Reversible Covalent Imine-Tethering for Selective Stabilization of 14-3-3 Hub Protein Interactions

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Supporting Information

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Table S1: The 14-3-3pred server allows the *in silico* analysis of potential 14-3-3 binding sites of Pin1.

Position	Peptide [-6:4]	ANN	PSSM	SVM	Consensus	phosphoS/T
18	EKRMSR[S]SGRV	0.655	0.376	-0.187	0.281	-
19	KRMSRS[S]GRVY	0.236	0.172	-0.741	-0.111	-
29	YYFNHI[T]NASQ	0.112	-0.118	-1.135	-0.380	-
32	NHITNA[S]QWER	0.344	0.017	-1.048	-0.229	-
38	SQWERP[S]GNSS	0.049	-0.032	-1.242	-0.408	-
41	ERPSGN[S]SSGG	0.062	-0.107	-1.383	-0.476	-
42	RPSGNS[S]SGGK	0.136	-0.226	-0.929	-0.340	-
43	PSGNSS[S]GGKN	0.147	0.033	-0.636	-0.152	-
58	PARVRC[S]HLLV	0.525	0.332	-0.614	0.081	-
65	HLLVKH[S]QSRR	0.142	-0.013	-1.085	-0.319	-
67	LVKHSQ[S]RRPS	0.214	-0.020	-0.469	-0.092	-
71	SQSRRP[S]SWRQ	0.552	0.712	-0.128	0.379	-
72	QSRRPS[S]WRQE	0.583	0.821	0.370	0.591	-
79	WRQEKI[T]RTKE	0.118	-0.035	-1.168	-0.362	-
81	QEKITR[T]KEEA	0.114	-0.035	-1.036	-0.319	-
98	YIQKIK[S]GEED	0.519	0.214	0.162	0.298	-
105	GEEDFE[S]LASQ	0.159	-0.151	-1.308	-0.433	-
108	DFESLA[S]QFSD	0.167	-0.173	-1.130	-0.379	-
111	SLASQF[S]DCSS	0.062	-0.163	-1.479	-0.527	-
114	SQFSDC[S]SAKA	0.069	-0.328	-1.607	-0.622	-
115	QFSDCS[S]AKAR	0.248	0.007	-0.605	-0.117	-
126	GDLGAF[S]RGQM	0.262	-0.048	-0.479	-0.088	-
138	KPFEDA[S]FALR	0.152	-0.078	-1.023	-0.316	-
143	ASFALR[T]GEMS	0.050	-0.369	-1.800	-0.706	-
147	LRTGEM[S]GPVF	0.528	0.730	0.209	0.489	-
152	MSGPVF[T]DSGI	0.200	-0.067	-0.747	-0.205	-
154	GPVFTD[S]GIHI	0.131	-0.120	-0.970	-0.320	-
162	IHIILR[T]E---	0.118	0.148	-1.347	-0.360	-

Table S2: Data collection and refinement statistics for binary peptide/14-3-3σΔC complex as indicated. Data collection statistics, as reported by aimless, the refinement statics were derived from the 'Table1' software of the phenix suite. Values in parenthesis represent the highest resolution shell.

	Pin1pS72	AS160
PDB code	7AOG	7NIX
X-ray source	DESY, Petra III, P11	DLS, I03
Data Collection		
Wavelength	1.0332	0.976284
Resolution range	62.65-1.5 (1.53-1.50)	104.98-1.9 (1.94-1.9)
Space group	C2221	P6122
Unit cell		
a, b, c (Å)	82.43 112.06 62.65	121.22 121.22 74.46
α, β, γ (°)	90.00 90.00 90.00	90.00 90.00 120.00
Unique reflections	44552 (1508)	25949 (1623)
Multiplicity	10.5 (3.8)	37.5 (38.7)
Completeness (%)	95.1 (64.3)	100 (100)
Mean I/sigma(I)	20.1 (1.6)	25.4 (3.3)
R-merge	0.055 (0.736)	0.104 (1.550)
R-meas	0.058 (0.859)	0.105 (1.570)
R-pim	0.017 (0.425)	0.017 (0.252)
CC1/2	0.999 (0.527)	1.000 (0.970)
Refinement		
Reflections used in refinement	44518 (3163)	25890 (2522)
Reflections used for R-free	2152 (162)	1244 (117)
R-work	0.1653 (0.2470)	0.1856 (0.2706)
R-free	0.1843 (0.2549)	0.2041 (0.3141)
No. of non-hydrogen atoms	2258	2133
macromolecules	1970	1908
ligands	4	60
solvent	284	197
RMS (bonds)	0.008	0.005
RMS (angles)	0.91	0.81
Ramachandran favored (%)	97.87	99.14
Ramachandran allowed (%)	2.13	0.86
Ramachandran outliers (%)	0	0
Rotamer outliers (%)	0	0
Clashscore	3.1	1.32
Average B-factor	24.98	34.39
macromolecules	23.54	33.34
ligands	30.26	49.65
solvent	34.88	42.4

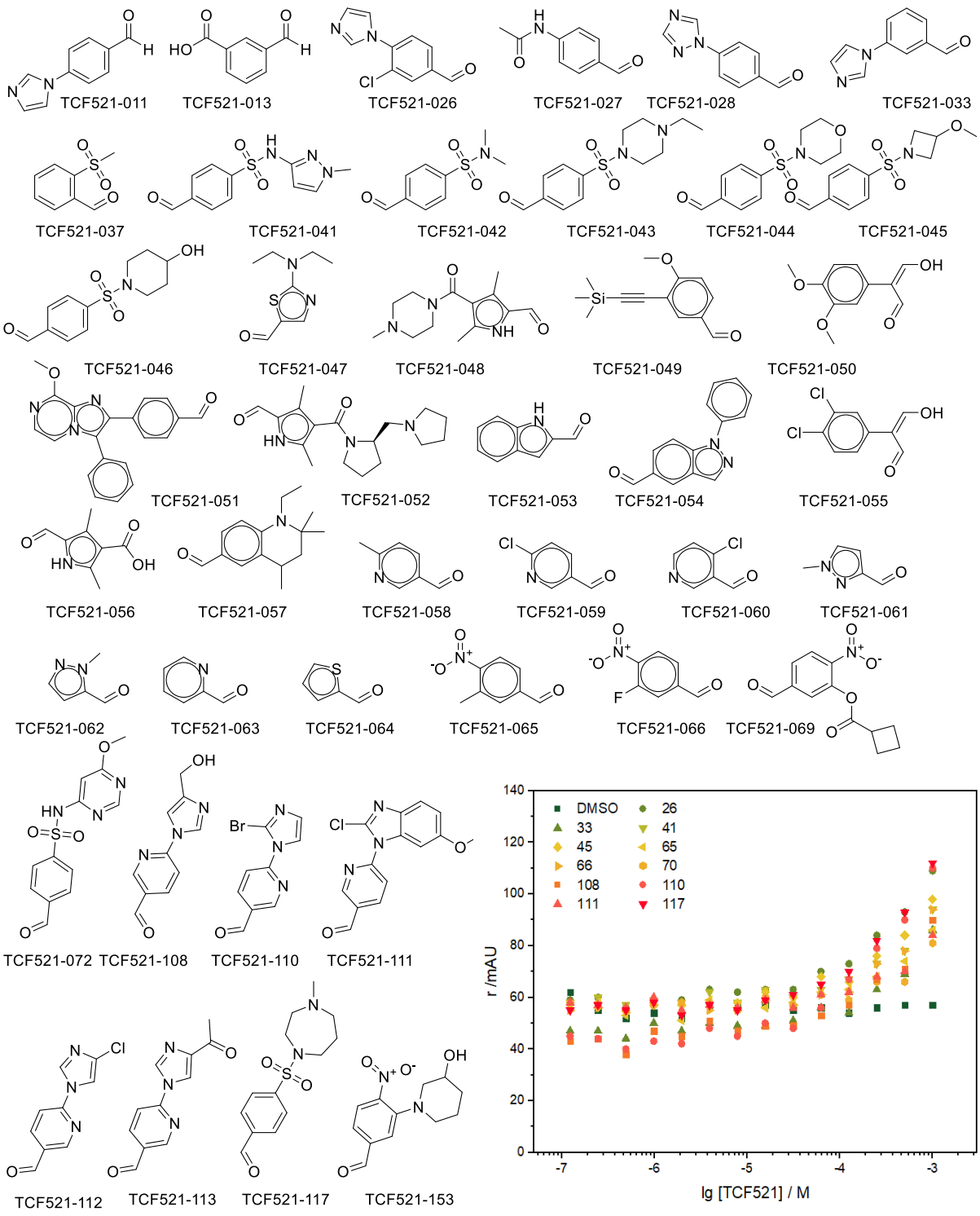


Figure S1: Imine tethering screen of an aldehyde library revealed 11 hit fragments. Stabilization of the Pin1_72/14-3-3σ complex was tested with FA compound titrations in presence of 50 µM 14-3-3σ and 100 nM Pin1_72.

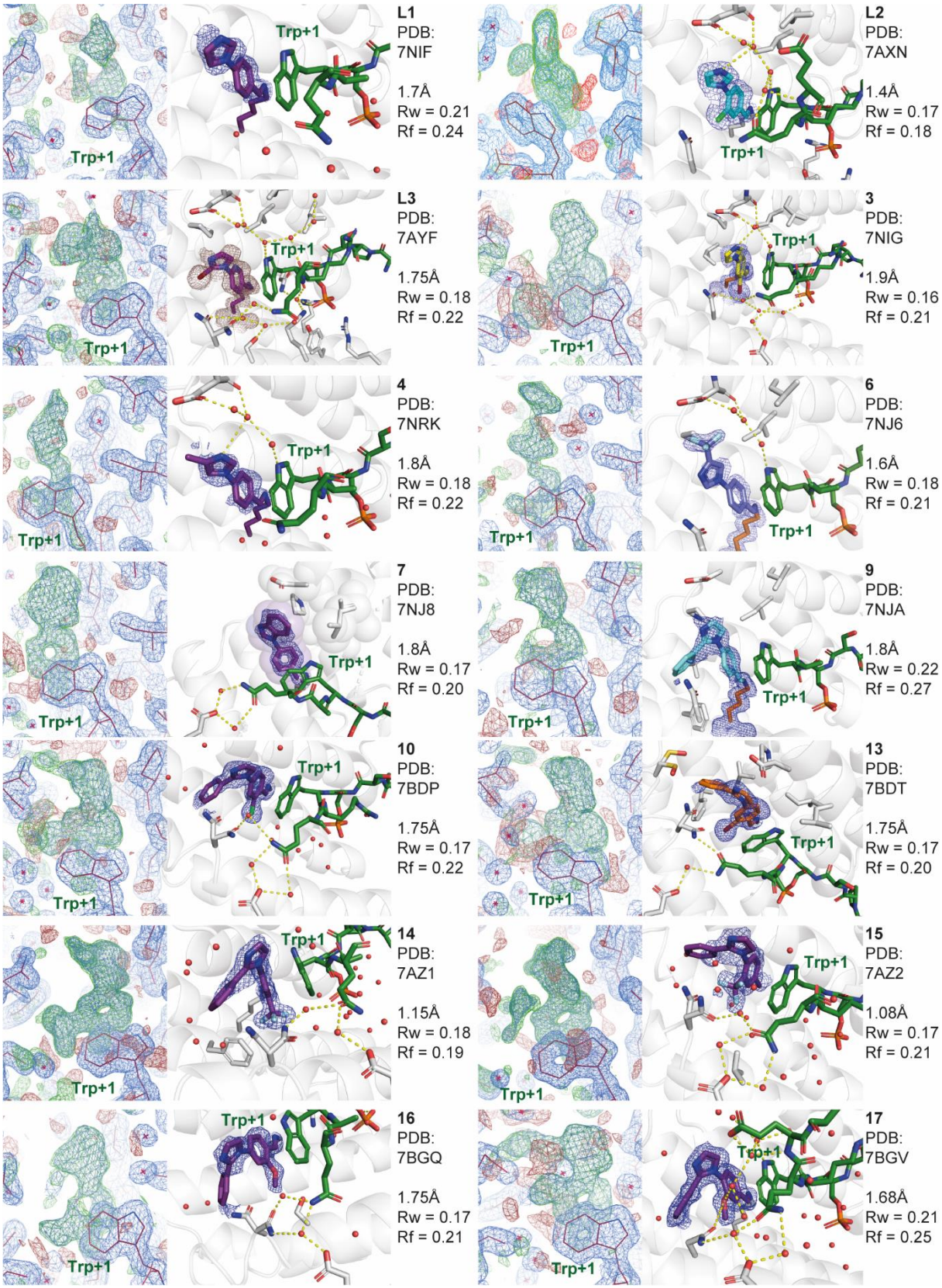


Figure S2 (continued on next page).

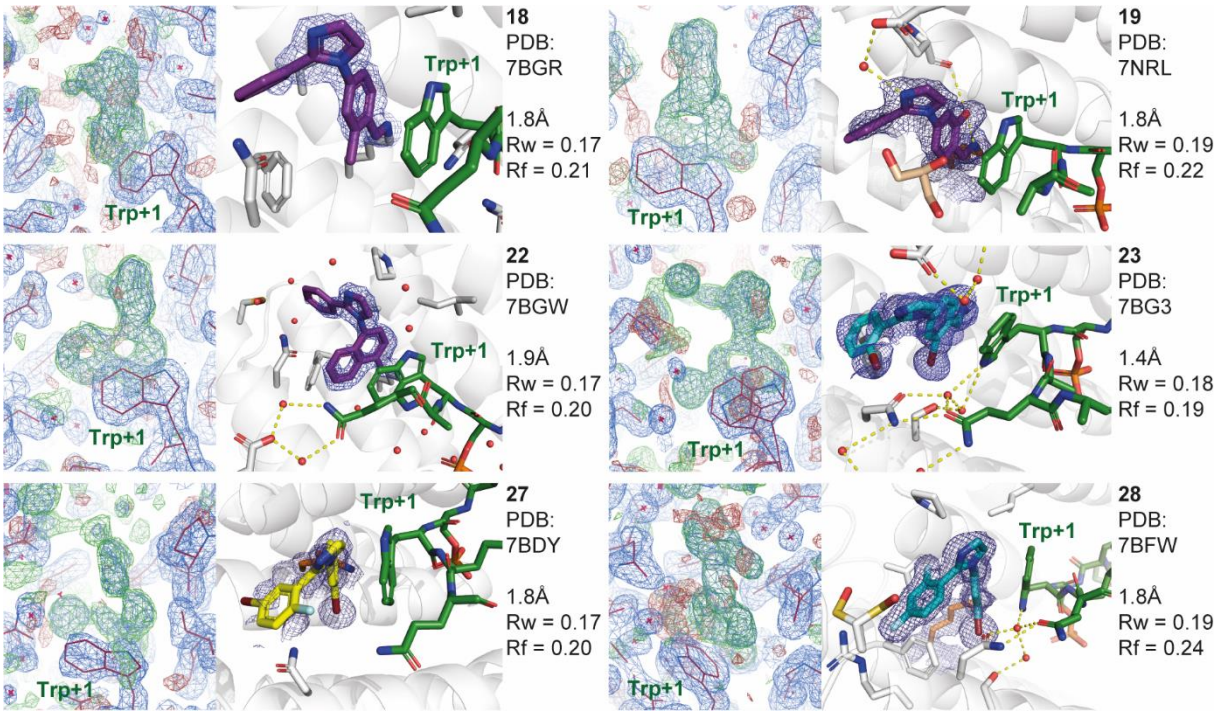


Figure S2: Ternary crystal structures of fragment/Pin1_72/14-3-3σΔC. Chemical structure and biophysical activity of the fragments are listed in Table 1 and additional crystallographic statistics are listed in Table S3. Indicated are the unbiased electron density in absence of fragment coordinates as provided by coot (left, blue 2Fo-Fc map: rmsd=1, green/red Fo-Fc map: rmsd= 2.5), the binding pose of the in-built fragment (middle, colour legend see below) and the compound number, PDB ID, high-resolution limit, R_{work} (Rw) and R_{free} (Rf) (right). 14-3-3σΔC: white cartoon and sticks; Pin1_72: green cartoon; water: red spheres; hydrogen bonds: yellow dashes. 2Fo-Fc electron density map at 1σ: blue mesh.

Table S3: Data collection and refinement statistics for ternary Pin1_72/14-3-3σΔC/fragment complex as indicated. Data collection statistics, as reported by aimless, the refinement statics were derived from the 'Table1' software of the phenix suite. Values in parenthesis represent the highest resolution shell.

Compound number	**L1**	**L2**	**L3**	**3**	**4**	**6**
PDB code	7NIF	7AXN	7AYF	7NIG	7NRK	7NJ6
X-ray source	homesource	DLS, I03	homesource	homesource	homesource	homesource
Data Collection						
Wavelength	1.54187	0.97625	1.54187	1.54187	1.54187	1.54187
Resolution range	33.19-1.71 (1.74-1.71)	66.16-1.4 (1.42-1.40)	33.16-1.75 (1.78-1.75)	41.68-1.90 (1.93-1.90)	33.98-1.75 (1.78-1.75)	33.16-1.59 (1.61-1.59)
Space group	C2221	C2221	C2221	C2221	C2221	C2221
Unit cell						
a, b, c (Å)	82.39 112.05 62.53	82.03 111.94 62.47	82.31 112.00 62.67	82.09 111.58 62.69	82.25 111.96 62.45	82.33 111.91 62.59
α, β, γ (°)	90.00 90.00 90.00	90.00 90.00 90.00	90.00 90.00 90.00	90.00 90.00 90.00	90.00 90.00 90.00	90.00 90.00 90.00
Unique reflections	29170 (761)	54952 (2050)	28956 (1089)	22988 (1103)	28225 (1321)	32592 (99)
Multiplicity	5.8 (3.5)	1.9 (1.9)	6.0 (3.7)	6.3 (5.7)	6.1 (3.5)	5.6 (1.1)
Completeness (%)	92.6 (48.7)	96.6 (73.8)	98.1 (74.0)	99.9 (97.5)	96.0 (82.3)	82.5 (5.1)
Mean I/sigma(I)	5.7 (1.1)	16.2 (2.1)	7.9 (1.8)	16.5 (5.9)	13.2 (2.3)	14.1 (1.7)
R-merge	0.210 (0.779)	0.014 (0.241)	0.161 (0.641)	0.082 (0.286)	0.099 (0.468)	0.082 (0.222)
R-meas	0.231 (0.919)	0.020 (0.341)	0.176 (0.738)	0.089 (0.315)	0.109 (0.548)	0.089 (0.314)
R-pim	0.129 (0.588)	0.014 (0.241)	0.070 (0.356)	0.035 (0.131)	0.043 (0.279)	0.036 (0.222)
CC1/2	0.987 (0.549)	1.000 (0.870)	0.993 (0.673)	0.998 (0.956)	0.997 (0.838)	0.998 (0.927)
Refinement						
Reflections used in refinement	29124 (1693)	54936 (4297)	28932 (2351)	22968 (2228)	28193 (2430)	32565 (483)
Reflections used for R-free	1461 (83)	2722 (234)	1482 (119)	1185 (103)	1409 (121)	1628 (25)
R-work	0.2109 (0.2671)	0.1655 (0.2436)	0.1752 (0.2182)	0.1649 (0.1739)	0.1829 (0.2411)	0.1814 (0.2495)
R-free	0.2392 (0.3134)	0.1822 (0.2639)	0.2176 (0.2674)	0.2053 (0.2133)	0.2157 (0.2693)	0.2070 (0.3067)
Number of non-hydrogen atoms	2239	2403	2297	2261	2200	2283
macromolecules	1883	2045	1904	1924	1865	1918
ligands	15	18	14	16	29	17
solvent	341	340	379	321	306	348
RMS (bonds)	0.004	0.015	0.006	0.005	0.004	0.003
RMS (angles)	0.69	1.37	0.76	0.81	0.7	0.61
Ramachandran favored (%)	97.76	97.84	98.21	98.21	98.2	98.2
Ramachandran allowed (%)	2.24	2.16	1.79	1.79	1.8	1.8
Ramachandran outliers (%)	0	0	0	0	0	0
Rotamer outliers (%)	0	0	0	0	0	0
Clashscore	3.76	1.73	1.06	2.88	1.35	1.84
Average B-factor	14.89	21.94	15.03	14.89	14.77	15.37
macromolecules	12.84	20.1	12.51	12.86	12.73	13.13
ligands	33.17	25.73	29.04	21.47	35.66	39.9
solvent	25.37	32.78	27.17	26.69	25.22	26.5

Table S3 (continued): Data collection and refinement statistics for ternary Pin1_72/14-3-3σΔC/fragment complex as indicated. Data collection statistics, as reported by aimless, the refinement statics were derived from the 'Table1' software of the phenix suite. Values in parenthesis represent the highest resolution shell.

Compound number	**7**	**9**	**10**	**13**	**14**	**15**
PDB code	7NJ8	7NJA	7BDP	7BDT	7AZ1	7AZ2
X-ray source	homesource	homesource	homesource	homesource	DLS, I03	DLS, I03
Data Collection						
Wavelength	1.54187	1.54187	1.54187	1.54187	0.97627	0.97627
Resolution range	34.07-1.80 (1.83-1.80)	25.58-1.75 (1.78-1.75)	41.63-1.75 (1.78-1.75)	33.97-1.75 (1.78-1.75)	45.44-1.15 (1.17-1.15)	66.24-1.08 (1.10-1.08)
Space group	C2221	C2221	C2221	C2221	C2221	C2221
Unit cell						
a, b, c (Å)	82.39 112.24 62.58	82.40 112.10 62.62	81.99 111.69 62.46	82.10 111.95 62.57	82.25 111.75 62.46	82.14 112.02 62.46
α, β, γ (°)	90.00 90.00 90.00	90.00 90.00 90.00	90.00 90.00 90.00	90.00 90.00 90.00	90.00 90.00 90.00	90.00 90.00 90.00
Unique reflections	26776 (1261)	28879 (1275)	28431 (1383)	26997 (888)	101961 (4964)	119445 (4601)
Multiplicity	6.2 (5.1)	5.5 (3.3)	6.1 (3.3)	6.4 (5.2)	1.9 (2.0)	1.9 (2.0)
Completeness (%)	98.4 (92.4)	97.5 (78.6)	97.1 (86.8)	91.9 (56.6)	100.0 (99.6)	97.4 (76.9)
Mean I/sigma(I)	17.7 (4.8)	5.5 (1.1)	17.3 (2.8)	16.2 (3.3)	11.8 (1.5)	14.1 (1.2)
R-merge	0.069 (0.292)	0.231 (0.737)	0.074 (0.401)	0.086 (0.449)	0.017 (0.386)	0.027 (0.684)
R-meas	0.075 (0.324)	0.254 (0.871)	0.080 (0.476)	0.094 (0.496)	0.024 (0.546)	0.039 (0.967)
R-pim	0.041 (0.185)	0.104 (0.449)	0.032 (0.251)	0.036 (0.205)	0.017 (0.386)	0.027 (0.684)
CC1/2	0.999 (0.952)	0.983 (0.453)	0.999 (0.836)	0.998 (0.865)	1.000 (0.737)	0.999 (0.425)
Refinement						
Reflections used in refinement	26759 (2512)	28844 (2401)	28411 (2540)	26989 (1826)	101942 (10061)	119245 (9834)
Reflections used for R-free	1356 (127)	1472 (138)	1427 (124)	1349 (90)	5038 (481)	5967 (514)
R-work	0.1709 (0.1856)	0.2204 (0.3435)	0.1725 (0.2553)	0.1694 (0.2143)	0.1827 (0.2843)	0.1846 (0.3308)
R-free	0.2006 (0.2225)	0.2659 (0.4053)	0.2164 (0.3028)	0.2015 (0.2572)	0.1925 (0.2969)	0.1973 (0.3373)
Number of non-hydrogen atoms	2278	2244	2247	2297	2380	2364
macromolecules	1908	1880	1899	1941	2011	1987
ligands	18	19	20	20	25	25
solvent	352	345	328	336	344	352
RMS(bonds)	0.008	0.007	0.006	0.006	0.005	0.005
RMS(angles)	0.95	0.88	0.8	0.82	0.76	0.73
Ramachandran favored (%)	98.2	98.2	98.21	98.21	97.8	98.22
Ramachandran allowed (%)	1.8	1.8	1.79	1.79	2.2	1.78
Ramachandran outliers (%)	0	0	0	0	0	0
Rotamer outliers (%)	0.5	0	0	0	0.47	0
Clashscore	2.65	3.22	3.98	3.37	3	2.28
Average B-factor	15.97	14.03	16.83	16.82	19.5	21.12
macromolecules	13.74	11.81	14.8	14.75	17.24	18.61
ligands	26.85	36.59	21.6	23.34	26.15	40.63
solvent	27.49	24.85	28.31	28.38	32.24	33.92

Table S3 (continued): Data collection and refinement statistics for ternary Pin1_72/14-3-3σΔC/fragment complex as indicated. Data collection statistics, as reported by aimless, the refinement statics were derived from the 'Table1' software of the phenix suite. Values in parenthesis represent the highest resolution shell.

Compound number	**16**	**17**	**18**	**19**	**22**	**23**
PDB code	7BGQ	7BGV	7BGR	7NRL	7BGW	7BG3
X-ray source	homesource	homesource	homesource	homesource	homesource	DESY, Petra III, P11
Data Collection						
Wavelength	1.54187	1.54187	1.54187	1.54187	1.54187	1.0332
Resolution range	34.36-1.75 (1.78-1.75)	41.26-1.68 (1.71-1.68)	34.32-1.8 (1.84-1.8)	41.26-1.80 (1.85-1.80)	34.42-1.90 (1.94-1.90)	66.29-1.4 (1.42-1.40)
Space group	C2221	C2221	C2221	C2221	C2221	C2221
Unit cell						
a, b, c (Å)	82.30 111.93 62.43	82.51 112.53 62.78	82.19 111.68 62.39	82.53 112.48 62.52	82.45 111.79 62.56	82.15 112.21 62.80
α, β, γ (°)	90.00 90.00 90.00	90.00 90.00 90.00	90.00 90.00 90.00	90.00 90.00 90.00	90.00 90.00 90.00	90.00 90.00 90.00
Unique reflections	29095 (1337)	31746 (1012)	26985 (1548)	27292 (1947)	22305 (1370)	57149 (2617)
Multiplicity	5.9 (3.4)	5.4 (1.8)	6.1 (4.8)	6.0 (4.5)	6.5 (6.2)	12.1 (7.1)
Completeness (%)	98.8 (82.8)	94.8 (61.5)	99.9 (98.1)	99.8 (97.7)	96.4 (92.5)	99.6 (93.8)
Mean I/sigma(I)	15.6 (4.0)	5.7 (0.9)	16.8 (4.2)	12.1 (3.2)	13.3 (7.2)	21.7 (2.5)
R-merge	0.070 (0.278)	0.145 (0.646)	0.076 (0.316)	0.100 (0.410)	0.094 (0.362)	0.056 (0.692)
R-meas	0.076 (0.323)	0.175 (0.912)	0.083 (0.355)	0.109 (0.464)	0.102 (0.394)	0.058 (0.692)
R-pim	0.030 (0.161)	0.096 (0.644)	0.033 (0.159)	0.044 (0.212)	0.039 (0.154)	0.016 (0.251)
CC1/2	0.999 (0.936)	0.991 (0.681)	0.998 (0.933)	0.997 (0.895)	0.997 (0.876)	0.999 (0.847)
Refinement						
Reflections used in refinement	29049 (2543)	31700 (2300)	26955 (2636)	27208 (2608)	22296 (2109)	57119 (5412)
Reflections used for R-free	1454 (129)	1653 (132)	1359 (149)	1377 (119)	1148 (94)	2838 (287)
R-work	0.1713 (0.2091)	0.2130 (0.2863)	0.1699 (0.2155)	0.1860 (0.2473)	0.1723 (0.2152)	0.1770 (0.2486)
R-free	0.2076 (0.2533)	0.2456 (0.3039)	0.2110 (0.2506)	0.2249 (0.2953)	0.2030 (0.2399)	0.1896 (0.2593)
Number of non-hydrogen atoms	2310	2250	2230	2304	2236	2353
macromolecules	1926	1888	1898	1909	1863	2022
ligands	24	23	21	84	26	22
solvent	360	339	311	311	347	309
RMS(bonds)	0.006	0.002	0.006	0.012	0.006	0.006
RMS(angles)	0.79	0.45	0.77	1.13	0.76	0.79
Ramachandran favored (%)	98.21	98.23	98.21	98.21	97.76	97.37
Ramachandran allowed (%)	1.79	1.77	1.79	1.79	2.24	2.63
Ramachandran outliers (%)	0	0	0	0	0	0
Rotamer outliers (%)	0	0	0	0	0	0
Clashscore	2.6	1.86	3.97	6.16	1.62	2.74
Average B-factor	16.39	17.06	16.47	16.66	14.32	21.34
macromolecules	13.99	15.23	14.47	13.98	12.12	19.65
ligands	31.53	19.6	31.96	42.24	26.41	29.21
solvent	28.26	27.06	27.64	26.14	25.22	31.87

Table S3 (continued): Data collection and refinement statistics for ternary Pin1_72/14-3-3σΔC/fragment complex as indicated. Data collection statistics, as reported by aimless, the refinement statics were derived from the 'Table1' software of the phenix suite. Values in parenthesis represent the highest resolution shell.

Compound Number	**27**	**28**
PDB code	7BDY	7BFW
X-ray source	homesource	homesource
Data Collection		
Wavelength	1.54187	1.54187
Resolution range	41.15-1.8 (1.85-1.8)	40.98-1.8 (1.85-1.80)
Space group	C2221	C2221
Unit cell		
a, b, c (Å)	82.19 111.89 62.49	81.96 112.15 62.53
α, β, γ (°)	90.00 90.00 90.00	90.00 90.00 90.00
Unique reflections	26632 (1829)	26869 (1895)
Multiplicity	6.2 (5.3)	5.2 (4.1)
Completeness (%)	98.4 (93.6)	99.2 (96.7)
Mean I/sigma(I)	24.3 (8.9)	6.7 (1.3)
R-merge	0.050 (0.149)	0.153 (0.698)
R-meas	0.54 (0.165)	0.170 (0.796)
R-pim	0.021 (0.069)	0.072 (0.373)
CC1/2	0.999 (0.988)	0.989 (0.861)
Refinement		
Reflections used in refinement	26620 (2496)	26833 (2557)
Reflections used for R-free	1347 (124)	1320 (131)
R-work	0.1658 (0.1909)	0.1898 (0.3161)
R-free	0.1991 (0.2517)	0.2405 (0.3619)
Number of non-hydrogen atoms	2340	2385
macromolecules	1950	1978
ligands	24	23
solvent	366	384
RMS(bonds)	0.007	0.007
RMS(angles)	0.81	0.82
Ramachandran favored (%)	98.24	98.25
Ramachandran allowed (%)	1.76	1.75
Ramachandran outliers (%)	0	0
Rotamer outliers (%)	0	0
Clashscore	2.58	3.56
Average B-factor	14.23	13
macromolecules	11.61	10.92
ligands	63.79	12.03

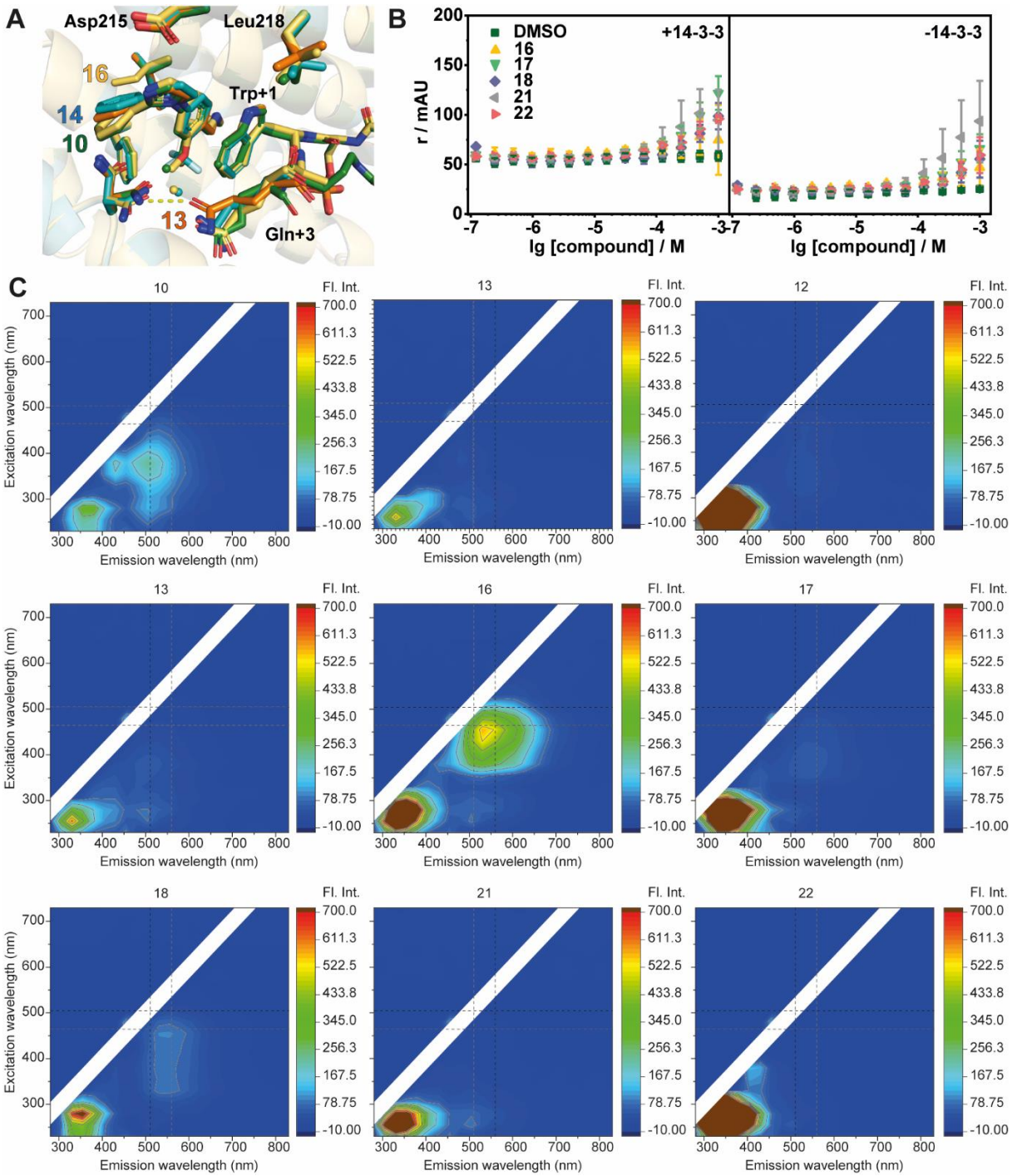


Figure S3: Details of fragments listed in Table 1. (A) Overlay of fragments **10** (green sticks), **13** (orange sticks), **14** (teal sticks) and **16** (yellow sticks) in complex with Pin1_72/14-3-3σΔC. **(B)** FA compound titrations show assay interference for **10, 16, 18, 21, 22** as the anisotropy increases in the absence of 14-3-3γ (-14-3-3) to a similar extent as with 14-3-3γ (+14-3-3). **(C)** Auto-fluorescence of compounds. Excitation/emission spectra of the fragments at a concentration of 1 mM. Spectra were measured for an $\lambda_{excitation}$-range of 230 to 730 nm and an $\lambda_{emission}$-range of 280 to 830 nm.

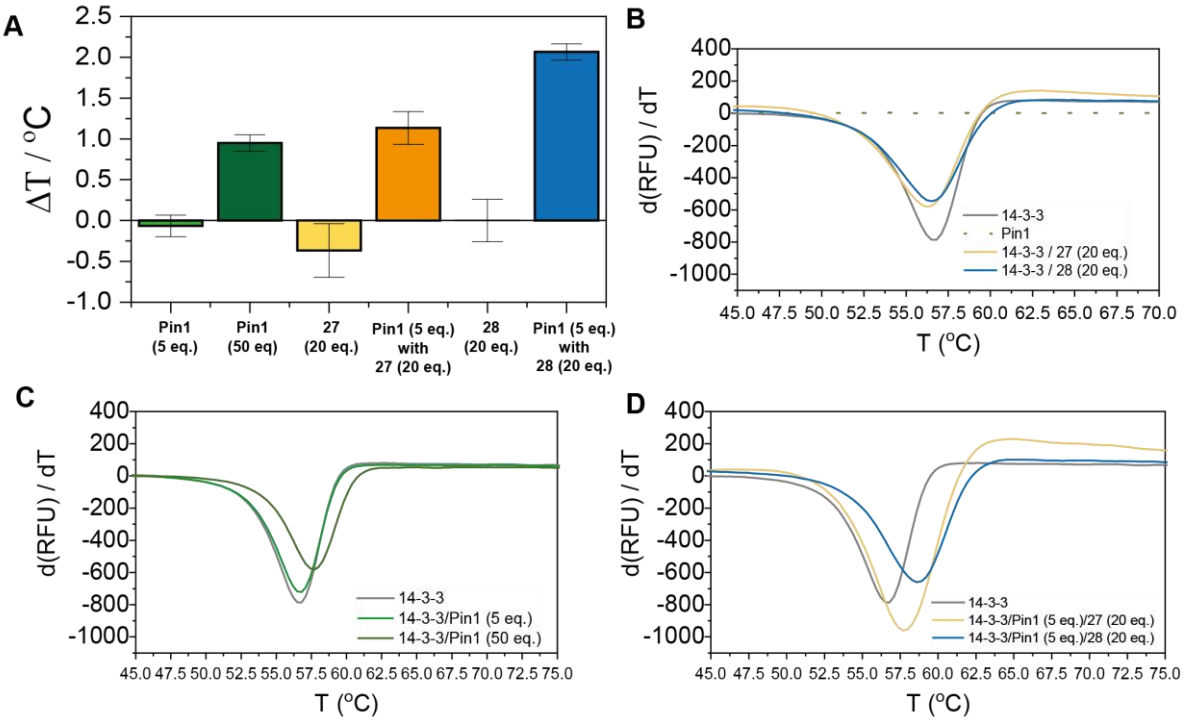


Figure S4: Thermal shift experiment with 14-3-3γ. (**A)** Shown are the changes in melting temperature of 14-3-3γ with addition of 5 equivalents (eq.) of Pin1 peptide (light green), 50 eq. Pin1 peptide (Dark green) and 20 eq. Fragment 27 (yellow). Notably, addition of 20 eq. of **28** resulted in no change in melting temperature. Additionally, change in melting temperature for 14-3-3/Pin1/**27** (orange) and14-3-3/Pin1/**28** (blue) is also shown. Values are given as the mean average and standard deviations (n=3) of three separate experiments performed using technical triplicates. (**B**) Differential melting curve of 14-3-3γ, Pin1 peptide, 20 eq. of **27** or **28** in the presence of 14-3-3γ. (**C**) Differential melting curve of 14-3-3γ, binary 14-3-3/Pin1 complex at 5 and 50 eq of Pin1 peptide. (**D**) Differential melting curve of 14-3-3γ, ternary 14-3-3/Pin1/fragment complex with 20 eq. fragment **27** or **28**. Differential melting curve are given as the mean average of one experiment.

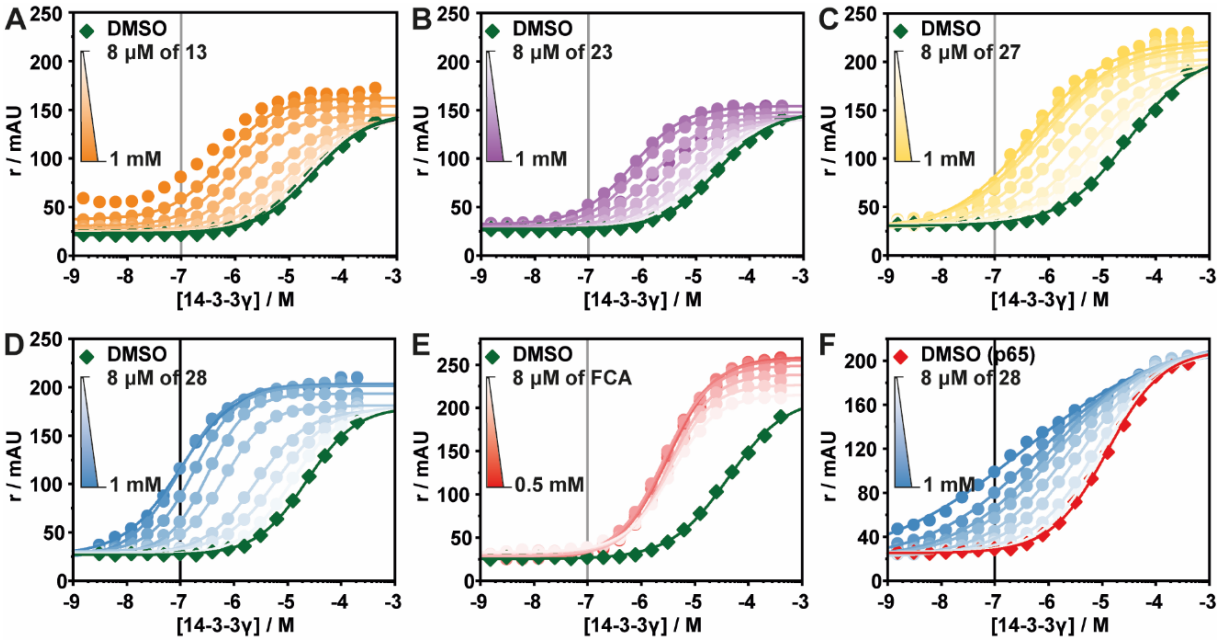


Figure S5: 2D Titrations of fragment **13, 23, 27** and **28** with Pin1_72 or p65_45 peptide and 14-3-3γ. Concentrations of fragment and 14-3-3 as indicated and 100nM Pin1_72 (n=1). (**A**) 2D titration of **13** with Pin1_72. (**B**) 2D titration of **14** with Pin1_72. (**C**) 2D titration of **27** with Pin1_72. (**D**) 2D titration of **28** with Pin1_72. (**E**) 2D titration of FCA with Pin1_72. (**F**) 2D titration of **28** with p65_45.

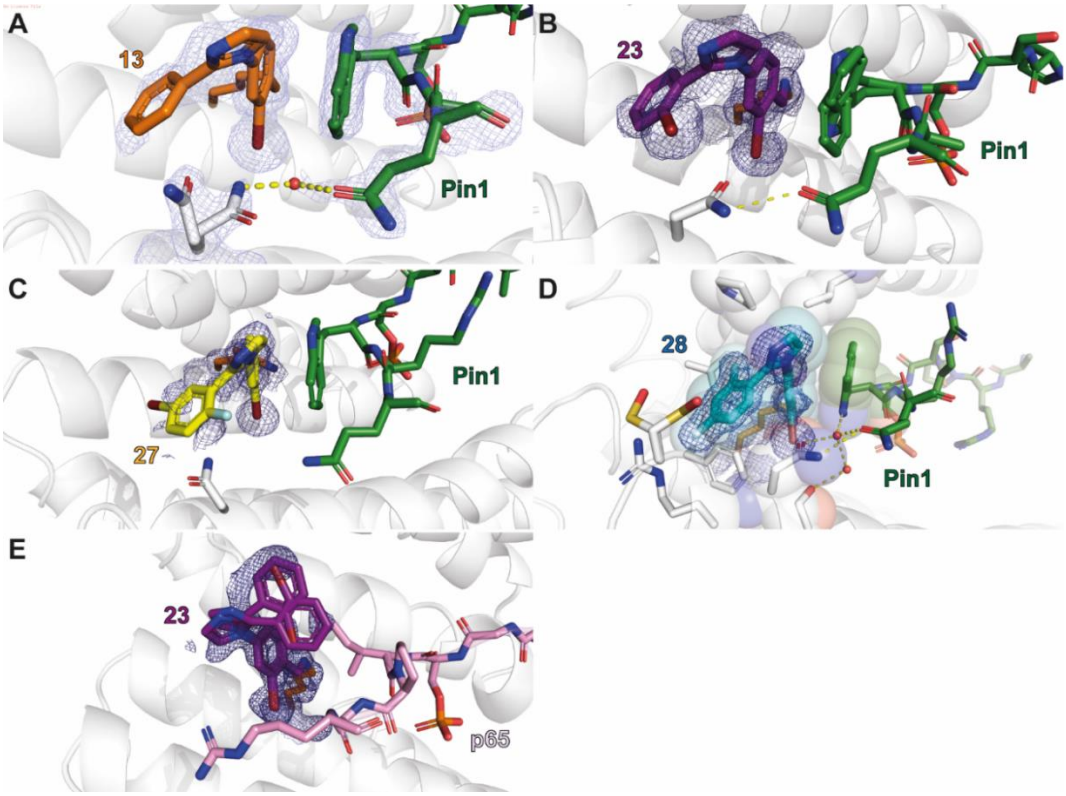


Figure S6: Ternary structures of **13**, **23**, **27** and **28** in complex with 14-3-3σΔC and Pin1_72 or p65. (A) Fragment **13** (orange sticks) with Pin1_72 (green sticks). (B) Fragment **23** (violet sticks) with Pin1_72 (green sticks). (C) Fragment **27** (Cyan sticks) with Pin1_72 (green sticks). (D) Fragment **28** (yellow sticks) with Pin1_72 (green sticks). 14-3-3σΔC is shown as white cartoon and sticks; the Pin1_72 is displayed as green cartoon. (E) Fragment 23 (violet sticks) with p65 (rosa sticks). The 2Fo-Fc electron density map (blue mesh) is contoured at 1σ.

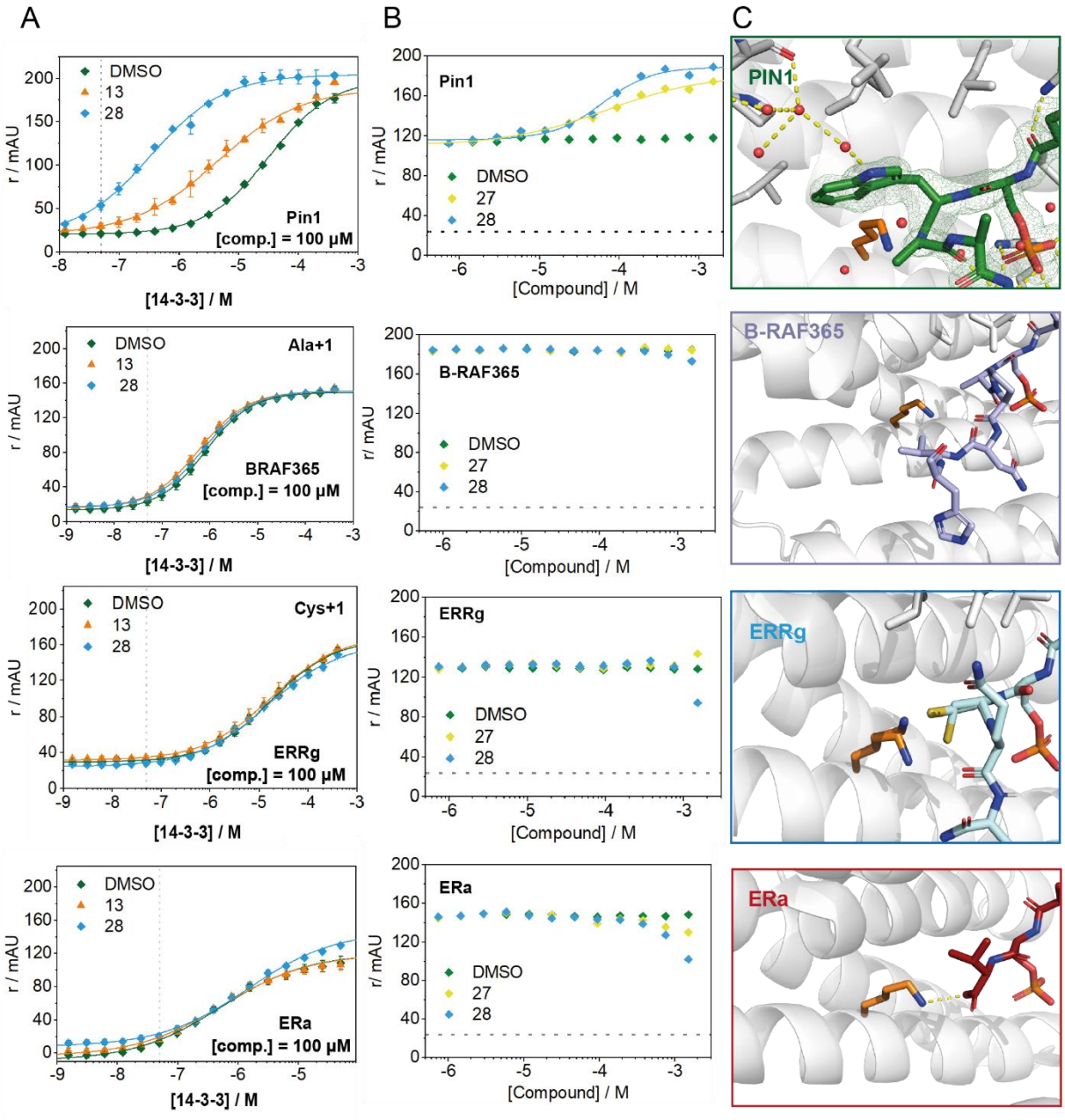


Figure S7: Competition assay. (A) Protein titration of 14-3-3 with 50 nM of Pin1, BRaf pS365, ERRγ or ER α and 100 µM of **13** or **28**. **(B)** Compound Titration after overnight incubation of 14-3-3γ and **27** or **28**, followed by addition of 100 nM of Pin1, BRaf pS365, ERRγ or ER α. The grey dotted line indicates the anisotropy of 100% free peptide. **(C)** Enlarged view of the interface of the binary 14-3-3/partner peptide complex for Pin1, BRaf pS365, ERRγ or ER α.

Table S4: Data collection and refinement statistics for the 13 and 23 in complex with p65_45 and 14-3-3σΔC. Data collection statistics, as reported by aimless, the refinement statics were derived from the 'Table1' software of the phenix suite. Values in parenthesis represent the highest resolution shell.

Compound number	**13**	**23**
PDB code	7NQP	7NSV
X-ray source	DESY, Petra III, P11	DESY, Petra III, P11
Data Collection		
Wavelength	1.0332	1.0332
Resolution range	66.38-1.24 (1.27-1.24)	66.35-1.33 (1.36-1.33)
Space group	C2221	C2221
Unit cell		
a, b, c (Å)	82.396 112.035 62.607	82.91 112.16 62.64
α, β, γ (°)	90 90 90	90 90 90
Unique reflections	81921 (5898)	66729 (4896)
Multiplicity	12.0 (8.1)	9.2 (9.1)
Completeness (%)	99.8 (97.8)	100 (100)
Mean I/sigma(I)	11.1 (1.0)	9.9 (1.1)
R-merge	0.102 (1.734)	0.103 (1.369)
R-meas	0.106 (1.851)	0.109 (1.452)
R-pim	0.030 (0.630)	0.036 (0.479)
CC1/2	0.998 (0.636)	0.998 (0.722)
Refinement		
Reflections used in refinement	81852 (7959)	66692 (6591)
Reflections used for R-free	4082 (403)	3339 (309)
R-work	0.1946 (0.3485)	0.2020 (0.3558)
R-free	0.2161 (0.3573)	0.2130 (0.3742)
Number of non-hydrogen atoms	2209	2164
macromolecules	1913	1887
ligands	20	46
solvent	276	231
RMS(bonds)	0.01	0.007
RMS(angles)	1.13	0.92
Ramachandran favored (%)	98.66	98.17
Ramachandran allowed (%)	1.34	1.83
Ramachandran outliers (%)	0	0
Rotamer outliers (%)	0.51	0
Clashscore	4.2	4.99
Average B-factor	20.27	20.12
macromolecules	18.32	18.33
ligands	33.3	51.15
solvent	32.79	28.57

Experimental Section – Biophysical Assays

Protein Expression and Purification

The 14-3-3 proteins were recombinantly expressed in BL21(DE3) cells using pPROEX HTb vectors encoding for the 14-3-3σΔC (ΔC17 truncated C-terminus) and 14-3-3γ isoforms and TB medium. At a culture density of OD_{600}=0.8-1, protein expression was initiated with 0.4mM IPTG for 18h at 18°C. The cells were isolated by centrifugation (10.000 xg, 15 min) and resuspended in lysis buffer (50 mM Tris/HCl pH8, 300 mM NaCl, 12.5 mM imidazole, 2 mM β-mercaptoethanol). A homogenizer was utilized for cell lysis, followed by centrifugation (40.000 xg, 30min) to clear the lysate. The proteins were purified using standard protocols for Ni-NTA-columns. The proteins were eluted with 250 mM imidazole (50 mM Tris/HCl pH8, 300 mM NaCl, 250 mM imidazole, 2 mM β-mercaptoethanol) and the full length 14-3-3γ was dialysis against 25mM HEPES pH7.5, 100mM NaCl, 10mM $MgCl_2$, 0.5mM Tris(2-carboxyethyl)phosphine) and stored at -80°C. The 14-3-3σΔC for crystallography required removal of the His6-tag by TEV protease; the TEV was removed with Ni-NTA-columns. To ensure highest purity, the 14-3-3σΔC was applied to a size exclusion chromatography (20 mM HEPES pH7.5, 150 mM NaCl, 2 mM β-mercaptoethanol) and stored at -80°C.

X-Ray Crystallography

All binary crystals prepared by mixing 12 mg/ml 14-3-3σΔC in a 1:2 ratio with acetylated peptide in 20 mM HEPES pH7.5, 2 mM $MgCl_2$, 2 mM β-mercaptoethanol, followed by overnight incubation. Pin1_72/14-3-3σΔC crystals were grown in a hanging drop set up, whereby the complexation solution was mixed in 1:1 ratio with precipitation buffer (95 mM HEPES pH7.1, 27-28% PEG400, 190 mM $CaCl_2$, 5% glycerol). B-Raf/14-3-3σΔC and Abl1/14-3-3σΔC crystals were grown in a sitting drop set up. The complexation solution was mixed in 1:1 ratio with precipitation buffer (95 mM HEPES pH7.5, 27-28% PEG400, 190 mM $CaCl_2$, 5% glycerol). For AS160/14-3-3σΔC crystals the complexation solution was mixed in a 1:1 ration with the Wizard Cryo™ crystallization screen (Rigaku, Bainbridge Island, US), resulting in crystal growth with 40% (v/v) MPD and 100 mM CHES/ Sodium hydroxide pH 9.5. All crystals were directly flash-frozen in liquid nitrogen and data acquisition took place at either the P11 beamline of PetraIII (DESY campus, Hamburg, Germany) or i-03/i-24 beamline of the diamond light source (Oxford, UK) or in-house.

Fragment screening was performed by crystal soaking, whereby a final concentration of 10 mM fragment was added to fully grown crystals (final DMSO ≤1%). The fragment/crystal mixtures incubated for seven days prior to data acquisition at the diamond light source (beamline i03, Oxford, UK), DESY (P11, Hamburg, Germany) or a homesource as indicated. The diffraction data were analyzed with the xia2/DIALS pipeline[1] and MolRep[2,3] was used for phasing. For solving the Pin1/14-3-3 and AS160/14-3-3 structures the PDB entry 4JDD served as search model. The search model for fragment soaks was the correlating binary structure (Pin1/14-3-3: 7AOG, p65/14-3-3: 6QHL). For model refinement Coot[4], Refmac5[5] and phenix.refine[6] were utilized in iterative cycles. The elbow software of the phenix suite[6] was used for ligand preparation based on fragment SMILES. Figures were generated with PyMOL© (V2.0.6, Schrodinger LLC).

Fluorescence Anisotropy (FA) Assays

Dissociation constants of binary complex formation were measured with a 1:1 dilution series of 14-3-3γ in the presence of 50 nM fluorescently labeled peptide (Table S5). Stabilization factors (SF) were measured by a 1:1 dilution series of 14-3-3γ in the presence of 50 nM fluorescently labeled peptide and 100 μM compound (or as indicated) or DMSO as control, with $SF = K_{D,DMSO}/K_{D,compound}$. For compound titrations, constant 10 μM of 14-3-3γ and 50 nM

of fluorescently labeled Pin1_72 peptide was used, whereby the compound was titrated in a 1:1 dilution series. All FA assays were measured in FA buffer (10 mM HEPES pH 7.4, 150 mM NaCl, 0.1% Tween20, 1% BSA) in Corning 384 well plates (black, round bottom, low binding). Plates were incubated overnight prior to anisotropy measurements with the Tecan Infinite 500 plate reader ($\lambda_{excitation}$ = 485 nm, $\lambda_{emission}$ = 535 nm).

Table S5: Overview of utilized peptide epitopes. Given are the names as mentioned in the main text, the binding site, the N-terminal modifications with either a fluorophore-linker construct or an acetylation (ace.) for crystallography, the binding sequence and the reference.

Name	Binding Site	N-term. modification	Binding Epitope	Reference/ Purchased
Pin1_71	pS71	FITC-Ahx	LVKHSQSRRP pS SWRQEK	Genscript
Pin1_72	pS72	FITC-Ahx/ ace.	LVKHSQSRRPS pS WRQEK	Genscript
p65	pS45pS281	FITC-βAla	EGRSAG pS IPGRRSGSGGGSGPSDREL pS EPMEFQ	[7]
p65_45	pS45	ace.	EGRSAG pS IPGRRS	[7]
B-Raf	pS729	FITC-Ahx	IHRSA pS EPSLN	Genscript
B-Raf	pS365	FITC-Ahx	RDRSS pS APNVH	Genscript
C-Raf	pT259	FITC-βAla	SQRQRST pS TPNVH	[8]
AS160	pT642	FITC-Ahx/ ace.	RRRAH pT FSHPP	Genscript
Abl1	pT735	FITC-Ahx	EWRSV pT LPRDL	Genscript
CFTR	pS753pS568	FITC-βAla	AILPRI pS VISTGPTLQARRRQ pS VLNLMT	[9]
Raptor	pS792	FITC-Ahx	MRRAS pS YSSLN	Genscript
ERα	pT594	FITC-O1Pen	AEGFPA pT V-COOH	[8]
Mypt1	pS472	FITC-βAla	GVTRSA pS SPRLSS	'Synthesized in-house'
ERRγ	pS179	FITC-Ahx	KRRRK pS CQA	[10]
TBC1D	pS237	FITC-Ahx	MRKSF pS QPGLR	Genscript

Excitation/Emission Scans

Excitation/Emission profiles of fragments were measured at a final concentraion of 1 mM in PBS buffer (137 mM NaCl, 2.7 mM KCl, 10 mM Na_2HPO_4, 1.8 mM KH_2PO_4). Measurements were performed with a Tecan Safire 2 plate reader ($\lambda_{excitation}$ = 230-730 nm; $\lambda_{emission}$ = 280-830 nm; step size: 25 nm; gain: 60; lagtime: 0 μs) in a Corning 384-well plate (black, round bottom, low volume, low binding).

Experimental Section – Chemistry

General materials

All reactions were prepared using AR or HPLC grade solvents without further purification. All reagents were purchase from Fluorochem, ABCR, Ak Scientific or Simga-Adrich and were used without further purification unless stated. Microwave reactions were performed using a Biotage Initiator Plus equipped with a handling robot. Solvents were removed in vacuo using a Büchi rotary evaporator and a diaphragm pump. DMF and CH_2Cl_2 were dried and purified by means of a MBRAUN Solvent Purification System (MB-SPS-800). All other solvents used were of chromatography or analytical grade and supplied by Biosolve or Sigma-Aldrich. TLC was carried out on aluminum-backed silica (Merck silica gel 60 F254) plates supplied by Merck. Visualization of the plates was achieved using an ultraviolet lamp (λ_{max} = 254 nm), 2,4-DNP, $KMnO_4$, anisaldehyde, bromine or ninhydrin. Column chromatography was either

performed manually using silica gel (60–63 um particle size), automated Grace Reveleris X2 or Biotage Isolera chromatograph with prepacked silica columns supplied by Buchi/Grace (40 µm particle size). LC–MS analysis was carried out with a system comprising a Phenomex kinetex® 2.6 µm EVO C18 50 x 2.1 mm column using ultrapure water with 0.1% formic acid (FA) and acetonitrile with 0.1% FA, in general with using a gradient of 5–100% MeCN in water (+ 0.1% HCOOH) over 10 min, connected to a Thermo Fisher LCQ Fleet Ion Trap Mass Spectrometer. The purity of the samples was assessed using a PDA and MS. Unless otherwise stated all final compounds were ≥95% pure as judged by HPLC. GCMS analysis was performed on a Phenomenex Zebron ZB-5MS 30 m × 0.25 mm × 0.25 mm column with a gradient of 80 °C for 1 min to 300 °C for 1 min with a rate of 30 °C/min in helium gas connected to a GCMS-QP2010 Plus Quadrupole Mass Spectrometer. High resolution mass spectra (HRMS) were recorded using a Waters ACQUITY UPLC I-Class LC system coupled to a Xevo G2 Quadrupole Time of Flight (Q-tof) mass spectrometer equipped with a Phenomex kinetex® 2.6 µm EVO C18 100 x 2.1 mm column. Proton (^{1}H) and carbon (^{13}C) NMR spectral data were collected on a 400 MHz Bruker Cryomagnet or 400 MHz Varian Gemini. Chemical shifts (δ) are quoted in parts per million (ppm) and referenced to the residual solvent peak. Coupling constants (*J*) are quoted in Hertz (Hz) and splitting patterns reported in an abbreviated manner: app. (apparent), s (singlet), d (doublet), t (triplet), q (quartet), and m (multiplet). Assignments were made with the aid of 2D COSY, HMQC, and HMBC experiments.

Synthetic Procedures and Characterization

General Procedure 1

To a microwave reaction tube was added 4-fluorobenzaldehyde derivative (1 eq), imidazole derivative (1.1 eq) and potassium carbonate (1.5 eq) in 2 mL of DMF. The reaction mixture was subject to microwave irradiation at 120 °C for 15 min. To the resulting reaction mixture was added water (50 mL) and was extracted with ethyl acetate (2 x 50 mL). The organic layers were combined, washed with water (3 x 100 mL) and brine (100 mL). The organic layer was then separated, dried over sodium sulphate and concentrated under vacuum. The resulting crude residue was then subject silica column chromatography (gradient; hexane/EtOAc) to afford the titled compound.

General Procedure 2

To a microwave reaction tube was added 4-fluorobenzaldehyde derivative (1 eq), imidazole derivative (1.1 eq) and potassium carbonate (1.5 eq) in 2 mL of DMF. The reaction mixture was subject to microwave irradiation at 120 °C for 15 min. To the resulting reaction mixture was added water (10 mL) and the reaction mixture was subject to 2 min of ultra-sonication. The resulting precipitate was then filtered under vacuum, washed with water (2x 3 mL) and dried under vacuum to afford the titled compound

Compound Characterization

3-bromo-4-(1H-imidazol-1-yl)benzaldehyde (1)

Fragment **1** was synthesized according to general synthesis procedure 2 using 3-bromo-4-fluorobenzaldehyde (203 mg, 1.00 mmol), K_2CO_3 (207 mg, 1.50 mmol) and imidazole (75 mg, 1.10 mmol) to afford an amorphous cream solid (120 mg, 48%); LRMS (ESI+) 250 *m/z* (M+H); HRMS (ESI) calcd for $C_{10}H_8BrN_2O$ (M+H), 250.9815; found 250.9810; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.1 (s, 1H), 8.4 (d, *J* = 1.7 Hz, 1H), 8.0 (dd, *J* = 8.1, 1.8 Hz, 1H), 8.0 (s, 1H), 7.7 (d, *J* = 8.0 Hz, 1H), 7.5 (s, 0H), 7.1 (s, 1H); ^{13}C NMR (101 MHz, DMSO) δ 191.8, 141.3, 138.1, 137.5, 135.2, 129.7, 129.6, 129.5, 121.3, 120.0.

2-chloro-4-(1H-imidazol-1-yl)benzaldehyde (2)
Fragment **2** was synthesized according to general synthesis procedure 1 using 2-chloro-4-fluorobenzaldehyde (159 mg, 1.00 mmol), K_2CO_3 (207 mg, 1.50 mmol) and imidazole (75 mg, 1.10 mmol) to afford an amorphous cream solid (38 mg, 18%); LRMS (ESI+) 207 *m/z* (M+H); HRMS (ESI) calcd for CH_8ClN_2O (M+H), 207.0325; found 207.0323; ^{1}H NMR (400 MHz, Acetone-d6) δ 10.42 (s, 1H), 8.34 (s, 1H), 8.03 (d, *J* = 8.5 Hz, 1H), 7.94 (d, *J* = 2.1 Hz, 1H), 7.89 – 7.76 (m, 2H), 7.17 (s, 1H); ^{13}C NMR (100 MHz, Acetone) δ 188.7, 143.2, 139.4, 136.6, 132.0, 131.9, 131.4, 122.5, 119.9, 118.4.

2-bromo-4-(1H-imidazol-1-yl)benzaldehyde (3)
Fragment **3** was synthesized according to general synthesis procedure 2 using 2-bromo-4-fluorobenzaldehyde (100 mg, 0.49 mmol), K_2CO_3 (74.9 mg, 0.54 mmol) and imidazole (36.9 mg, 0.54 mmol) to afford an amorphous cream solid (91 mg, 74%); LRMS (ESI+) 250 *m/z* (M+H); HRMS (ESI) calcd for $C_{10}H_8BrN_2O$ (M+H), 250.9815; found 250.9813; ^{1}H NMR (400 MHz, Acetone) δ 8.34 (s, 1H), 8.11 (d, *J* = 2.2 Hz, 1H), 8.01 (d, *J* = 8.5 Hz, 1H), 7.88 (dd, *J* = 8.5, 2.1 Hz, 1H), 7.81 (s, 1H), 7.17 (s, 1H), 2.05 (p, *J* = 2.2 Hz, 3H); ^{13}C NMR (101 MHz, Acetone) δ 190.7, 143.2, 136.6, 132.5, 132.2, 132.0, 128.2, 125.7, 120.5, 118.4.

4-(4-methyl-1H-imidazol-1-yl)benzaldehyde (4)
Fragment **4** was synthesized according to general synthesis procedure 1 using 4-fluorobenzaldehyde (200 mg, 1.61 mmol), K_2CO_3 (245 mg, 1.77 mmol) and 4-methylimidazole (146 mg, 1.77 mmol). The reaction mixture was diluted with water (50 mL) and was extracted with ethylacetate (2x50 mL). The resulting mixture was washed with sodium chloride solution (100 mL). The material was absorbed to silica and subject to automated column chromatography (0 – 100% Hexane:EtOAc) to afford the titled compound as cream solid (70 mg, 21.3%); LRMS (ESI+) *m/z* 187 (M+H); HRMS (ESI) calcd for $C_{11}H_{11}N_2O$ (M+H), 187.0871; found 187.0869; ^{1}H NMR (400 MHz, Acetone) δ 10.05 (s, 1H), 8.15 (s, 1H), 8.05 (d, *J* = 8.7 Hz, 2H), 7.82 (d, *J* = 8.6 Hz, 2H), 7.44 (s, 1H), 2.21 (s, 3H). ^{13}C NMR (101 MHz, Acetone) δ 191.7, 142.7, 140.9, 135.6, 135.5, 132.2(2C), 120.9 (2C), 114.6, 13.9
NB: structural isomer [4-(3-methyl-1H-imidazol-1-yl)benzaldehyde] observed as a 7.4% impurity (based on proton NMR).

4-(4-(trifluoromethyl)-1H-imidazol-1-yl)benzaldehyde (6)
Fragment **6** was synthesized according to general synthesis procedure 1 using 4-fluorobenzaldehyde (200 mg, 1.61 mmol), K_2CO_3 (245 mg, 1.17 mmol) and 4-(trifluoromethyl)-1H-imidazol (241 mg, 1.17 mmol) to afford an amorphous cream solid (109 mg, 39%); LRMS (ESI+) 241 *m/z* (M+H); HRMS (ESI) calcd for $C_{11}H_8F_3N_2O$ (M+H), 241.0586; found 241.0589; ^{1}H NMR (400 MHz, Acetone-d6) δ 10.11 (s, 1H), 8.42 (s, 1H), 8.31 (s, 1H), 8.13 (d, *J* = 8.6 Hz, 2H), 7.98 (d, *J* = 8.6 Hz, 2H); ^{13}C NMR (101 MHz, Acetone) δ 191.8, 141.6, 138.1, 136.8, 134.1, (q, *J* = 38.5 Hz), 132.1 (2C), 122.8, (q, *J* = 266.3), 122.5 (2C), 119.5 (q, *J* = 4.0 Hz).

4-(1H-benzo[d]imidazol-1-yl)benzaldehyde (7)
Fragment **7** was synthesized according to general synthesis procedure 2 using using 4-fluorobenzaldehyde (100 mg, 0.81 mmol), K_2CO_3 (123 mg, 0.89 mmol) and benzimidazole (105 mg, 0.89 mmol) to afford the titled compound as a brown solid (45 mg, 25%); LRMS (ESI+) *m/z* 223 (M+H); HRMS (ESI) calcd for $C_{14}H_{11}N_2O$ (M+H), 223.0871; found 223.0860; ^{1}H NMR (400 MHz, Chloroform-d) δ 10.11 (s, 1H), 8.19 (s, 1H), 8.12 (d, *J* = 8.5 Hz, 2H), 7.90 (dt, *J* = 7.1, 3.6 Hz, 1H), 7.74 (d, *J* = 8.4 Hz, 2H), 7.62 (dt, *J* = 6.7, 3.5 Hz, 1H), 7.43 – 7.34

(m, 2H); ^{13}C NMR (100 MHz, CDCl3) δ 190.8, 144.5, 141.9, 141.5, 135.4, 133.1(2C), 131.8, 124.4, 123.9(2C), 123.6, 121.2, 110.6.

1-(4-formylphenyl)-1H-imidazole-2-carboxylic acid (8)
Fragment **2** was synthesized according to general synthesis procedure 2 using 2-chloro-4-fluorobenzaldehyde (124 mg, 1.00 mmol), K_2CO_3 (207 mg, 1.50 mmol) and imidazole (123 mg, 1.10 mmol) to afford the titled compound as an off-white amorphous solid (18 mg, 8 %); LRMS (ESI+) 217 *m/z* (M+H); HRMS (ESI) calcd for $C_{11}H_8N_2O_3$ (M+H), 217.0608; found 217.0612; ^{1}H NMR (400 MHz, Acetone-d_6) δ 10.08 (s, 1H), 8.27 (s, 1H), 8.09 (d, *J* = 8.4 Hz, 2H), 7.89 (d, *J* = 8.4 Hz, 2H), 7.17 (s, 1H); ^{13}C NMR (100 MHz, Acetone) δ 192.7, 143.6, 137.4, 136.8, 133.1 (2C), 132.7, 122.4 (2C), 119.3.

4-(2-phenyl-1H-imidazol-1-yl)benzaldehyde (9)
Fragment **9** was synthesized according to general synthesis procedure 2 using 4-fluorobenzaldehyde (100 mg, 0.81 mmol), K_2CO_3 (122.5 mg, 0.89 mmol) and 2-phenylimidazole (127.8 mg, 0.89 mmol) to afford the titled compound as an amorphous yellow oil (0.8 mg, 0.4%); LRMS (ESI+) *m/z* 249 (M+H); HRMS (ESI) calcd for $C_{16}H_{13}N_2O$ (M+H), 248.1028; found 249.1024; ^{1}H NMR (400 MHz, DMSO-d6) δ 10.04 (s, 1H), 7.98 (d, *J* = 8.4 Hz, 2H), 7.61 (d, *J* = 1.3 Hz, 1H), 7.58 – 7.46 (m, 2H), 7.46 – 7.27 (m, 5H), 7.24 (d, *J* = 1.3 Hz, 1H). ^{13}C NMR (101 MHz, DMSO) δ 192.7, 146.3, 143.2, 135.7, 131.2 (2C), 130.6, 129.7, 129.0, 128.90 (2C), 128.85 (2C), 126.9 (2C), 123.9.

2-chloro-4-(2-phenyl-1H-imidazol-1-yl)benzaldehyde (10)
Fragment **10** was synthesized according to general synthesis procedure 2 using 3-chloro-4-fluorobenzaldehyde (100 mg, 0.81 mmol), K_2CO_3 (116 mg, 0.84 mmol) and 2-phenylimidazole (90.1 mg, 0.63 mmol) to afford the titled compound as a brown solid (45 mg, 20%); LRMS (ESI+) *m/z* 283 (M+H); HRMS (ESI) calcd for $C_{16}H_{12}ClN_2O$ (M+H), 283.0638; found 283.0639; ^{1}H NMR (400 MHz, Acetone) δ 10.41 (s, 1H), 7.92 (d, *J* = 8.3 Hz, 1H), 7.57 (d, *J* = 2.0 Hz, 1H), 7.51 (d, *J* = 1.4 Hz, 1H), 7.42 (dd, *J* = 7.5, 2.1 Hz, 2H), 7.39 (d, *J* = 2.0 Hz, 1H), 7.38 – 7.29 (m, 3H), 7.21 (d, *J* = 1.4 Hz, 1H). ^{13}C NMR (101 MHz, Acetone) δ 189.0, 147.3, 144.8, 138.5, 132.5, 131.4, 131.2, 130.5, 129.7(2C), 129.5, 129.2(2C), 128.4, 126.0, 123.6.

3-chloro-4-(2-phenyl-1H-imidazol-1-yl)benzaldehyde (11)
Fragment **11** was synthesized according to general synthesis procedure 2 using 3-chloro-4-fluorobenzaldehyde (159 mg, 1.00 mmol), K_2CO_3 (207 mg, 1.50 mmol) and 2-phenylimidazole (159 mg, 1.10 mmol) to afford an amorphous cream solid (31 mg, 11%); LRMS (ESI+) *m/z* 283 (M+H); HRMS (ESI) calcd for $C_{16}H_{12}ClN_2O$ (M+H), 283.0638; found 283.0647; ^{1}H NMR (400 MHz, Acetone-d_6) δ 10.12 (s, 1H), 8.12 (d, *J* = 1.7 Hz, 1H), 8.04 (dd, *J* = 8.0, 1.7 Hz, 1H), 7.77 (d, *J* = 8.1 Hz, 1H), 7.39 (dd, *J* = 7.4, 2.0 Hz, 2H), 7.35 (d, *J* = 1.3 Hz, 1H), 7.31 – 7.21 (m, 4H); ^{13}C NMR (100 MHz, Acetone) δ 191.2, 147.8, 142.1, 138.9, 133.4, 132.0, 131.61, 131.59, 130.3, 129.7, 129.3, 129.2 (2C), 128.5 (2C), 123.8.

3-bromo-4-(2-phenyl-1H-imidazol-1-yl)benzaldehyde (12)
Fragment **12** was synthesized according to general synthesis procedure 2 using 3-bromo-4-fluorobenzaldehyde (100 mg, 0.49 mmol), K_2CO_3 (75 mg, 0.54 mmol) and 2-phenylimidazole (78 mg, 0.54 mmol) to afford the titled compound as an amorphous brown oil (9.7 mg, 6%); LRMS (ESI+) *m/z* 327 (M+H); HRMS (ESI) calcd for $C_{16}H_{12}BrN_2O$ (M+H), 327.0133; found 327.0139; ^{1}H NMR (400 MHz, $CDCl_3$) δ 10.44 (s, 1H), 9.35 (d, *J* = 8.6 Hz, 1H), 8.00 (d, *J* = 7.5 Hz, 1H), 7.76 (t, *J* = 7.6 Hz, 1H), 7.70 – 7.57 (m, 1H), 7.49 (d, *J* = 7.5 Hz, 1H), 7.30 (d, *J*

= 7.6 Hz, 2H), 7.23 – 7.01 (m, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 192.7, 135.7, 132.0, 131.7, 130.4, 130.1, 128.8, 128.7, 128.5 (2C), 127.9 (2C), 125.5, 124.7, 123.4.

2-bromo-4-(2-phenyl-1H-imidazol-1-yl)benzaldehyde (13)
Fragment **13** was synthesized according to general synthesis procedure 1 using 2-bromo-4-fluorobenzaldehyde (100 mg, 0.49 mmol), K_2CO_3 (75 mg, 0.54 mmol) and 2-phenylimidazole (78.1 mg, 0.54 mmol to afford the titled compound as a brown solid (64 mg, 40%); LRMS (ESI+) *m/z* 327 (M+H); HRMS (ESI) calcd for $C_{16}H_{12}BrN_2O$ (M+H), 327.0133; found 327.0130; ^{1}H NMR (400 MHz, Acetone-d_6) δ 10.32 (s, 1H), 7.92 (d, *J* = 8.3 Hz, 1H), 7.76 (d, 1H), 7.53 (s, 1H), 7.48 – 7.31 (m, 6H), 7.22 (s, 1H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 190.5, 147.1, 143.9, 132.7, 130.9, 130.4, 130.3, 129.7, 129.3, 129.0 (2C), 128.7 (2C), 127.5, 125.2, 122.2.

4-(2-phenyl-1H-imidazol-1-yl)-2-(trifluoromethyl)benzaldehyde (14)
Fragment **14** was synthesized according to general synthesis procedure 2 using 4-Fluoro-2-(trifluoromethyl)benzaldehyde (100 mg, 0.52 mmol), K_2CO_3 (79 mg, 0.57 mmol) and 2-phenylimidazole (82.6 mg, 0.57 mmol) to afford the titled compound as a brown solid (75 mg, 46%); LRMS (ESI+) *m/z* 317 (M+H); HRMS (ESI) calcd for $C_{16}H_{12}BrN_2O$ (M+H), 317.0902; found 317.0906. ^{1}H NMR (400 MHz, Acetone) δ 10.36 (d, *J* = 2.3 Hz, 1H), 8.19 (d, *J* = 8.3 Hz, 1H), 7.88 (s, 1H), 7.78 (d, *J* = 8.3 Hz, 1H), 7.63 (s, 1H), 7.51 – 7.31 (m, 6H), 7.27 (s, 1H); ^{13}C NMR (100 MHz, Acetone-d_6) δ 189.4 (q, *J* = 2.1 Hz), 148.3, 144.6, 134.4 (d, *J* = 1.5 Hz), 133.1, 132.7 (d, *J* = 33.1 Hz), 132.2 , 131.6, 130.7 (2C), 130.5, 130.1 (2C), 125.5 (q, *J* = 6.0 Hz), 124.5.

4-(2-phenyl-1H-imidazol-1-yl)-3-(trifluoromethyl)benzaldehyde (15)
Fragment **15** was synthesized according to general synthesis procedure 2 using 4-Fluoro-3-(trifluoromethyl)benzaldehyde (100 mg, 0.52 mmol), K_2CO_3 (96 mg, 0.69 mmol) and 2-phenylimidazole (75 mg, 0.52 mmol) to afford the titled compound as a brown solid (9 mg, 5.4%); LRMS (ESI+) *m/z* 317 (M+H); HRMS (ESI) calcd for $C_{17}H_{12}F_3N_2O$ (M+H), 317.0902; found 317.0921; ^{1}H NMR (400 MHz, DMSO-d6) δ 10.17 (s, 1H), 8.46 (d, *J* = 1.8 Hz, 1H), 8.28 (dd, *J* = 8.1, 1.8 Hz, 1H), 7.81 (d, *J* = 8.1 Hz, 1H), 7.51 (brs, 1H), 7.29 – 7.24 (m, 6H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 192.0, 147.2, 140.9 (d, *J* = 1.7 Hz, *quartet not completely resolved), 137.1, 134.4, 132.9, 130.2, 129.2, 128.9 (2C), 127.9 (2C), 127.2 (q, *J* = 31.2 Hz), 125.4, 122.9 (d, *J* = 274.2 Hz, *quartet not completely resolved).

2-methoxy-4-(2-phenyl-1H-imidazol-1-yl)benzaldehyde (16)
Fragment **16** was synthesized according to general synthesis procedure 2 using 2-methoxy-4-fluorobenzaldehyde (100 mg, 0.65 mmol), K_2CO_3 (89.7 mg, 0.65 mmol) and 2-phenylimidazole (62.4 mg, 0.43 mmol) to afford the titled compound as a brown solid (22.9 mg, 19%); LRMS (ESI+) *m/z* 279 (M+H); HRMS (ESI) calcd for $C_{17}H_{15}N_2O_2$ (M+H), 279.1134; found 279.1133; ^{1}H NMR (400 MHz, $CDCl_3$) δ 10.40 (s, 1H), 7.84 (d, *J* = 8.3 Hz, 1H), 7.39 (dd, *J* = 7.6, 2.0 Hz, 1H), 7.33 – 7.24 (m, 3H), 7.21 (s, 1H), 6.90 (dd, *J* = 8.2, 1.2 Hz, 1H), 6.72 (d, *J* = 1.9 Hz, 1H), 3.70 (s, 2H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 207.1, 188.6, 162.2, 146.9, 144.6, 130.1, 123.0, 129.8, 128.9 (2C), 128.5 (2C), 124.0, 122.2, 117.6, 109.5, 56.0.

3-methoxy-4-(2-phenyl-1H-imidazol-1-yl)benzaldehyde (17)
Fragment **17** was synthesized according to general synthesis procedure 1 using 3-methoxy-4-fluorobenzaldehyde (100 mg, 0.65 mmol), K_2CO_3 (99 mg, 0.71 mmol) and 2-phenylimidazole (103 mg, 0.71 mmol) to afford the titled compound as a brown solid (7 mg, 10%); LRMS (ESI+)

m/z 279 (M+H); HRMS (ESI) calcd for $C_{17}H_{15}N_2O_2$ (M+H), 279.1134; found 279.1144; ^{1}H NMR (400 MHz, Acetone) δ 10.07 (s, 1H), 7.65 (d, *J* = 6.6 Hz, 2H), 7.54 (d, *J* = 8.2 Hz, 1H), 7.43 – 7.36 (m, 2H), 7.33 – 7.22 (m, 4H), 7.20 (s, 1H), 3.70 (s, 3H). ^{13}C NMR (100 MHz, Acetone) δ 192.1, 155.7, 147.9, 138.9, 133.6, 132.1, 129.9, 129.5, 129.1, 128.9 (2C), 128.4 (2C), 124.1, 124.0, 112.8, 56.4.

3-methyl-4-(2-phenyl-1H-imidazol-1-yl)benzaldehyde (18)
Fragment **18** was synthesized according to general synthesis procedure 1 using 4-Fluoro-3-methylbenzaldehyde (100 mg, 0.72 mmol), K_2CO_3 (133.4 mg, 0.97 mmol) and 2-phenylimidazole (72.4 mg, 0.72 mmol) to afford the titled compound as an amorphous brown oil (12 mg, 6.6%); LRMS (ESI+) 263 *m/z* (M+H); HRMS (ESI) calcd for $C_{17}H_{15}N_2O$ (M+H), 263.1179; found 263.1185; ^{1}H NMR (400 MHz, Acetone) δ 10.09 (s, 1H), 7.91 (d, *J* = 7.1 Hz, 2H), 7.56 (d, *J* = 8.7 Hz, 1H), 7.39 (dd, *J* = 7.8, 1.9 Hz, 2H), 7.32 – 7.20 (m, 4H), 2.03 (s, 3H). ^{13}C NMR (101 MHz, Acetone) δ 192.3, 143.9, 137.8, 137.2, 133.1, 131.8, 130.3, 129.6, 129.2, 129.1 (2C), 129.0, 128.4(2C), 123.6, 17.5.

2-hydroxy-4-(2-phenyl-1H-imidazol-1-yl)benzaldehyde (19)
Boron tribromide (1 M in DCM, 5 mL) was added dropwise to a solution of 2-methoxy-4-(2-phenyl-1H-imidazol-1-yl)benzaldehyde (**16**, 30 mg, 0.11 mmol) in anhydrous DCM (3 mL). The reaction was stirred at rt overnight after addition under an argon atmosphere. The reaction was quenched using ice water, followed by additional 5 M hydrochloride solution until the pH reached 1. The product was extracted using ethylacetate (2x50 mL). The resulting organic layer was washed with saturated sodium chloride solution (100 mL). The crude residue was absorbed to silica and subject to column chromatography (0-20% EtOAc:methanol) to afford the titled compound as a yellow solid (5 mg, 17%); LRMS (ESI+) *m/z* 265 (M+H); HRMS (ESI) calcd for $C_{16}H_{13}N_2O_2$ (M+H), 265.0977; found 265.0974; ^{1}H NMR (400 MHz, Acetone-d6) δ 10.09 (s, 1H), 7.92 – 7.75 (m, 1H), 7.56 – 7.40 (m, 3H), 7.39 – 7.26 (m, 3H), 7.20 (d, J = 1.3 Hz, 1H), 7.05 – 6.81 (m, 2H).13C NMR (101 MHz, Acetone) δ 198.0, 187.1, 136.5, 132.6, 131.3, 130.4, 130.3 (2C), 130.02, 130.01 (2C), 124.5, 122.4, 122.1, 119.2, 115.8.

4-(2-phenyl-1H-imidazol-1-yl)-3-(trifluoromethoxy)benzaldehyde (20)
Fragment **20** was synthesized according to general synthesis procedure 1 using 4-Fluoro-3-(trifluoromethoxy)benzaldehyde (100 mg, 0.48 mmol), K_2CO_3 (66 mg, 0.48 mmol) and 2-phenylimidazole (46 mg, 0.32 mmol) to afford the titled compound as an amorphous brown oil (64 mg, 60%); LRMS (ESI+) *m/z* 333 (M+H); HRMS (ESI) calcd for $C_{17}H_{12}F_3N_2O_2$ (M+H), 333.0851; found 333.0837; ^{1}H NMR (400 MHz, Acetone-d_6) δ 10.15 (s, 1H), 8.10 (d, *J* = 8.1 Hz, 1H), 8.00 (s, 1H), 7.83 (d, *J* = 8.1 Hz, 1H), 7.42 (s, 1H), 7.38 (d, *J* = 5.7 Hz, 2H), 7.33 – 7.28 (m, 4H); ^{13}C NMR (101 MHz, Acetone-d_6) δ 191.1, 148.1, 144.8 (q, *J* = 1.6 Hz), 138.7, 136.8, 131.4, 131.4, 130.4, 129.9, 129.4, 129.1(2C), 128.6(2C), 124.0, 122.4, 121.0 (q, *J* = 259.2 Hz).

3-phenoxy-4-(2-phenyl-1H-imidazol-1-yl)benzaldehyde (21)
Fragment **21** was synthesized according to general synthesis procedure 1 using 3-phenoxy-4-fluorobenzaldehyde (100 mg, 0.46 mmol), K_2CO_3 (64 mg, 0.46 mmol) and 2-phenylimidazole (44.5 mg, 0.31 mmol to afford the titled compound as an amorphous brown oil (17.5 mg, 17%); LRMS (ESI+) *m/z* 341 (M+H); HRMS (ESI) calcd for $C_{22}H_{17}N_2O_2$ (M+H), 341.1290; found 341.1291; ^{1}H NMR (400 MHz, Acetone-d_6) δ 10.00 (s, 1H), 7.79 (dd, *J* = 21.9, 8.0 Hz, 2H), 7.45 (d, *J* = 7.4 Hz, 2H), 7.39 (s, 1H), 7.36 – 7.27 (m, 6H), 7.21 – 7.14 (m, 2H), 6.66 (d, *J* = 8.0 Hz, 2H); ^{13}C NMR (101 MHz, Acetone) δ 191.7, 155.5, 154.1, 148.1, 138.6, 134.8, 132.3, 131.1 (2C), 130.5, 130.0, 129.2, 129.1 (2C), 128.6 (2C), 125.8, 125.7, 124.0, 120.6(2C), 117.8.

4-(2-phenyl-1H-imidazol-1-yl)-1-naphthaldehyde (22)
Fragment **22** was synthesized according to general synthesis procedure 1 using 4-fluoro-1-naphtaldehyde (100 mg, 0.57 mmol), K_2CO_3 (87mg, 0.63 mmol) and 2-phenylimidazole (91 mg, 0.63 mmol to afford the titled compound as a yellow solid (44 mg, 26%); LRMS (ESI+) *m/z* 299 (M+H); HRMS (ESI) calcd for $C_{20}H_{15}N_2O$ (M+H), 299.1184; found 299.1183; ^{1}H NMR (400 MHz, Acetone-d_6) δ 10.52 (s, 1H), 9.35 (d, *J* = 8.6 Hz, 1H), 8.29 (d, *J* = 7.5 Hz, 1H), 7.84 – 7.77 (m, 2H), 7.66 (t, *J* = 7.7 Hz, 1H), 7.52 – 7.44 (m, 2H), 7.38 – 7.31 (m, 3H), 7.23 – 7.09 (m, 3H); ^{13}C NMR (100 MHz, Acetone) δ 193.9, 148.4, 141.7, 136.8, 133.0, 132.2, 131.4, 131.3, 130.6, 130.1, 129.3, 129.2, 129.0 (2C), 128.4 (2C), 126.0, 125.9, 125.3, 124.0,

2-bromo-4-(2-(2-bromophenyl)-1H-imidazol-1-yl)benzaldehyde (23)
Fragment **23** was synthesized according to general synthesis procedure 2 using 3-bromo-4-fluorobenzaldehyde (120 mg, 0.6 mmol), K_2CO_3 (80 mg, 0.6 mmol) and 2-(2-Bromophenyl)-1H-imidazole (90 mg, 0.4 mmol) to afford an amorphous off white solid (94 mg, 58%); LRMS (ESI+) *m/z* 407 (M+H, 100%, Br^{79}, Br^{81}); HRMS (ESI) calcd for $C_{16}H_{11}Br_2N_2O$ (M+H), 404.9238; found 404.9237; ^{1}H NMR (400 MHz, Acetone-d_6) δ 10.25 (s, 1H), 7.84 (d, *J* = 8.4 Hz, 1H), 7.67 (s, 1H), 7.65 – 7.59 (m, 5H), 7.52 (t, *J* = 7.5 Hz, 2H), 7.43 (t, *J* = 7.7 Hz, 2H), 7.37 (d, *J* = 8.3 Hz, 2H), 7.26 (s, 1H); 13C NMR (100 MHz, Acetone) δ 190.8, 146.2, 144.0, 134.0, 133.70, 133.67, 133.1, 132.2, 131.3, 130.5, 130.3, 128.7, 126.9, 124.9, 124.5, 121.9.

2-bromo-4-(2-(4-bromophenyl)-1H-imidazol-1-yl)benzaldehyde (24)
Fragment **24** was synthesized according to general synthesis procedure 2 using 3-bromo-4-fluorobenzaldehyde (300 mg, 1.5 mmol), K_2CO_3 (210 mg, 1.5 mmol) and 2-(4-Bromophenyl)-1H-imidazole (220 mg, 1.0 mmol) to afford an amorphous cream solid (19 mg, 5%); LRMS (ESI+) *m/z* 407 (M+H, 100%, Br^{79}, Br^{81}); HRMS (ESI) calcd for $C_{16}H_{11}Br_2N_2O$ (M+H), 404.9238; found 404.9229; ^{1}H NMR (400 MHz, Acetone-d6) δ 10.33 (s, 1H), 7.95 (d, J = 8.3 Hz, 1H), 7.82 (d, J = 2.0 Hz, 1H), 7.54 (d, J = 8.9 Hz, 3H), 7.47 (dd, J = 8.4, 2.0 Hz, 1H), 7.38 (d, J = 8.5 Hz, 2H), 7.23 (s, 1H); 13C NMR (100 MHz, Acetone) δ 190.9, 146.2, 144.5, 133.8, 132.4 (2C), 131.63, 131.60, 131.4 (2C), 130.7, 130.5, 127.3, 126.6, 124.1, 123.3.

2-bromo-4-(2-(4-hydroxyphenyl)-1H-imidazol-1-yl)benzaldehyde (25)
Fragment **25** was synthesized according to general synthesis procedure 1 using 3-bromo-4-fluorobenzaldehyde (300 mg, 1.5 mmol), K_2CO_3 (210 mg, 1.5 mmol) and 4-(1H-Imidazol-2-yl)phenol (160 mg, 1.0 mmol) to afford an amorphous cream solid (66 mg, 19%); LRMS (ESI+) *m/z* 343 (M+H); HRMS (ESI) calcd for $C_{16}H_{12}BrN_2O_2$ (M+H), 343.0082; found 343.0085; ^{1}H NMR (400 MHz, Acetone-d_6) δ 11.71 (s, 1H), 10.23 (s, 1H), 8.11 (d, *J* = 8.7 Hz, 2H), 7.92 (d, *J* = 8.7 Hz, 1H), 7.31 (d, *J* = 2.4 Hz, 1H), 7.30 – 7.21 (m, 4H), 7.16 (dd, *J* = 8.7, 2.4 Hz, 1H), 7.09 (s, 1H). 13C NMR (100 MHz, Acetone) δ 191.3, 164.8, 156.2, 147.1, 133.4, 131.3, 130.55, 130.45, 129.3, 128.8 (2C), 123.6, 122.5 (2C), 119.0, 118.8.

2-bromo-4-(2-(pyridin-3-yl)-1H-imidazol-1-yl)benzaldehyde (26)
Fragment **26** was synthesized according to general synthesis procedure 2 using 3-bromo-4-fluorobenzaldehyde (300 mg, 1.5 mmol), K_2CO_3 (210 mg, 1.5 mmol) and 3-(1H-Imidazol-2-yl)-pyridine (150 mg, 1.0 mmol) to afford an amorphous cream solid (120 mg, 37%); LRMS (ESI+) *m/z* 328 (M+H,); HRMS (ESI) calcd for $C_{15}H_{10}BrN_3O$ (M+H), 328.0085; found 328.0077; ^{1}H NMR (400 MHz, Acetone-d_6) δ 10.33 (s, 1H), 8.63 (d, *J* = 2.2 Hz, 1H), 8.54 (dd, *J* = 4.9, 1.6 Hz, 1H), 7.95 (d, *J* = 8.3 Hz, 1H), 7.85 (d, *J* = 2.0 Hz, 1H), 7.75 (d, *J* = 8.0 Hz, 1H), 7.60 (d, *J* = 1.3 Hz, 1H), 7.51 (dd, *J* = 8.3, 2.0 Hz, 1H), 7.34 (dd, *J* = 8.0, 4.8 Hz, 1H),

7.28 (d, *J* = 1.3 Hz, 1H). 13C NMR (100 MHz, Acetone) δ 190.0, 149.4, 149.3, 143.8, 143.4, 135.6, 133.0, 130.9, 130.8, 130.0, 126.5, 126.5, 125.8, 123.4, 123.1.

2-bromo-4-(2-(5-bromo-2-fluorophenyl)-1H-imidazol-1-yl)benzaldehyde (27)

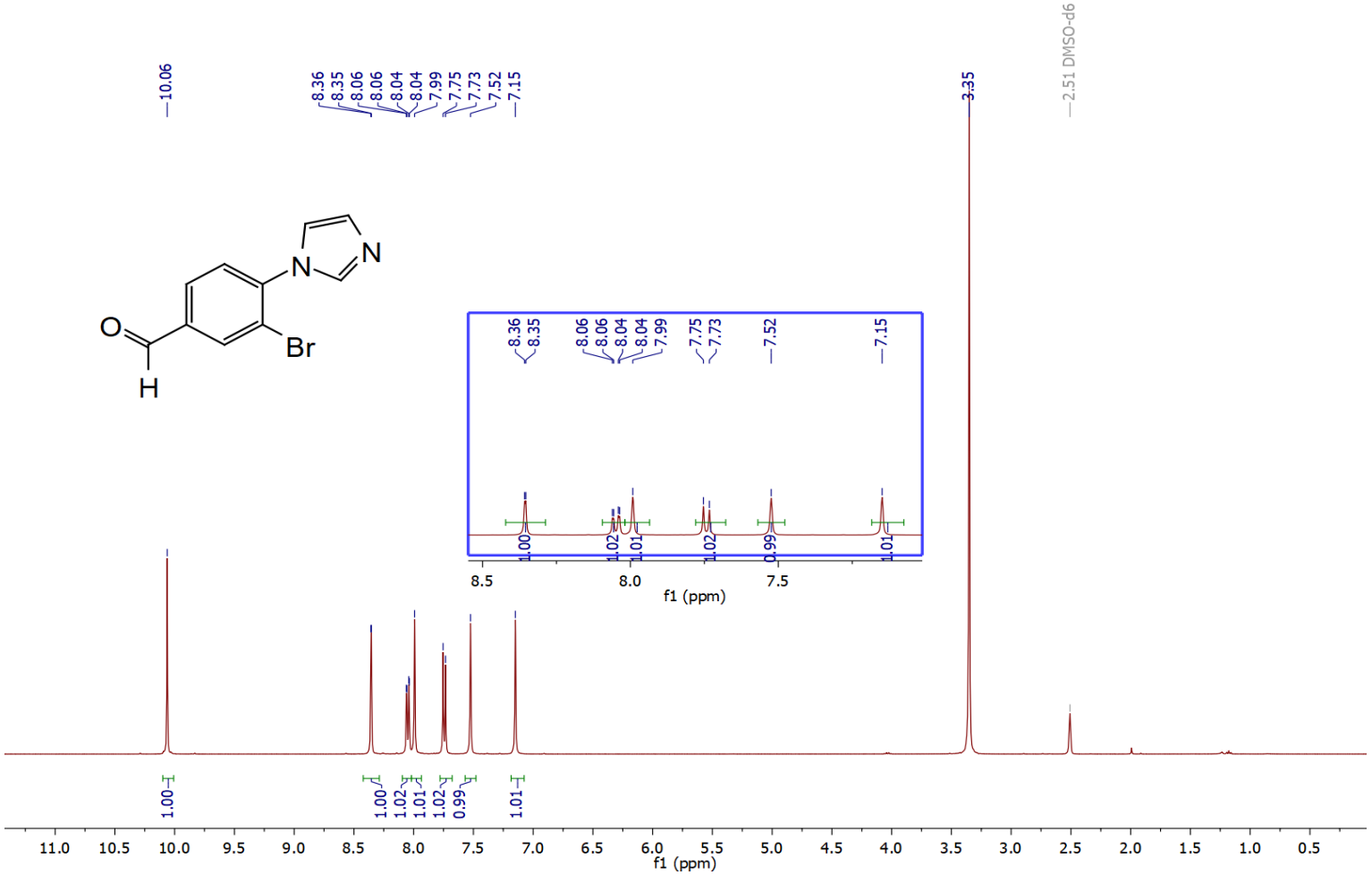
Fragment **27** was synthesized according to general synthesis procedure 1 using 3-bromo-4-fluorobenzaldehyde (203 mg, 1.0 mmol), K_2CO_3 (152 mg, 1.1 mmol) and 2-(2,4-difluorophenyl)-1H-imidazole (176 mg, 1.1 mmol) to afford an amorphous beige solid (80 mg, 19%); LRMS (ESI+) *m/z* 425 (M+H, 100%. Br^{79}, Br^{81}); HRMS (ESI) calcd for $C_{16}H_{10}Br_2FN_2O$ (M+H), 422.9144; found 422.9134; ^{1}H NMR (399 MHz, Acetone-d_6) δ 10.30 (s, 1H), 7.91 (d, *J* = 8.3 Hz, 1H), 7.87 (dd, *J* = 6.4, 2.5 Hz, 1H), 7.79 (s, 1H), 7.68 (d, *J* = 11.6 Hz, 2H), 7.47 (d, *J* = 8.3 Hz, 1H), 7.31 (s, 1H), 7.18 – 6.96 (m, 1H). ^{13}C NMR (100 MHz, Acetone-d6) δ 189.9, 159.7, 157.2, 143.3 (d, *J* = 2.2 Hz), 140.4 (d, *J* = 1.3Hz), 134.7 (d, *J* = 3.0 Hz), 134.4 (d, *J* = 8.5 Hz), 132.7, 130.5 (d, *J* = 29.6 Hz), 129.2, 126.3, 124.0, 122.7, 121.1 (d, *J* = 16.2 Hz), 117.9 (d, J = 23.6 Hz), 116.6 (d, *J* = 3.4 Hz).

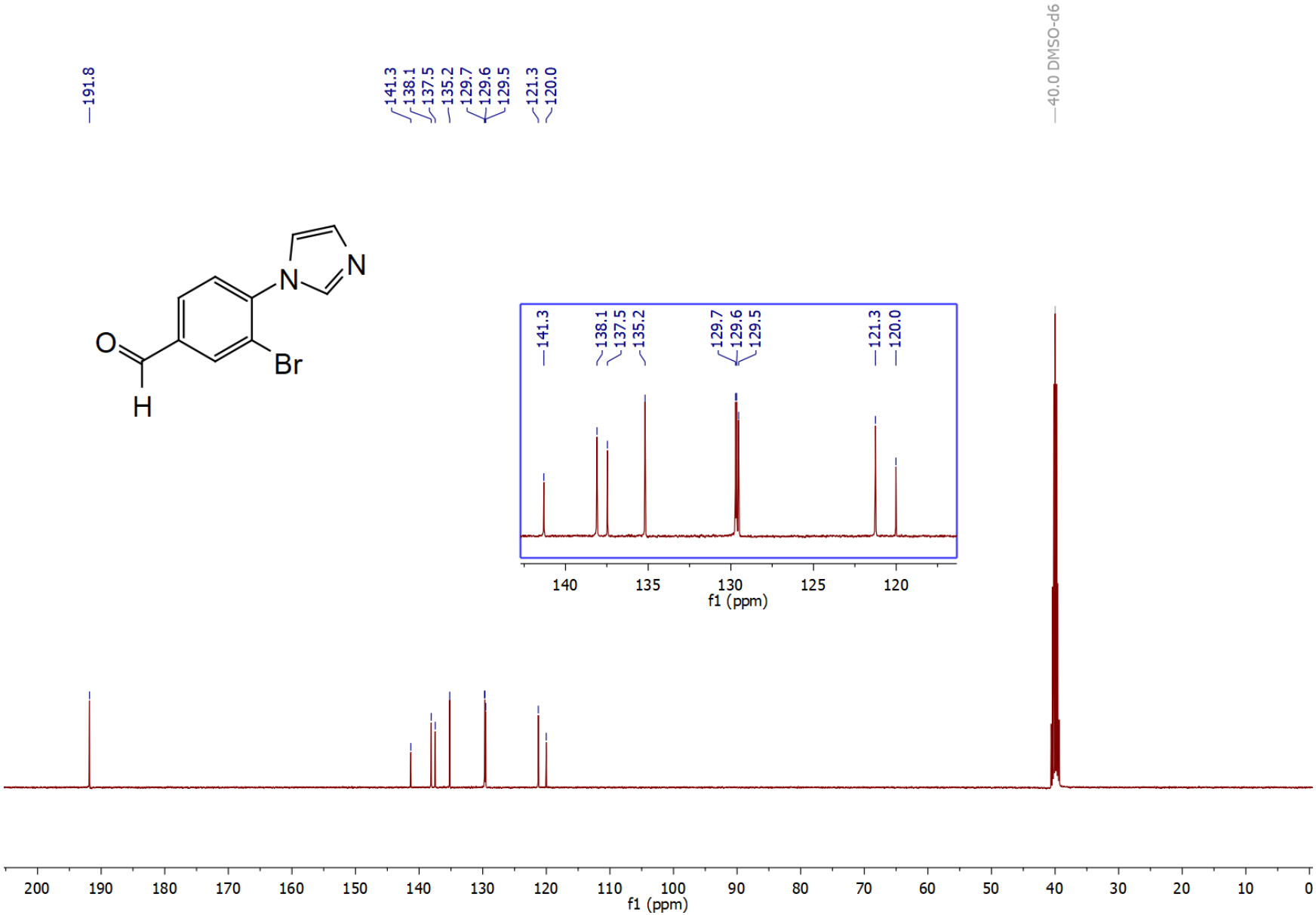
2-bromo-4-(2-(2,4-difluorophenyl)-1H-imidazol-1-yl)benzaldehyde (28)

Fragment **28** was synthesized according to general synthesis procedure 1 using 3-bromo-4-fluorobenzaldehyde (203 mg, 1.0 mmol), K_2CO_3 (152 mg, 1.1 mmol) and 2-(2,4-difluorophenyl)-1H-imidazole (198 mg, 1.1 mmol) to afford an amorphous beige solid (62 mg, 17%); LRMS (ESI+) *m/z* 363 (M+H,); HRMS (ESI) calcd for $C_{16}H_{10}BrF_2N_2O$ (M+H), 362.9945; found 362.9946; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.17 (s, 1H), 7.84 (d, *J* = 8.4 Hz, 1H), 7.78 (dd, *J* = 16.9, 1.8 Hz, 2H), 7.68 (td, *J* = 8.2, 6.3 Hz, 1H), 7.35 (ddd, *J* = 8.3, 2.1, 0.7 Hz, 1H), 7.30 (d, *J* = 1.4 Hz, 1H), 7.28 – 7.20 (m, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 190.7, 163.0 (dd, *J* = 249.4, 12.3 Hz), 159.1 (dd, *J* = 250.3, 12.8 Hz), 142.6 (d, *J* = 1.4 Hz), 140.3 (d, *J* = 1.0 Hz), 133.6 (dd, *J* = 10.0, 4.0 Hz), 132.0, 131.0, 129.8, 129.0, 125.9, 123.9, 122.7, 115.1 (dd, *J* = 14.9, 3.8 Hz), 112.4 (dd, *J* = 21.7, 3.5 Hz), 104.5 (t, *J* = 26.0 Hz).

NMR Spectra

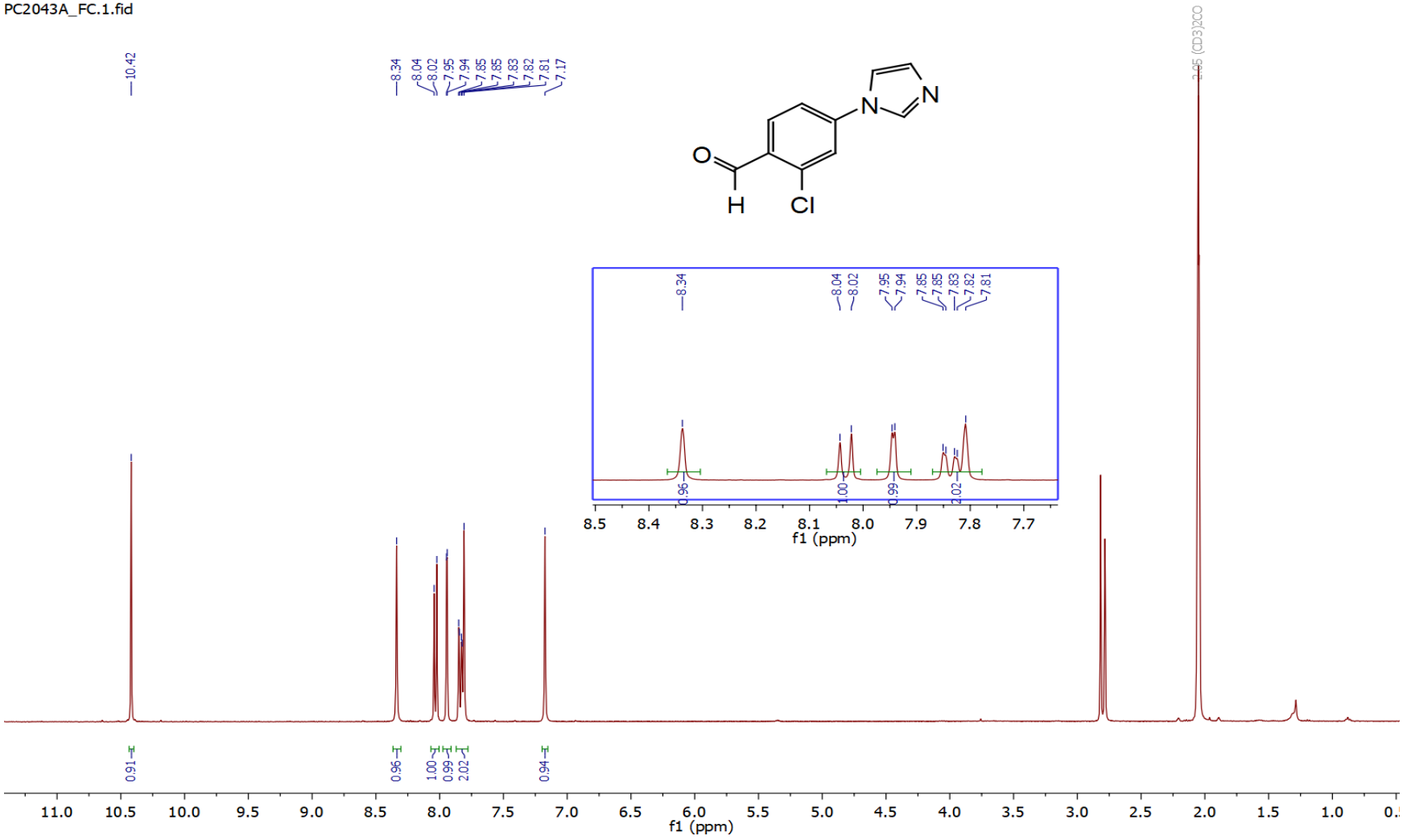
Fragment 1

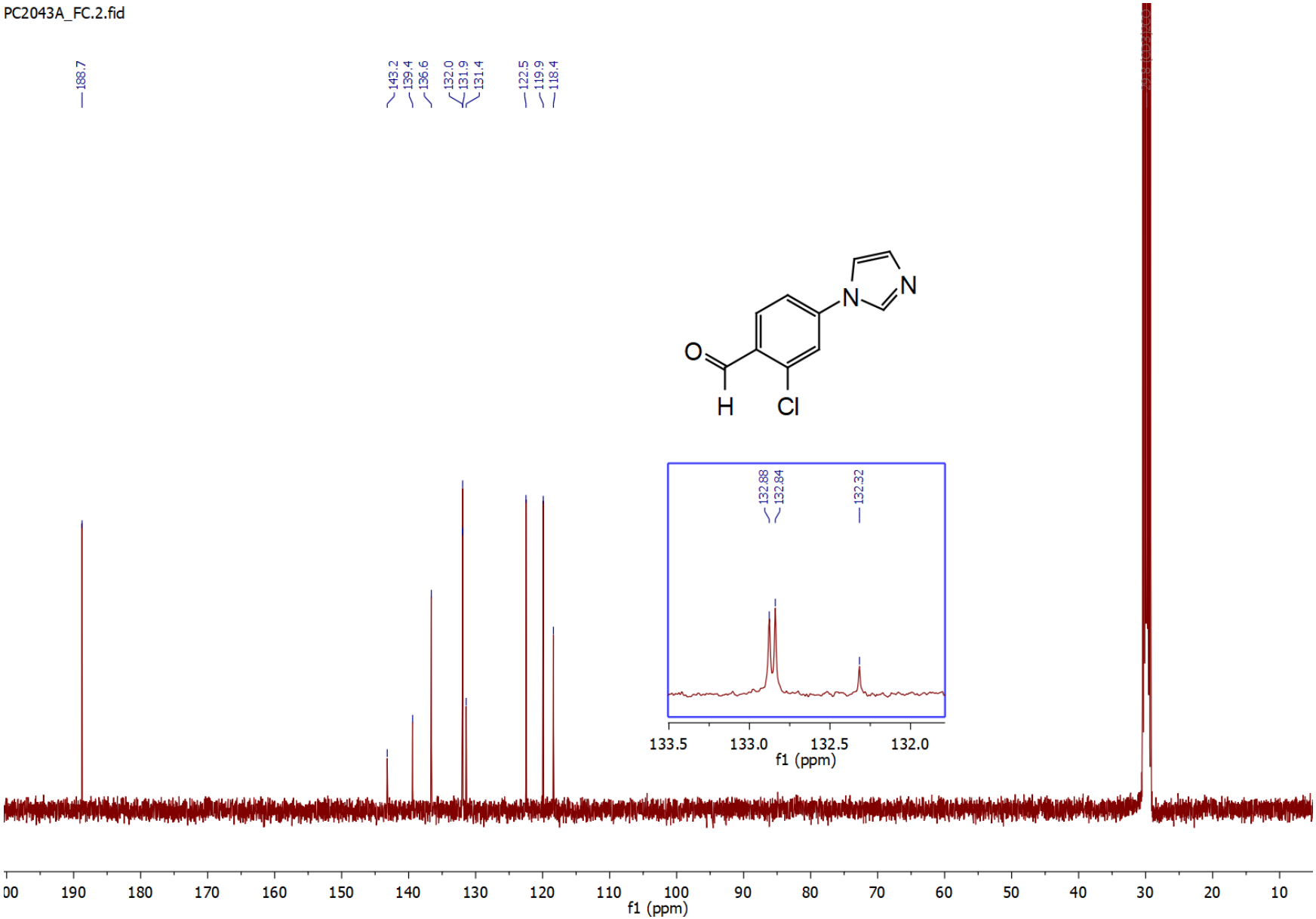



191.8
141.3
138.1
137.5
135.2
129.7
129.6
129.5
121.3
120.0
40.0 DMSO-d6
O
H
N
N
Br
f1 (ppm)
f1 (ppm)

Fragment 2

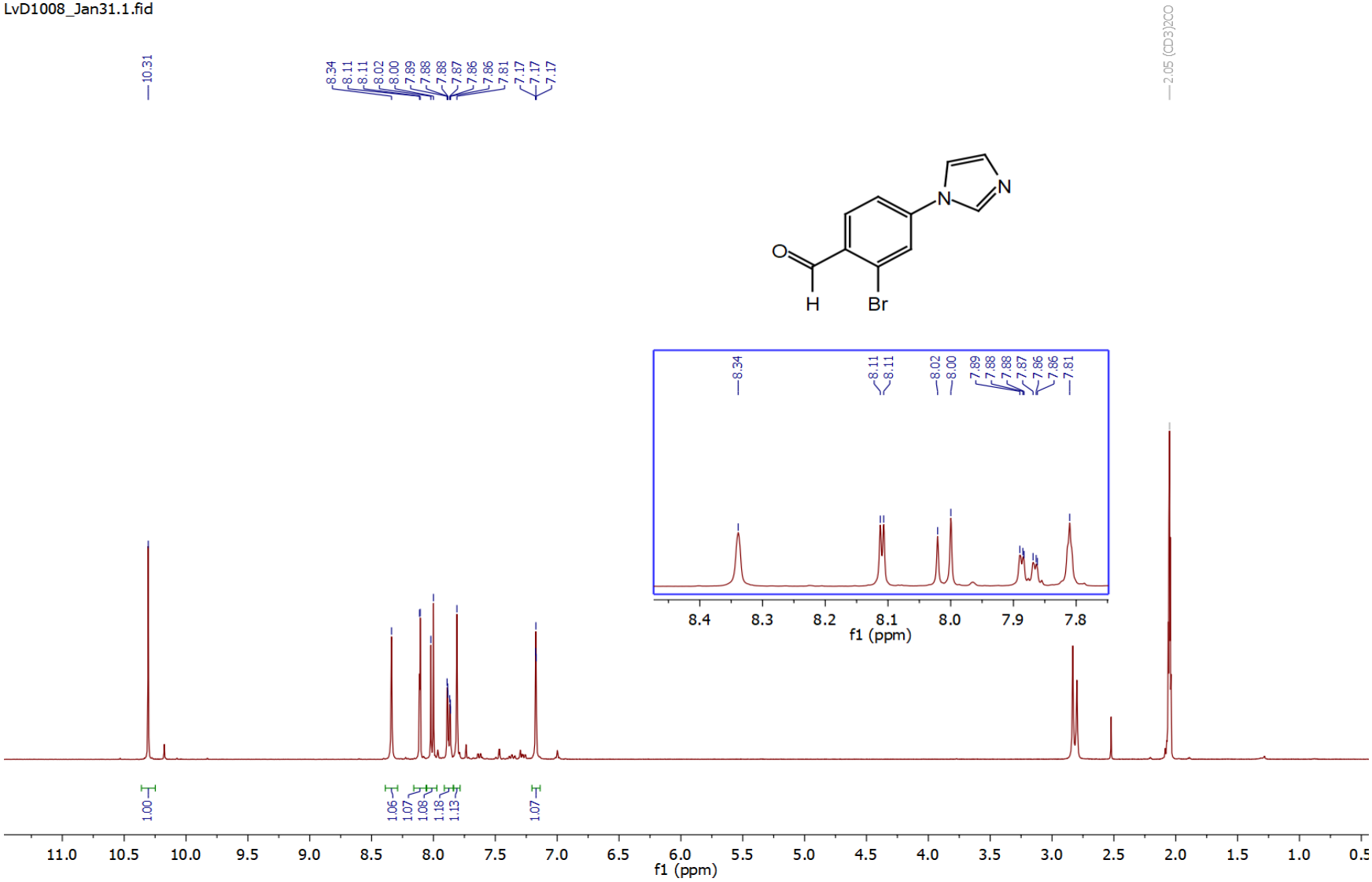
PC2043A_FC.1.fid

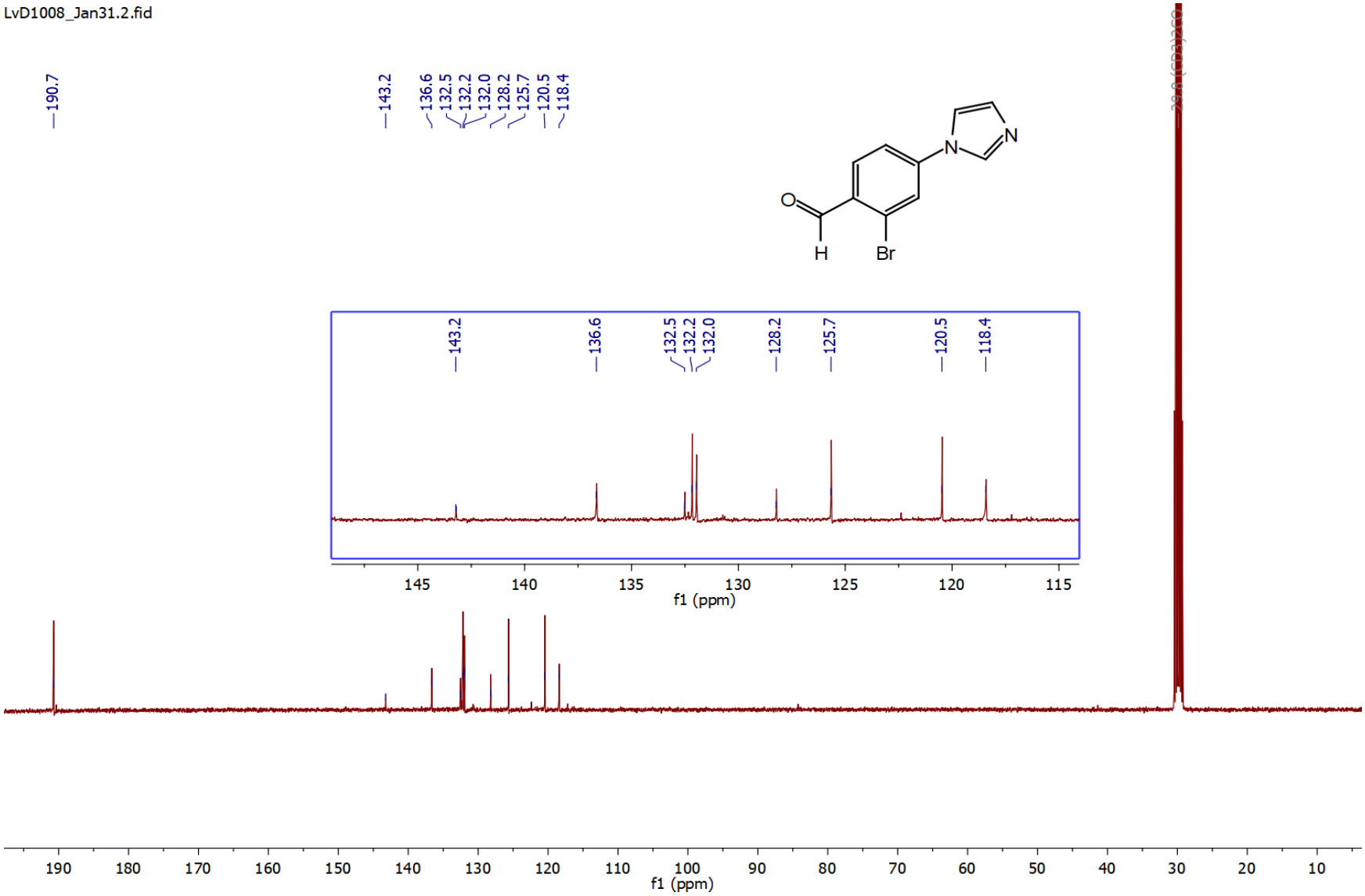



PC2043A_FC.2.fid
188.7
143.2
139.4
136.6
132.0
131.9
131.4
122.5
119.9
118.4
O
H
Cl
N
N
132.88
132.84
132.32
133.5
133.0
132.5
132.0
f1 (ppm)
f1 (ppm)

Fragment 3

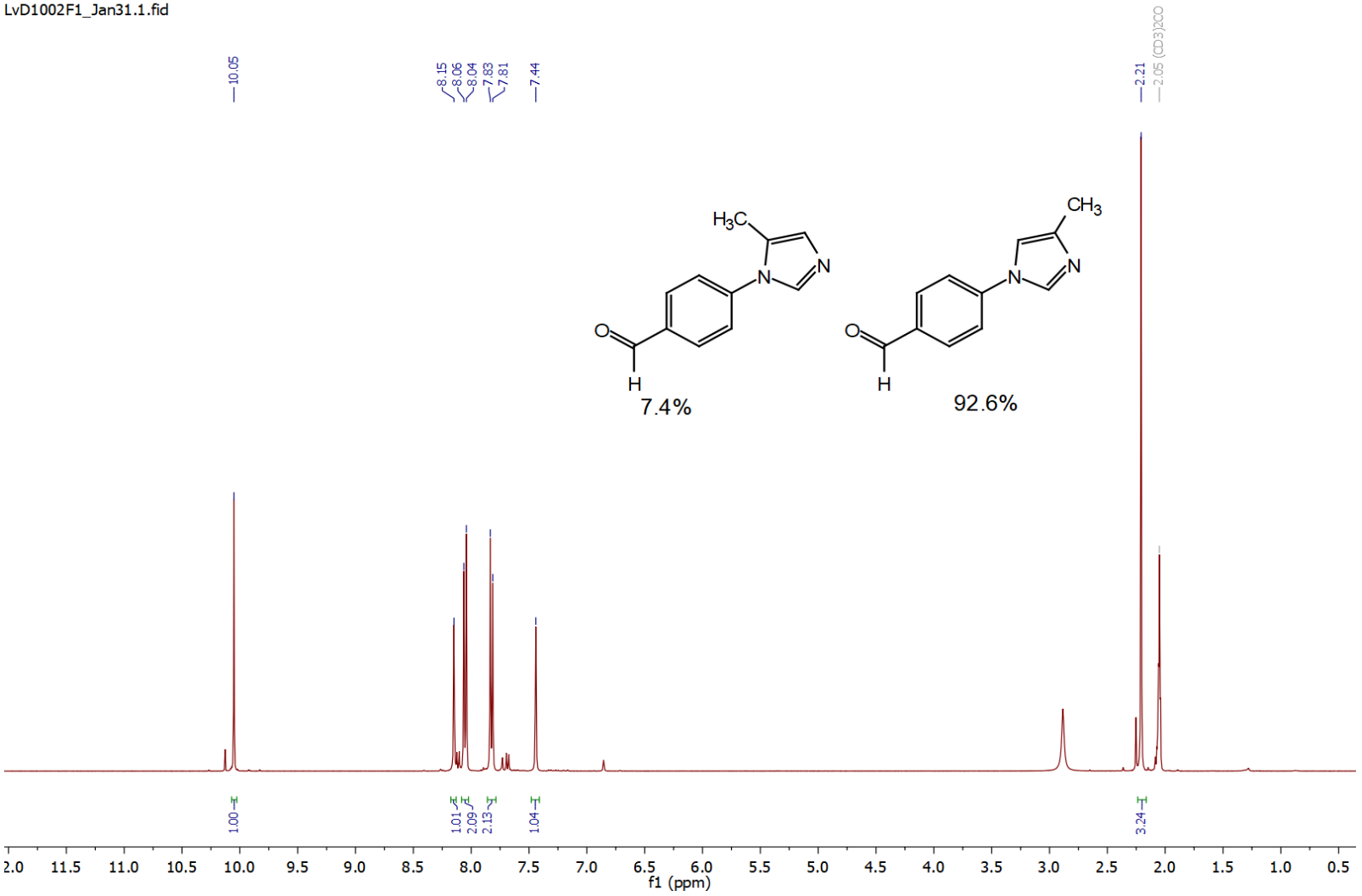
LvD1008_Jan31.1.fid

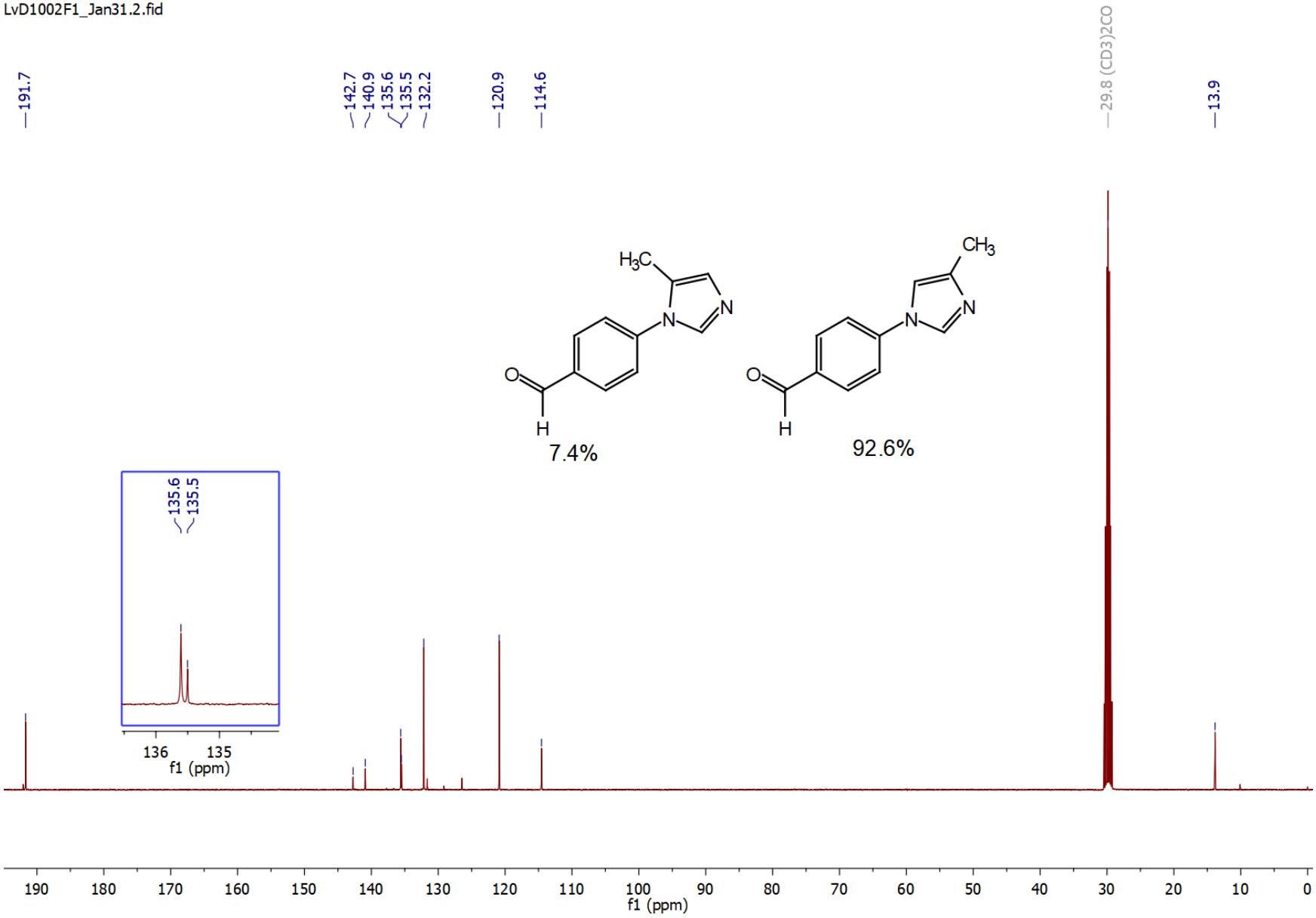



LvD1008_Jan31.2.fid
190.7
143.2
136.6
132.5
132.2
132.0
128.2
125.7
120.5
118.4
O
N
N
H
Br
f1 (ppm)
f1 (ppm)

Fragment 4

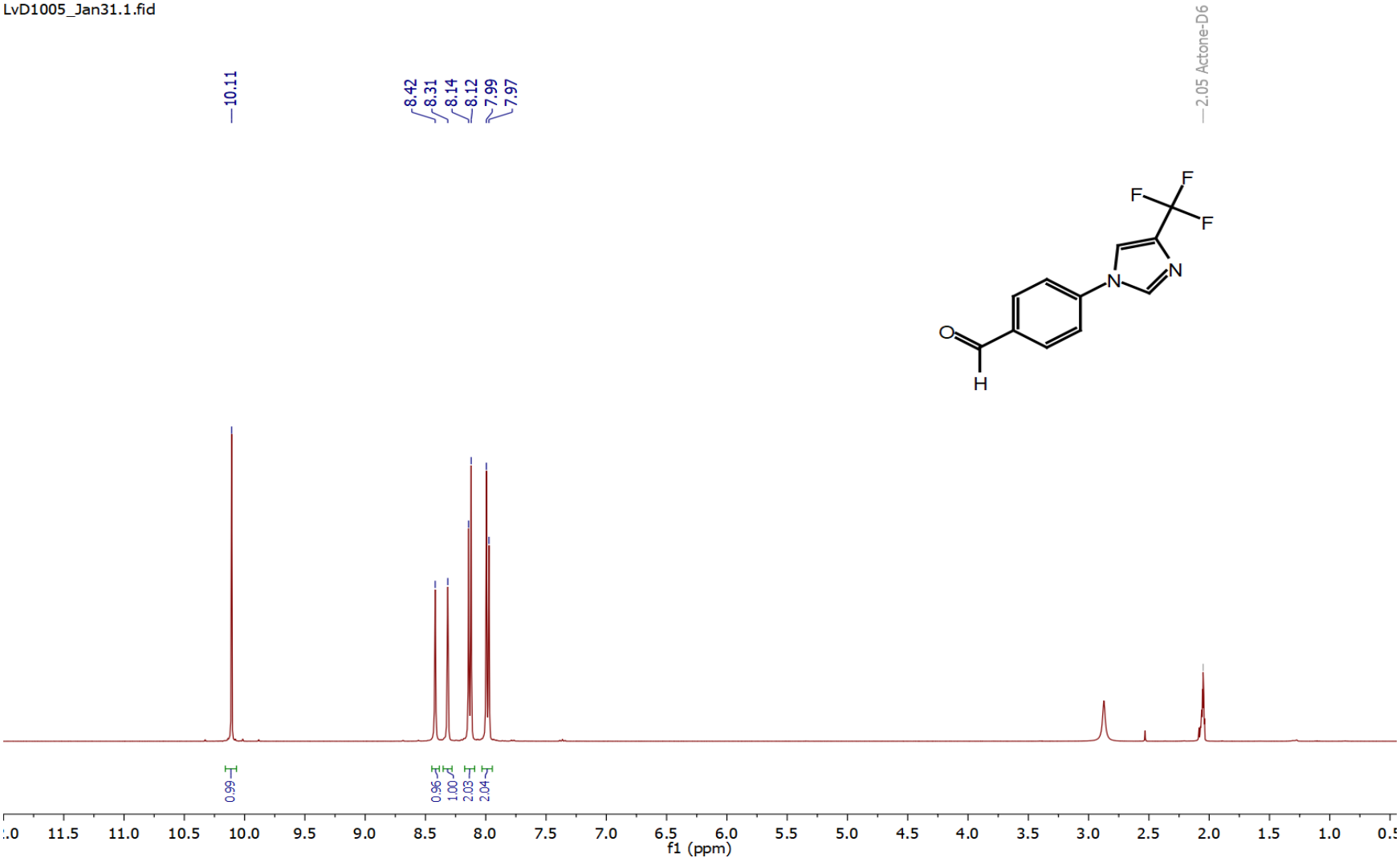
LvD1002F1_Jan31.1.fid

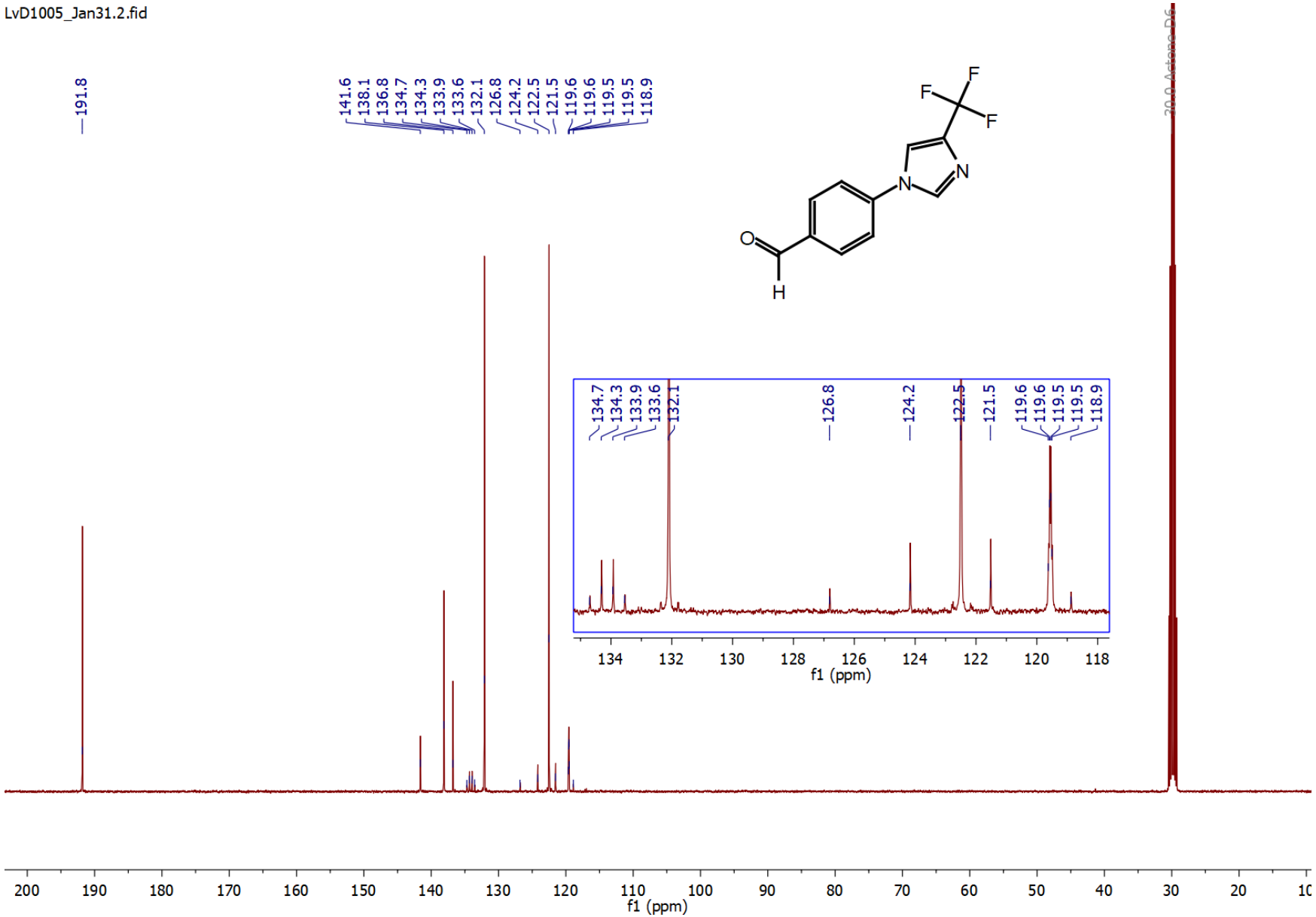



LvD1002F1_Jan31.2.fid
191.7
142.7
140.9
135.6
135.5
132.2
120.9
114.6
29.8 (CD3)2CO
13.9
H3C
N
N
O
H
7.4%
CH3
N
N
O
H
92.6%
135.6
135.5
136
135
f1 (ppm)
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

Fragment 6

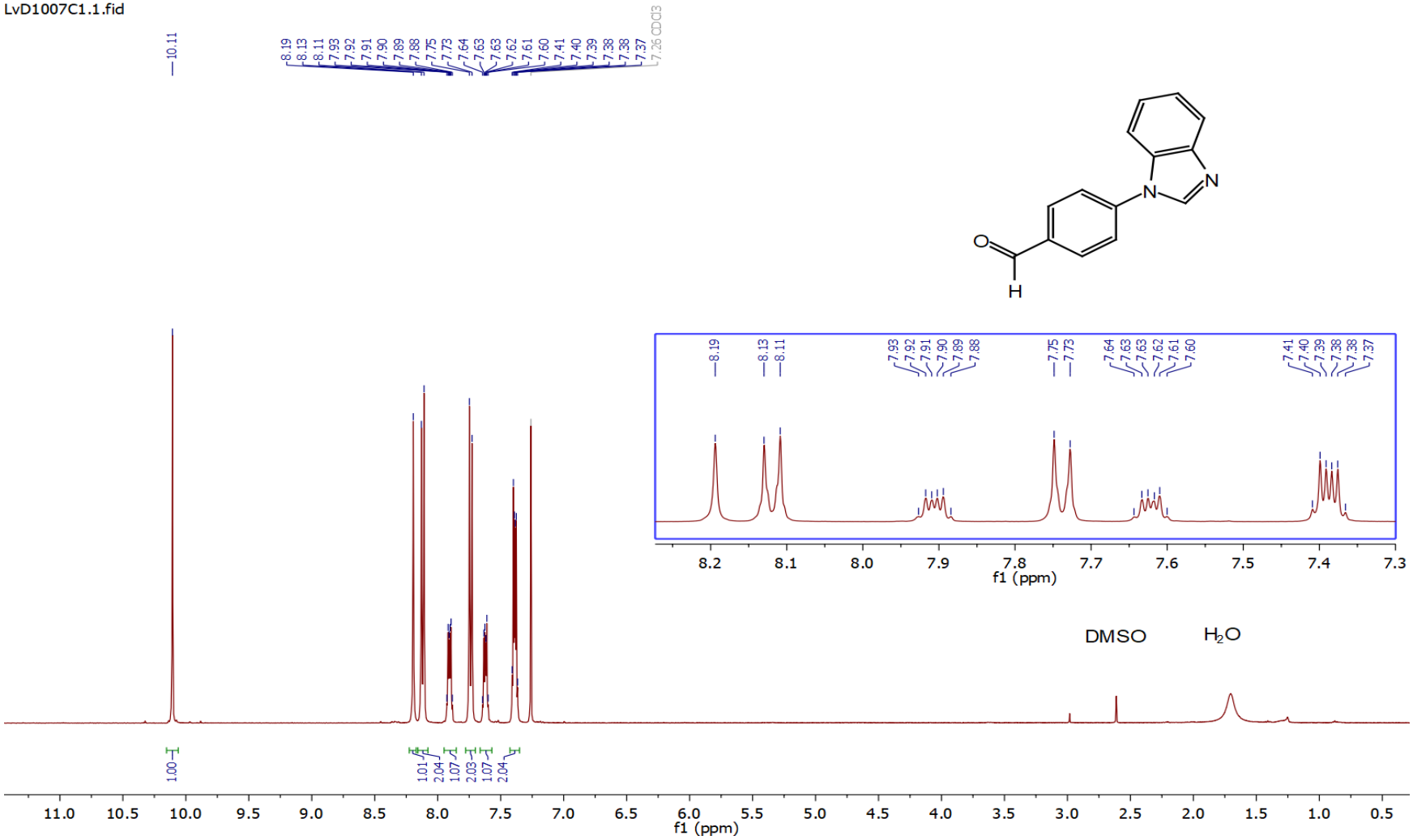
LvD1005_Jan31.1.fid

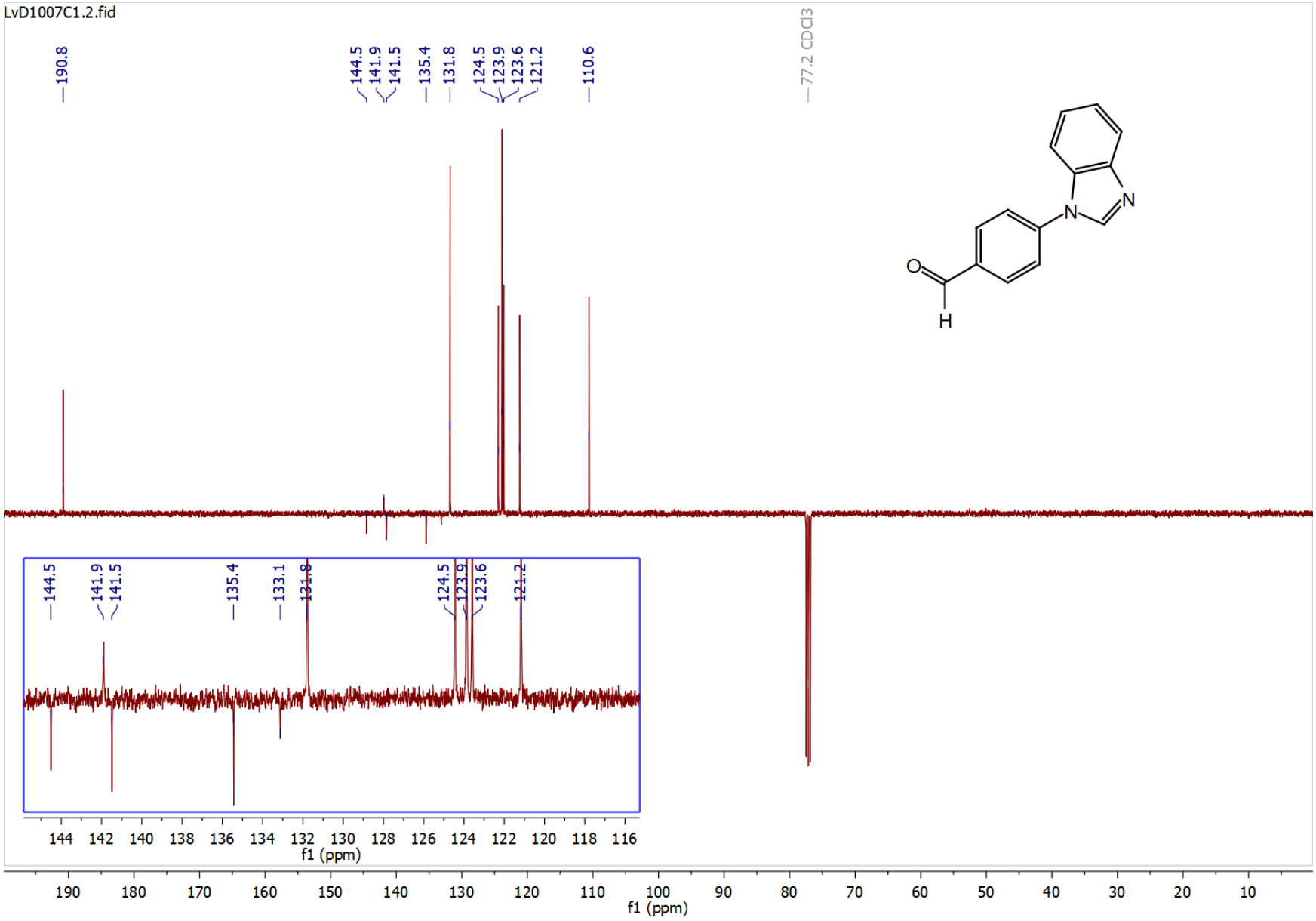



LvD1005_Jan31.2.fid
191.8
141.6
138.1
136.8
134.7
134.3
133.9
133.6
132.1
126.8
124.2
122.5
121.5
119.6
119.6
119.5
119.5
118.9
F
F
F
N
N
O
H
f1 (ppm)
f1 (ppm)

Fragment 7

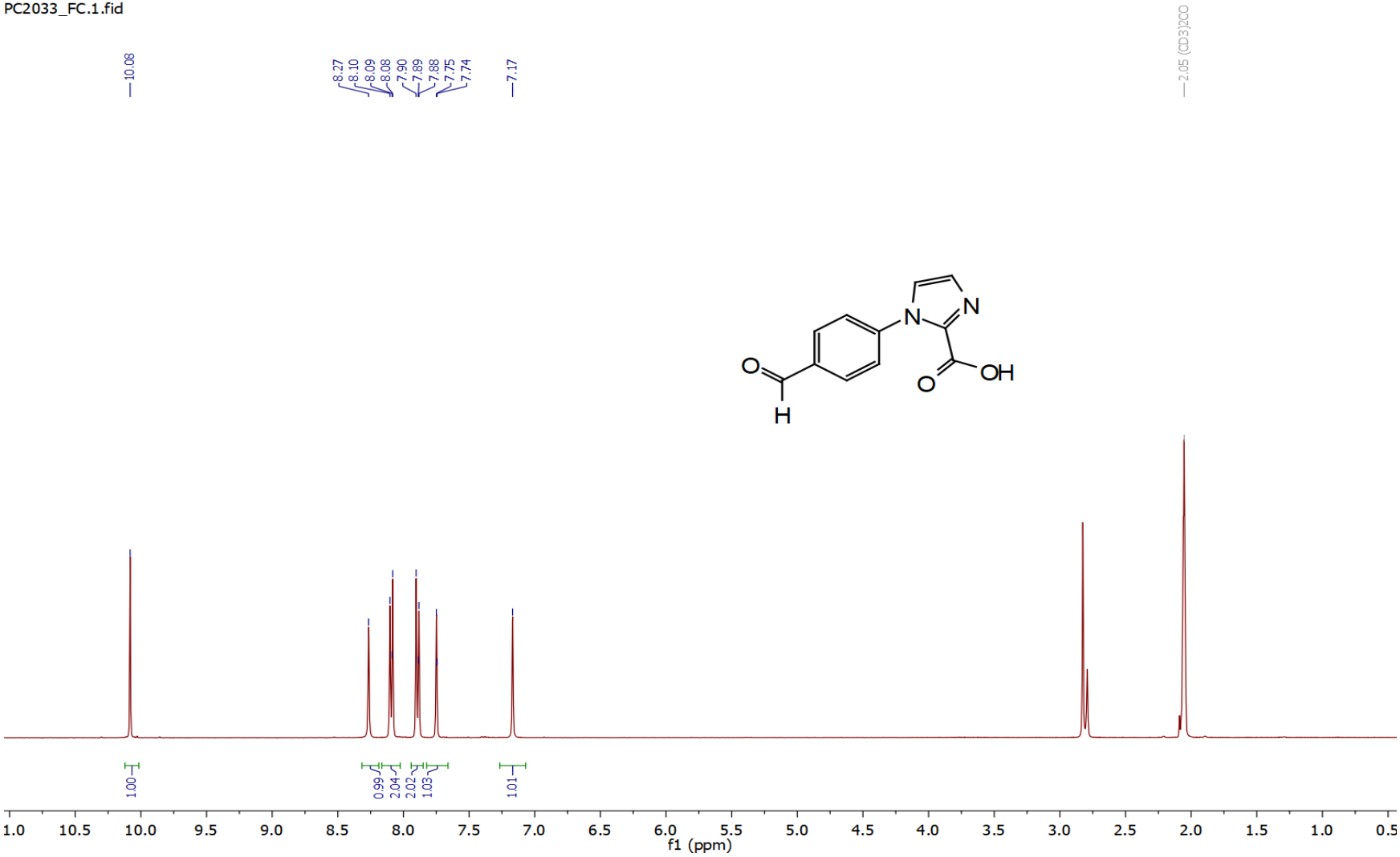
LvD1007C1.1.fid

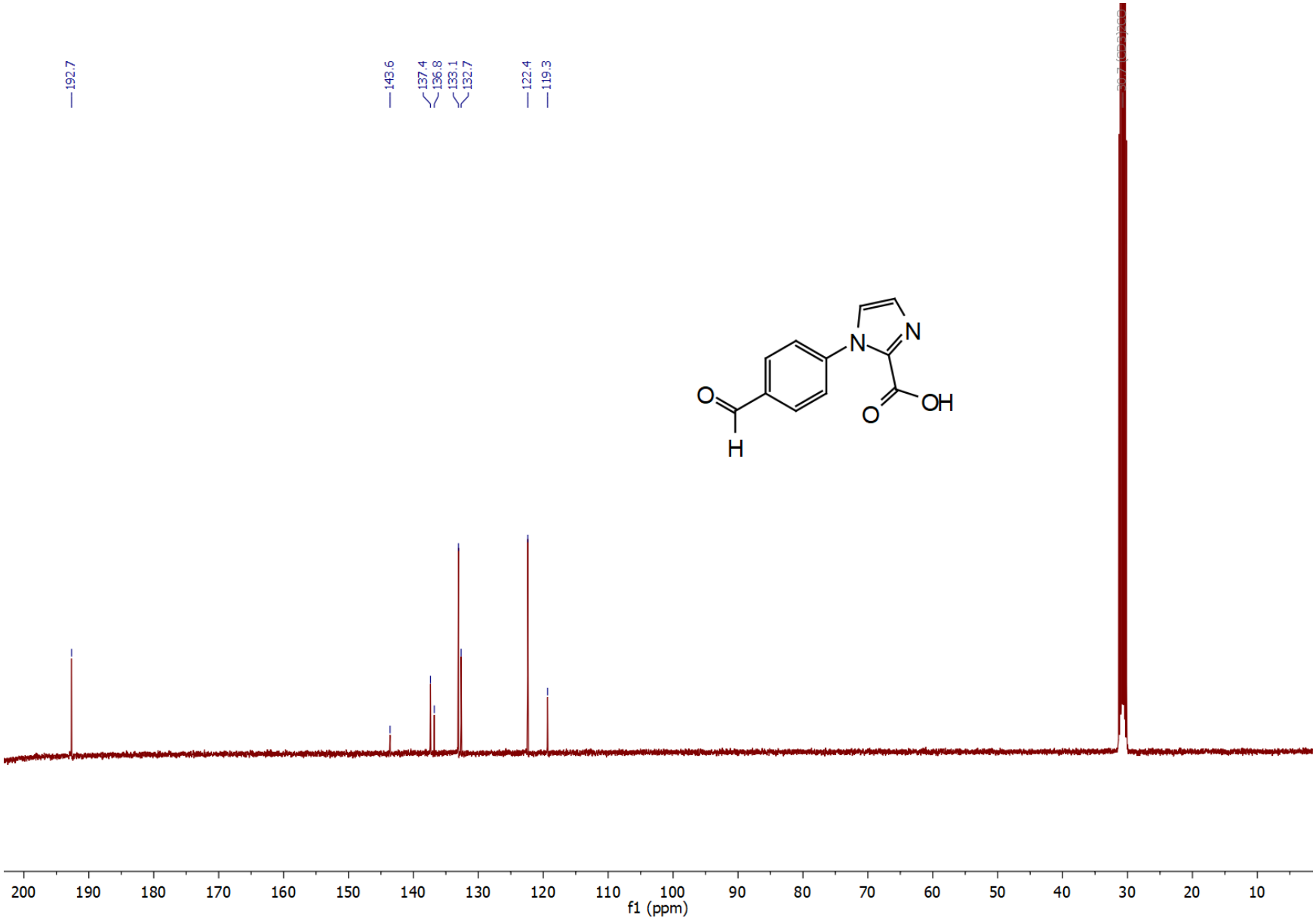



LvD1007C1.2.fid
190.8
144.5
141.9
141.5
135.4
131.8
124.5
123.9
123.6
121.2
110.6
77.2 CDCl3
O
H
N
N
144.5
141.9
141.5
135.4
133.1
131.8
124.5
123.9
123.6
121.2
144 142 140 138 136 134 132 130 128 126 124 122 120 118 116
f1 (ppm)
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

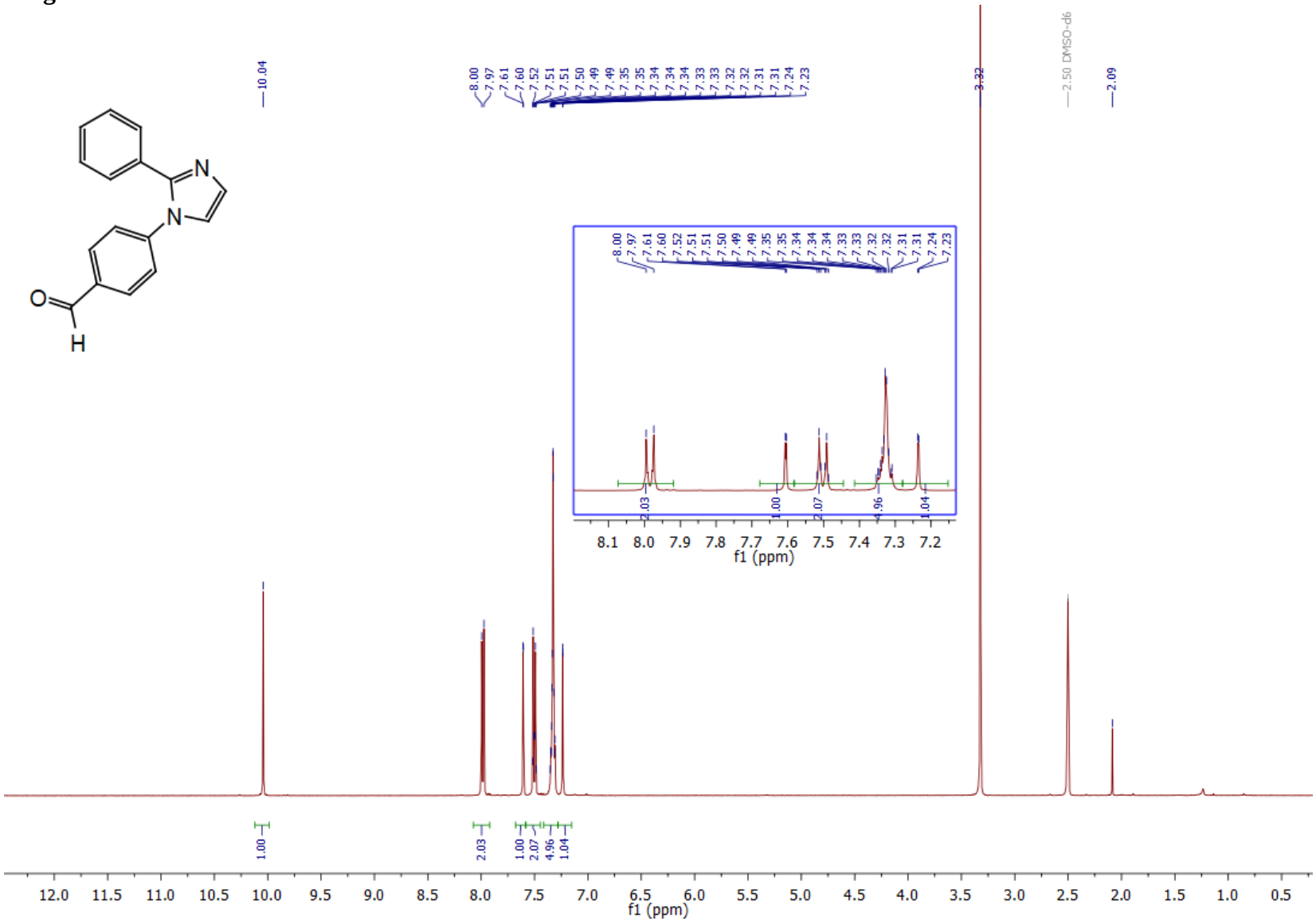
Fragment 8

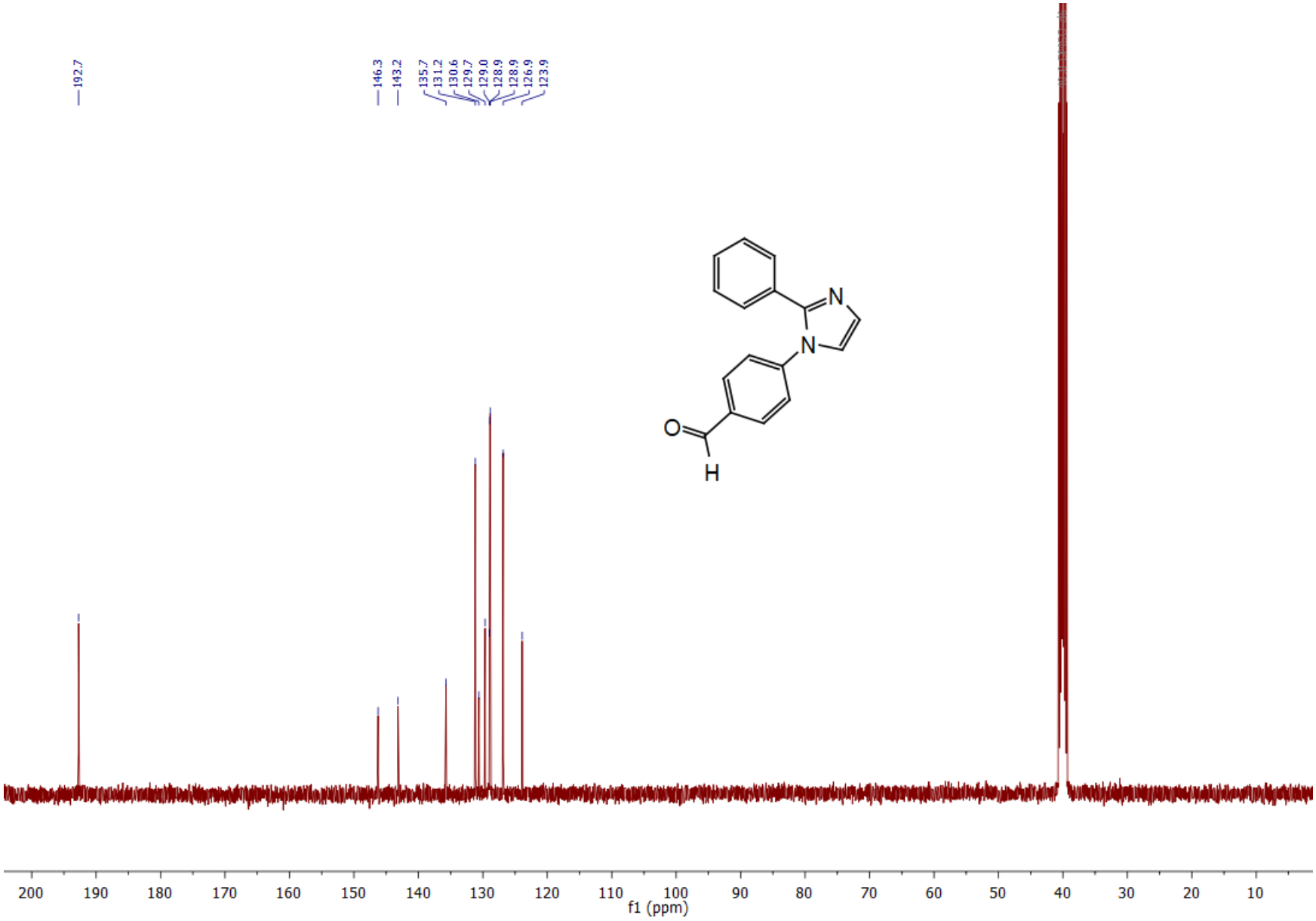
PC2033_FC.1.fid




192.7
143.6
137.4
136.8
133.1
132.7
122.4
119.3
O
H
N
N
O
OH
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

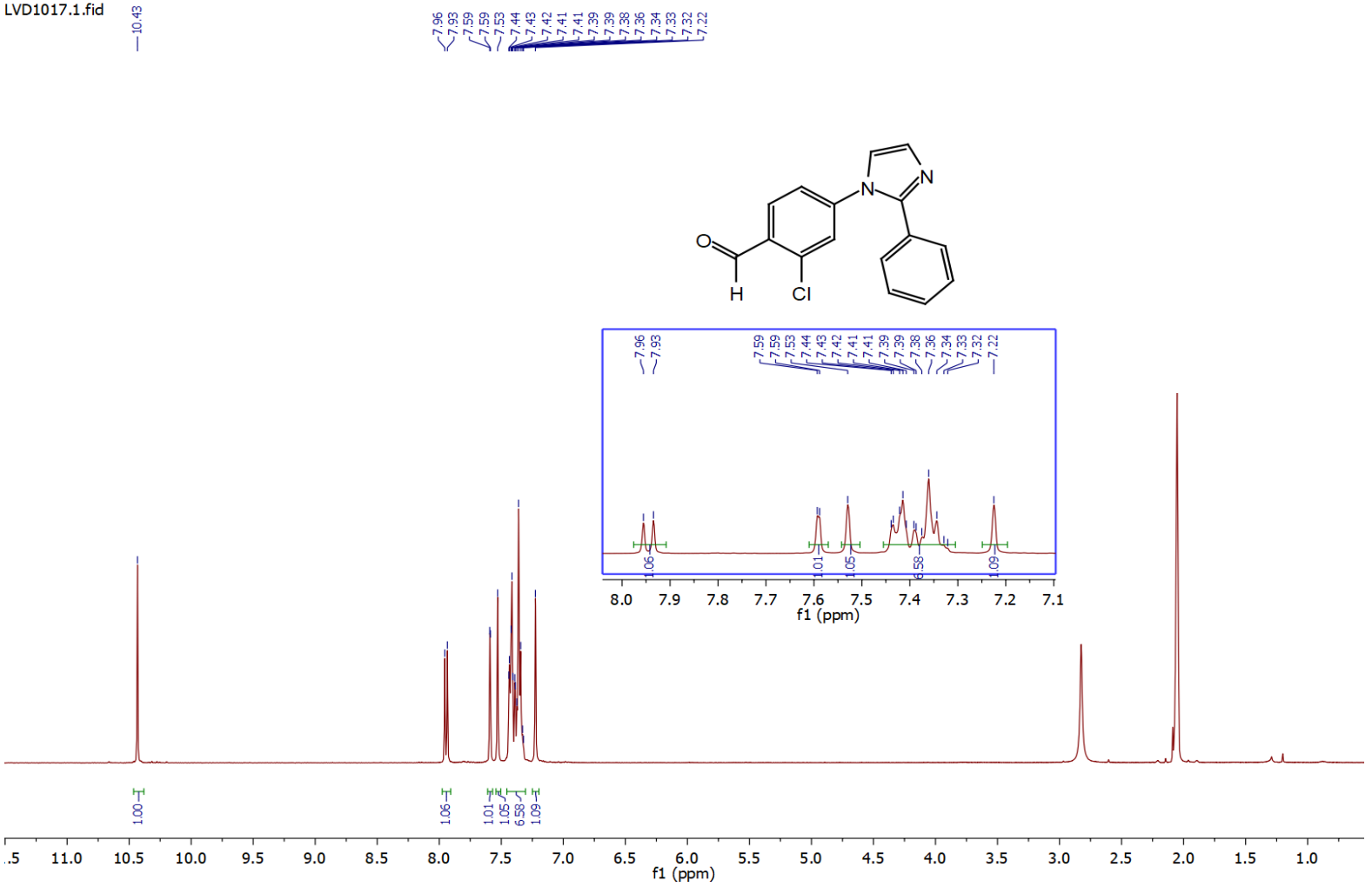
Fragment 9

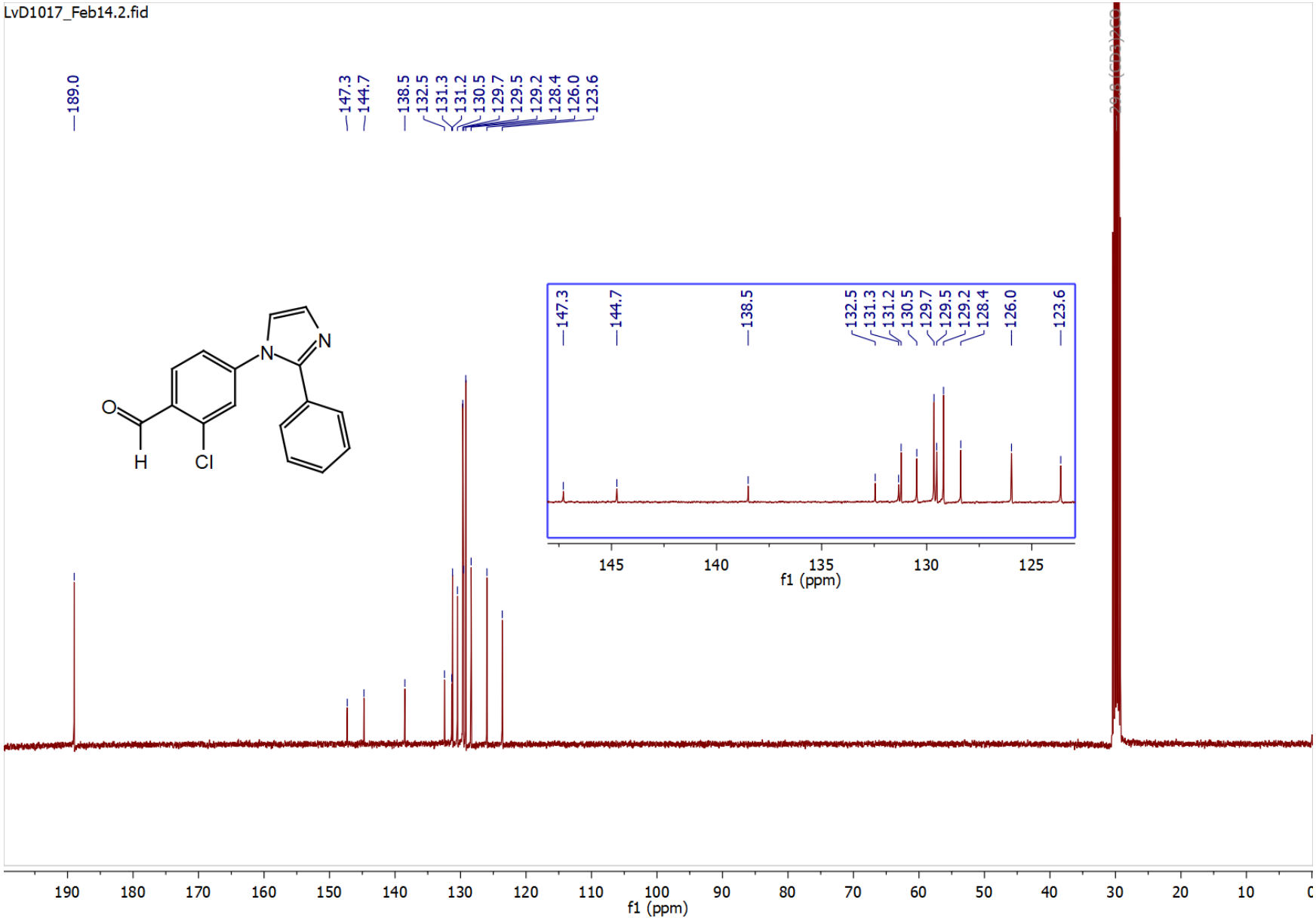

10.04
8.00
7.97
7.61
7.60
7.52
7.51
7.51
7.50
7.49
7.49
7.35
7.34
7.34
7.34
7.33
7.33
7.32
7.32
7.31
7.24
7.23
3.32
2.50 DMSO-d6
2.09
N
N
O
H
1.00
2.03
1.00
2.07
4.96
1.04
f1 (ppm)


192.7
146.3
143.2
135.7
131.2
130.6
129.7
129.0
128.9
128.9
126.9
123.9
N
N
O
H
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

Fragment 10

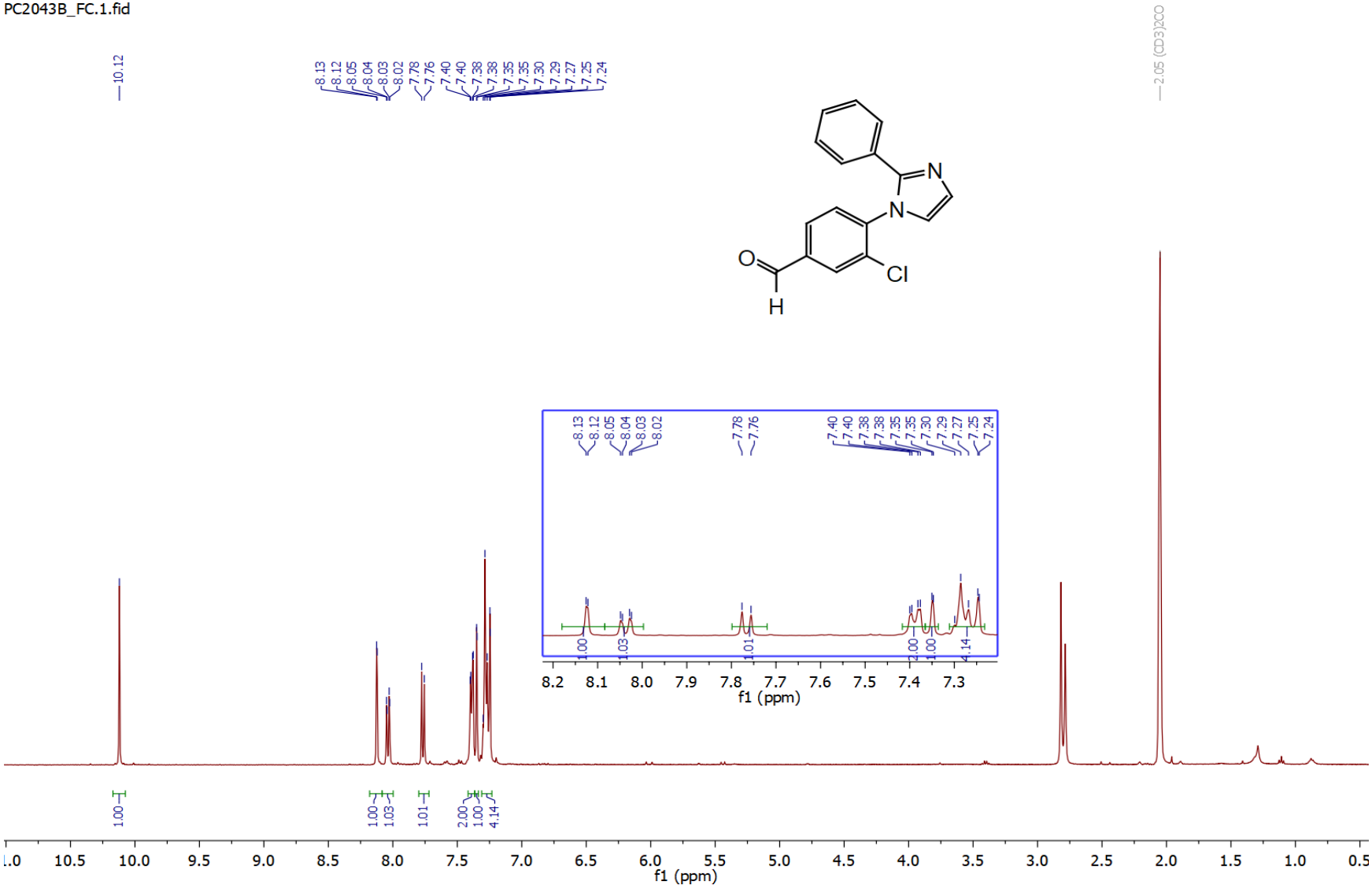
LVD1017.1.fid

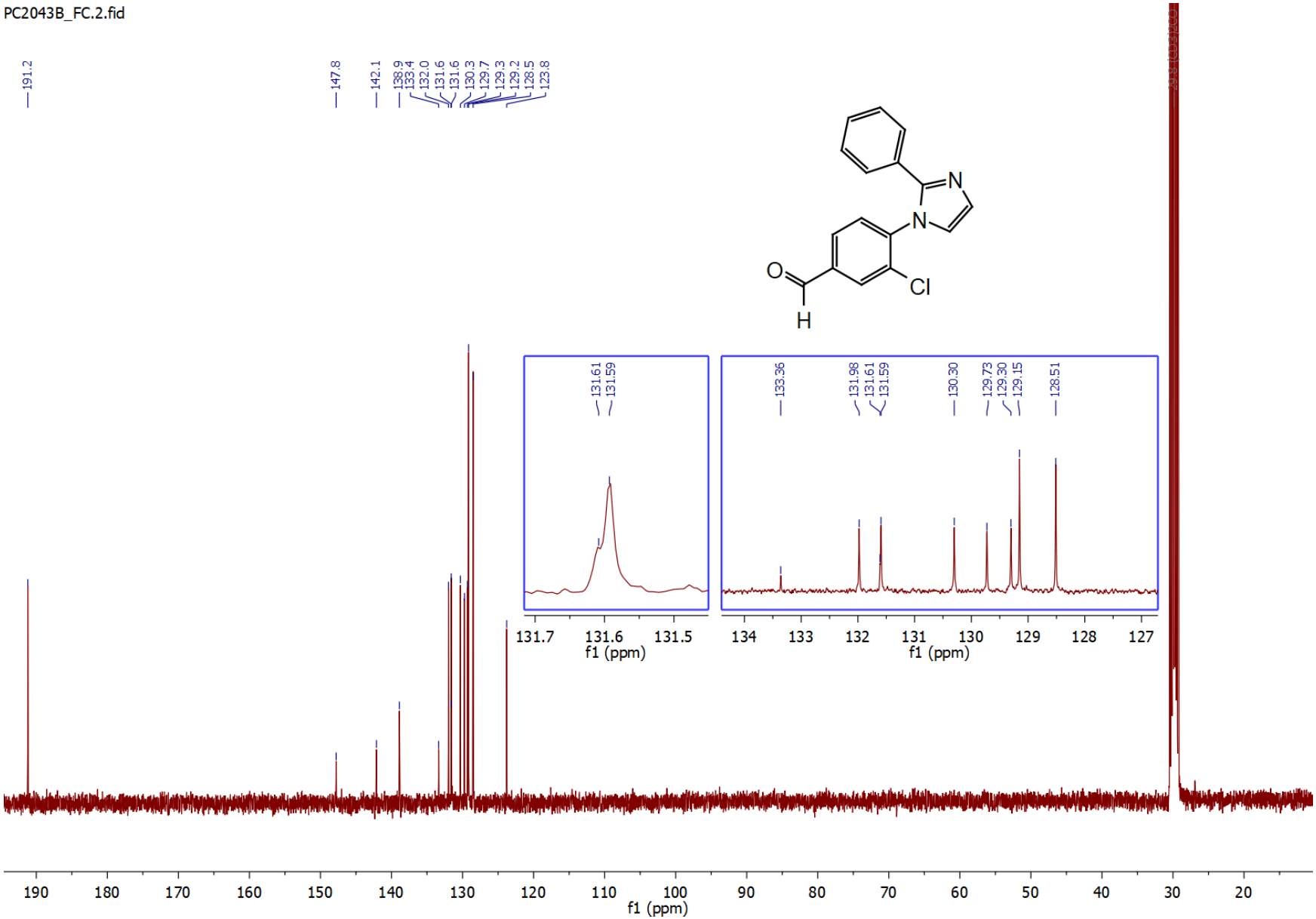



LvD1017_Feb14.2.fid
189.0
147.3
144.7
138.5
132.5
131.3
131.2
130.5
129.7
129.5
129.2
128.4
126.0
123.6
O
H
Cl
N
N
f1 (ppm)

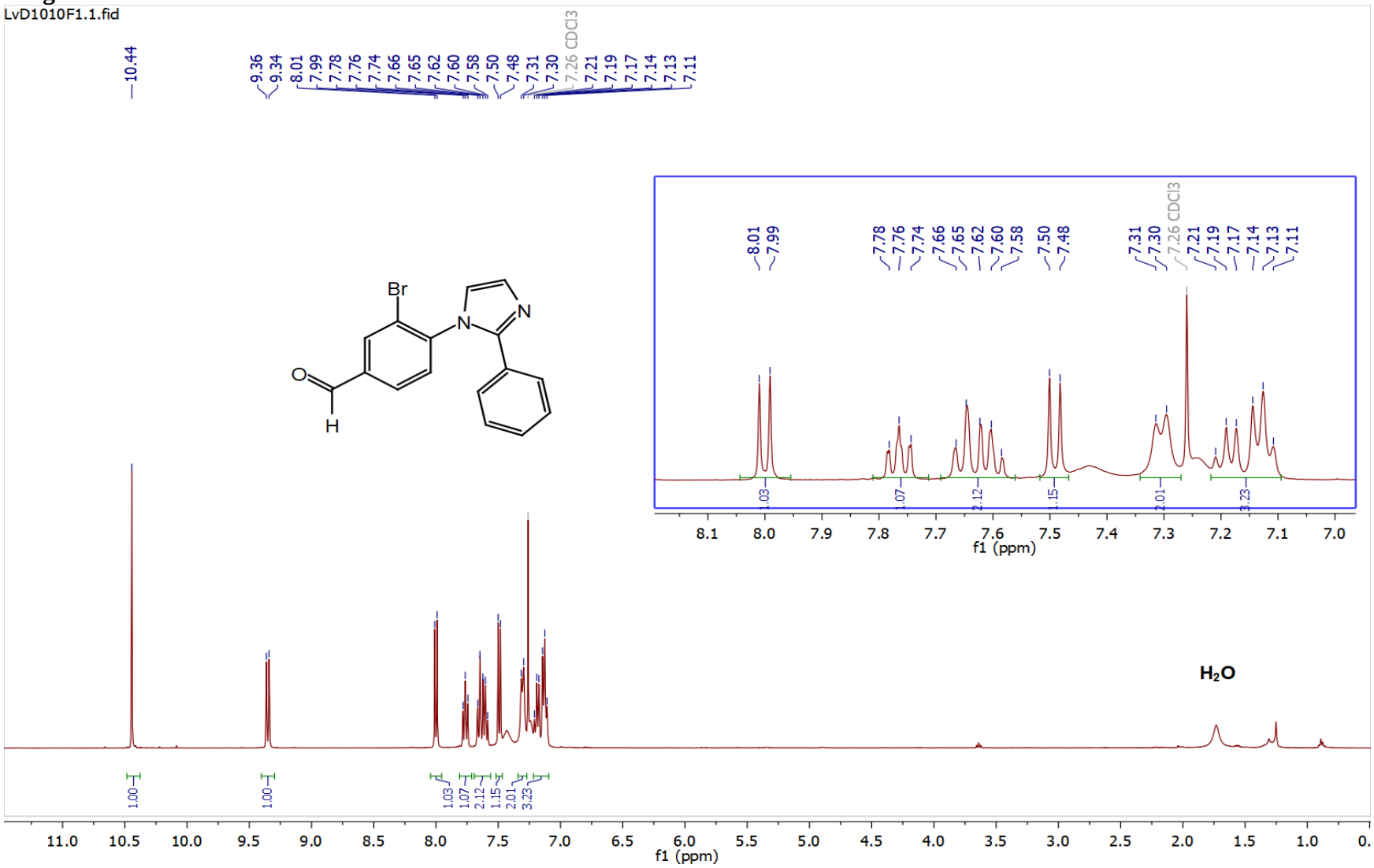
Fragment 11

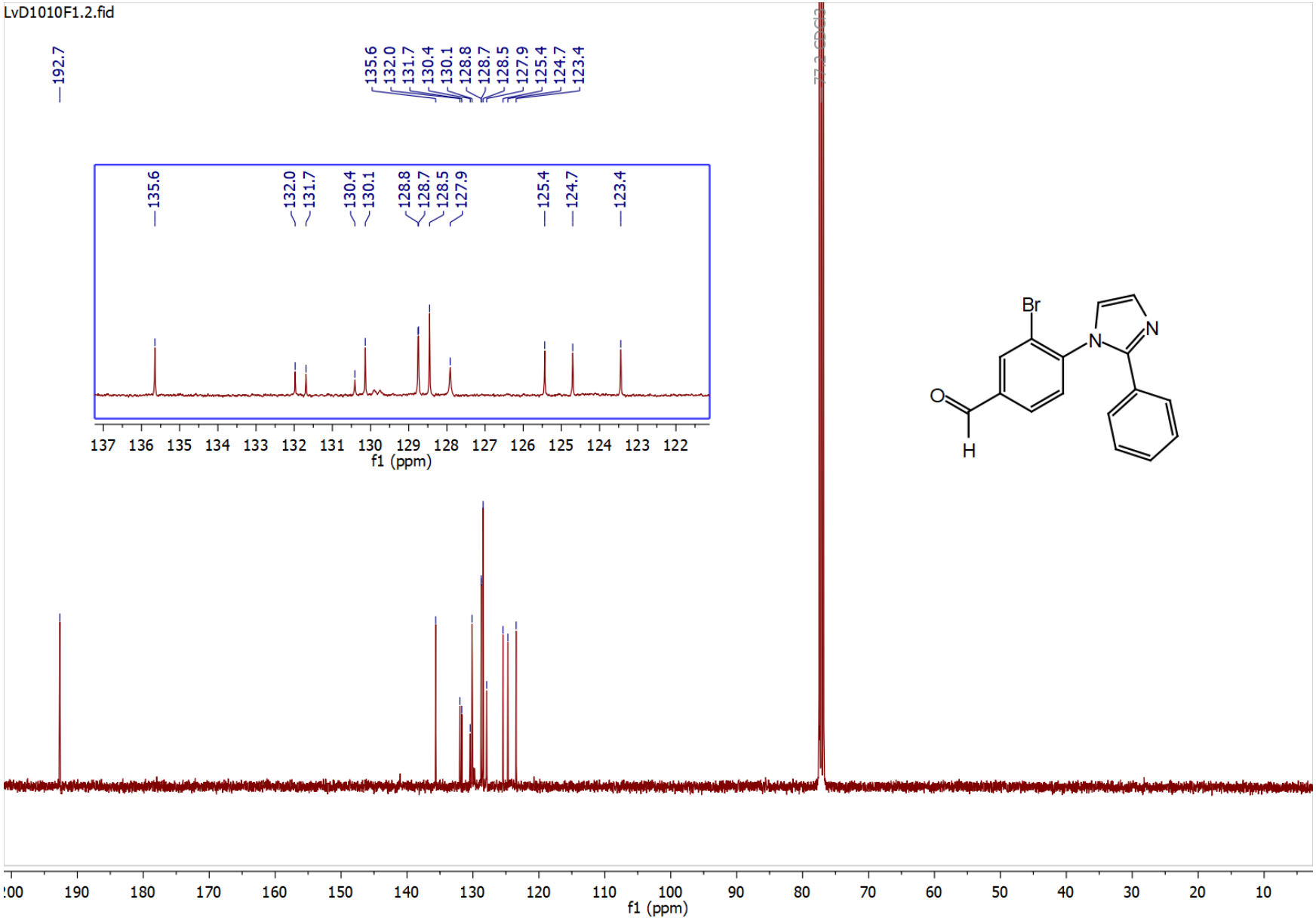
PC2043B_FC.1.fid



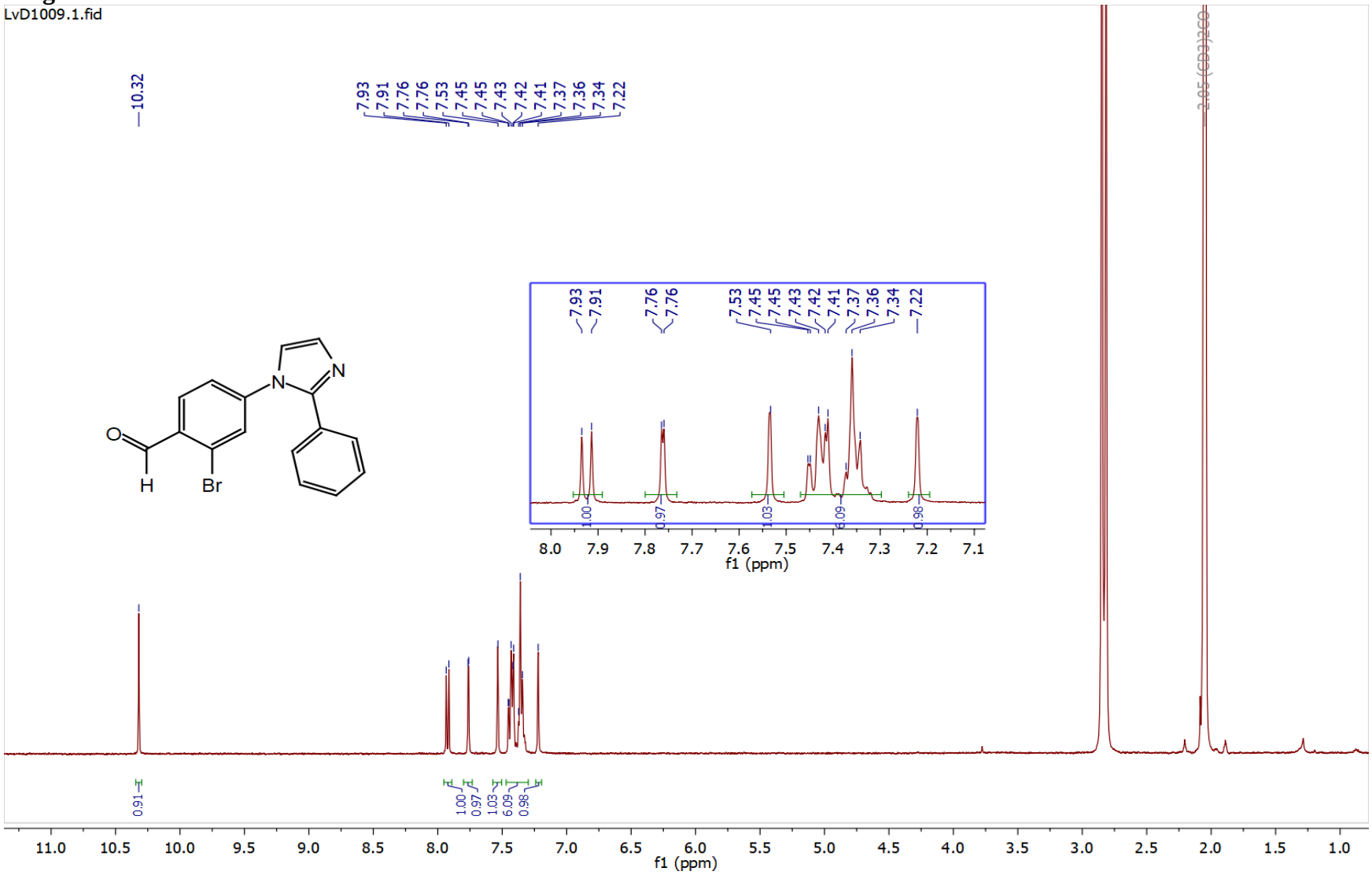

PC2043B_FC.2.fid
191.2
147.8
142.1
138.9
133.4
132.0
131.6
131.6
130.3
129.7
129.3
129.2
128.5
123.8
N
N
O
H
Cl
131.61
131.59
133.36
131.98
131.61
131.59
130.30
129.73
129.30
129.15
128.51
131.7
131.6
131.5
f1 (ppm)
134
133
132
131
130
129
128
127
f1 (ppm)
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
f1 (ppm)

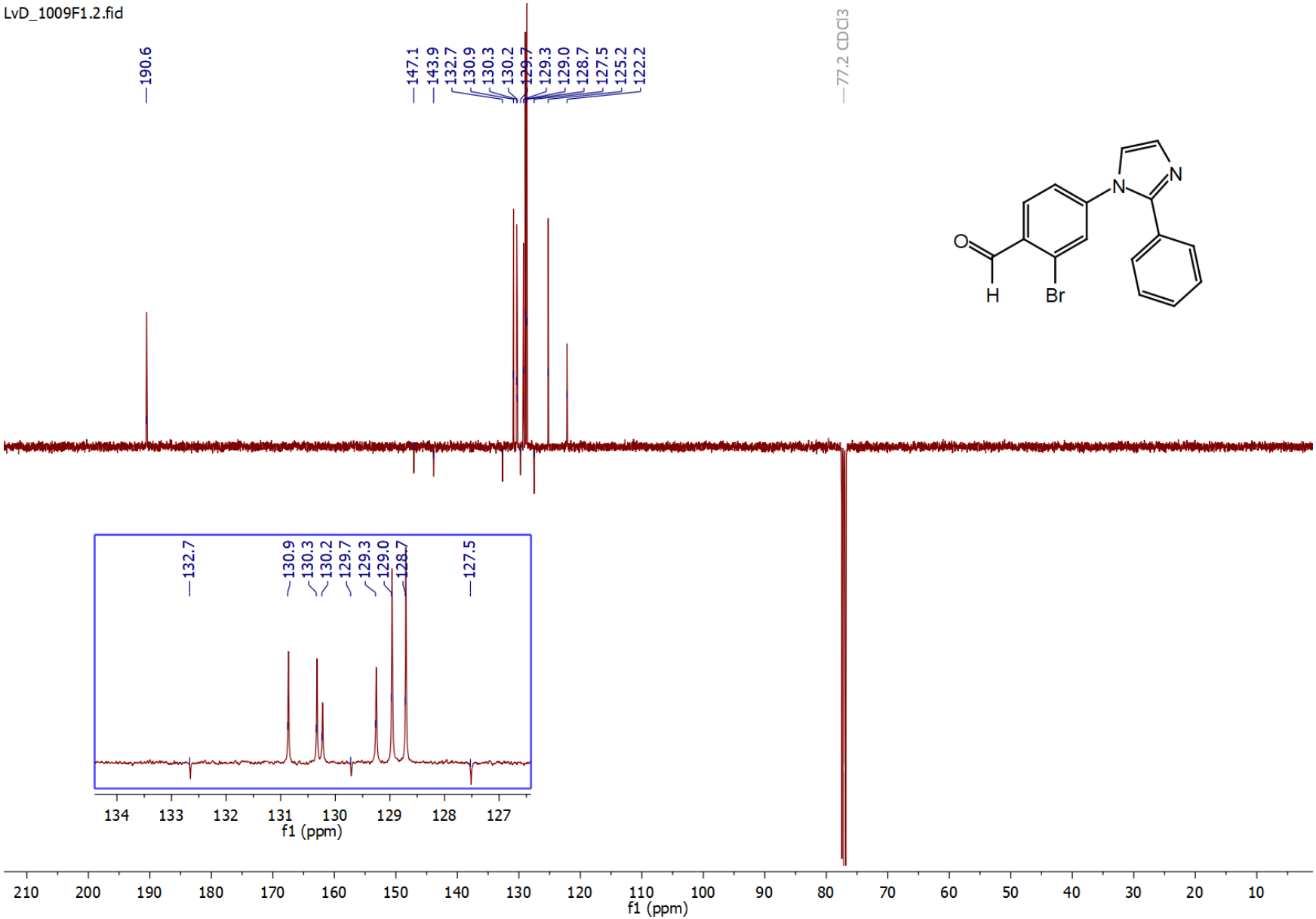
Fragment 12


LvD1010F1.1.fid
10.44
9.36
9.34
8.01
7.99
7.78
7.76
7.74
7.66
7.65
7.62
7.60
7.58
7.50
7.48
7.31
7.30
7.26 CDCl3
7.21
7.19
7.17
7.14
7.13
7.11
Br
N
N
O
H
H2O
1.00
1.00
1.03
1.07
2.12
1.15
2.01
3.23
f1 (ppm)

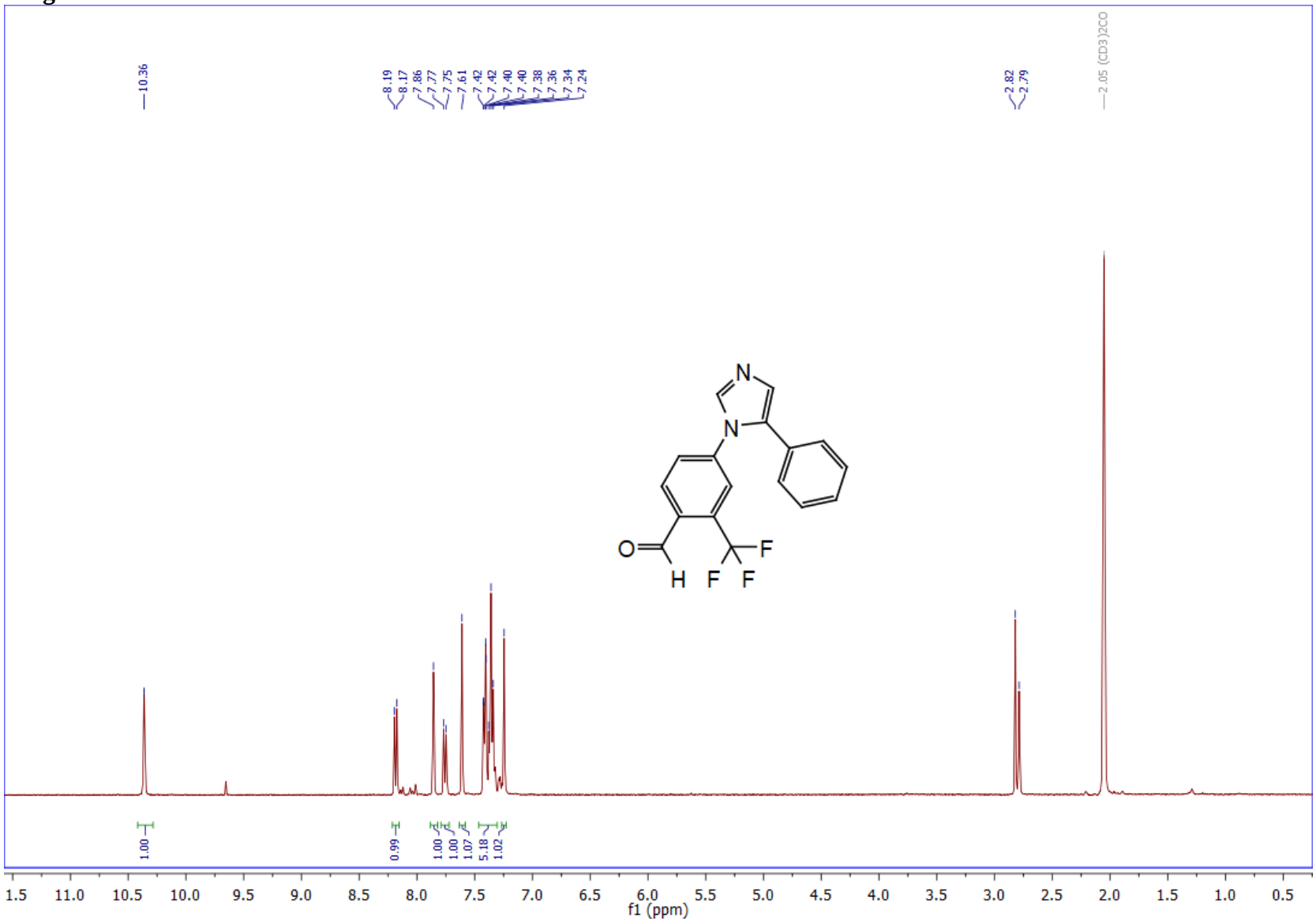

LvD1010F1.2.fid
192.7
135.6
132.0
131.7
130.4
130.1
128.8
128.7
128.5
127.9
125.4
124.7
123.4
77.2 CDCl3
f1 (ppm)
Br
N
N
O
H

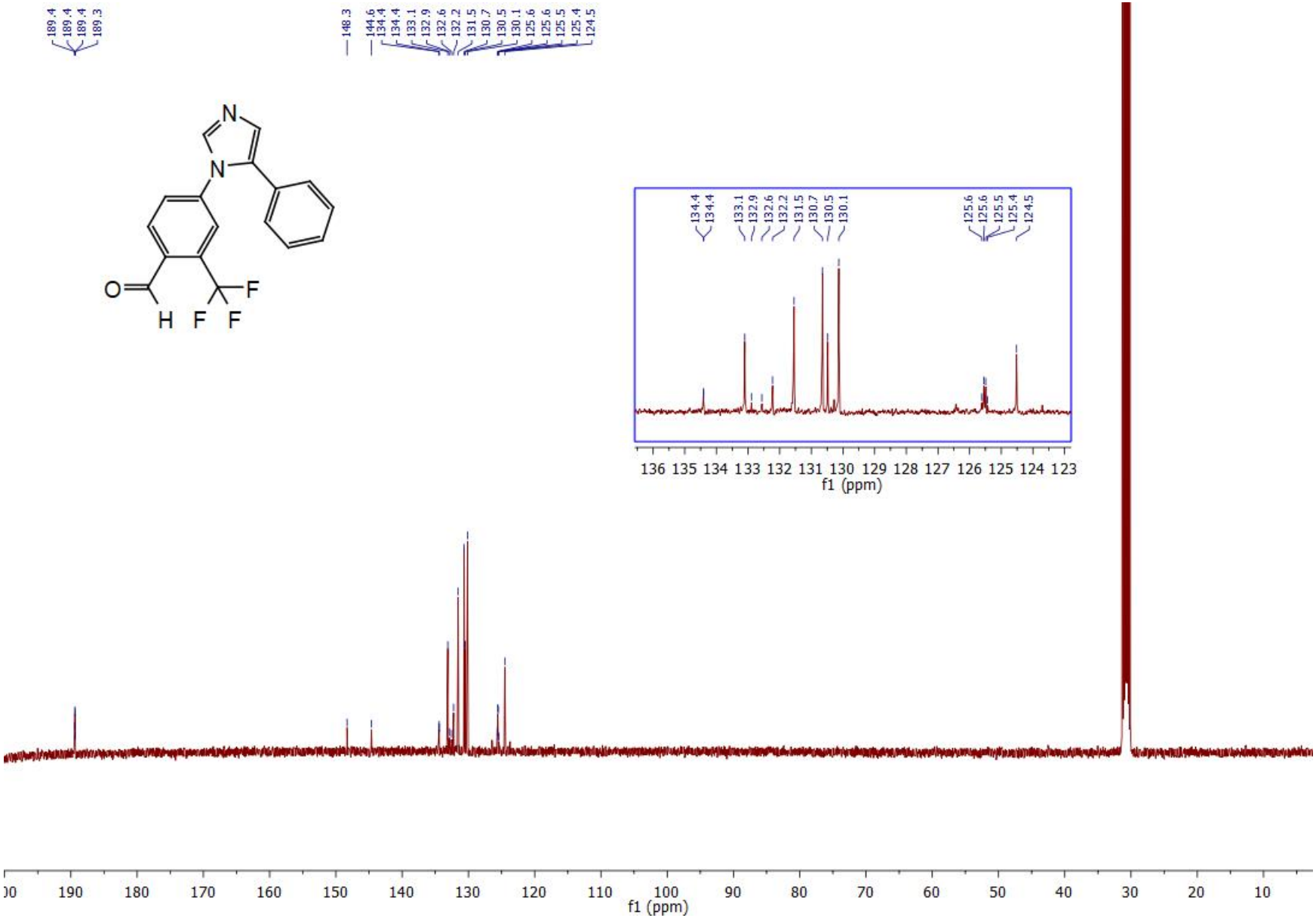
Fragment 13


LvD1009.1.fid
10.32
7.93
7.91
7.76
7.76
7.53
7.45
7.45
7.43
7.42
7.41
7.37
7.36
7.34
7.22
2.05 (CD3)2CO
O
H
Br
N
N
f1 (ppm)


LvD_1009F1.2.fid
190.6
147.1
143.9
132.7
130.9
130.3
130.2
129.7
129.3
129.0
128.7
127.5
125.2
122.2
77.2 CDCl3
O
H
Br
N
N
132.7
130.9
130.3
130.2
129.7
129.3
129.0
128.7
127.5
134
133
132
131
130
129
128
127
f1 (ppm)
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

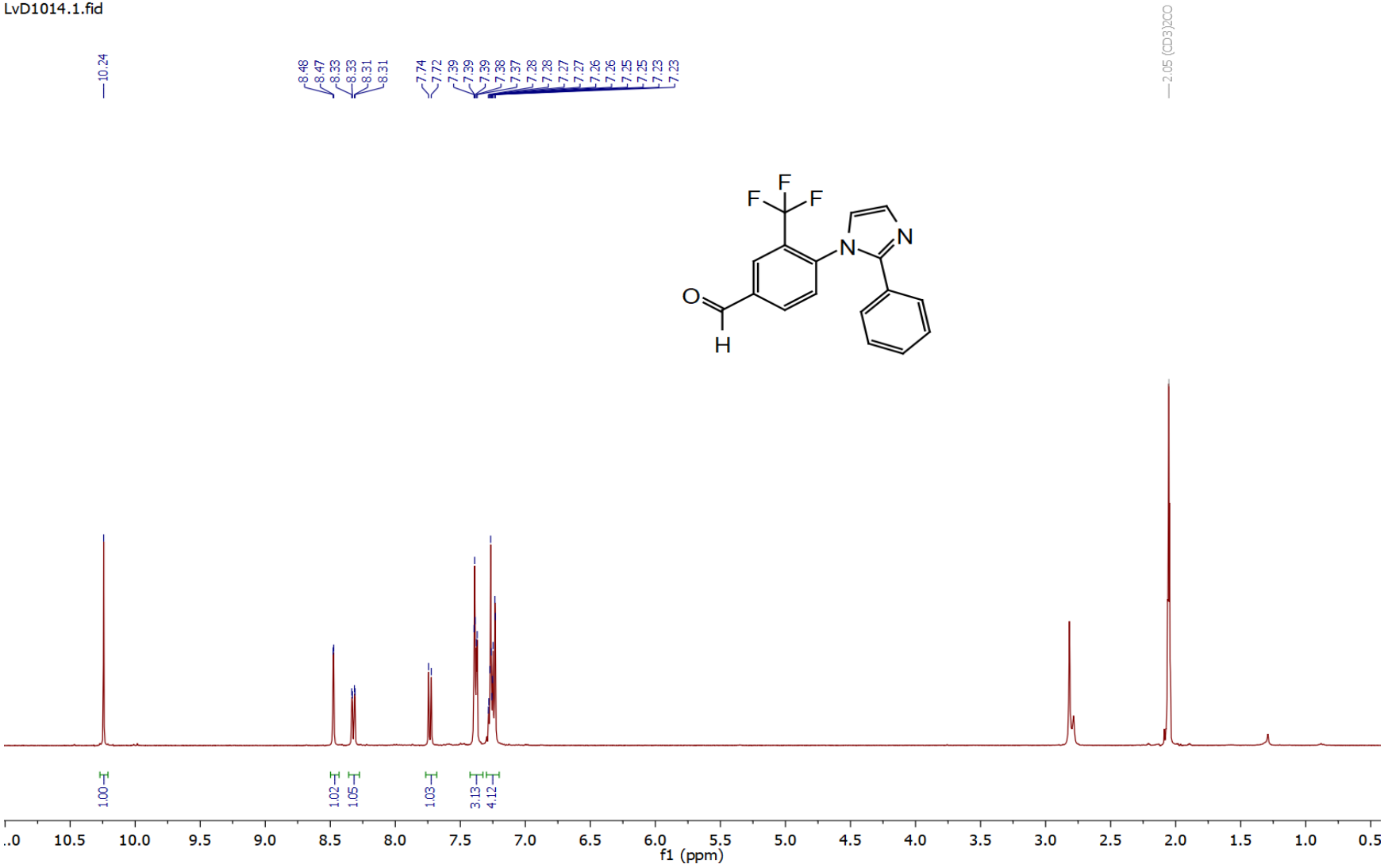
Fragment 14

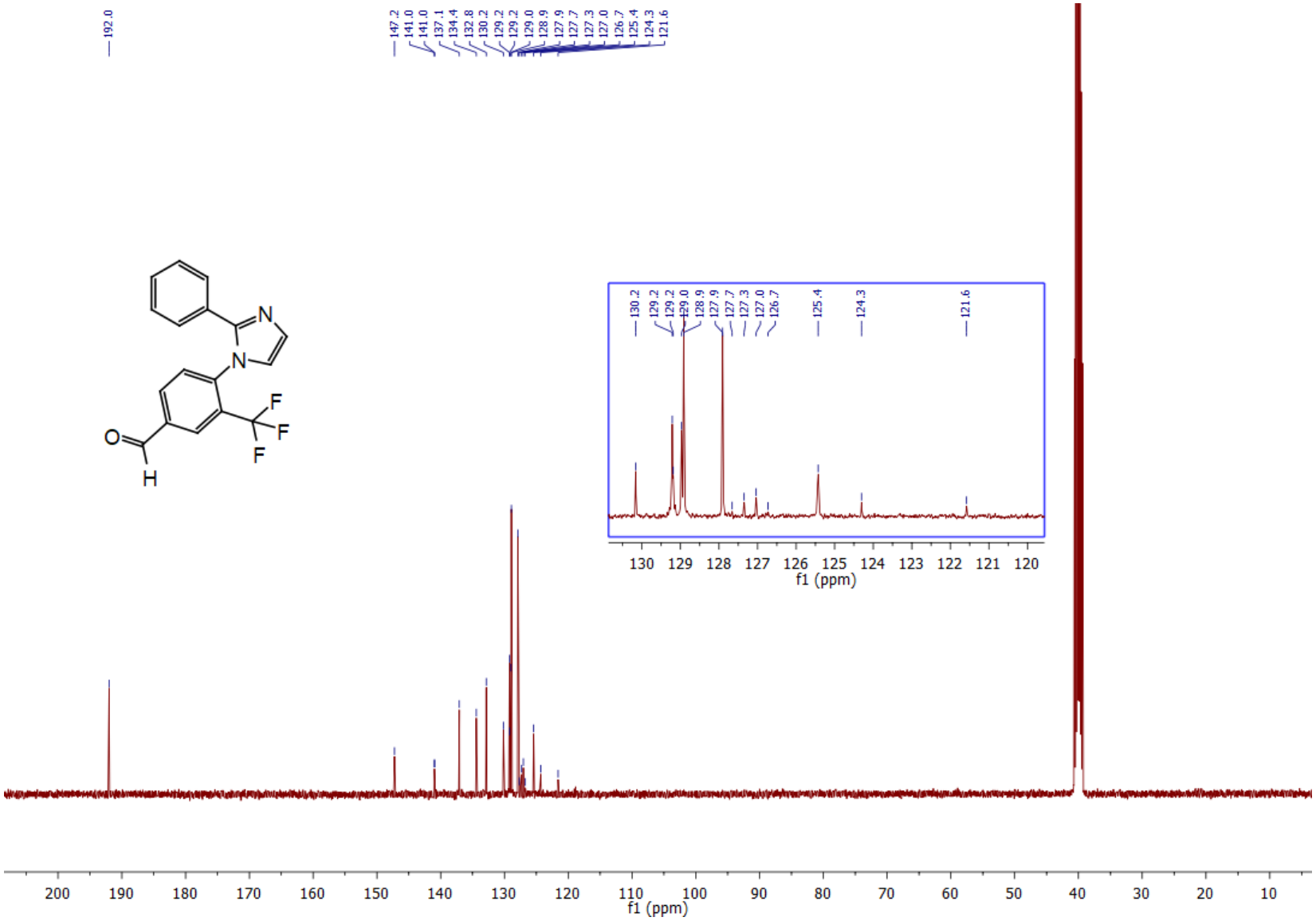

10.36
8.19
8.17
7.86
7.77
7.75
7.61
7.42
7.42
7.40
7.40
7.38
7.36
7.34
7.24
2.82
2.79
2.05 (CD3)2CO
N
N
O
H
F
F
F
1.00
0.99
1.00
1.00
1.07
5.18
1.02
1.5
11.0
10.5
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
f1 (ppm)


189.4
189.4
189.4
189.3
148.3
144.6
134.4
134.4
133.1
132.9
132.6
132.2
131.5
130.7
130.5
130.1
125.6
125.6
125.5
125.4
124.5
N
N
O
H
F
F
F
134.4
134.4
133.1
132.9
132.6
132.2
131.5
130.7
130.5
130.1
125.6
125.6
125.5
125.4
124.5
136 135 134 133 132 131 130 129 128 127 126 125 124 123
f1 (ppm)
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

Fragment 15

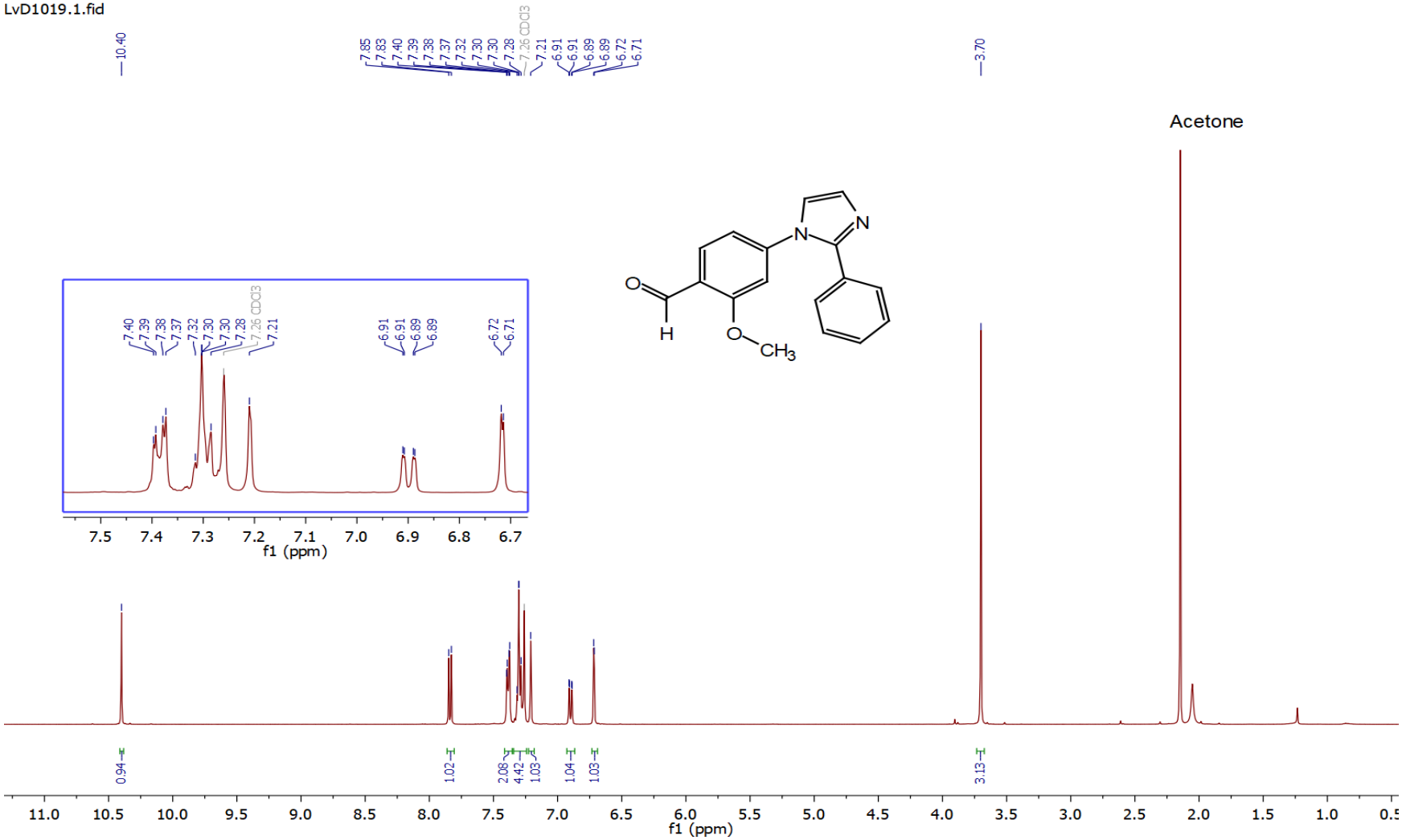
LvD1014.1.fid

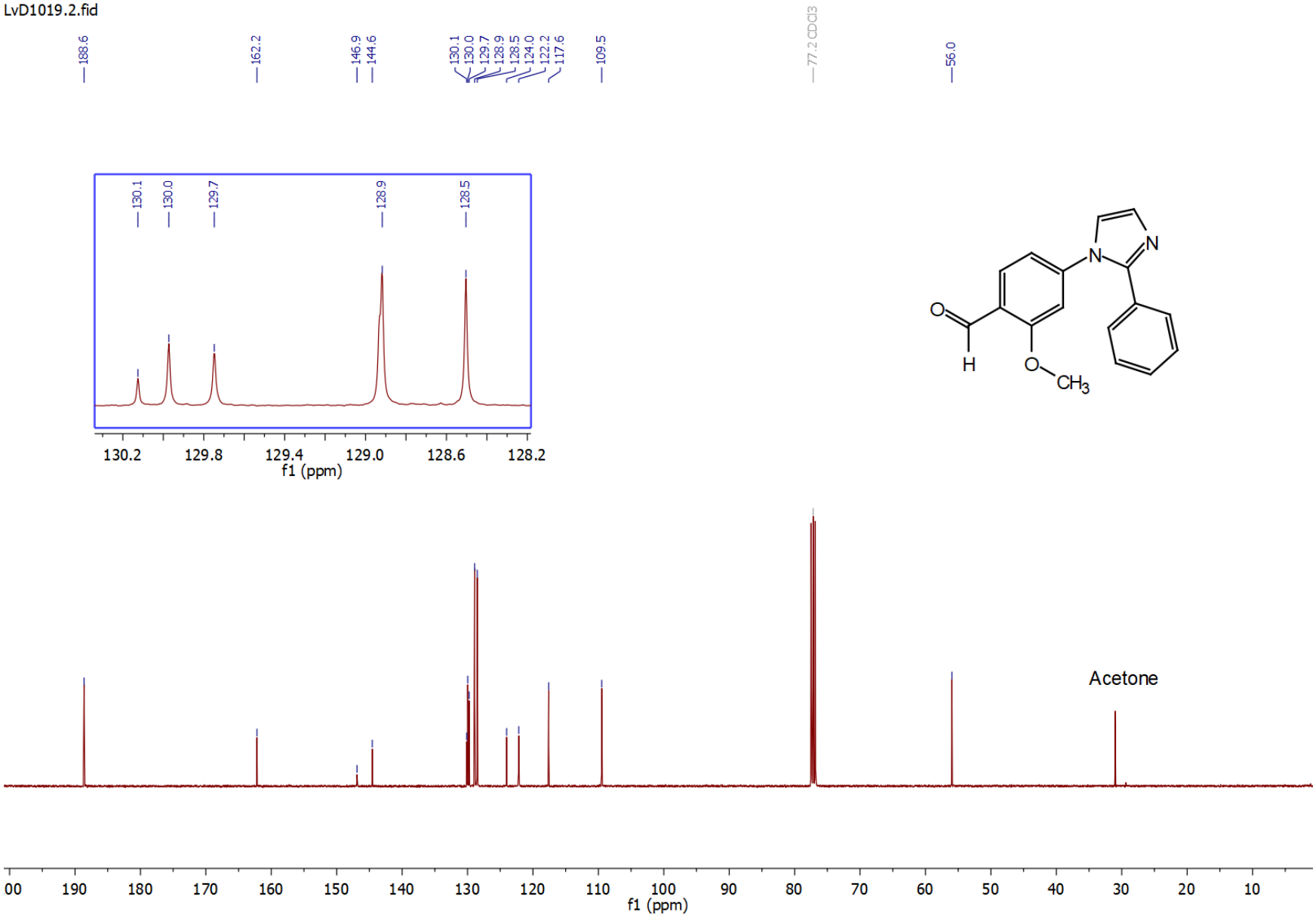



192.0
147.2
141.0
141.0
137.1
134.4
132.8
130.2
129.2
129.2
129.0
128.9
127.9
127.7
127.3
127.0
126.7
125.4
124.3
121.6
N
N
F
F
F
O
H
130.2
129.2
129.2
129.0
128.9
127.9
127.7
127.3
127.0
126.7
125.4
124.3
121.6
130 129 128 127 126 125 124 123 122 121 120
f1 (ppm)
200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

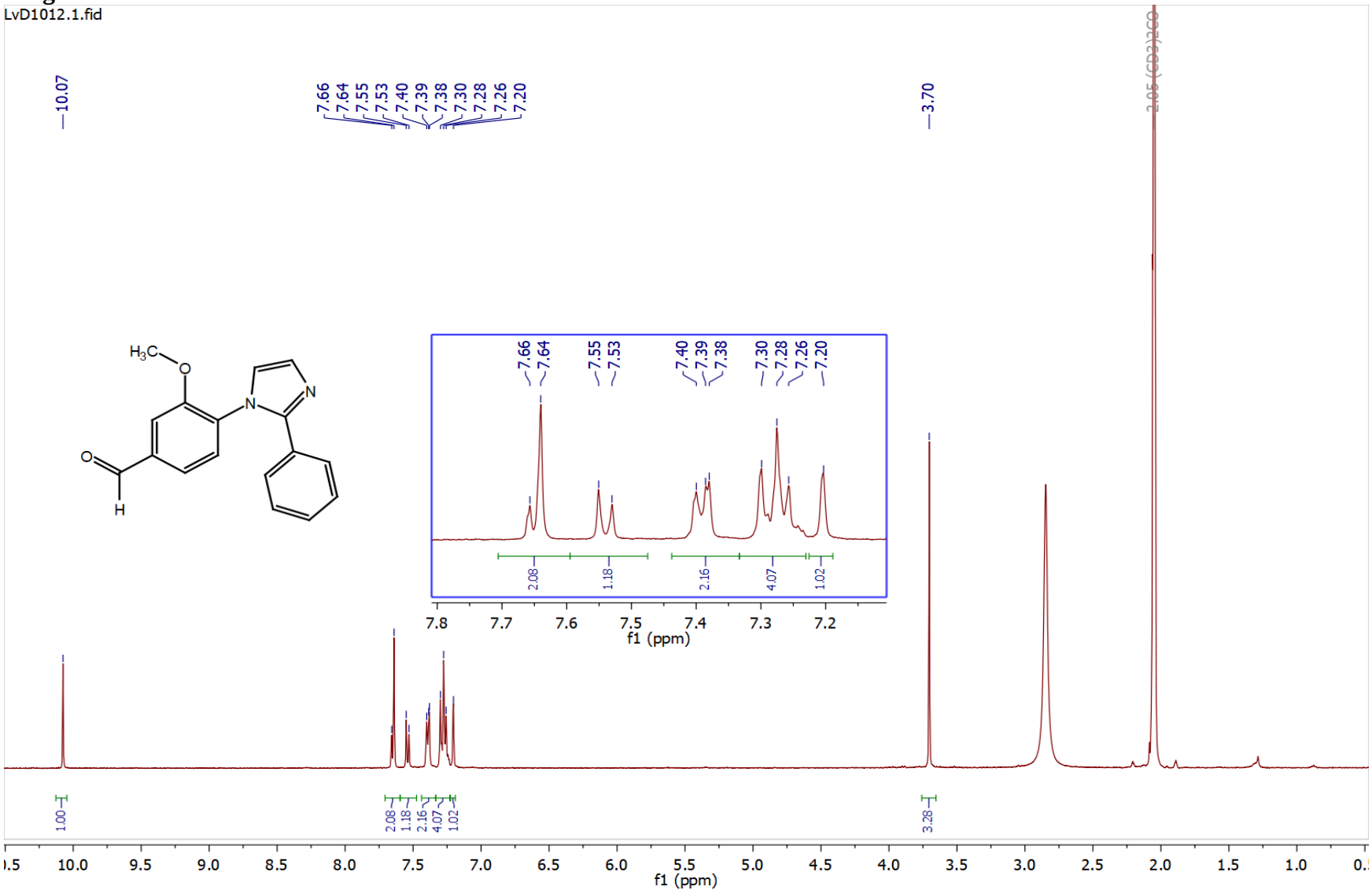
Fragment 16

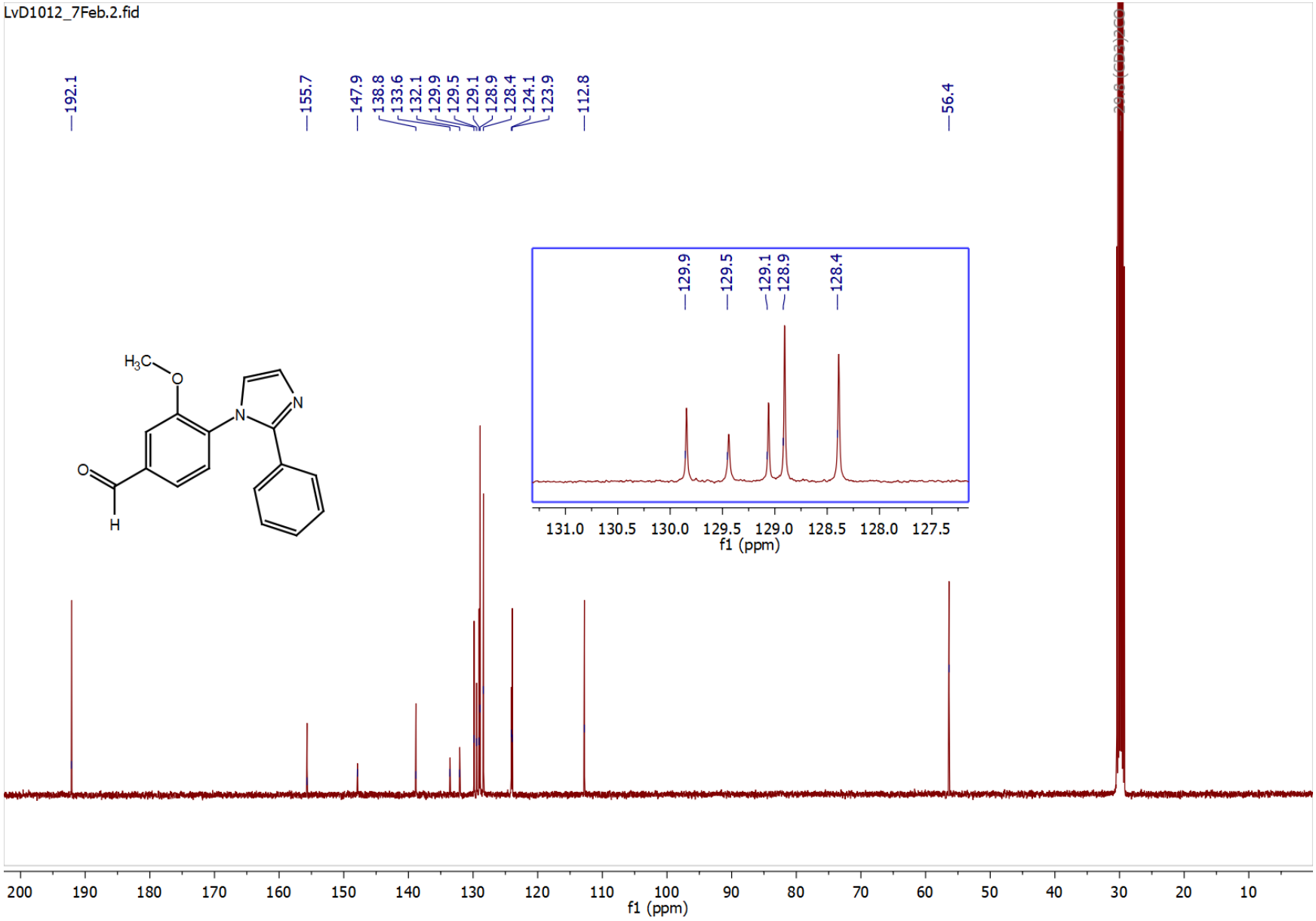
LvD1019.1.fid



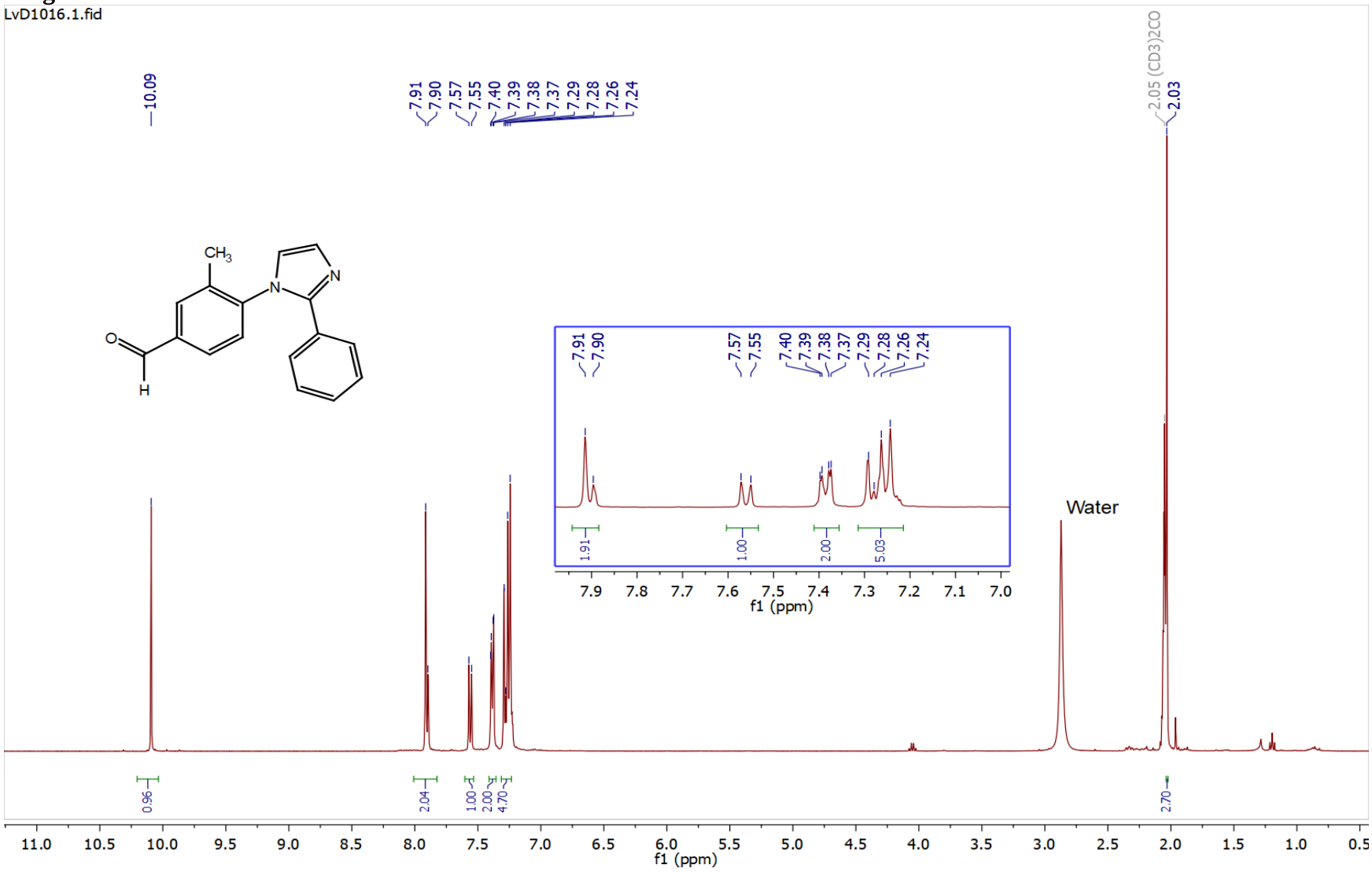

LvD1019.2.fid
188.6
162.2
146.9
144.6
130.1
130.0
129.7
128.9
128.5
124.0
122.2
117.6
109.5
77.2 CDCl3
56.0
130.1
130.0
129.7
128.9
128.5
130.2
129.8
129.4
129.0
128.6
128.2
f1 (ppm)
O
H
O
CH3
N
N
Acetone
00
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

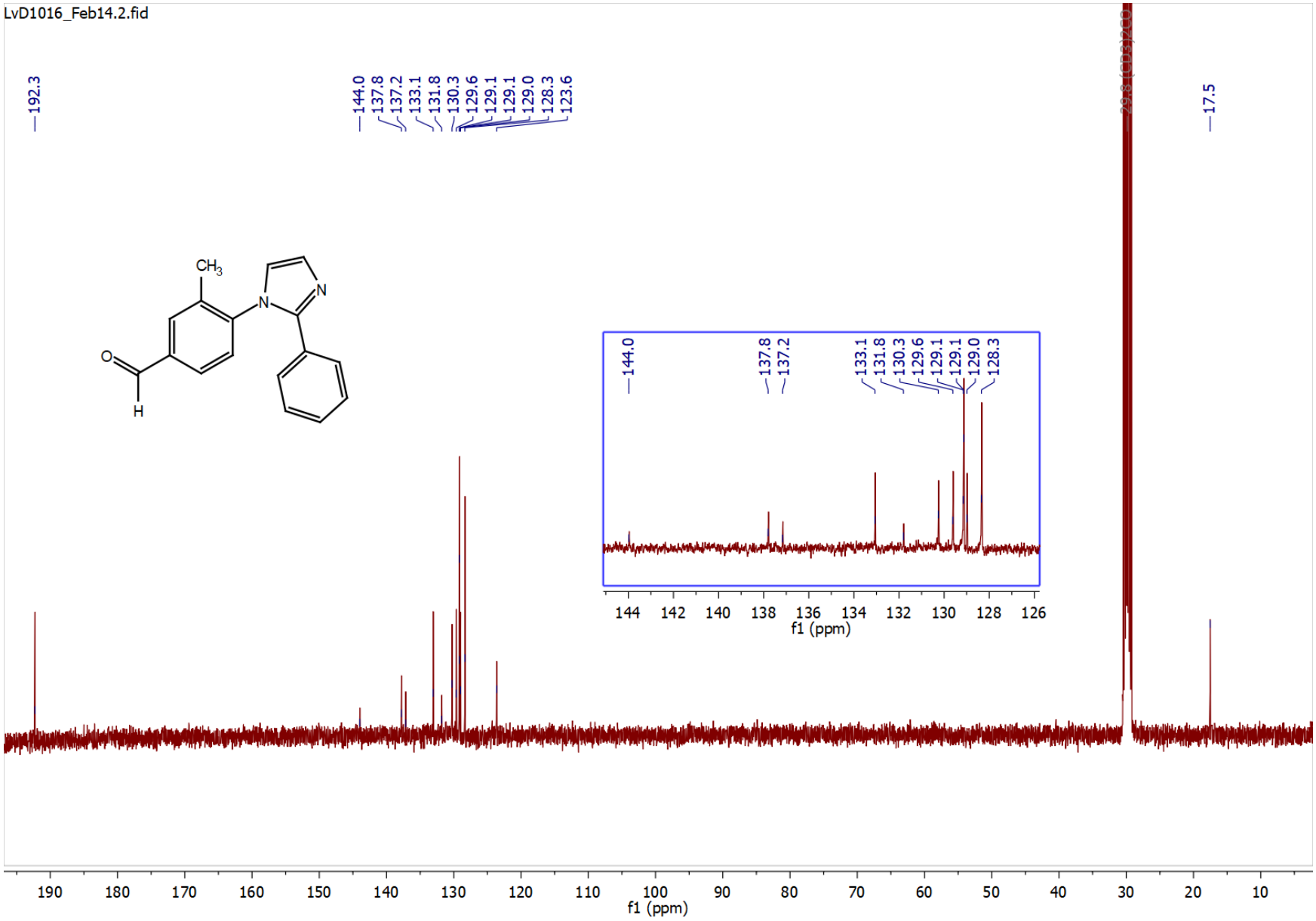
Fragment 17


LvD1012.1.fid
10.07
7.66
7.64
7.55
7.53
7.40
7.39
7.38
7.30
7.28
7.26
7.20
3.70
2.05 (CD3)2CO
H3C
O
N
N
O
H
2.08
1.18
2.16
4.07
1.02
7.8
7.7
7.6
7.5
7.4
7.3
7.2
f1 (ppm)
1.00
3.28
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
f1 (ppm)


LvD1012_7Feb.2.fid
192.1
155.7
147.9
138.8
133.6
132.1
129.9
129.5
129.1
128.9
128.4
124.1
123.9
112.8
56.4
129.9
129.5
129.1
128.9
128.4
131.0 130.5 130.0 129.5 129.0 128.5 128.0 127.5
f1 (ppm)
200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

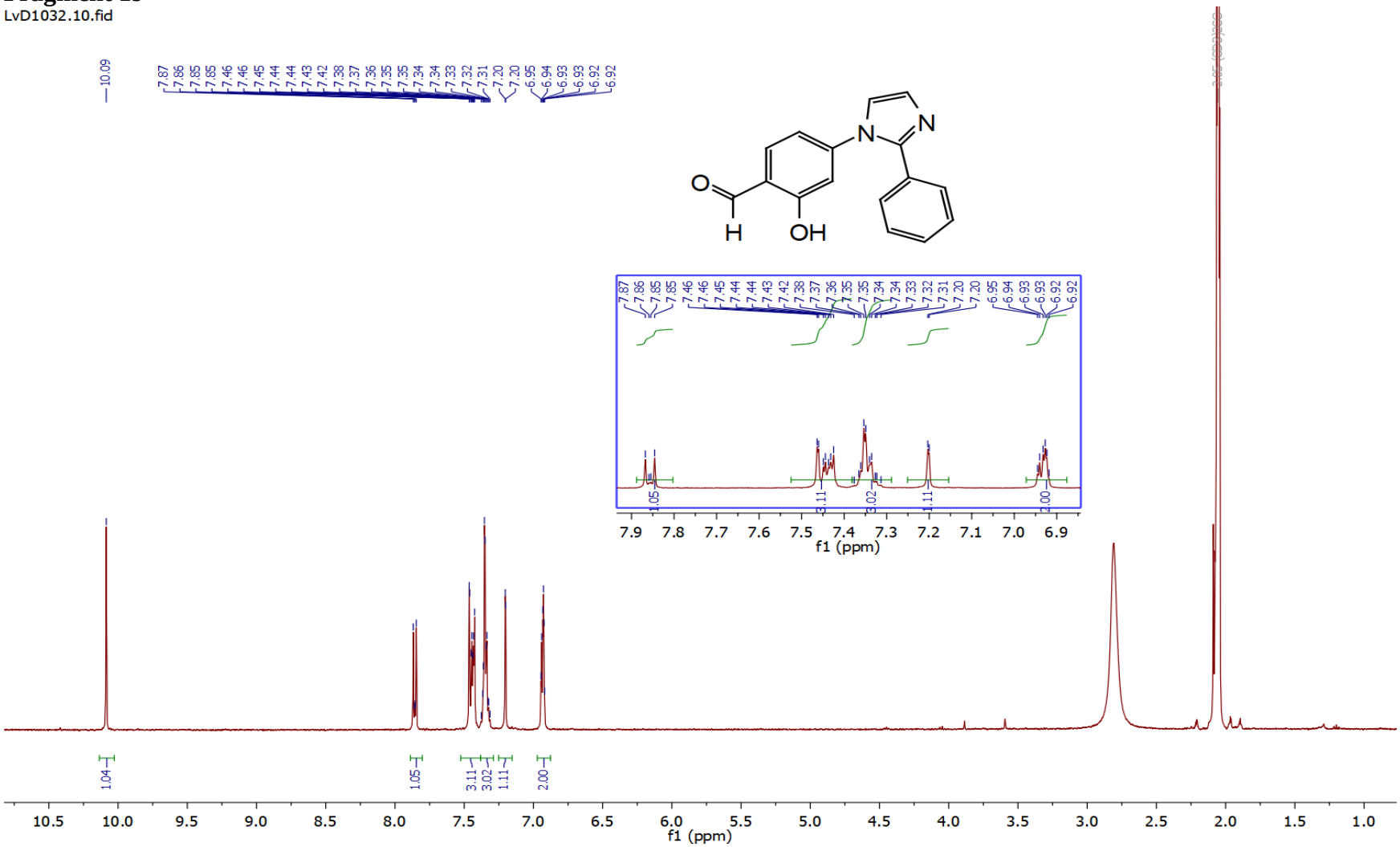
Fragment 18

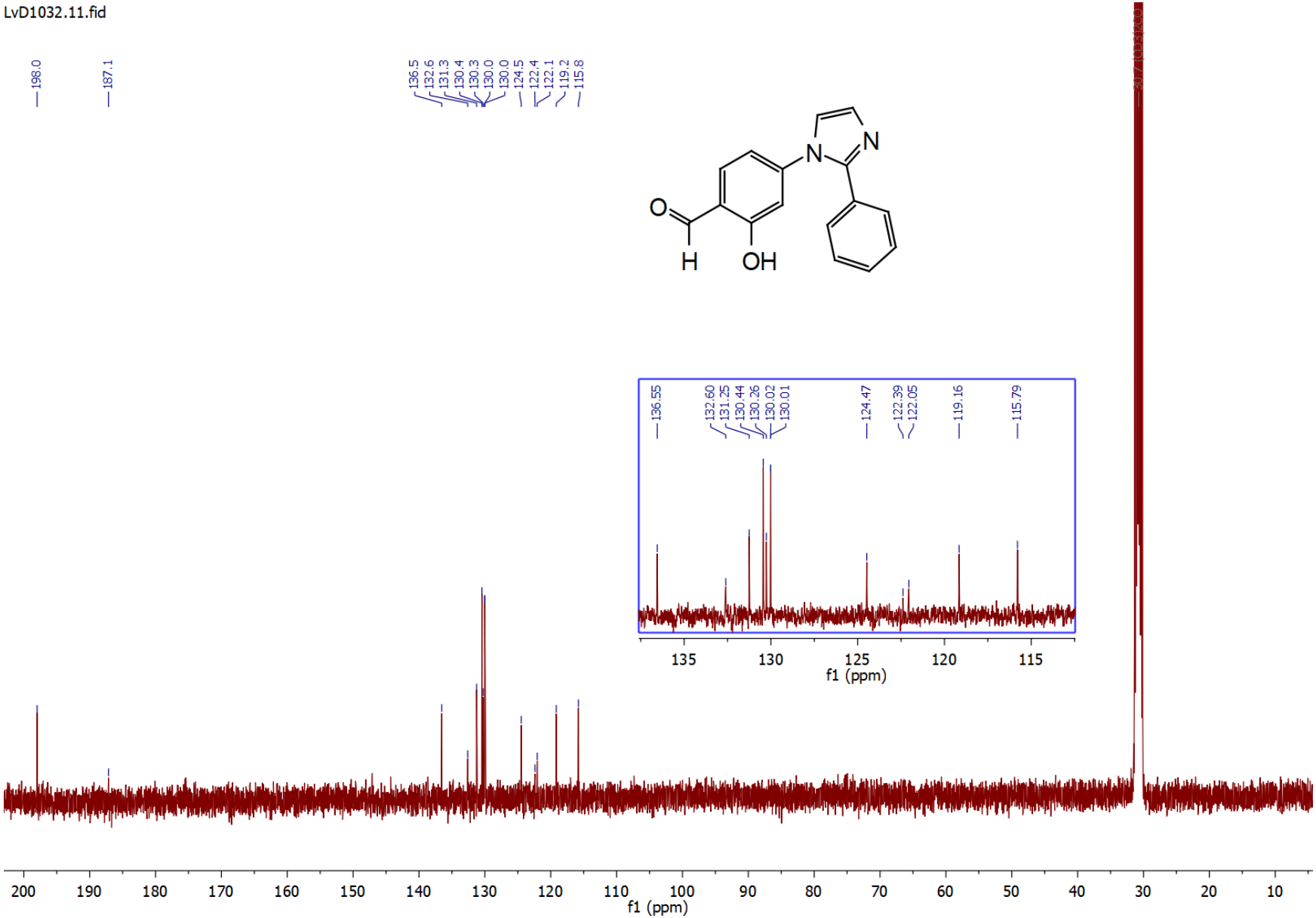

LvD1016.1.fid
10.09
7.91
7.90
7.57
7.55
7.40
7.39
7.38
7.37
7.29
7.28
7.26
7.24
2.05 (CD3)2CO
2.03
Water
CH3
N
N
O
H
1.91
1.00
2.00
5.03
f1 (ppm)
0.96
2.04
1.00
2.00
4.70
2.70
f1 (ppm)


LvD1016_Feb14.2.fid
192.3
144.0
137.8
137.2
133.1
131.8
130.3
129.6
129.1
129.1
129.0
128.3
123.6
17.5
CH3
N
N
O
H
144.0
137.8
137.2
133.1
131.8
130.3
129.6
129.1
129.1
129.0
128.3
144 142 140 138 136 134 132 130 128 126
f1 (ppm)
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

Fragment 19

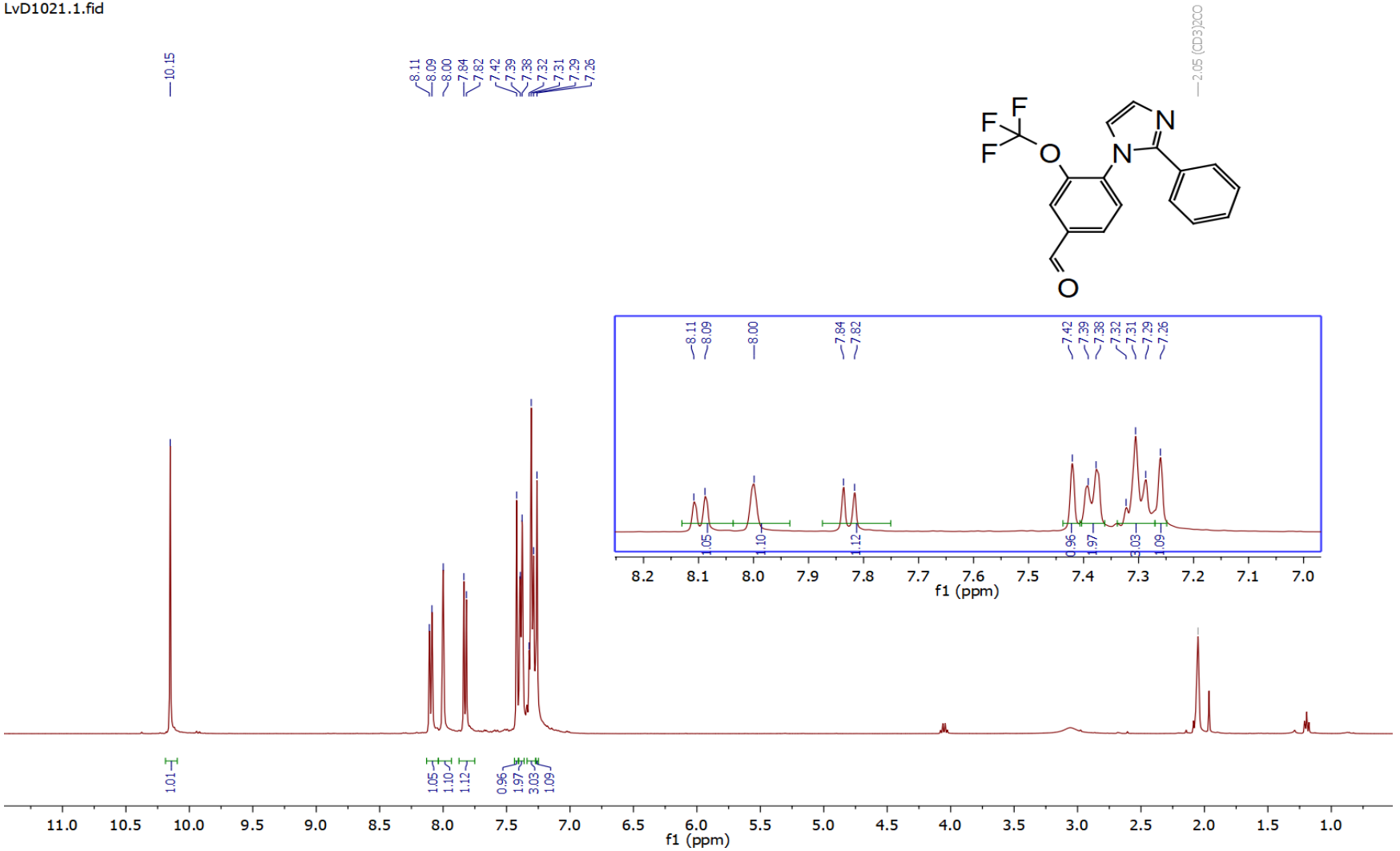
LvD1032.10.fid

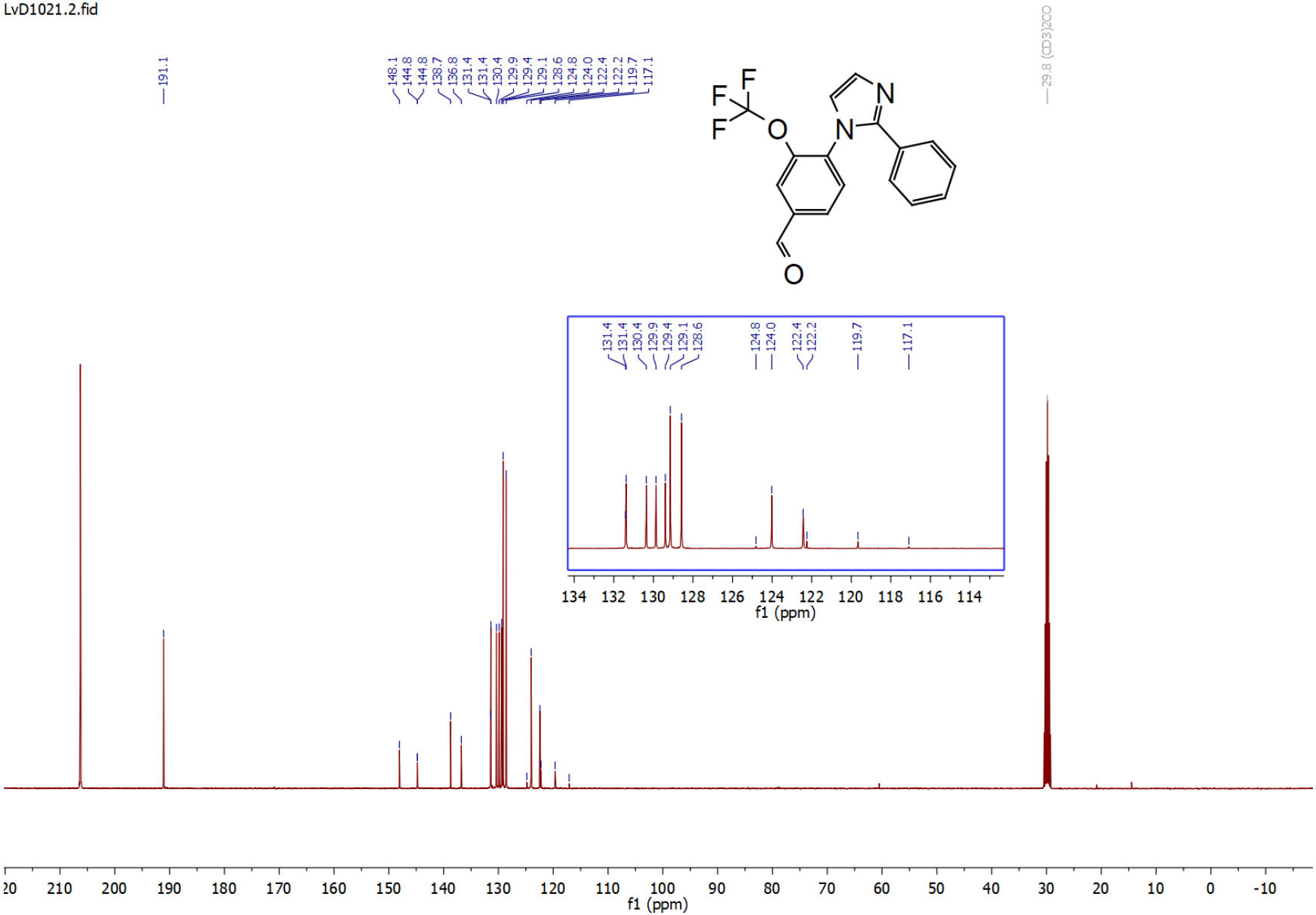



LvD1032.11.fid
198.0
187.1
136.5
132.6
131.3
130.4
130.3
130.0
130.0
124.5
122.4
122.1
119.2
115.8
O
H
OH
N
N
136.55
132.60
131.25
130.44
130.26
130.02
130.01
124.47
122.39
122.05
119.16
115.79
135
130
125
120
115
f1 (ppm)
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

Fragment 20

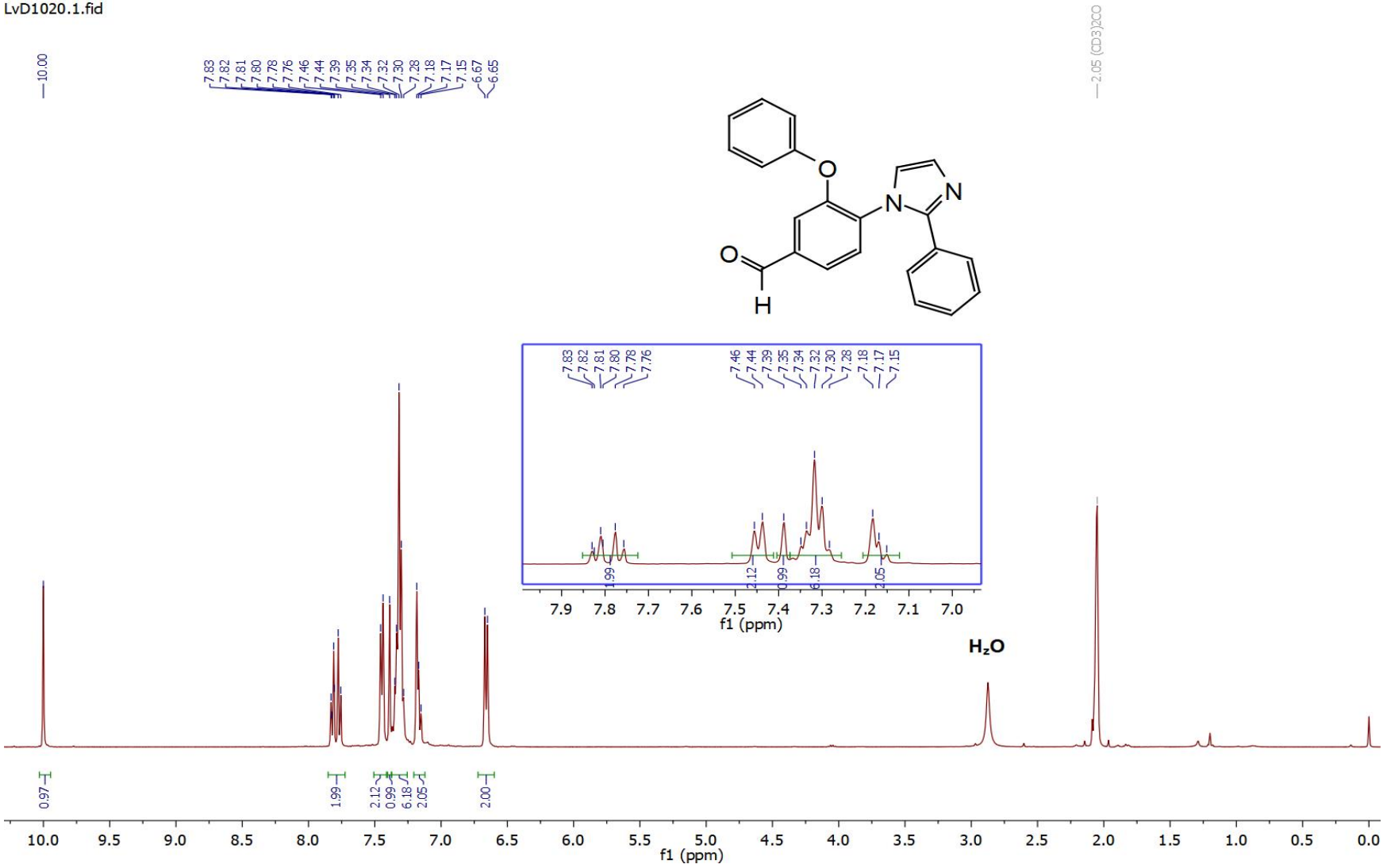
LvD1021.1.fid

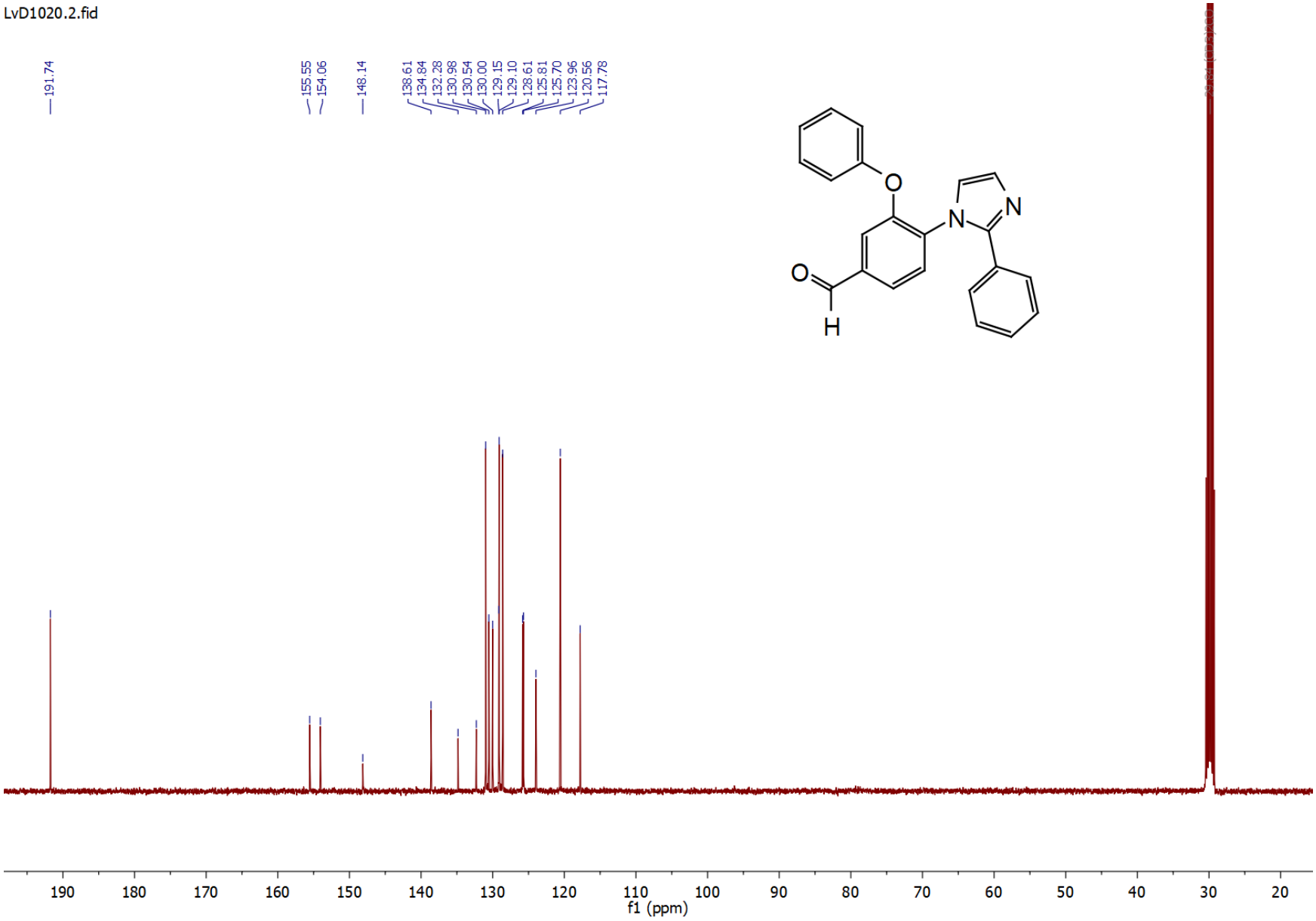



LvD1021.2.fid
191.1
148.1
144.8
144.8
138.7
136.8
131.4
131.4
130.4
129.9
129.4
129.1
128.6
124.8
124.0
122.4
122.2
119.7
117.1
29.8 (CD3)2CO
F
F
F
O
N
N
O
131.4
131.4
130.4
129.9
129.4
129.1
128.6
124.8
124.0
122.4
122.2
119.7
117.1
134 132 130 128 126 124 122 120 118 116 114
f1 (ppm)
210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10
f1 (ppm)

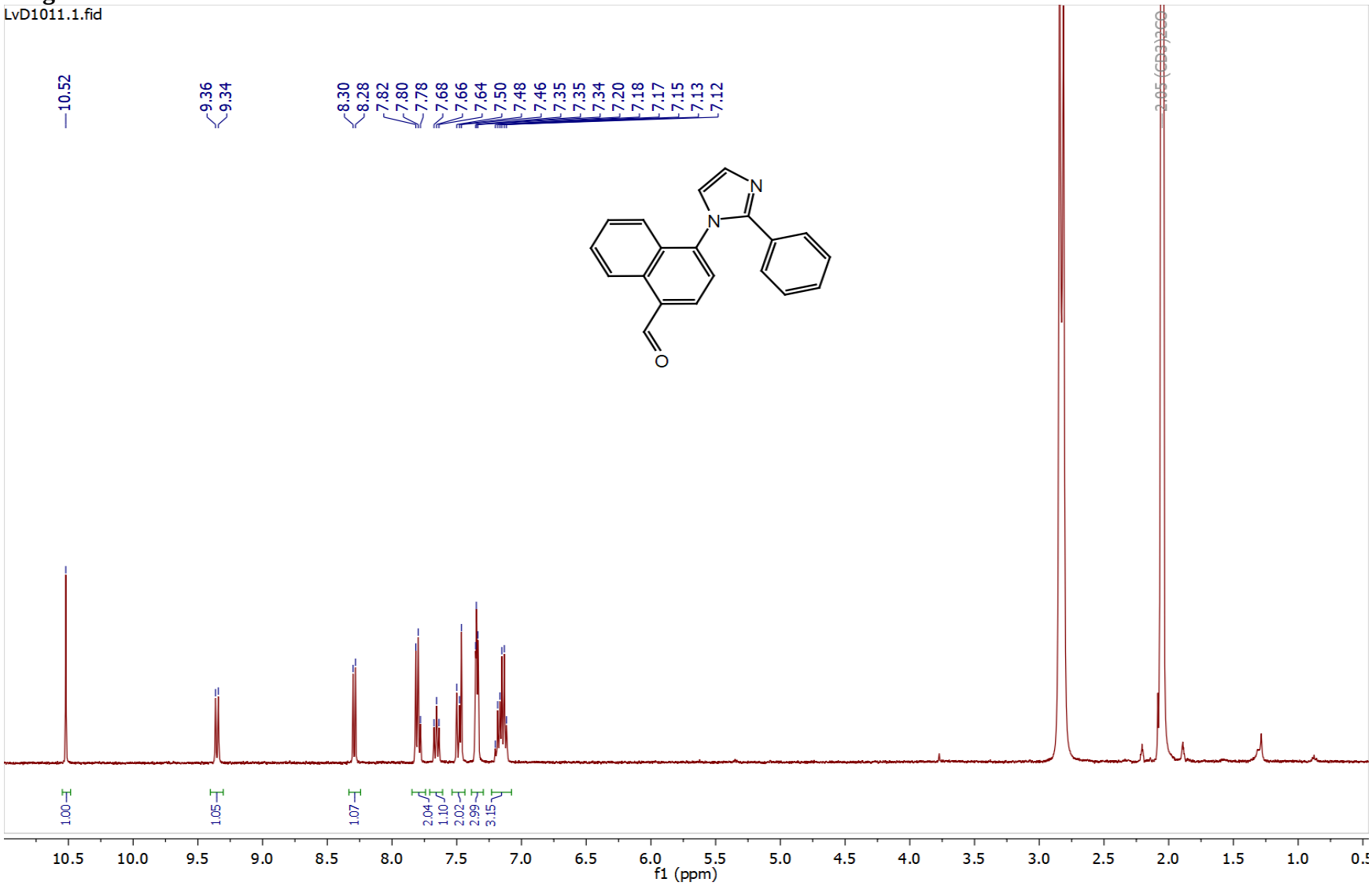
Fragment 21

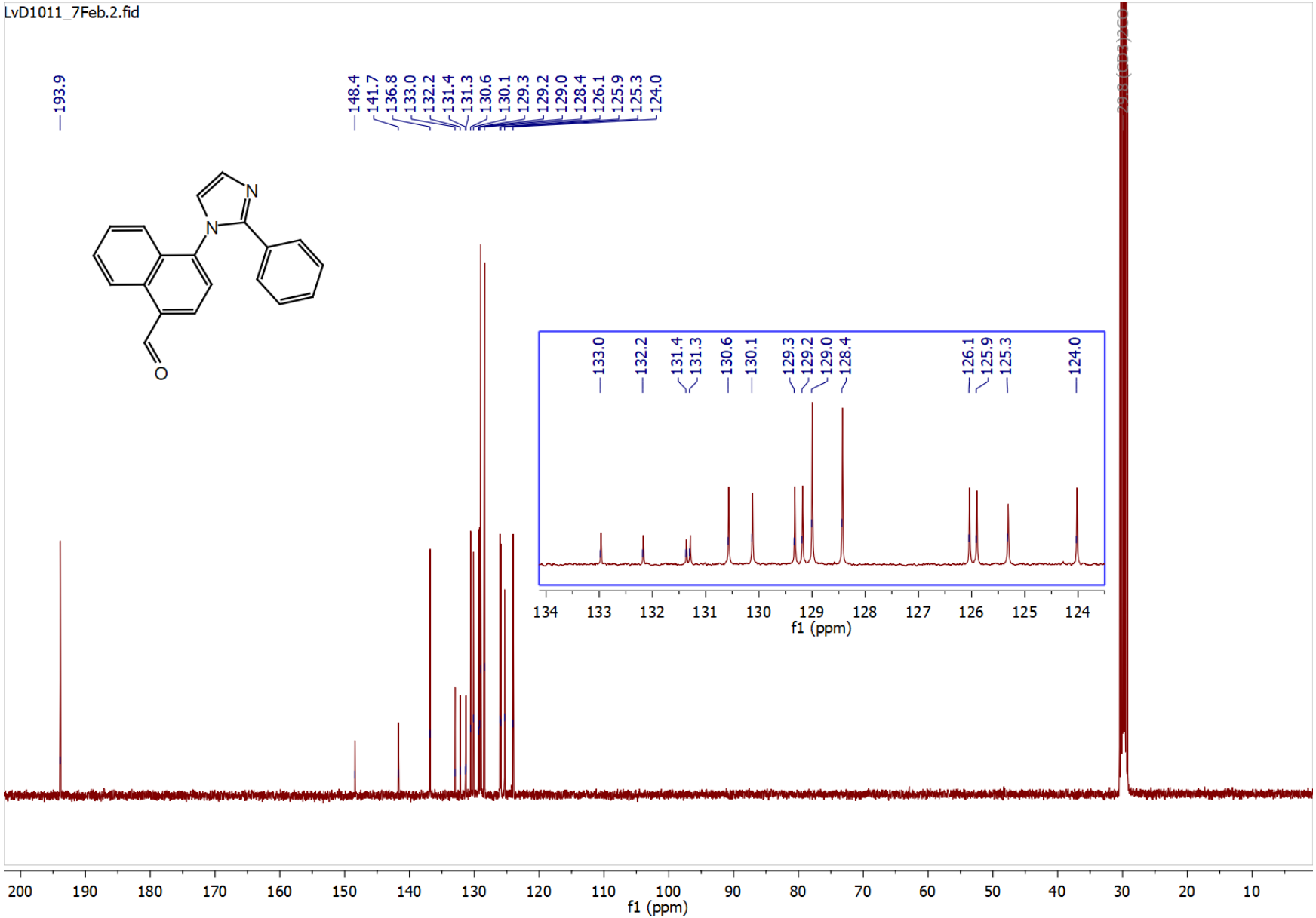
LvD1020.1.fid




LvD1020.2.fid
191.74
155.55
154.06
148.14
138.61
134.84
132.28
130.98
130.54
130.00
129.15
129.10
128.61
125.81
125.70
123.96
120.56
117.78
O
N
N
O
H
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
f1 (ppm)

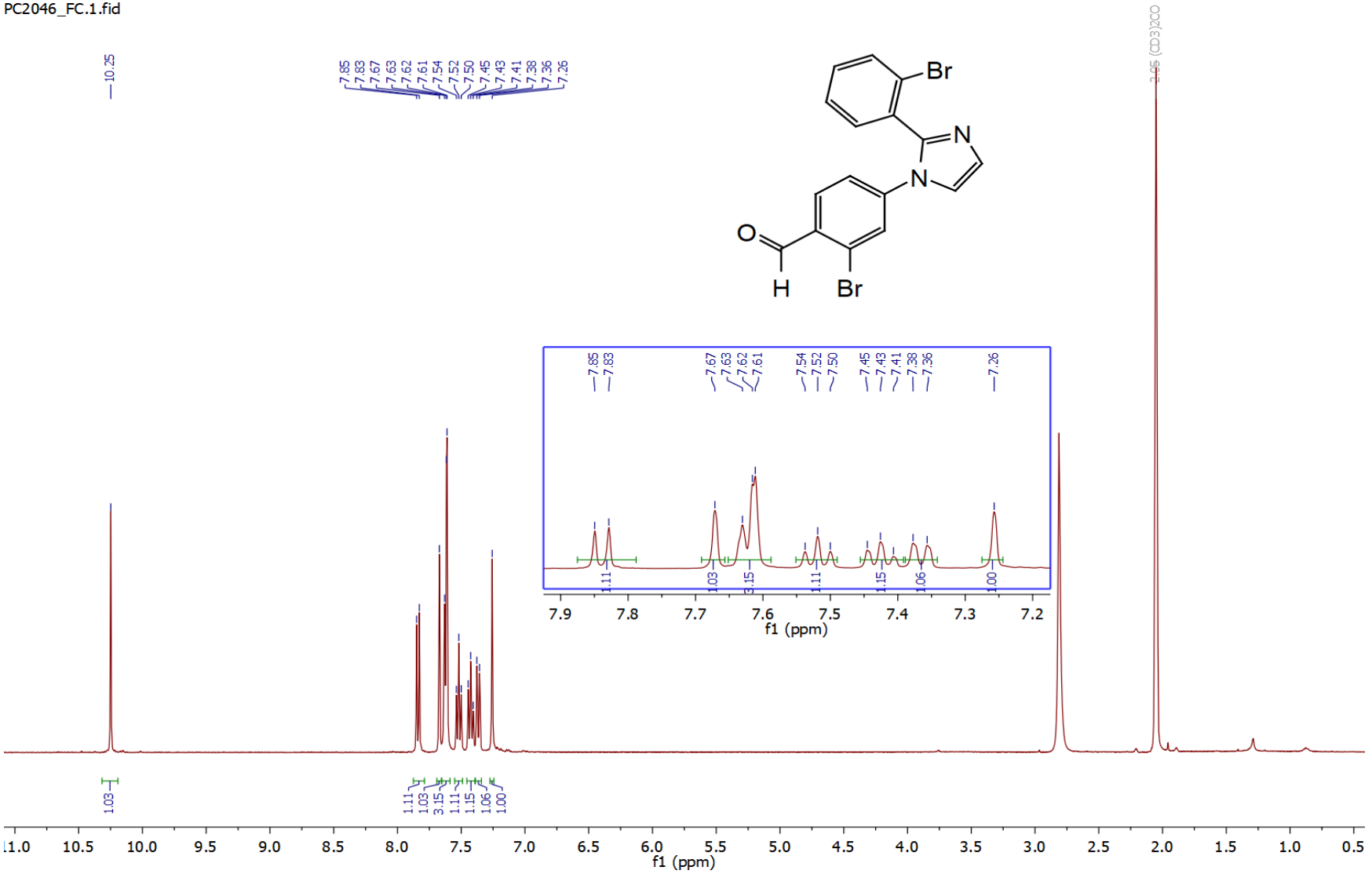
Fragment 22

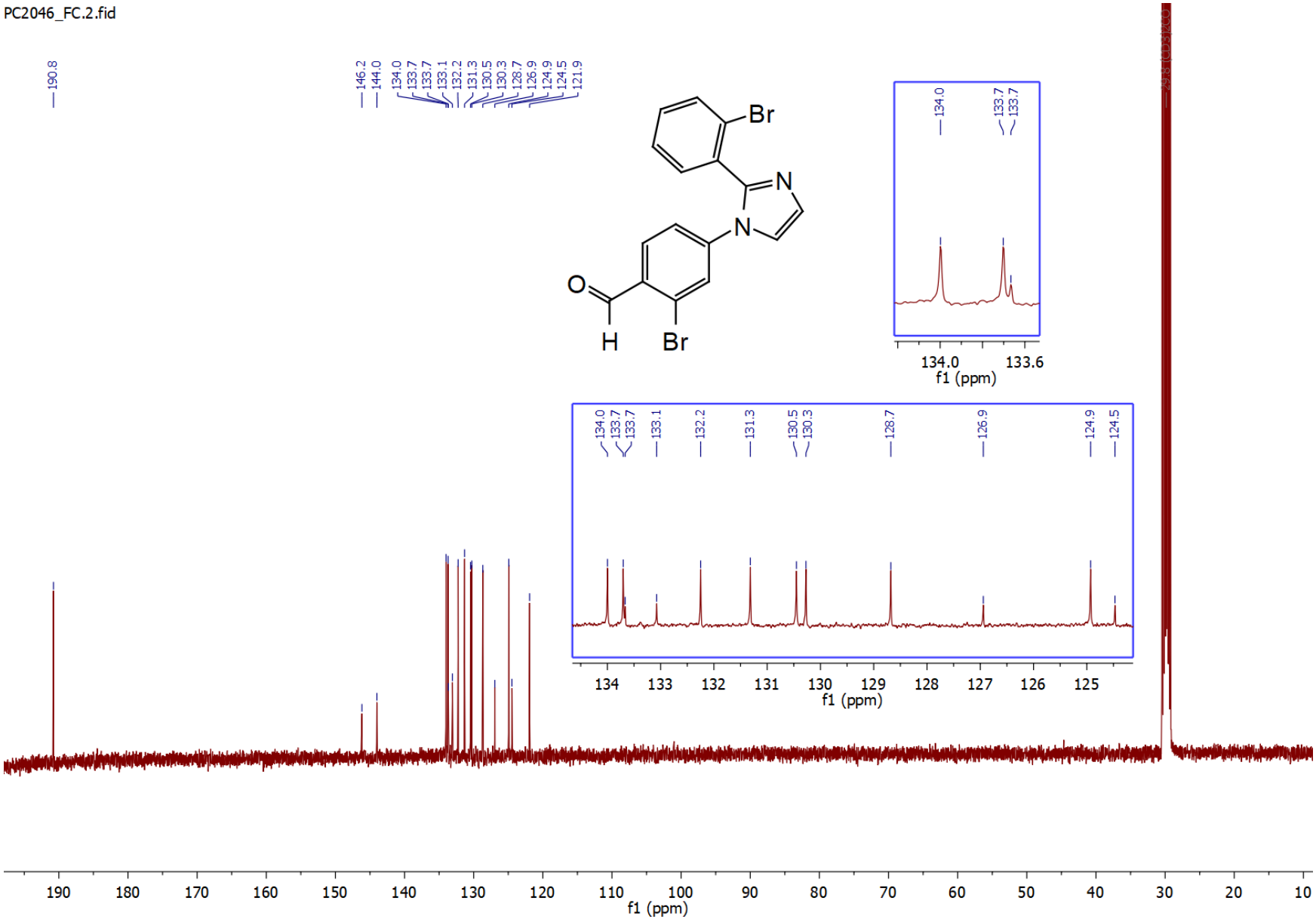

LvD1011.1.fid
10.52
9.36
9.34
8.30
8.28
7.82
7.80
7.78
7.68
7.66
7.64
7.50
7.48
7.46
7.35
7.35
7.34
7.20
7.18
7.17
7.15
7.13
7.12
2.05 (CD3)2CO
N
N
O
1.00
1.05
1.07
2.04
1.10
2.02
2.99
3.15
10.5
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
f1 (ppm)


LvD1011_7Feb.2.fid
193.9
148.4
141.7
136.8
133.0
132.2
131.4
131.3
130.6
130.1
129.3
129.2
129.0
128.4
126.1
125.9
125.3
124.0
N
N
O
f1 (ppm)
f1 (ppm)

Fragment 23

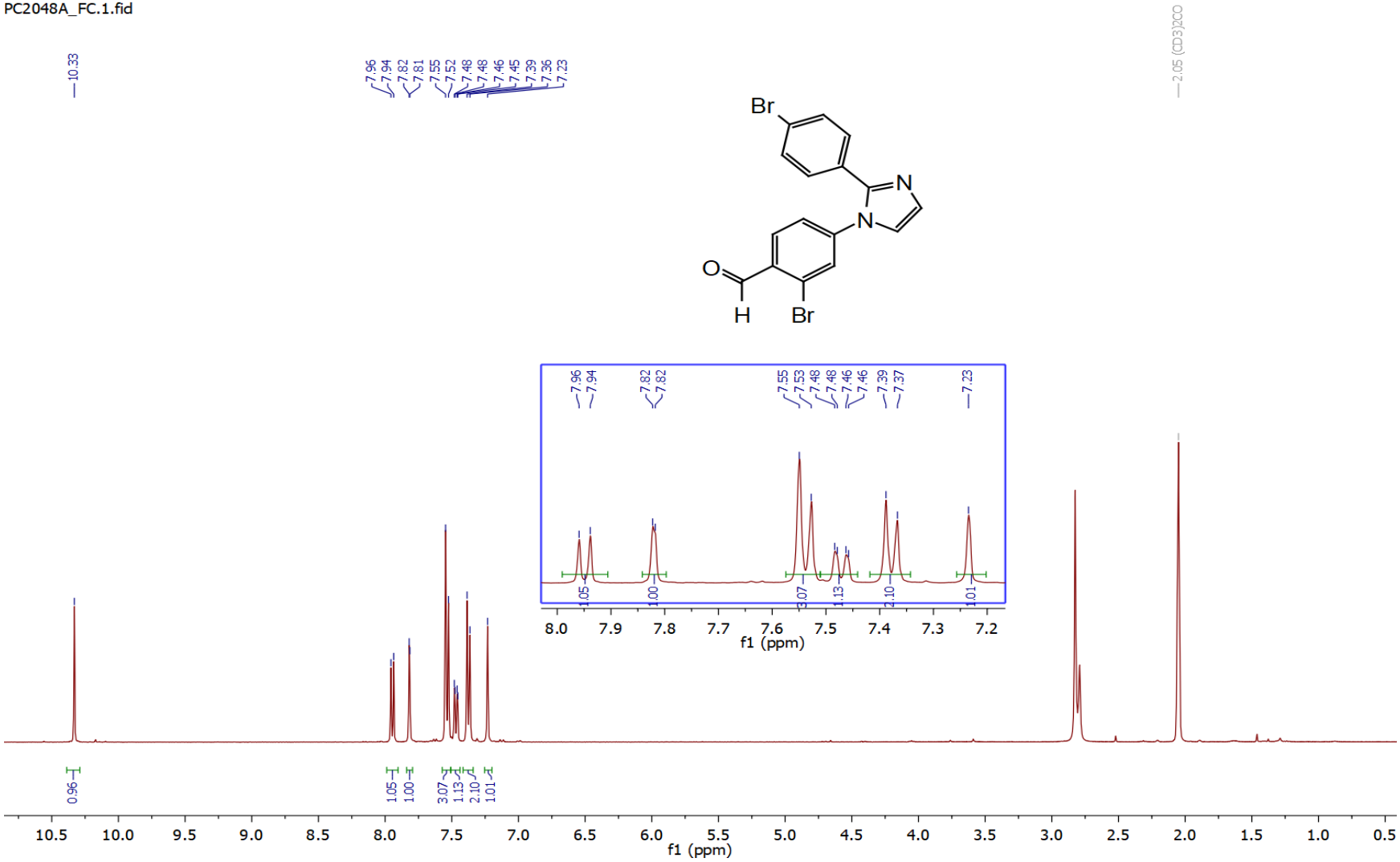
PC2046_FC.1.fid

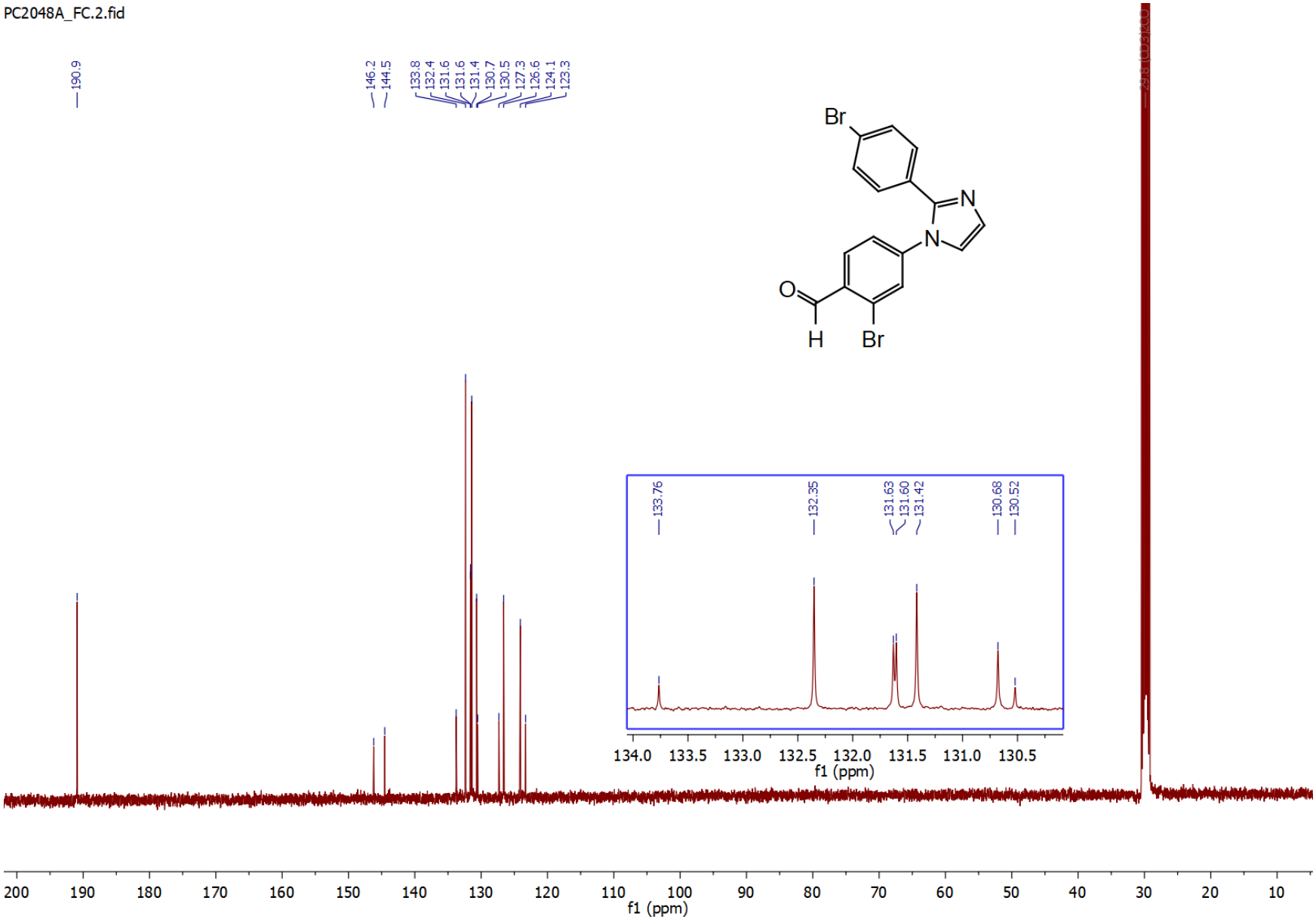



PC2046_FC.2.fid
190.8
146.2
144.0
134.0
133.7
133.7
133.1
132.2
131.3
130.5
130.3
128.7
126.9
124.9
124.5
121.9
Br
N
N
O
H
Br
134.0
133.7
133.7
134.0
133.6
f1 (ppm)
134.0
133.7
133.7
133.1
132.2
131.3
130.5
130.3
128.7
126.9
124.9
124.5
134
133
132
131
130
129
128
127
126
125
f1 (ppm)
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

Fragment 24

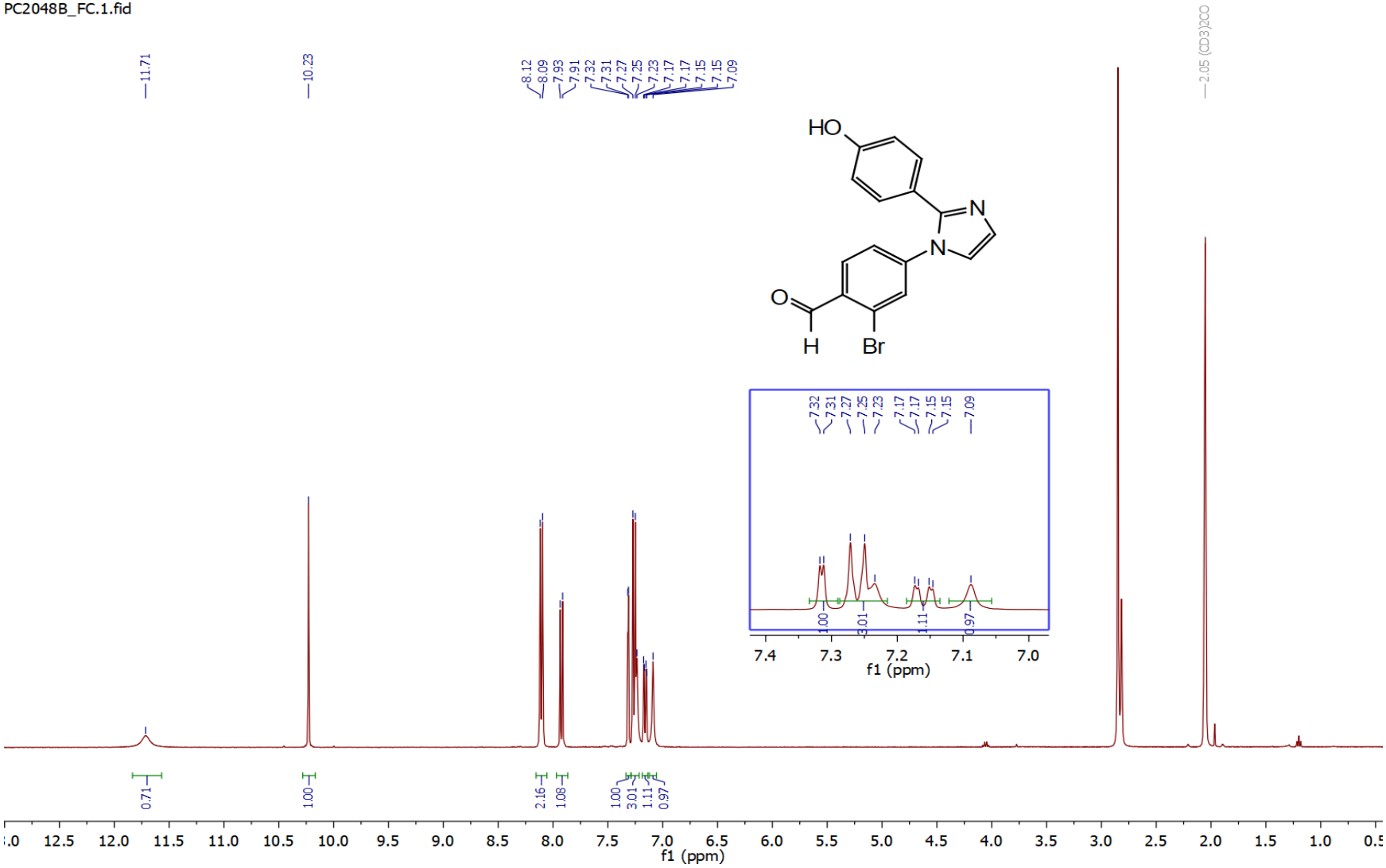
PC2048A_FC.1.fid

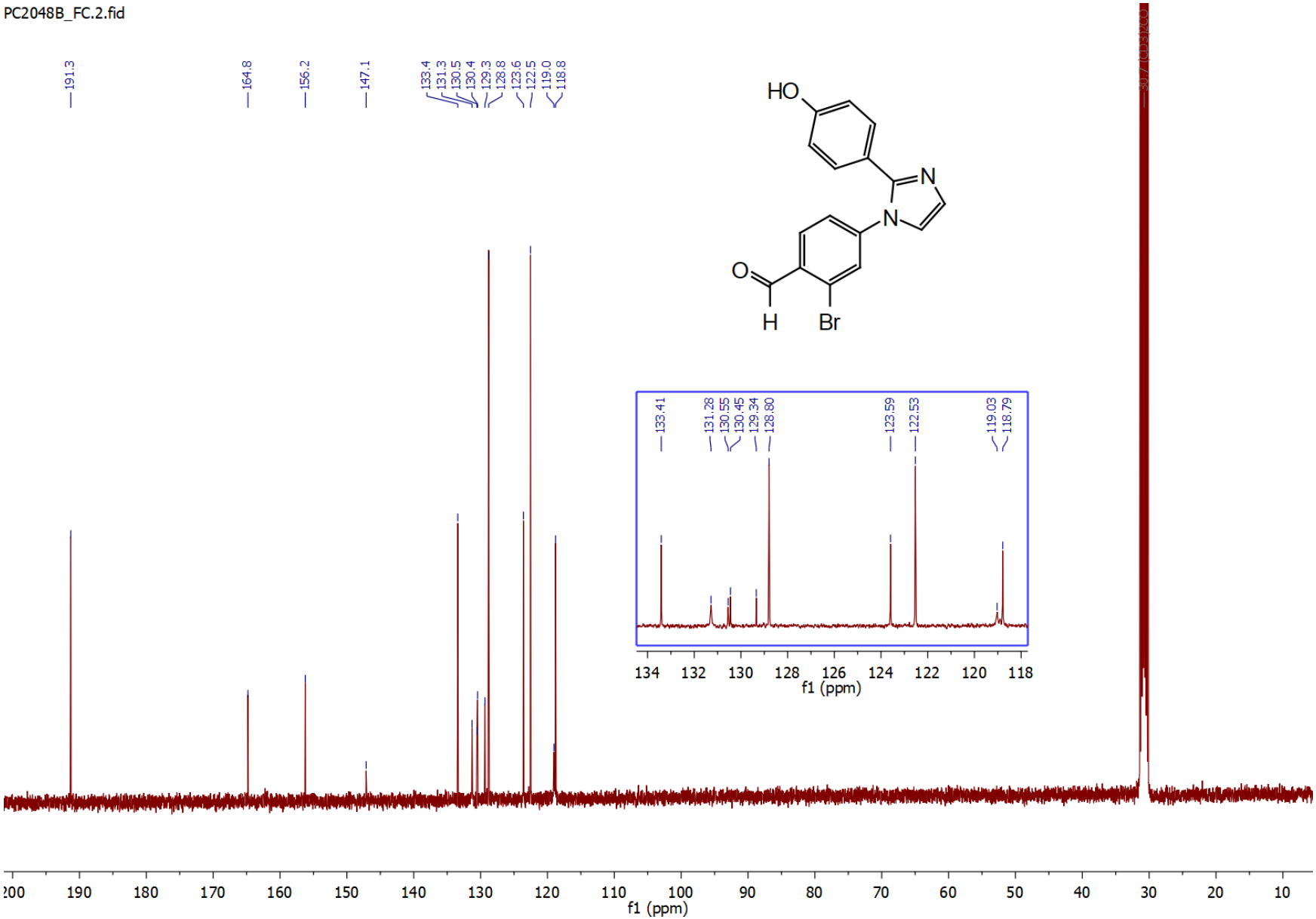



PC2048A_FC.2.fid
190.9
146.2
144.5
133.8
132.4
131.6
131.6
131.4
130.7
130.5
127.3
126.6
124.1
123.3
Br
N
N
O
H
Br
133.76
132.35
131.63
131.60
131.42
130.68
130.52
134.0 133.5 133.0 132.5 132.0 131.5 131.0 130.5
f1 (ppm)
200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

Fragment 25

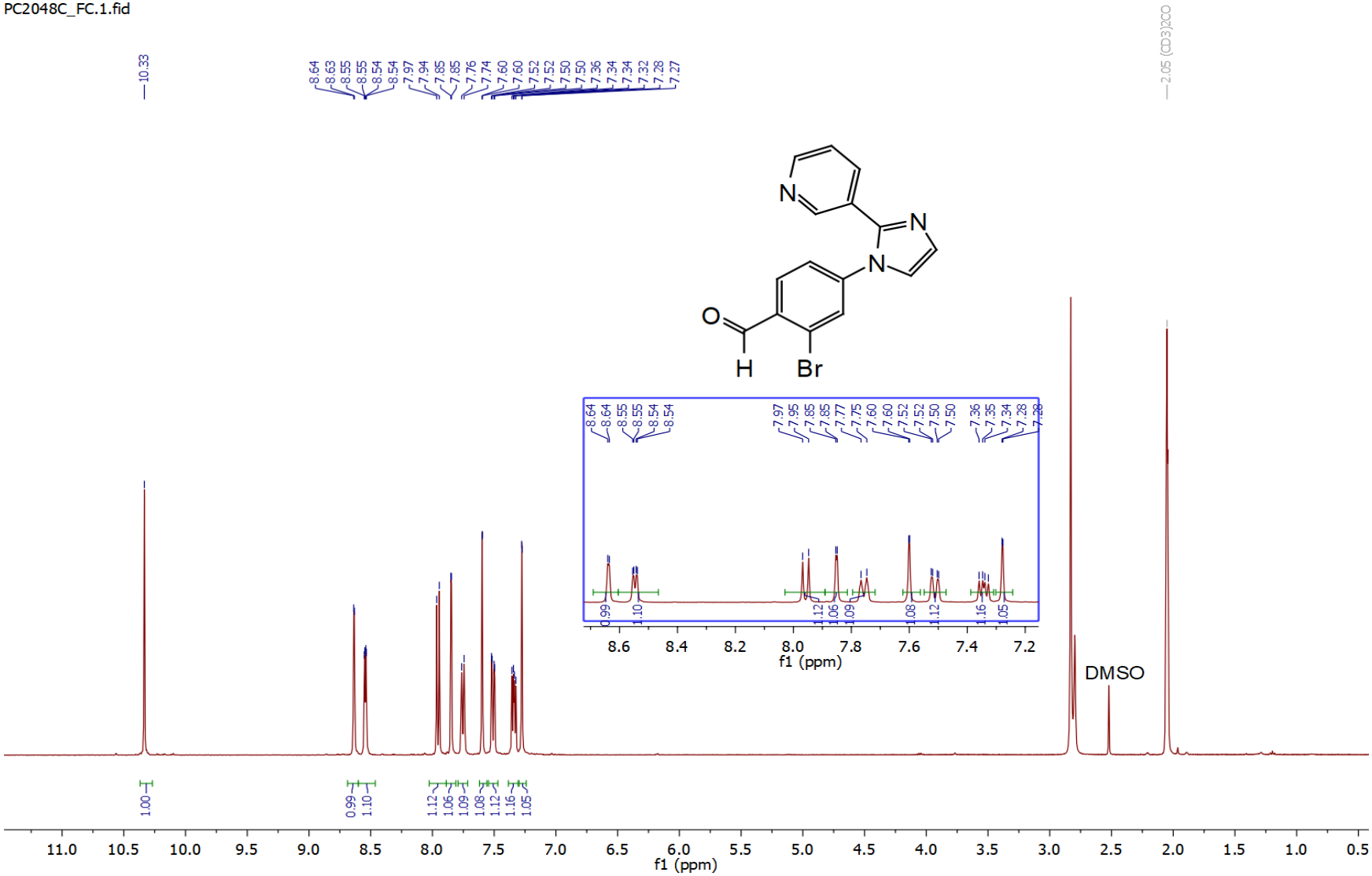
PC2048B_FC.1.fid



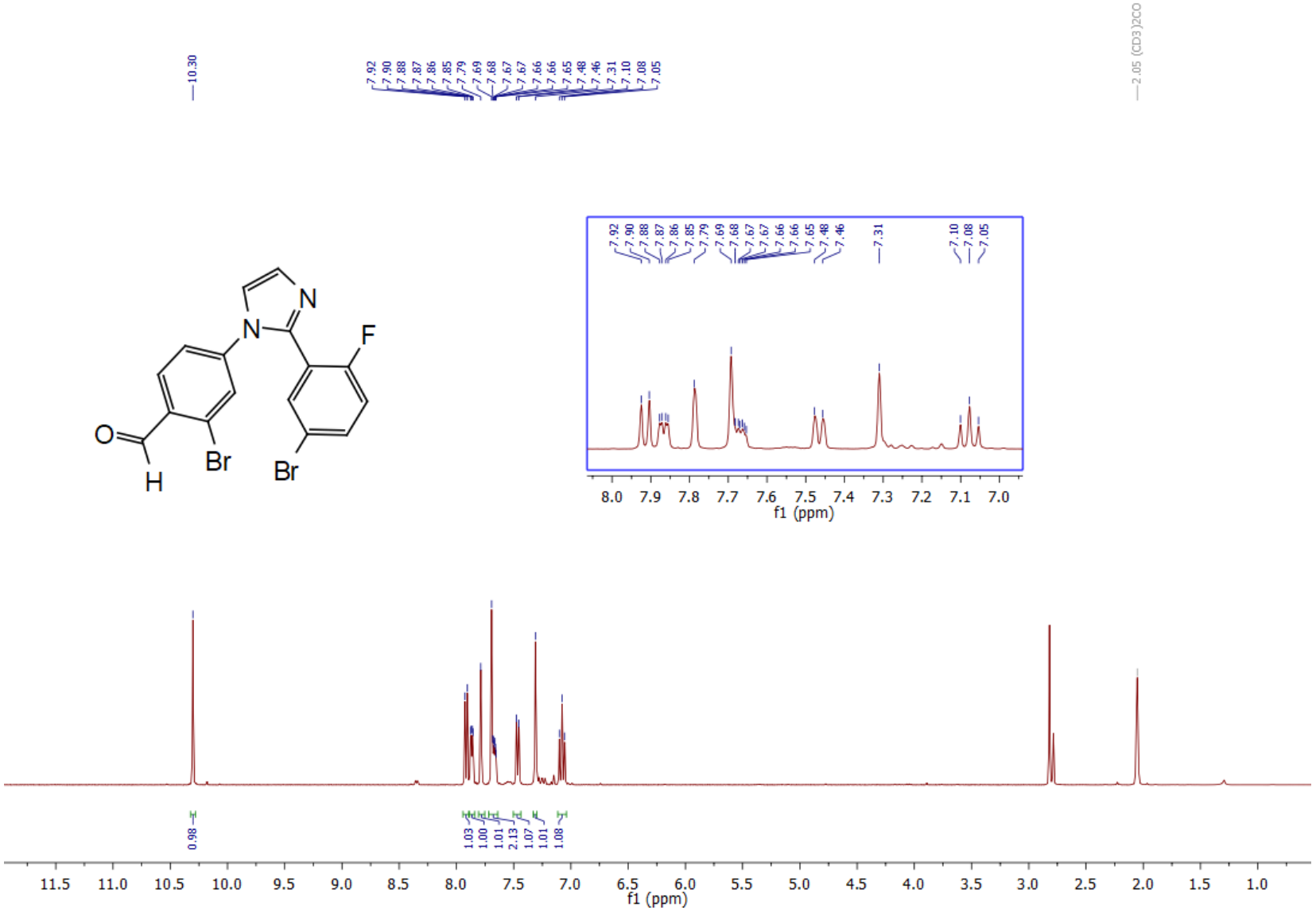

PC2048B_FC.2.fid
191.3
164.8
156.2
147.1
133.4
131.3
130.5
130.4
129.3
128.8
123.6
122.5
119.0
118.8
HO
N
N
O
H
Br
133.41
131.28
130.55
130.45
129.34
128.80
123.59
122.53
119.03
118.79
134 132 130 128 126 124 122 120 118
f1 (ppm)
200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)

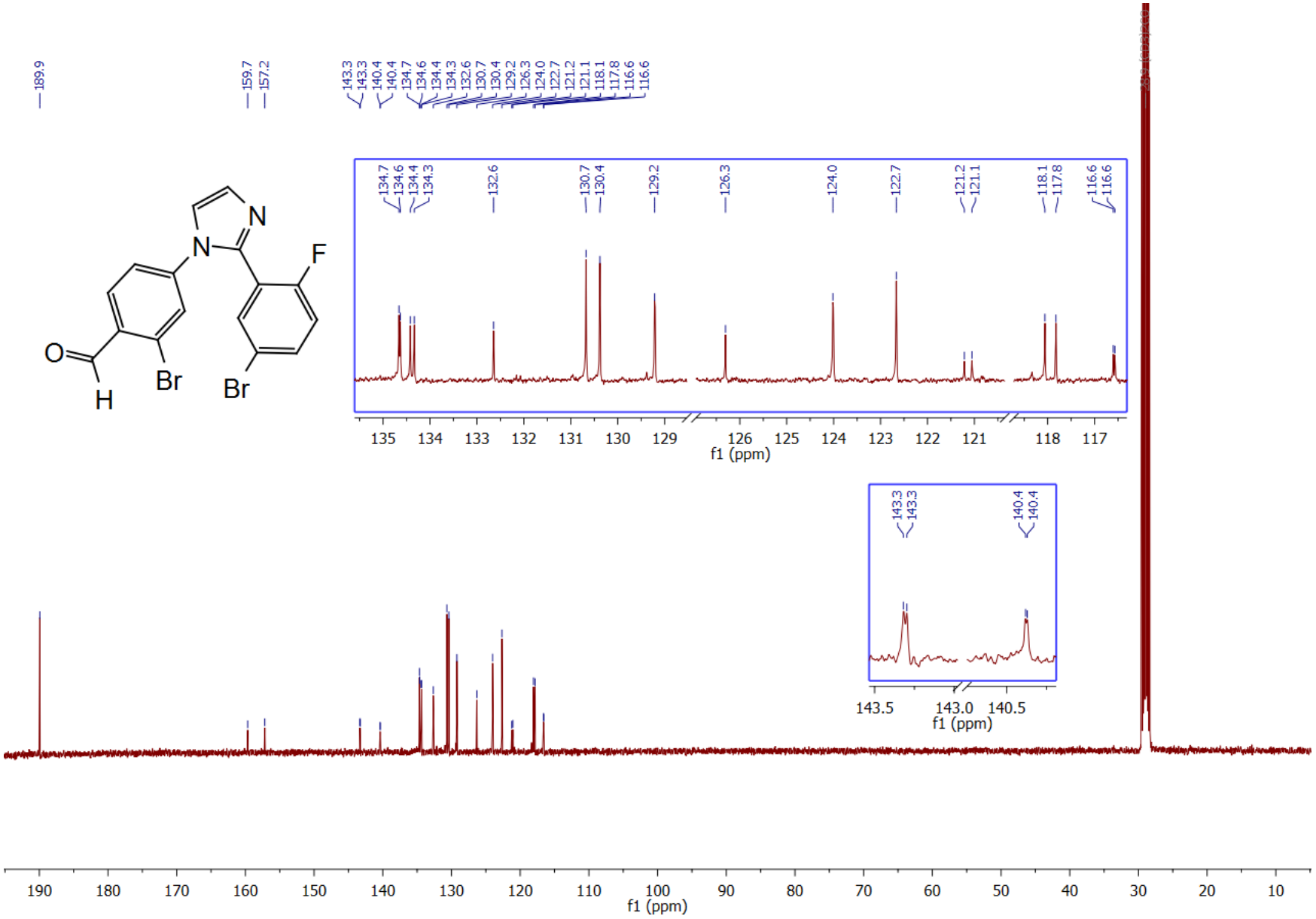
Fragment 26

PC2048C_FC.1.fid

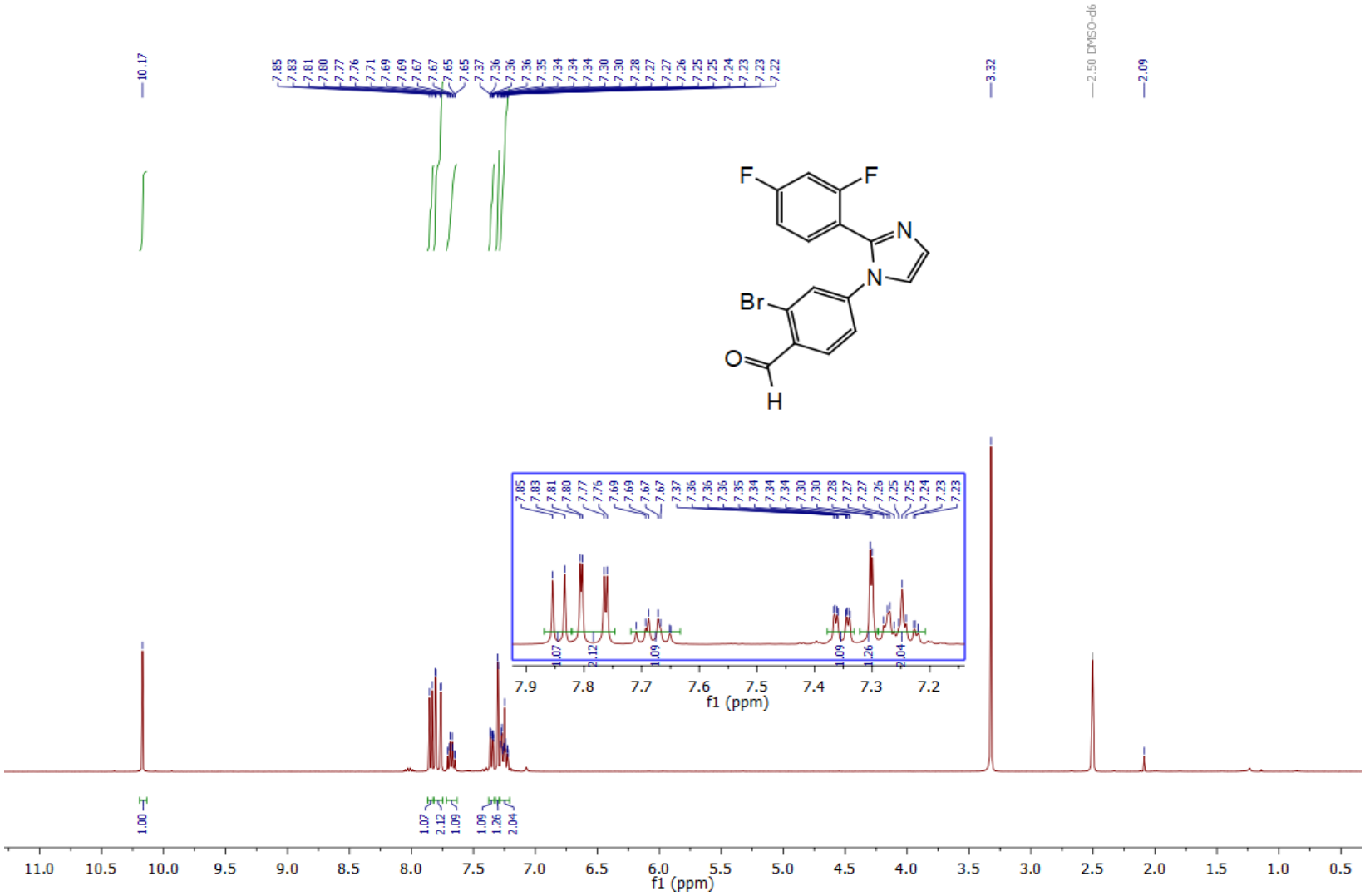


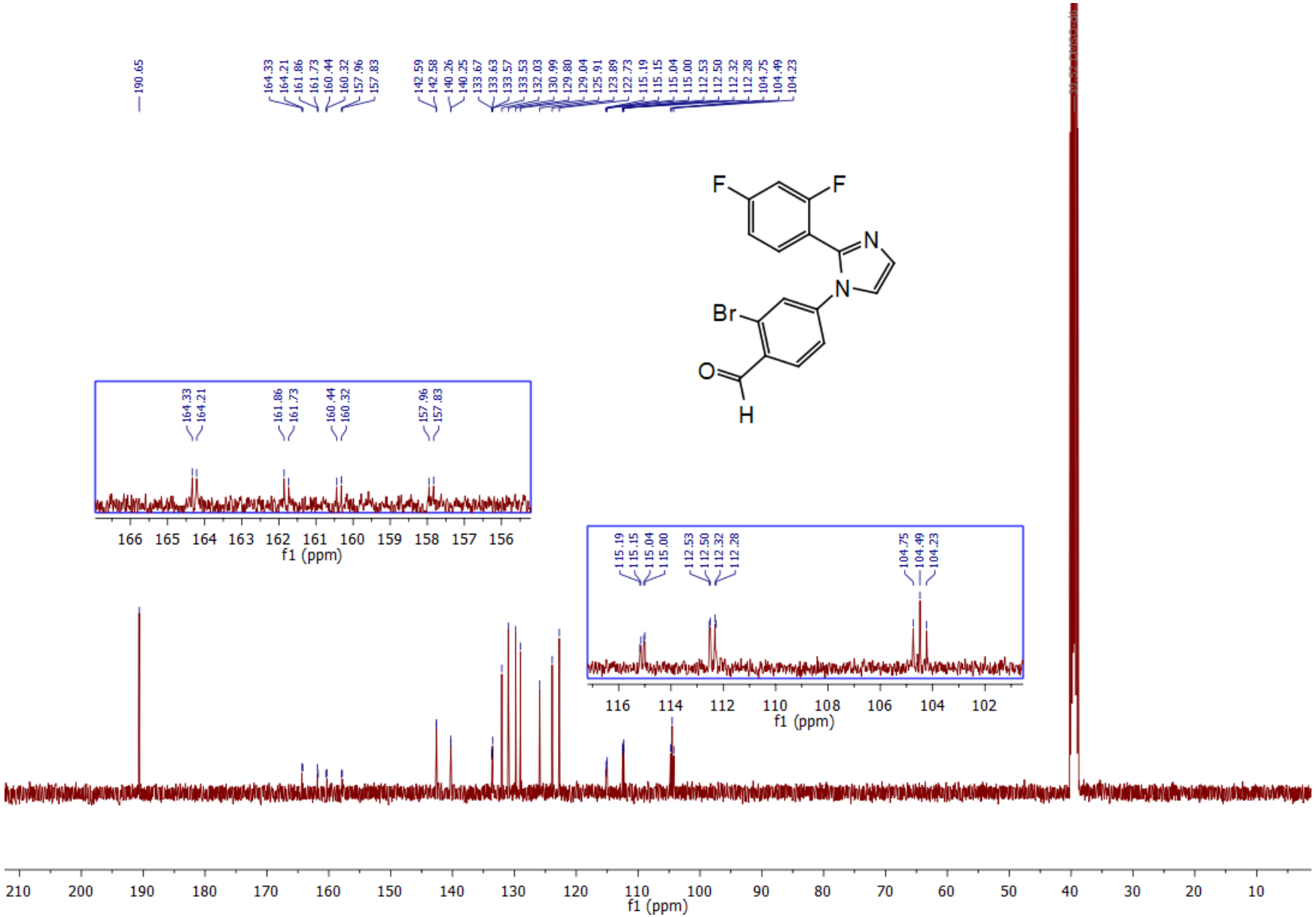
Fragment 27


10.30
7.92
7.90
7.88
7.87
7.86
7.85
7.79
7.69
7.68
7.67
7.67
7.66
7.66
7.65
7.48
7.46
7.31
7.10
7.08
7.05
2.05 (CD3)2CO
N
N
F
O
H
Br
Br
8.0 7.9 7.8 7.7 7.6 7.5 7.4 7.3 7.2 7.1 7.0
f1 (ppm)
0.98
1.03
1.00
1.01
2.13
1.07
1.01
1.08
11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0
f1 (ppm)


189.9
159.7
157.2
143.3
143.3
140.4
140.4
134.7
134.6
134.4
134.3
132.6
130.7
130.4
129.2
126.3
124.0
122.7
121.2
121.1
118.1
117.8
116.6
116.6
O
H
Br
Br
N
N
F
f1 (ppm)
f1 (ppm)
f1 (ppm)

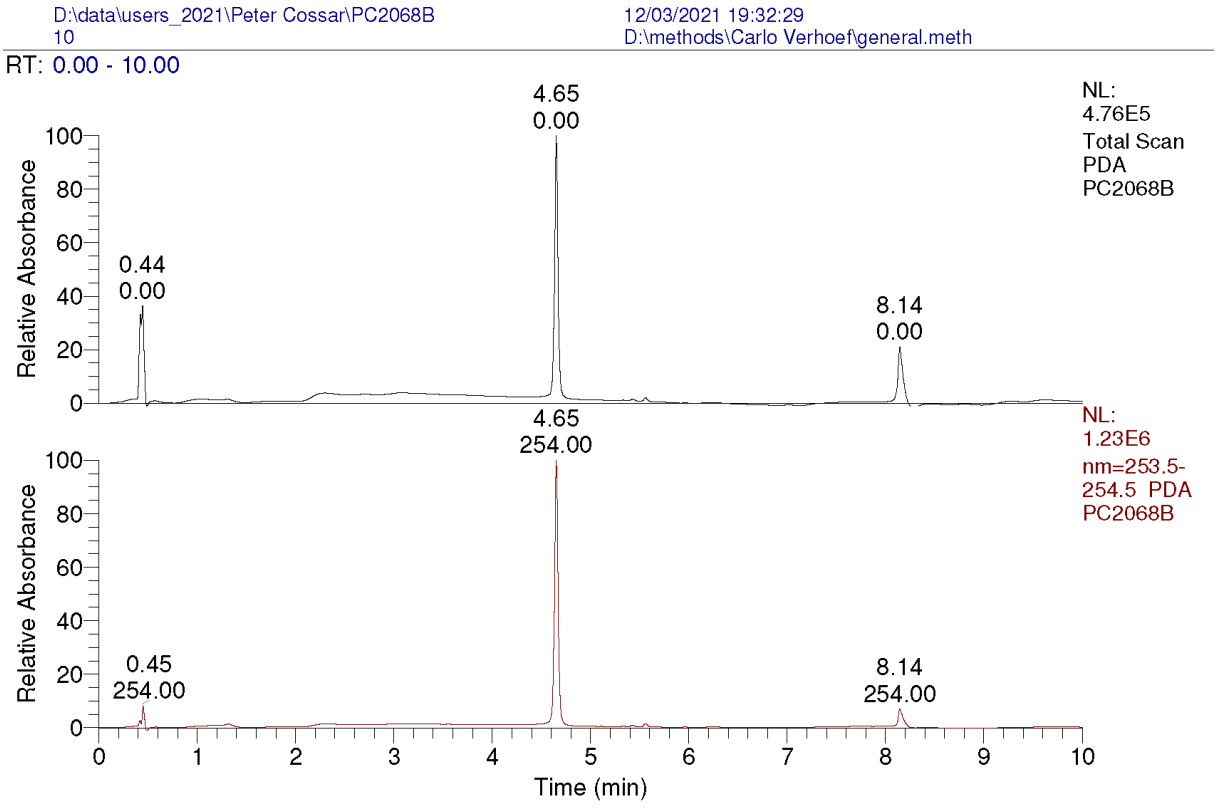
Fragment 28

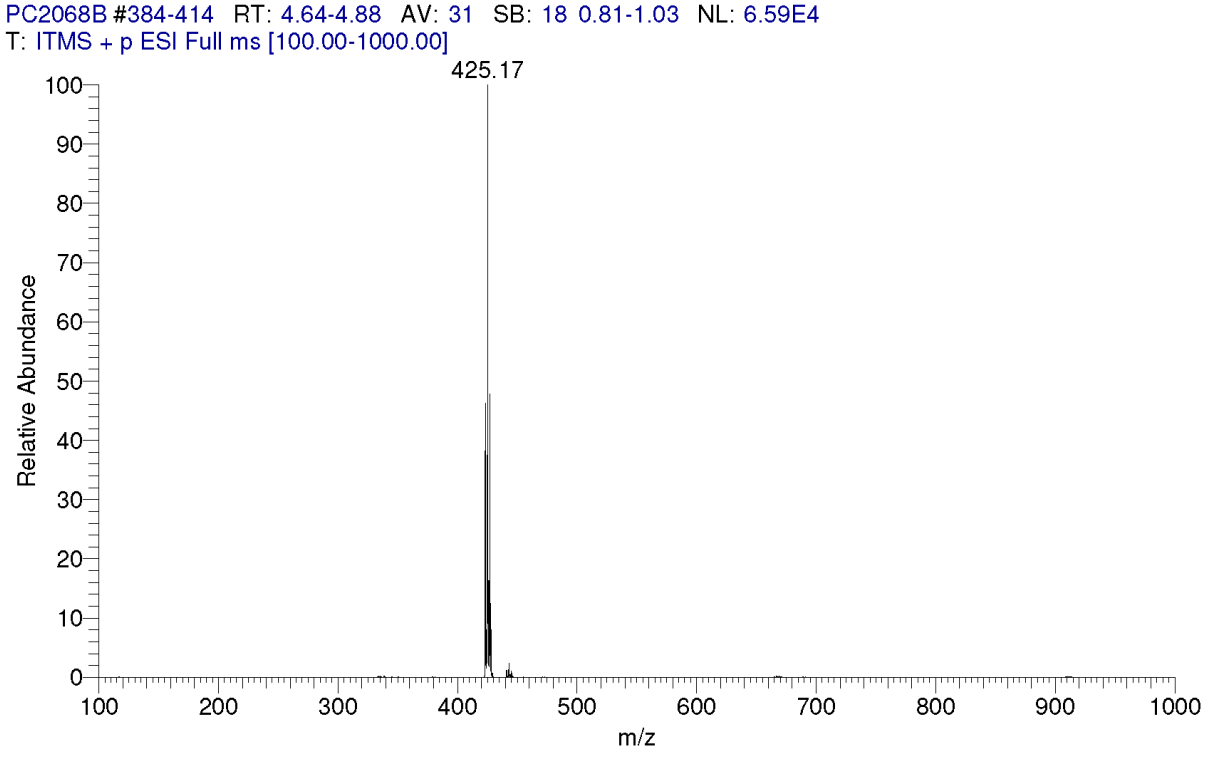

10.17
7.85
7.83
7.81
7.80
7.77
7.76
7.71
7.69
7.69
7.67
7.67
7.65
7.65
7.37
7.36
7.36
7.35
7.34
7.34
7.34
7.30
7.30
7.28
7.27
7.27
7.26
7.25
7.25
7.24
7.23
7.22
3.32
2.50 DMSO-d6
2.09
F
F
N
N
Br
O
H
1.00
1.07
2.12
1.09
1.09
1.26
2.04
f1 (ppm)


190.65
164.33
164.21
161.86
161.73
160.44
160.32
157.96
157.83
142.59
142.58
140.26
140.25
133.67
133.63
133.57
133.53
132.03
130.99
129.80
129.04
125.91
123.89
122.73
115.19
115.15
115.04
115.00
112.53
112.50
112.32
112.28
104.75
104.49
104.23
F
F
N
N
Br
O
H
f1 (ppm)

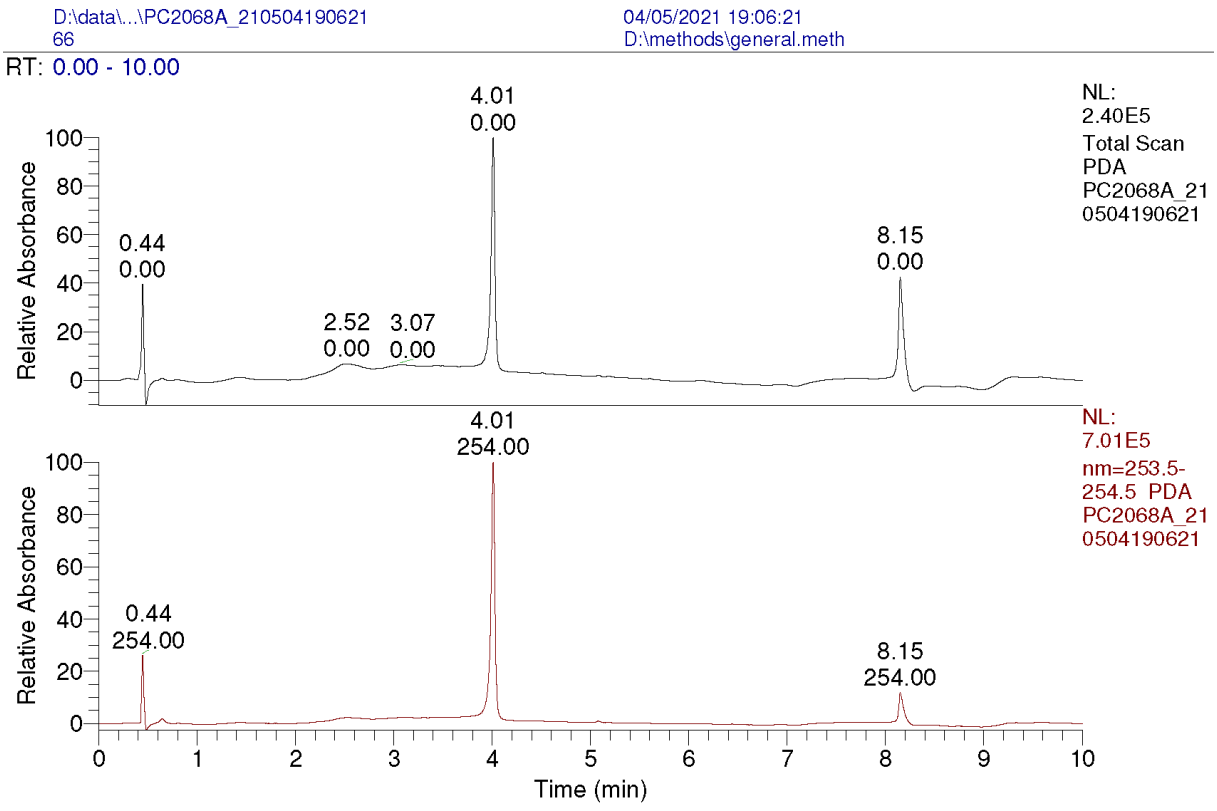
HPLC

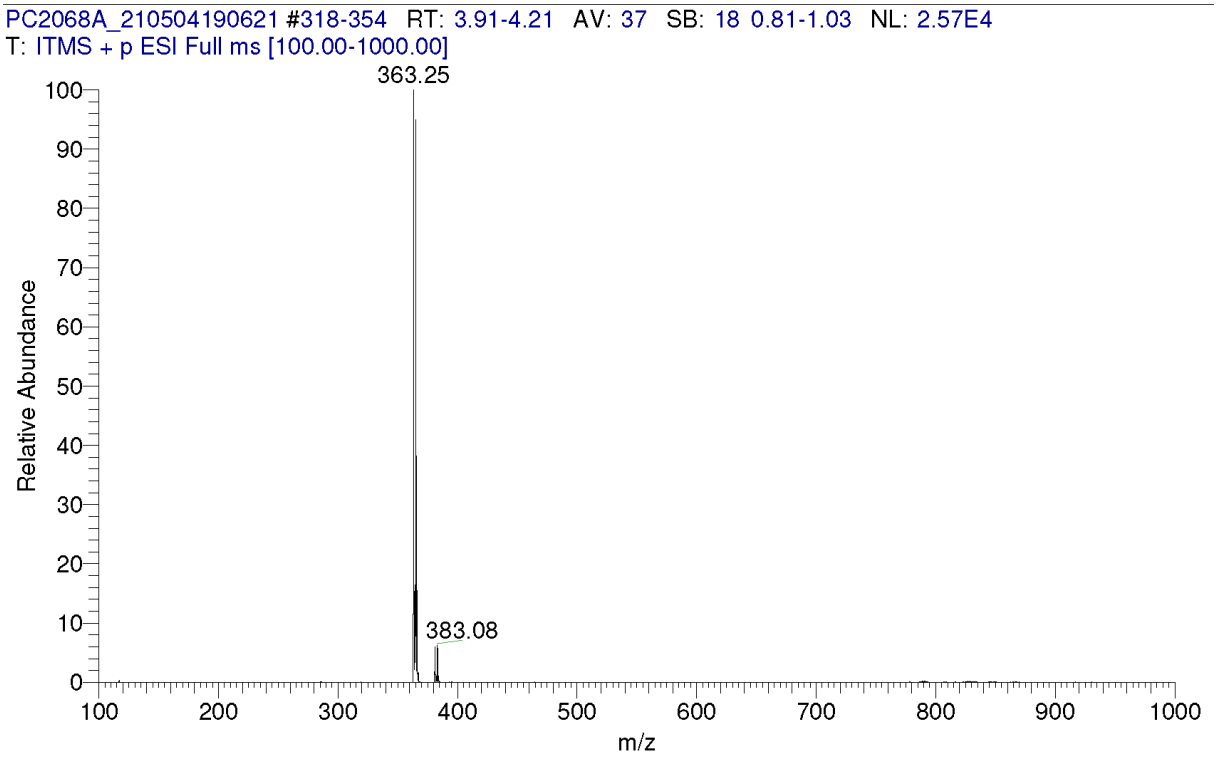
Fragment 27





Fragment 28


D:\data\...\PC2068A_210504190621
66
04/05/2021 19:06:21
D:\methods\general.meth
RT: 0.00 - 10.00
NL:
2.40E5
Total Scan
PDA
PC2068A_21
0504190621
4.01
0.00
0.44
0.00
2.52
0.00
3.07
0.00
8.15
0.00
Relative Absorbance
NL:
7.01E5
nm=253.5-
254.5 PDA
PC2068A_21
0504190621
4.01
254.00
0.44
254.00
8.15
254.00
Time (min)


PC2068A_210504190621 #318-354 RT: 3.91-4.21 AV: 37 SB: 18 0.81-1.03 NL: 2.57E4
T: ITMS + p ESI Full ms [100.00-1000.00]
363.25
383.08
Relative Abundance
m/z

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