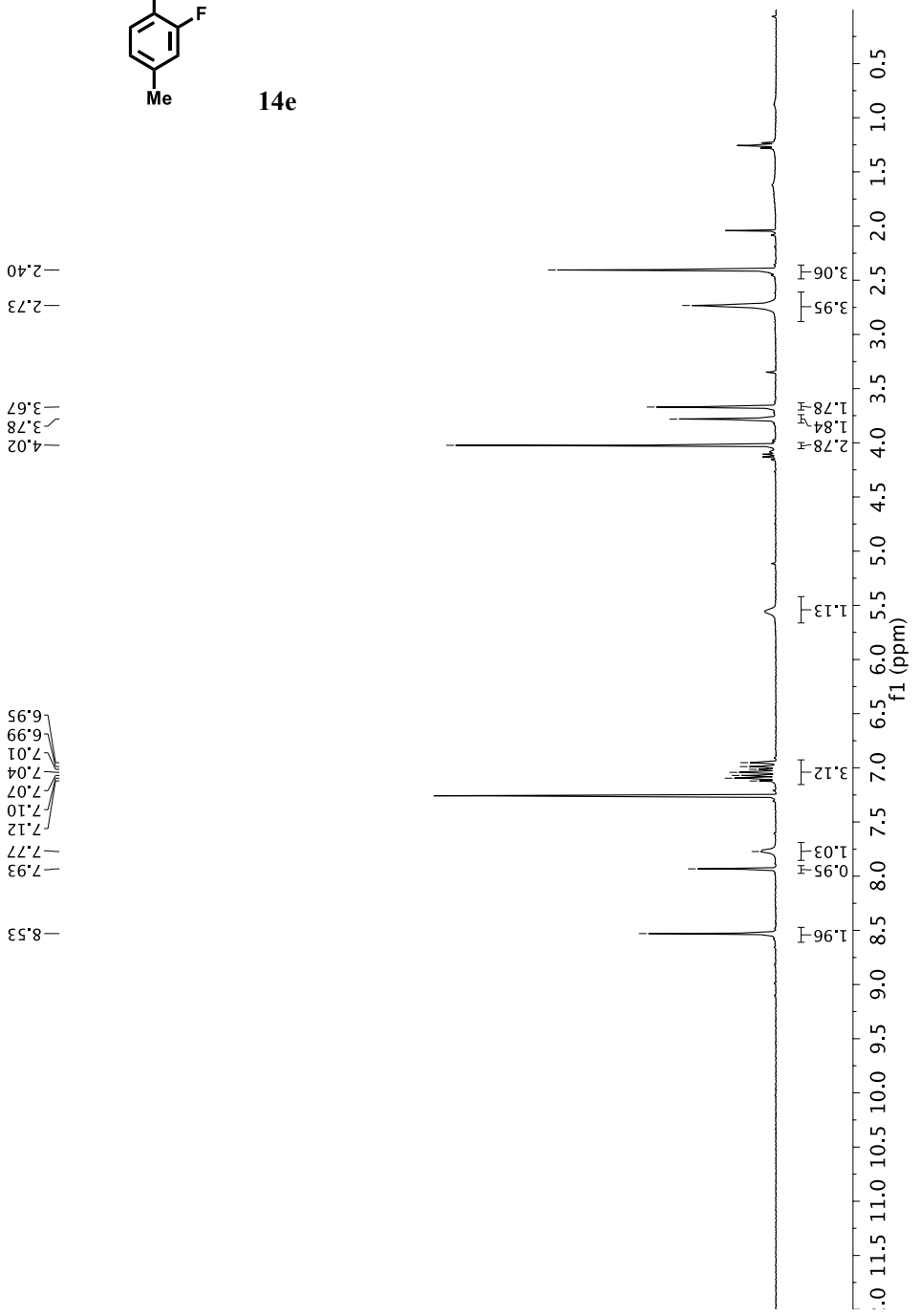

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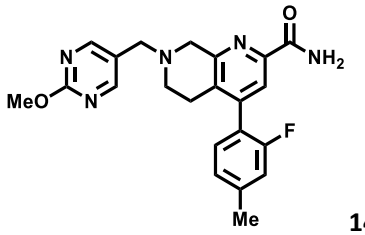

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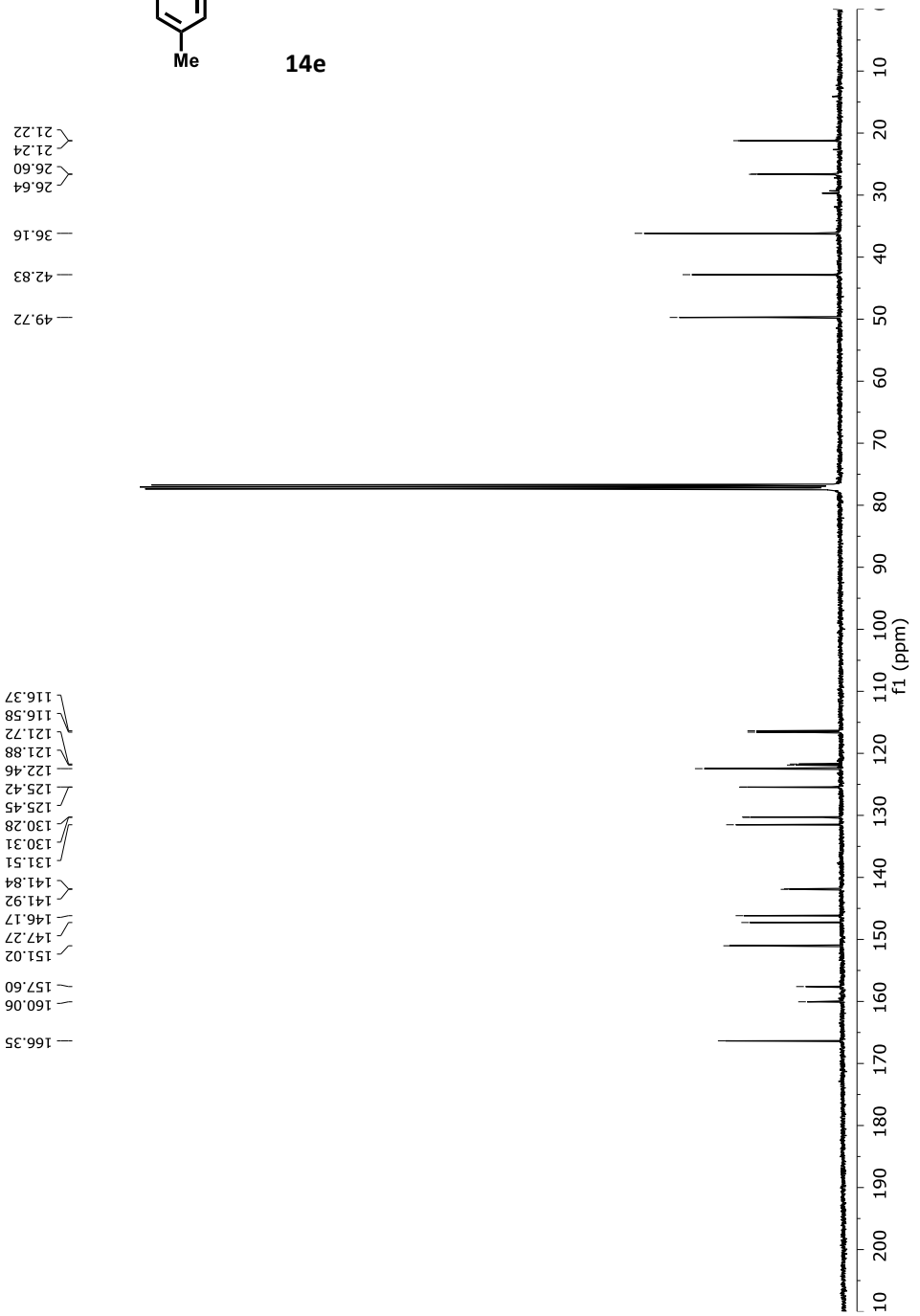
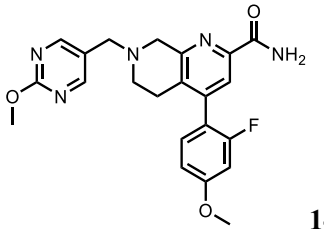


^{1}H spectrum of 14f



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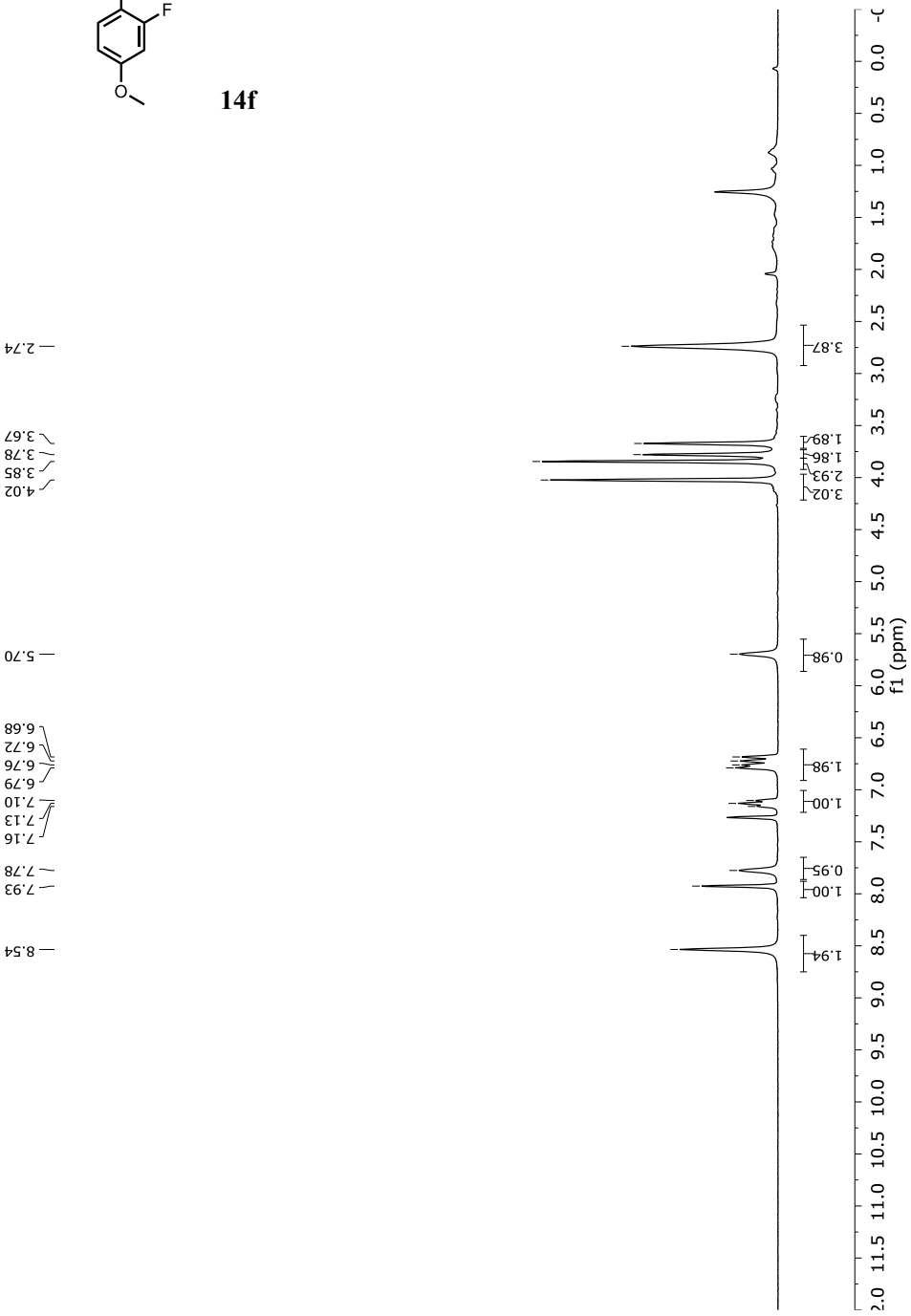

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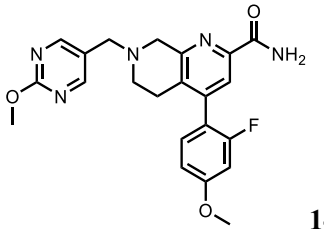

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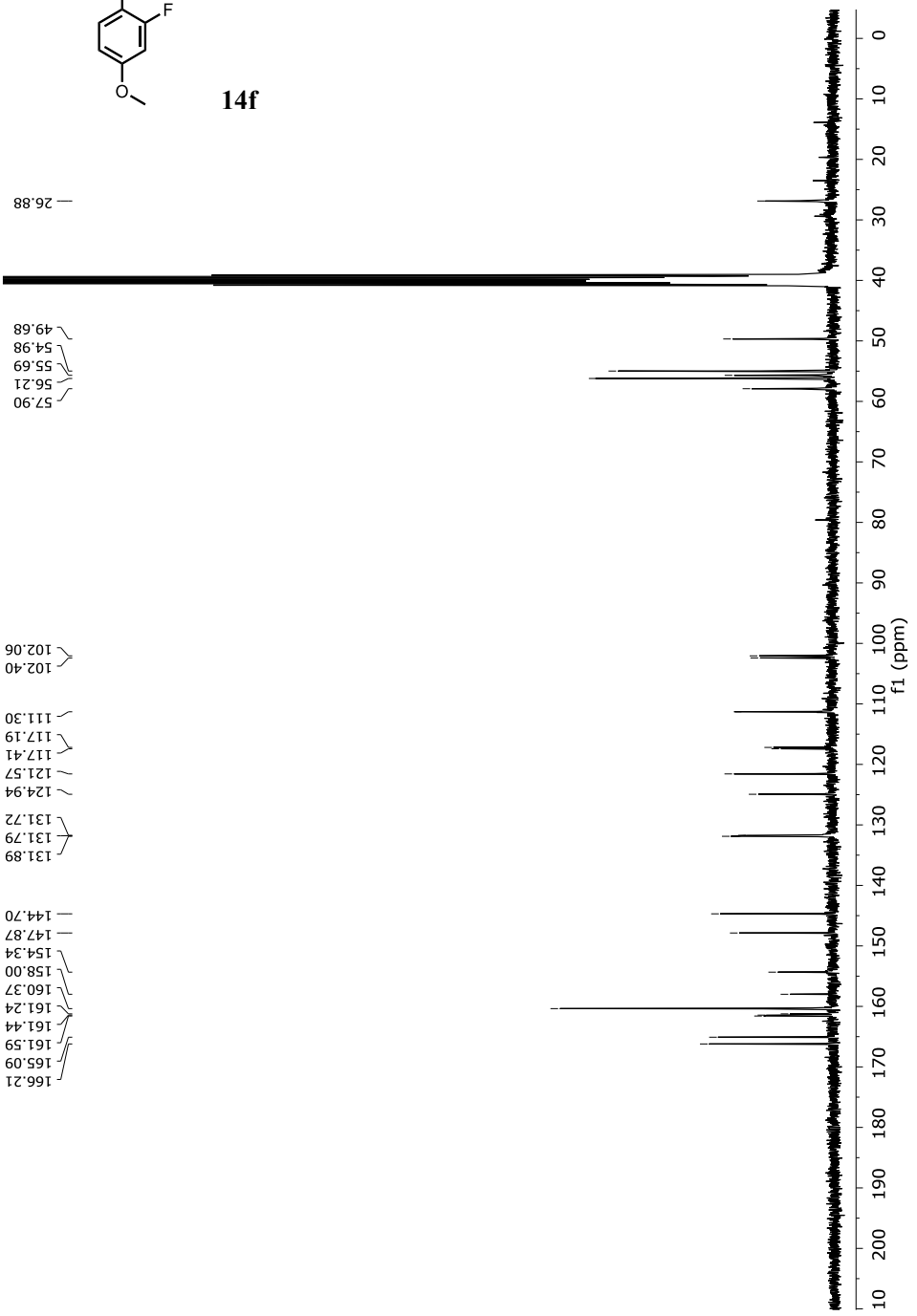
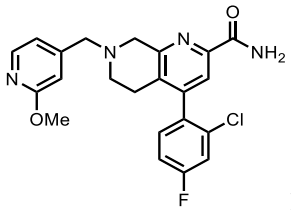


^{1}H spectrum of 14g



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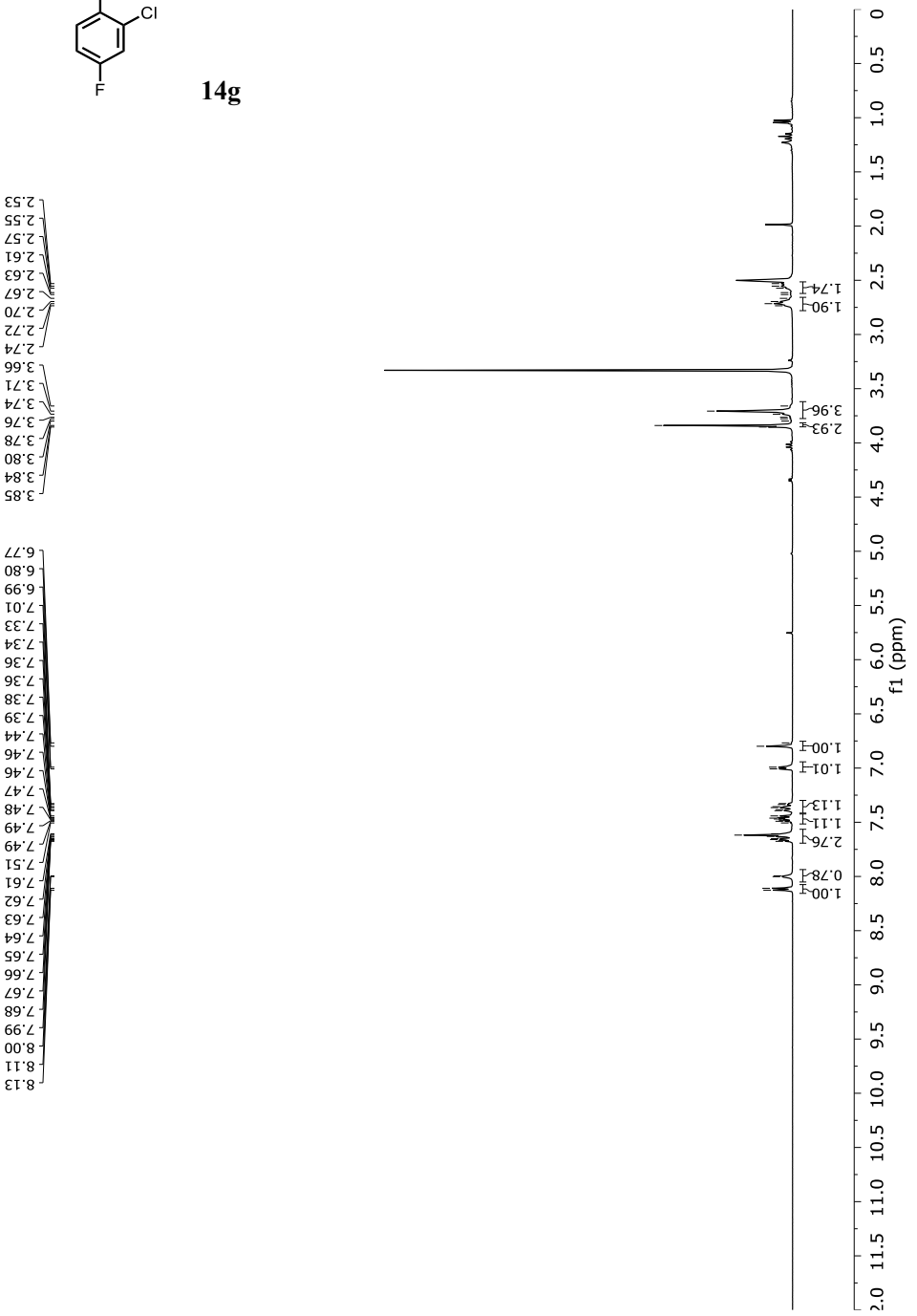
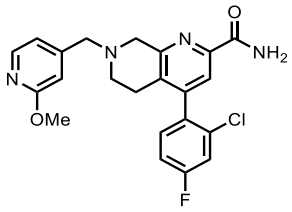


^{13}C spectrum of 14g



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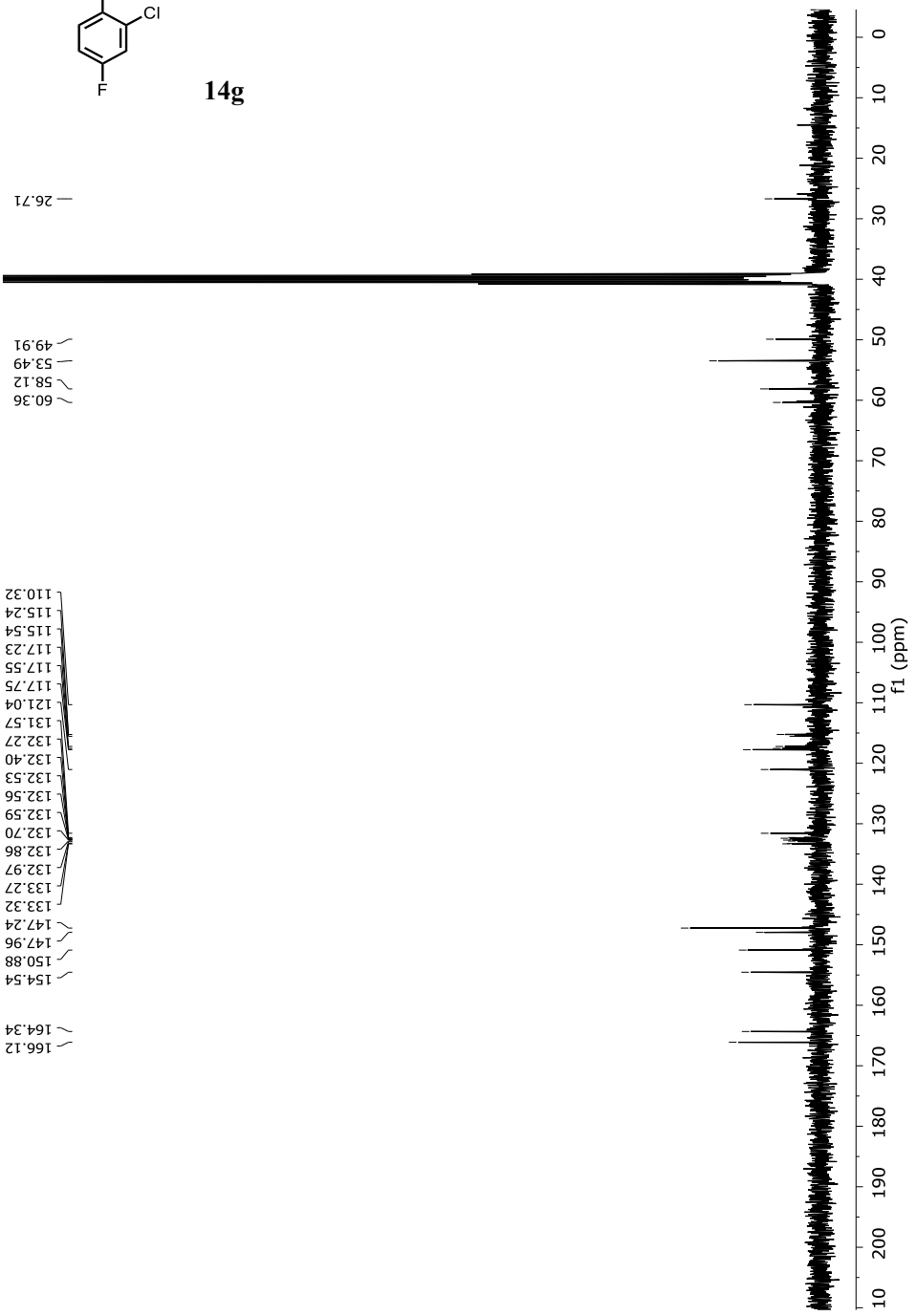
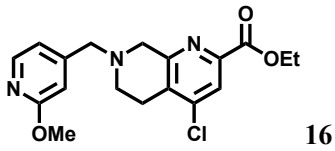
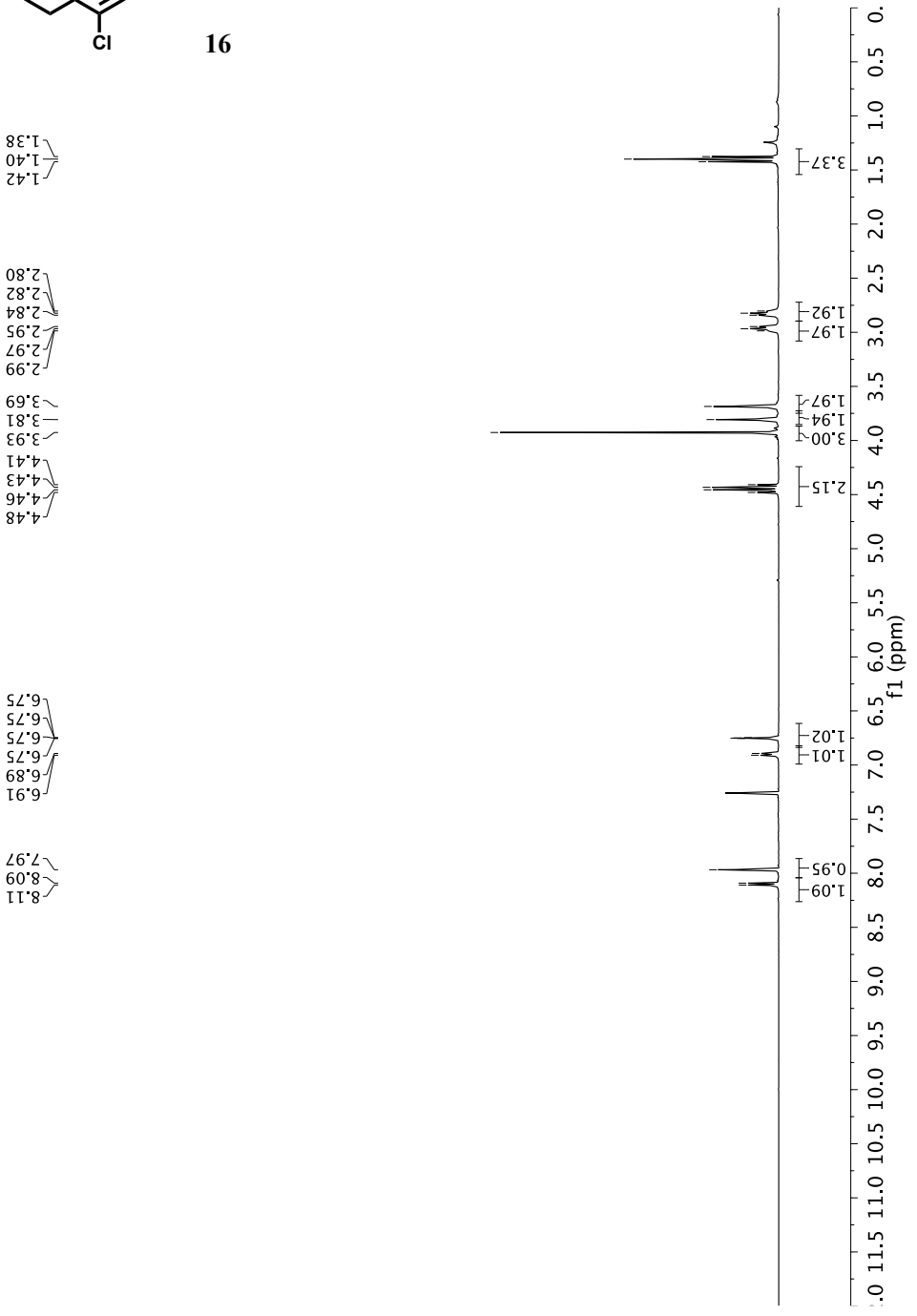


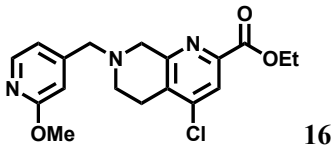
^{1}H spectrum of 16



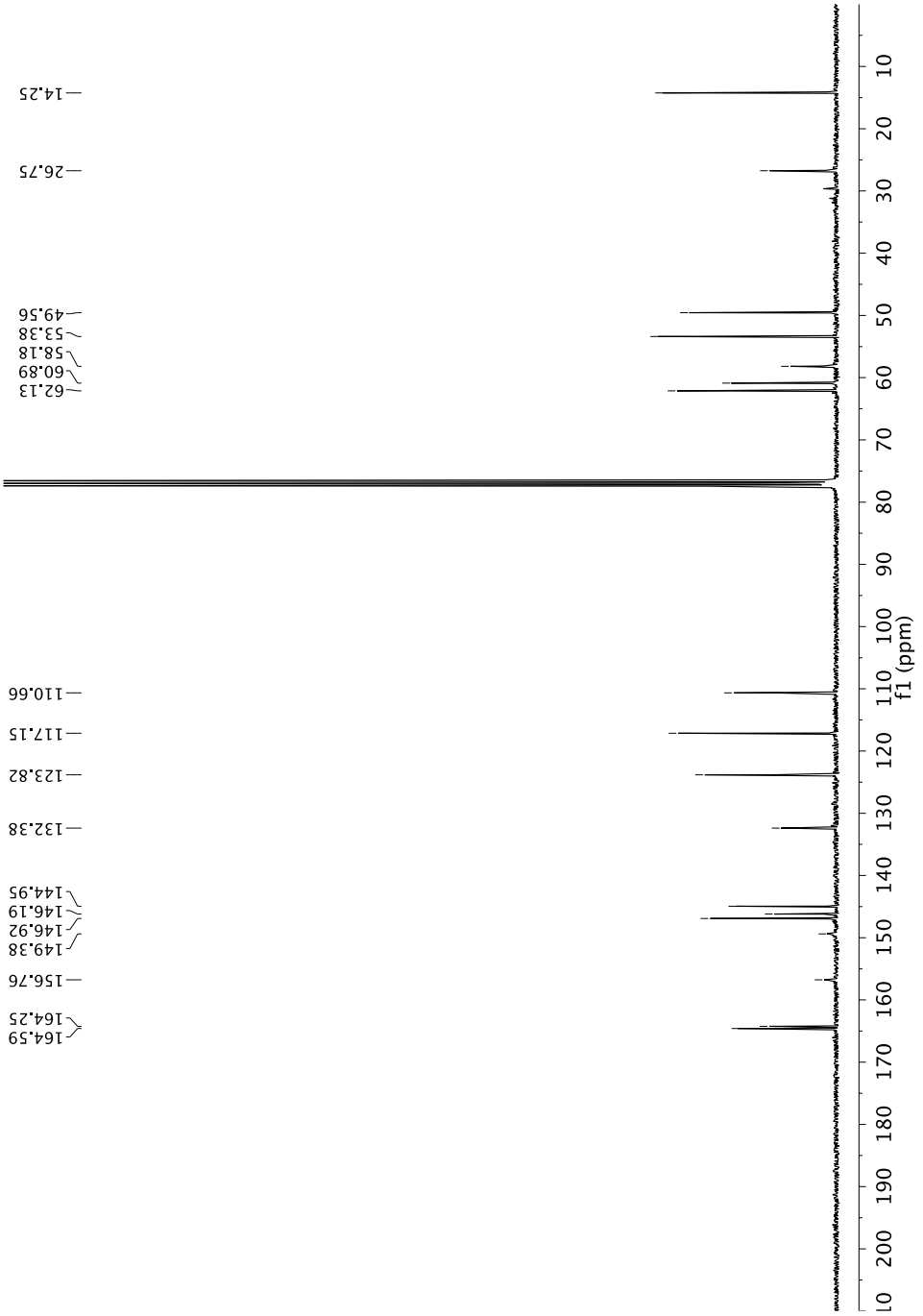
16



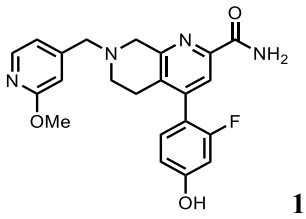
^{13}C spectrum of 16



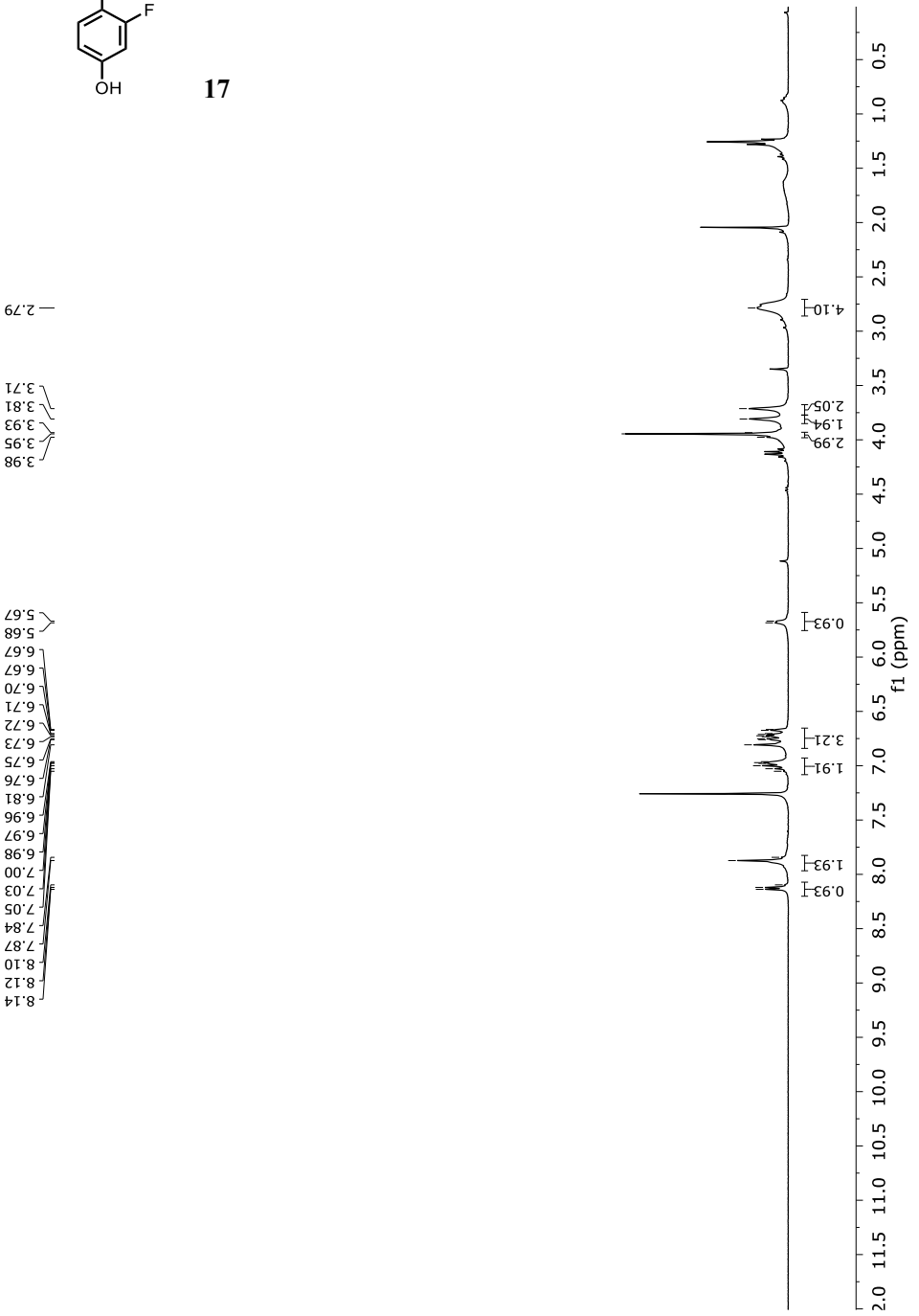
16



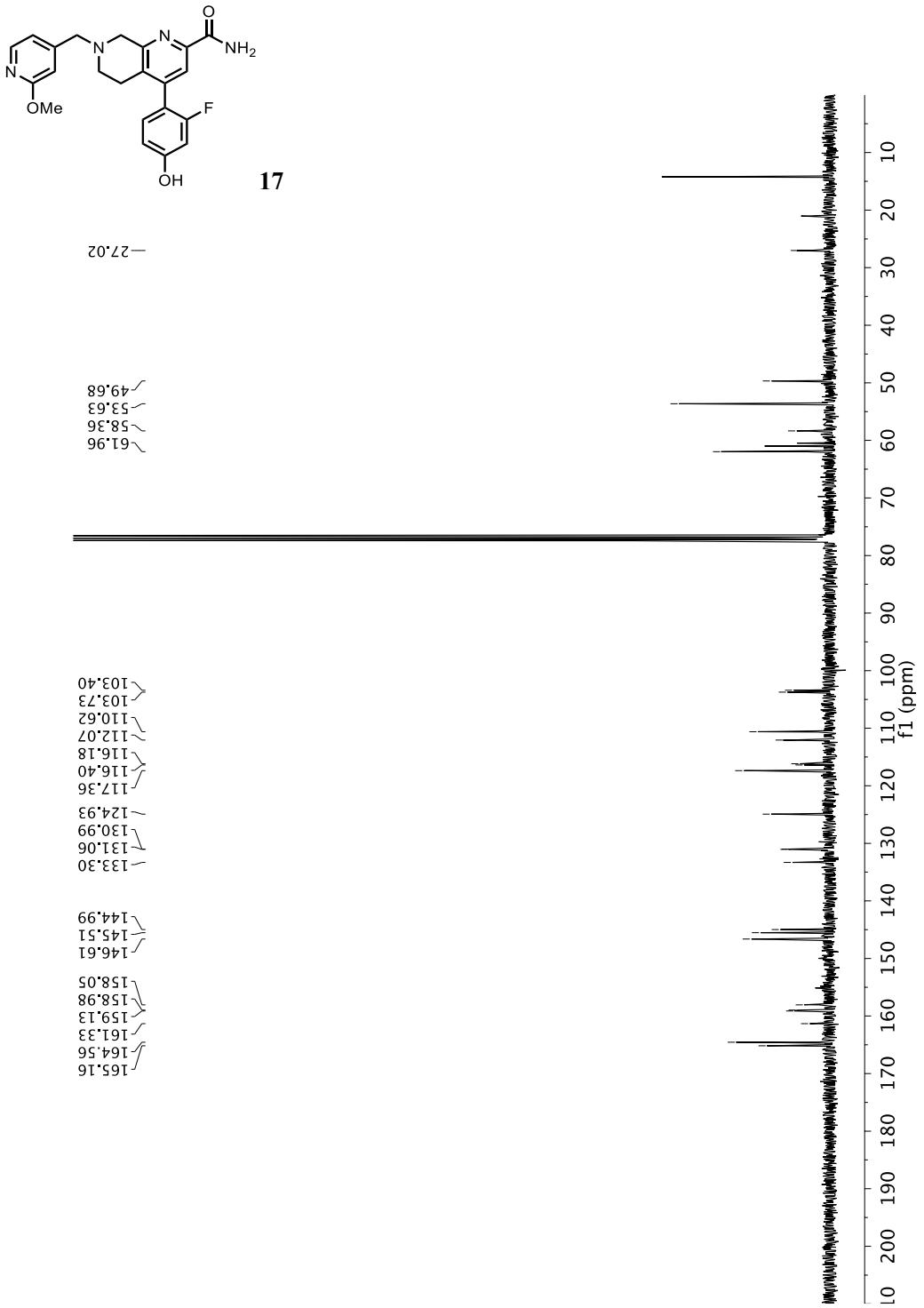
^{1}H spectrum of 17



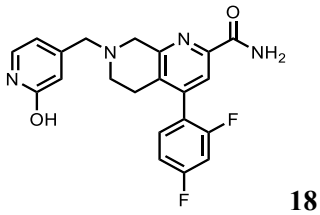
17



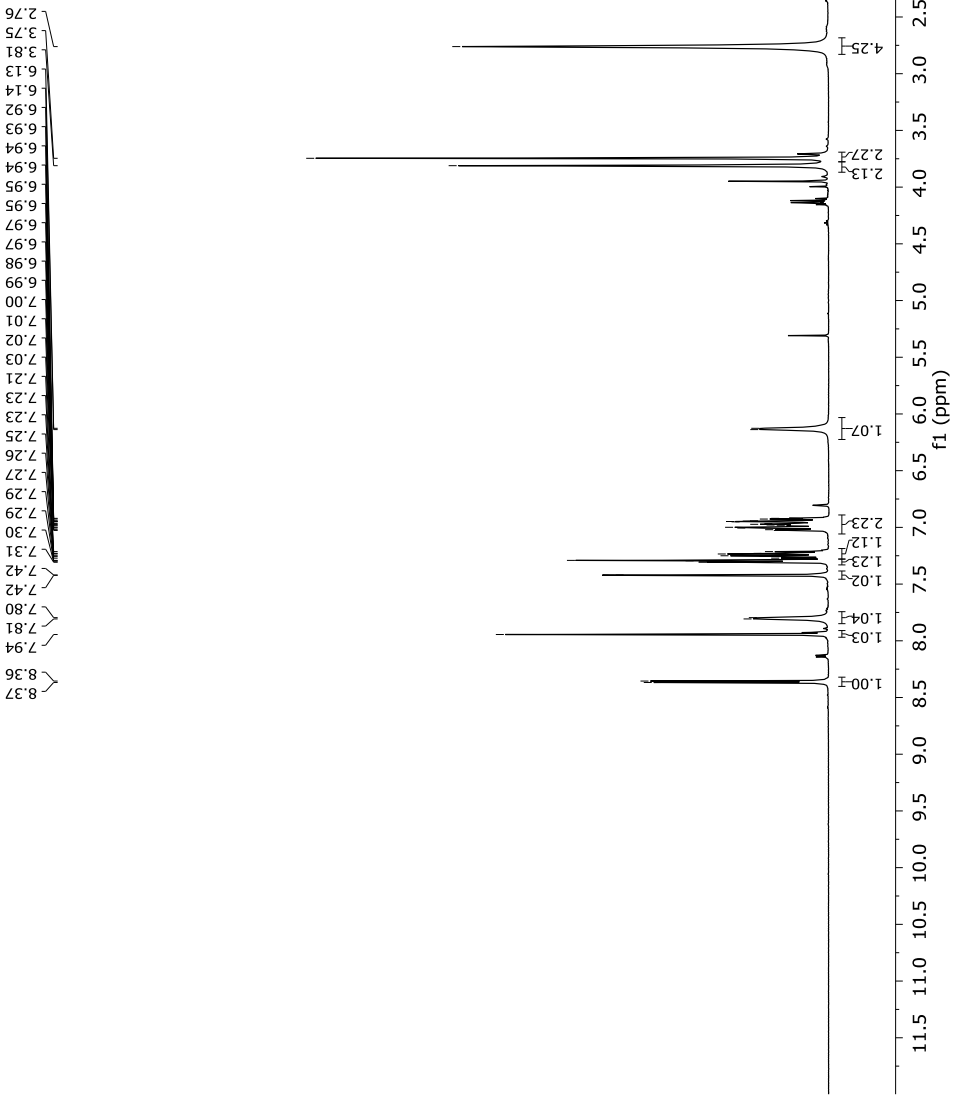
^{13}C spectrum of 17



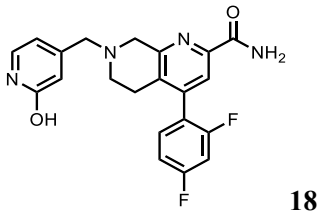
^{1}H spectrum of 18



18



^{13}C spectrum of 18



18