Supporting Information

For

Exploring 2-mercapto-*N*-arylacetamide analogs as promising anti-melanogenic agents: In vitro and in vivo evaluation

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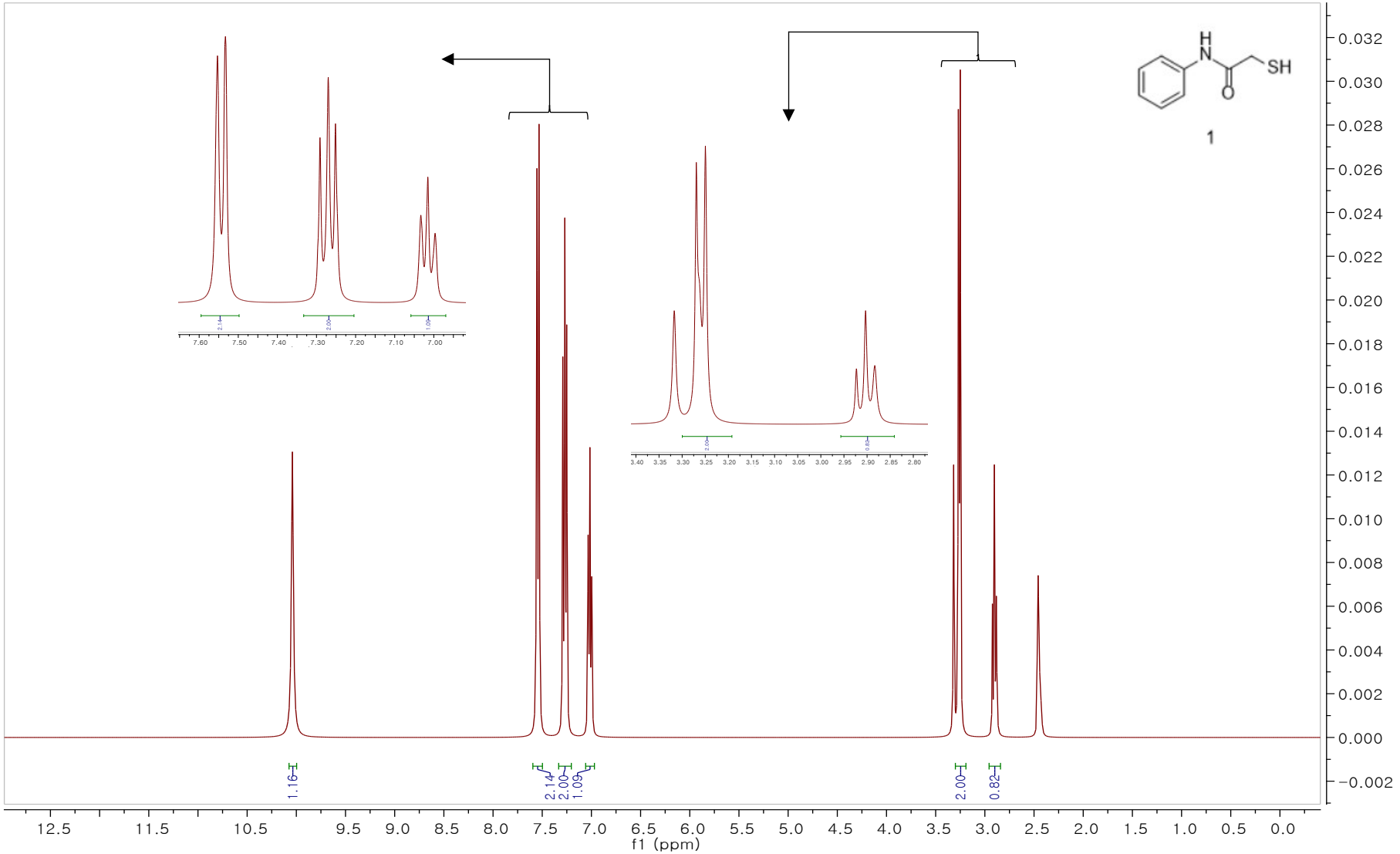
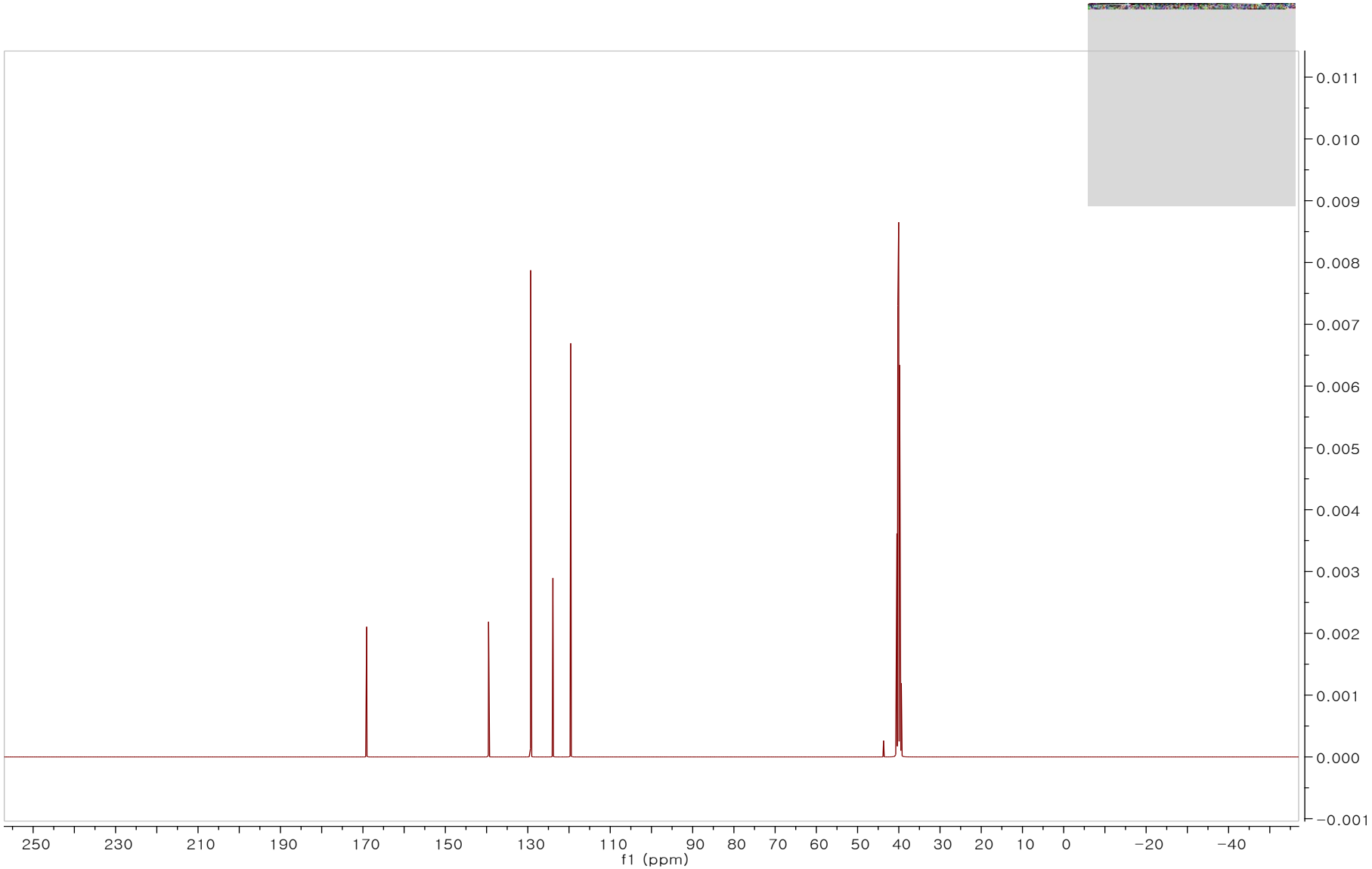


S1. ^{1}H NMR spectrum of analog **1**



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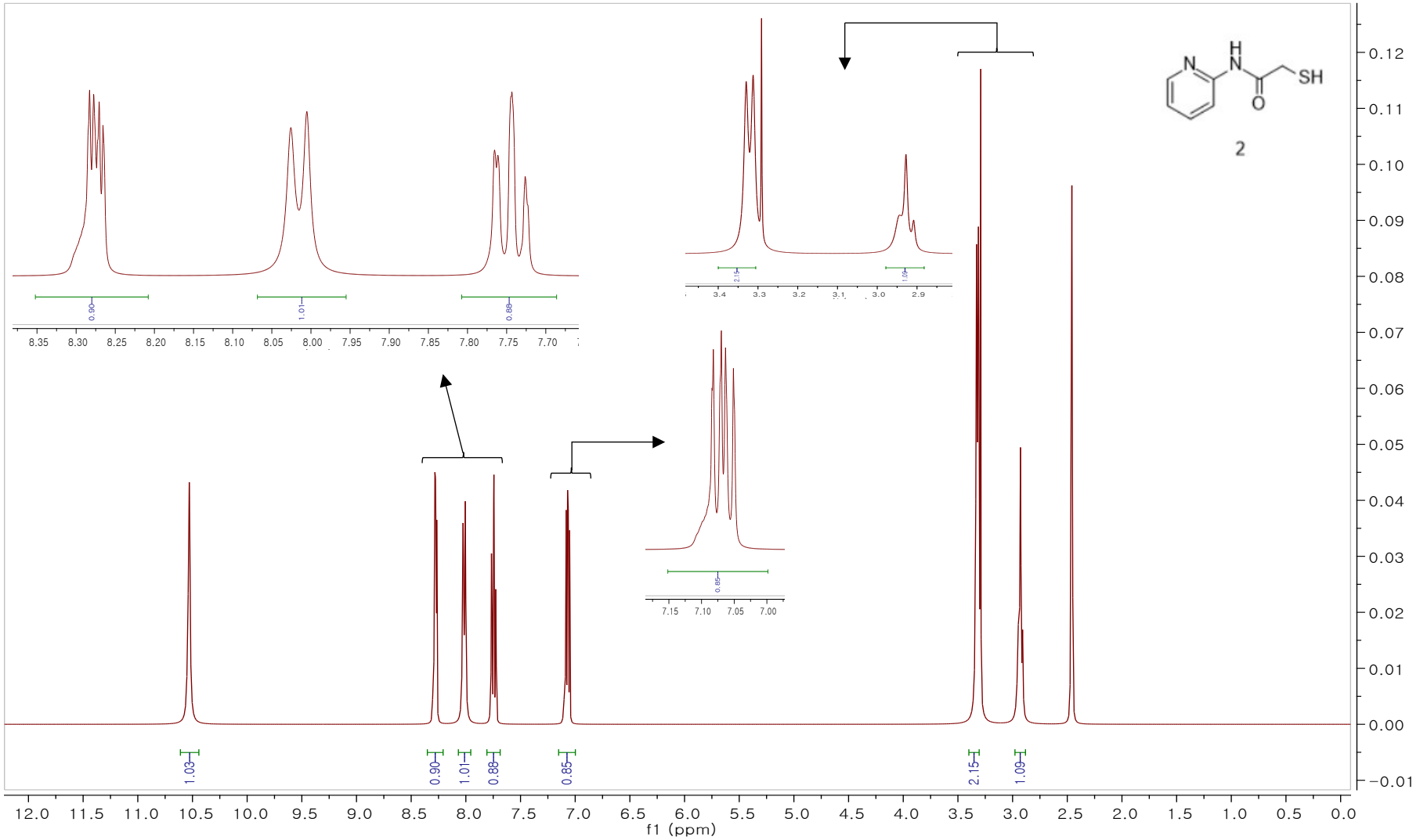
SH
2
1.03
0.90
1.01
0.88
0.85
2.15
1.09
f1 (ppm)

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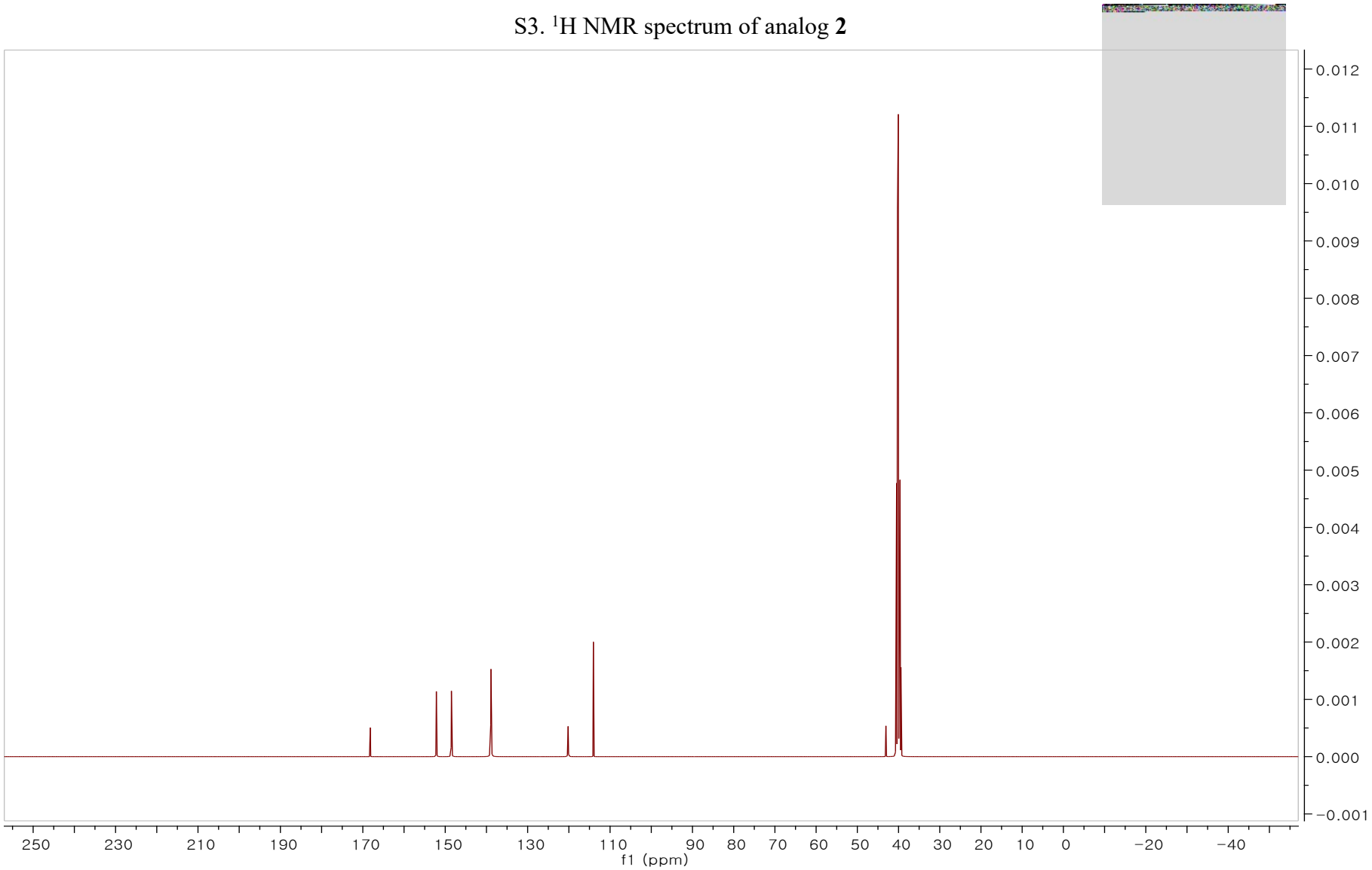

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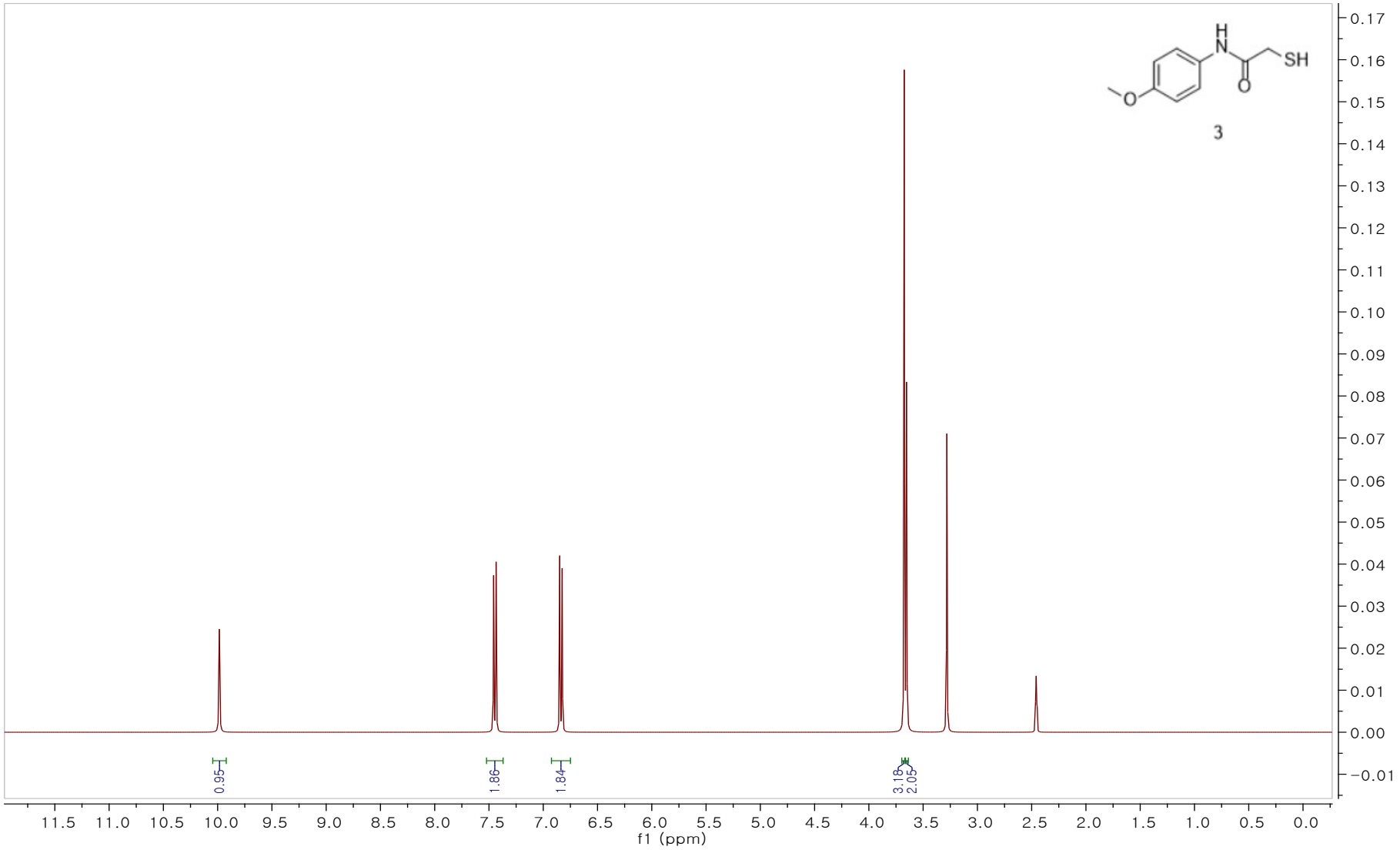
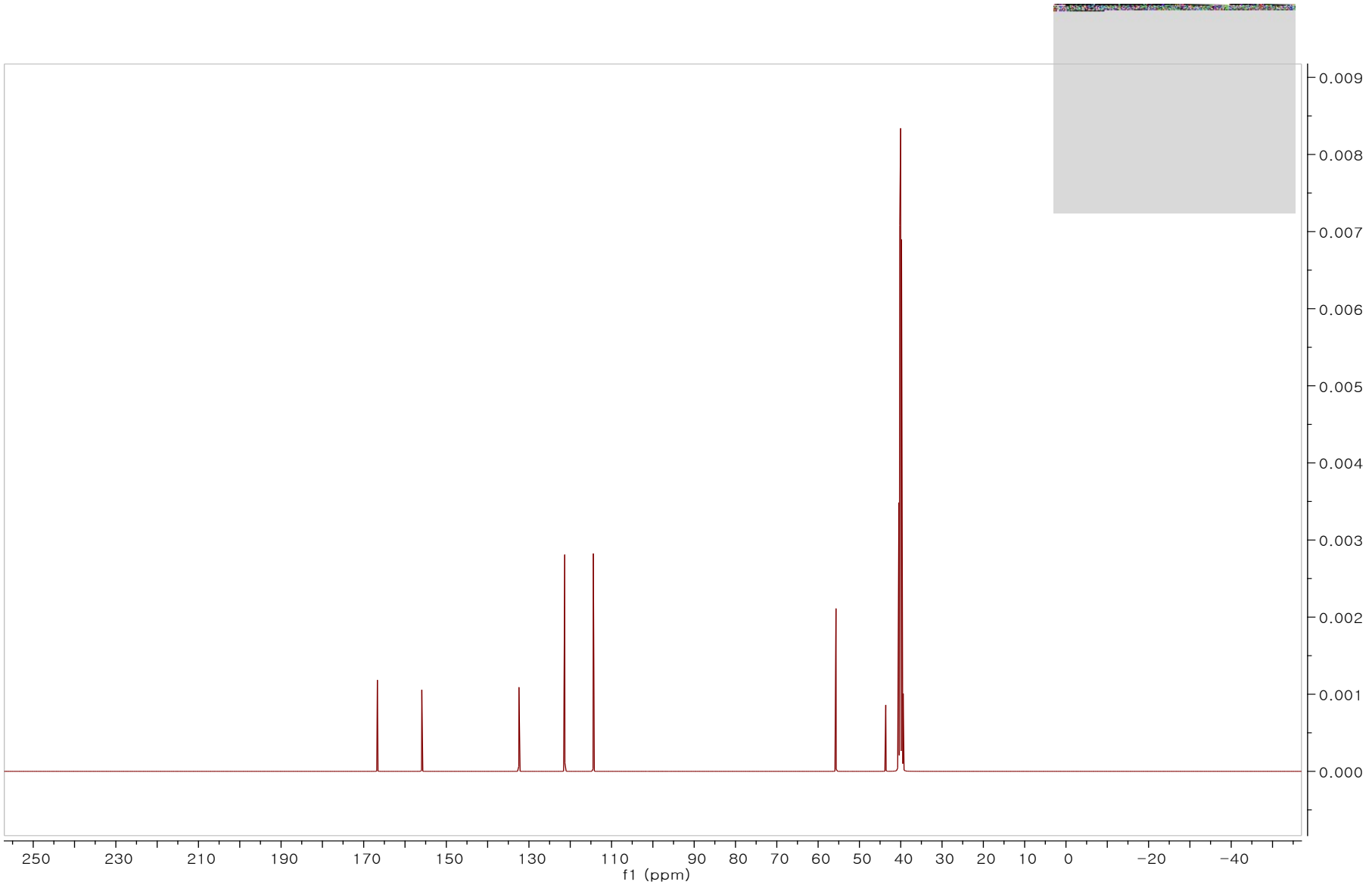
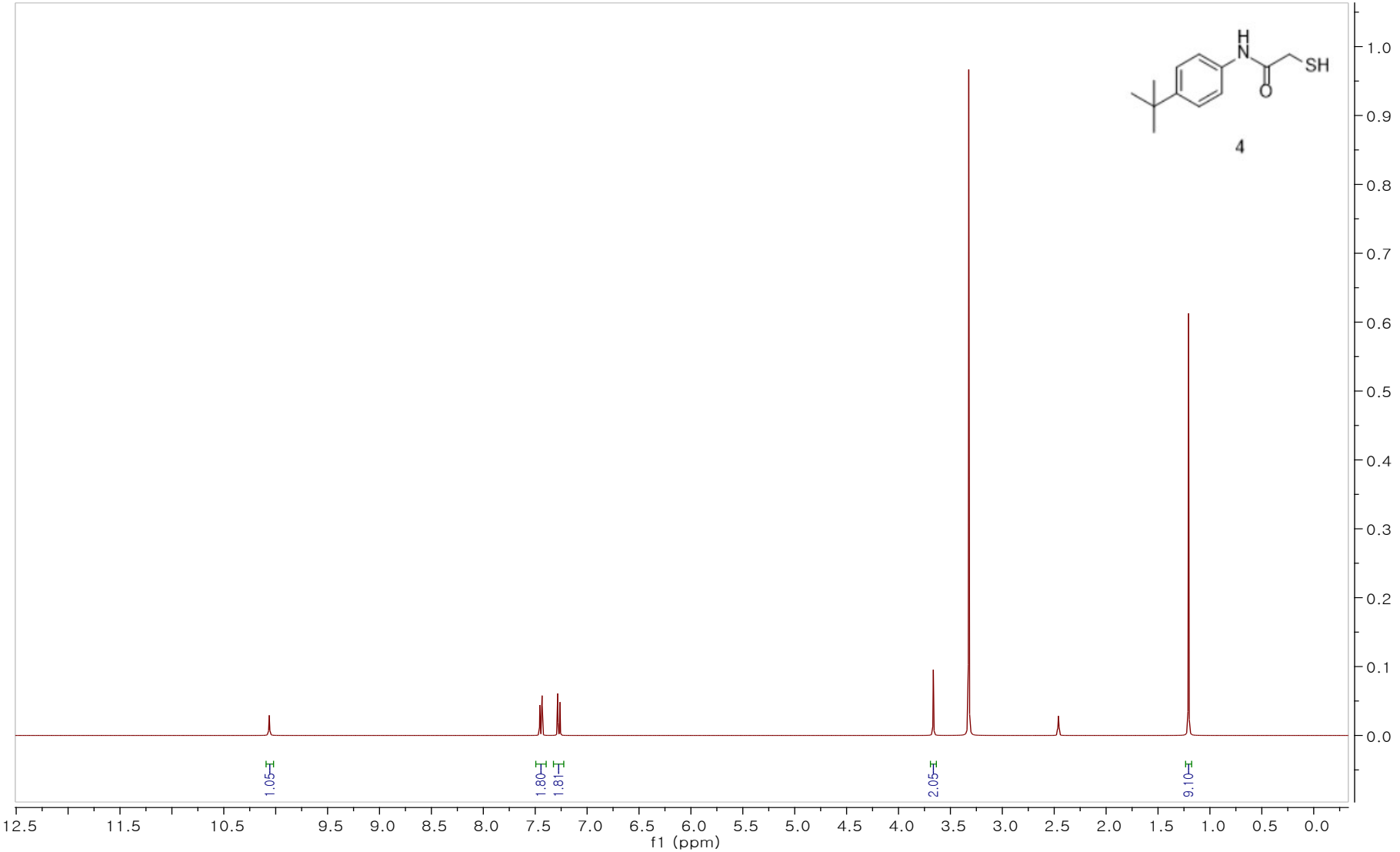


S5. ^{1}H NMR spectrum of analog **3**



S6. ^{13}C NMR spectrum of analog **3**



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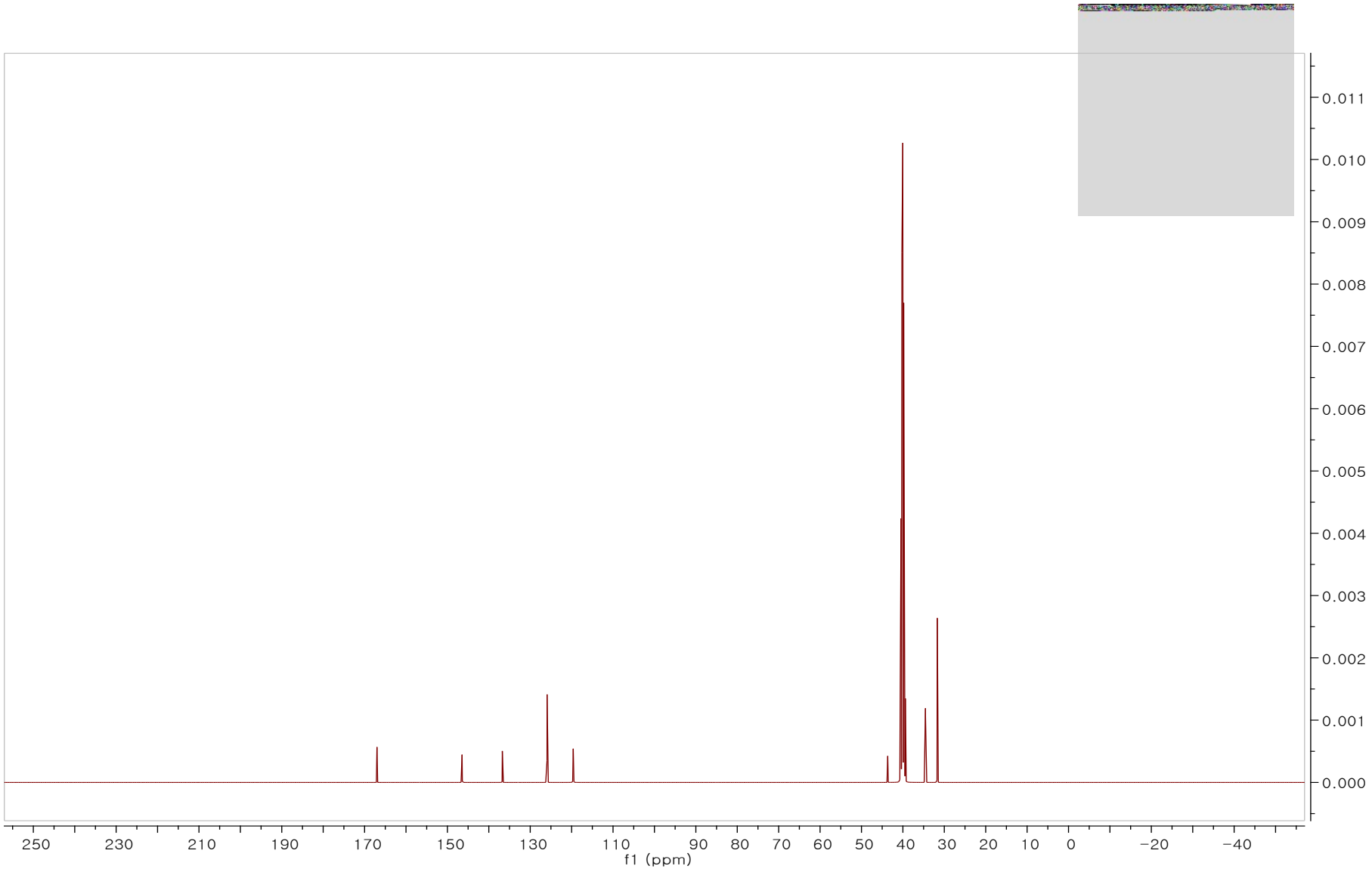

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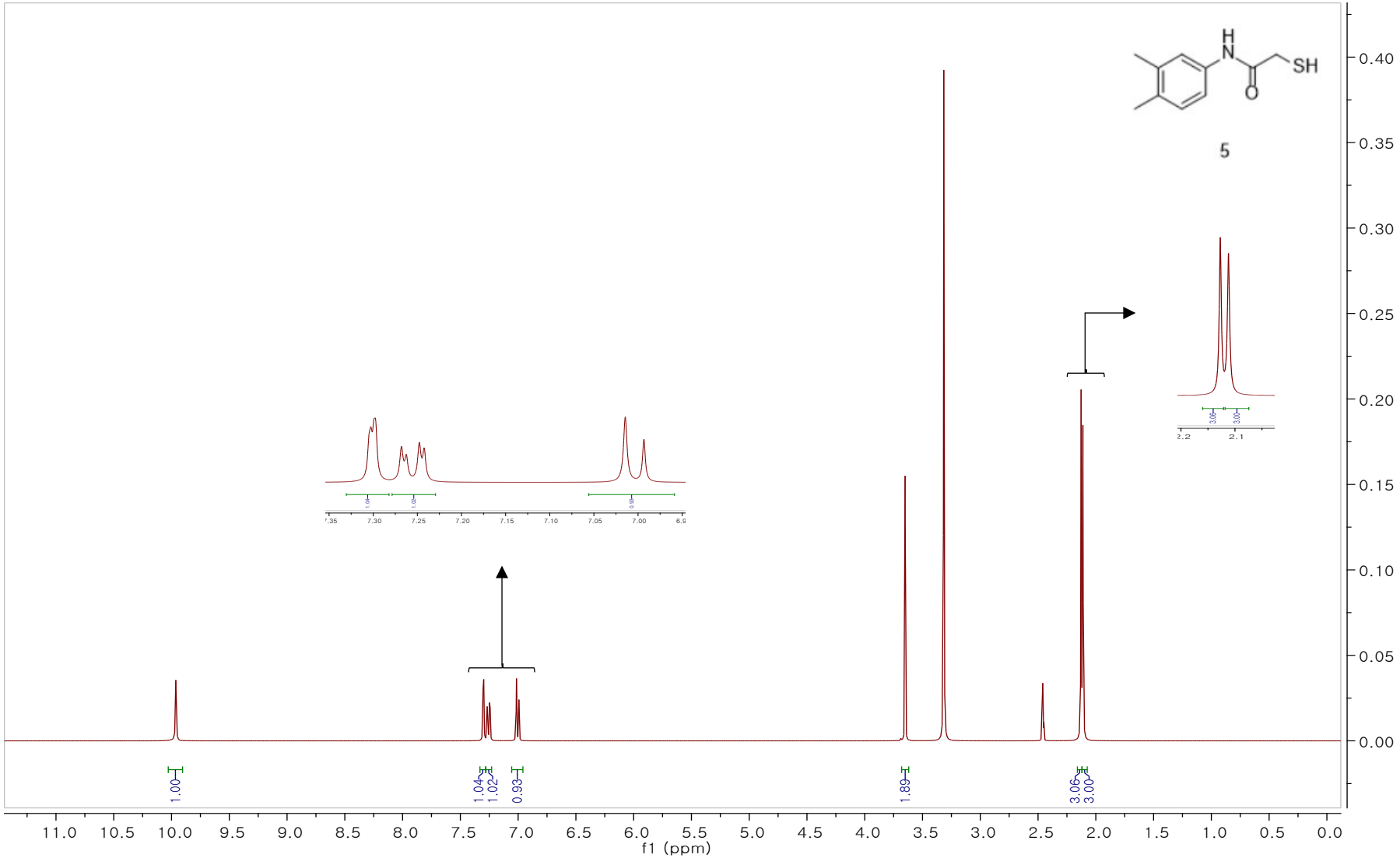

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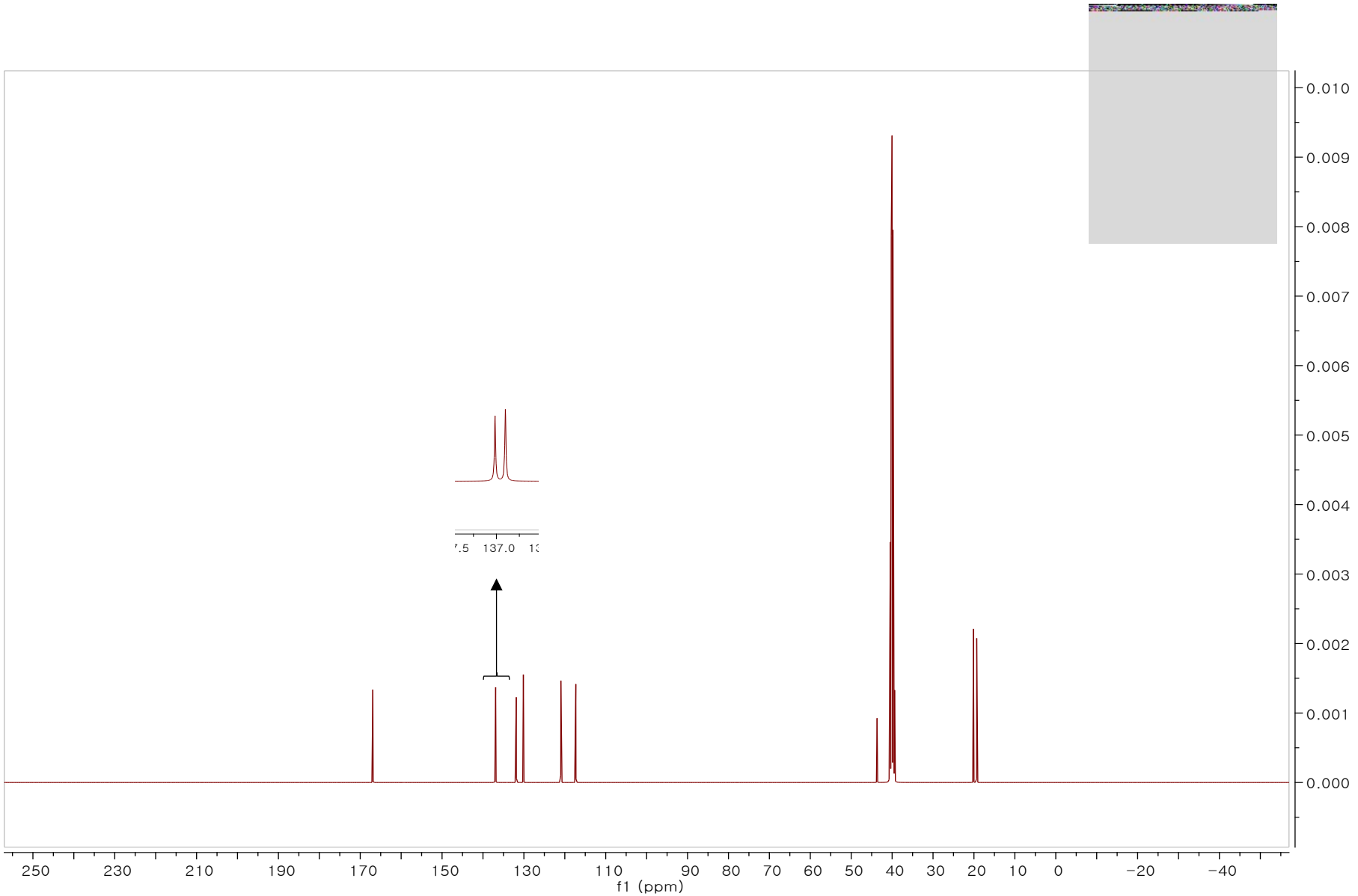

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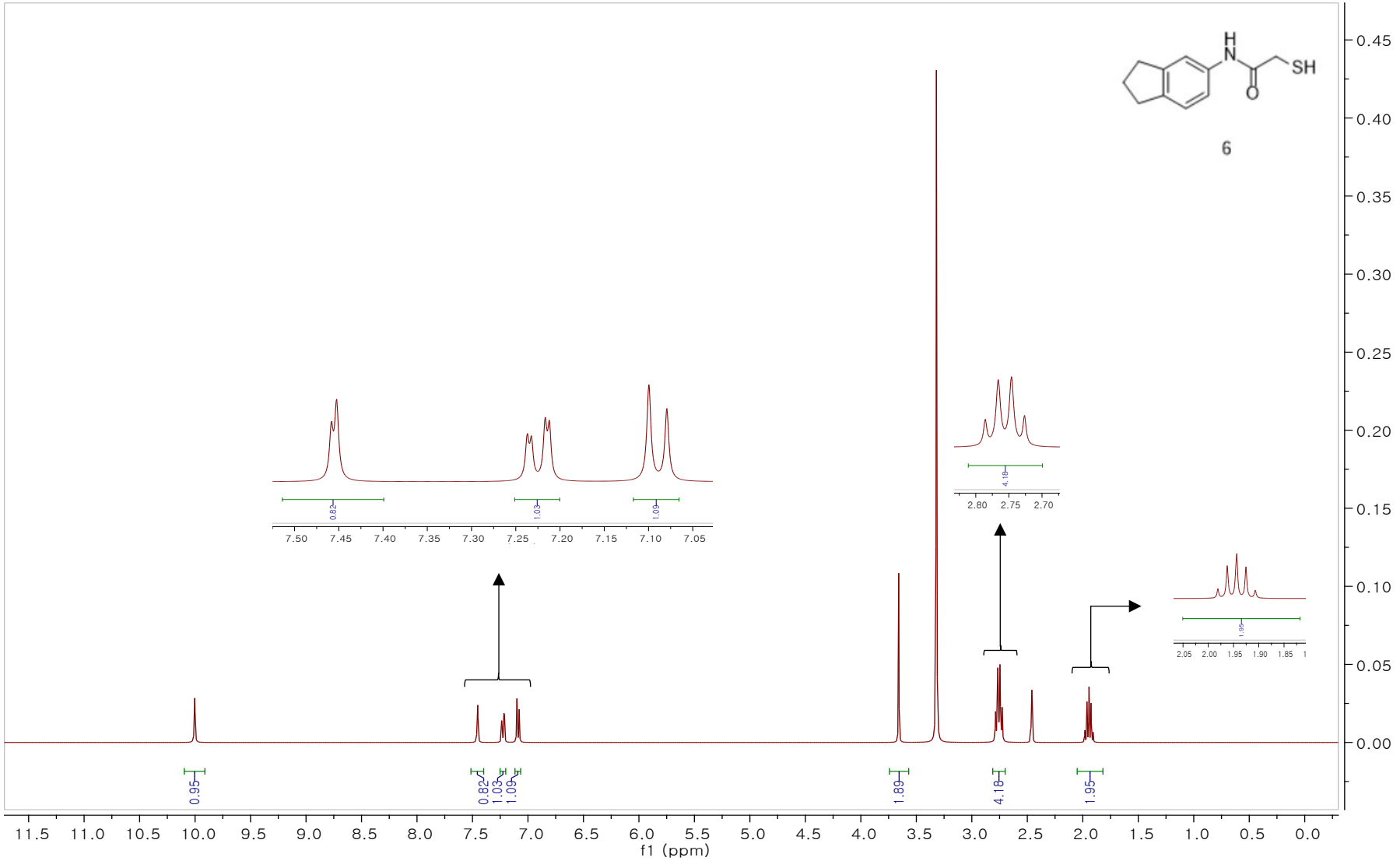

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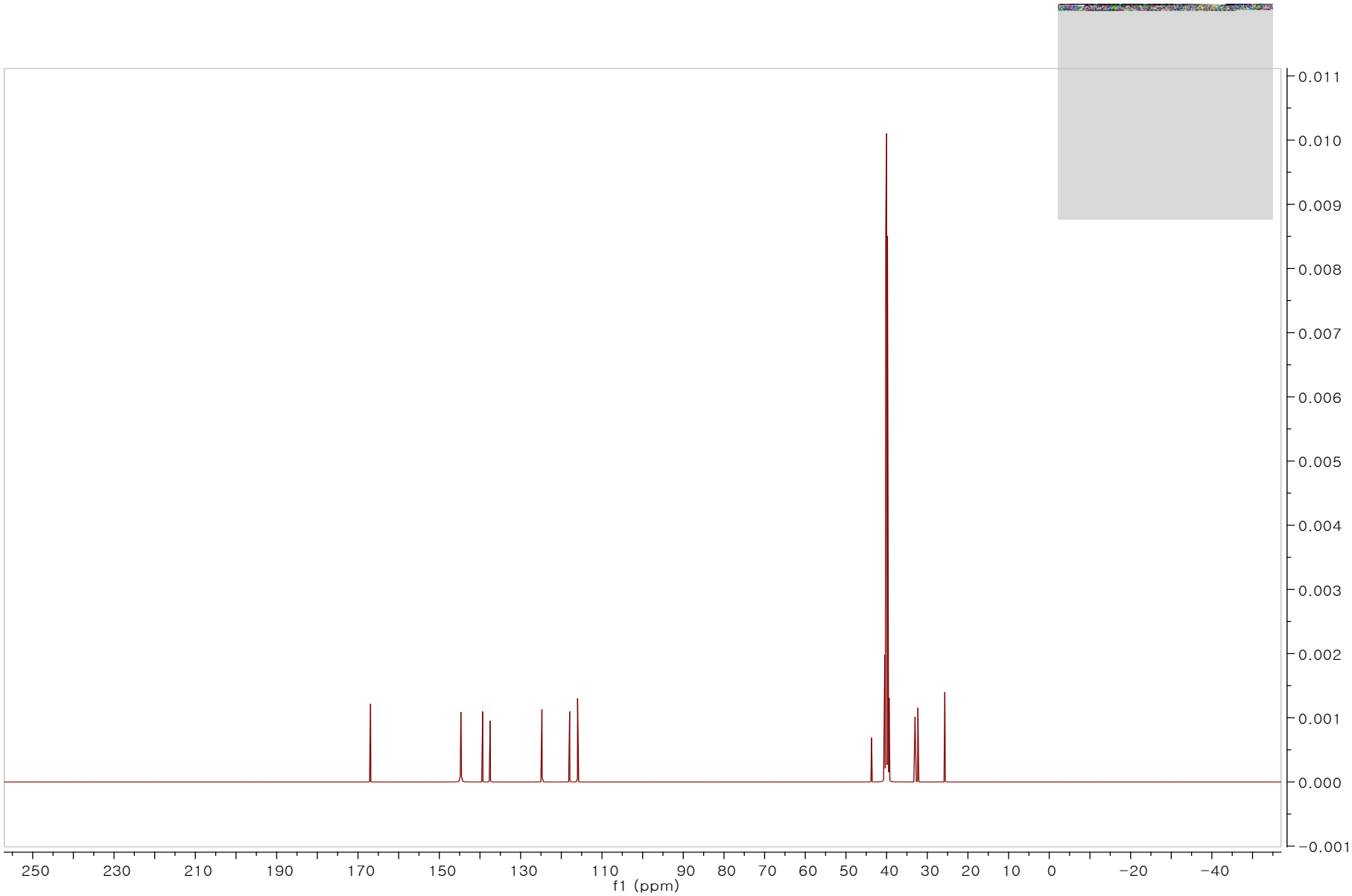

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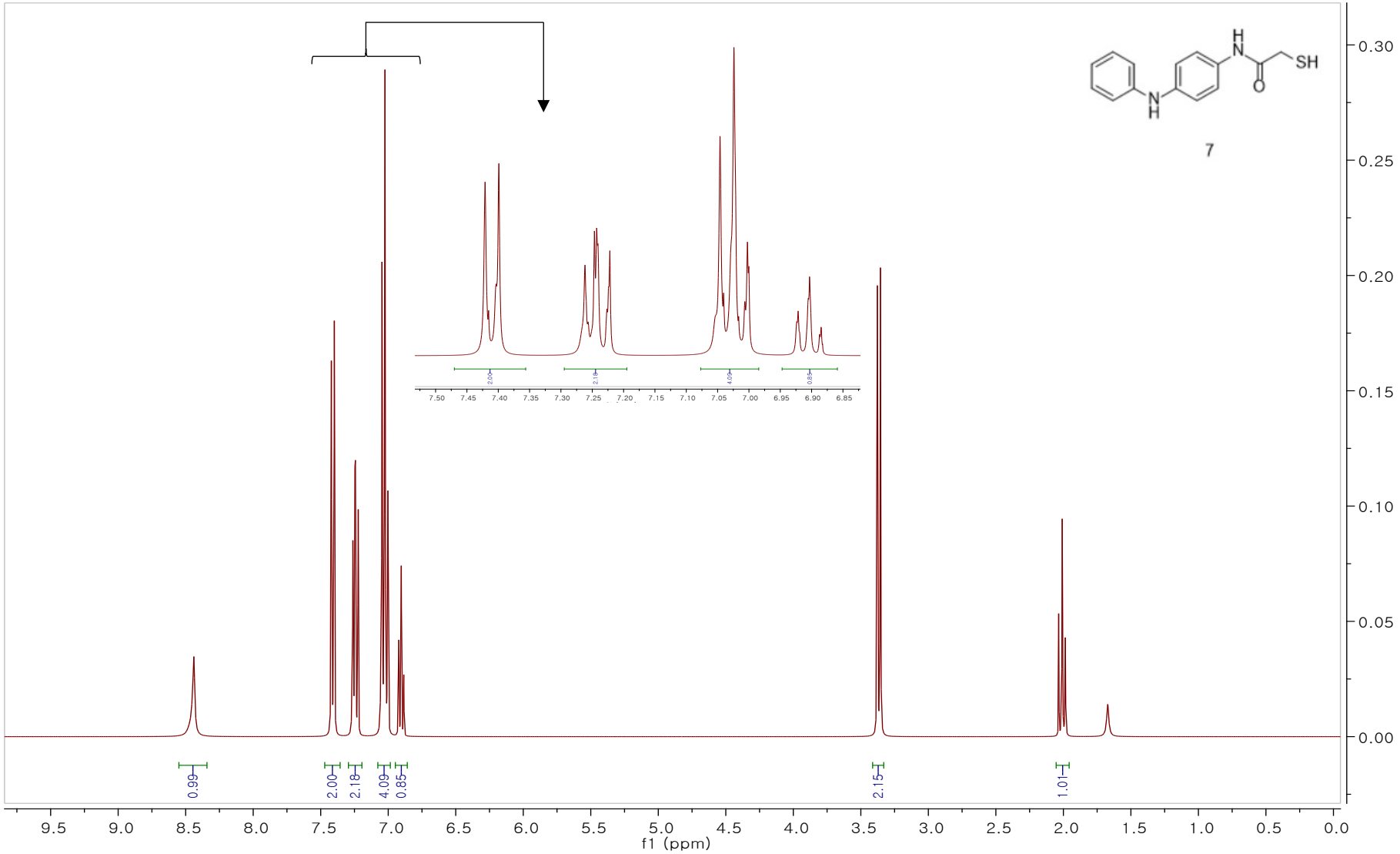

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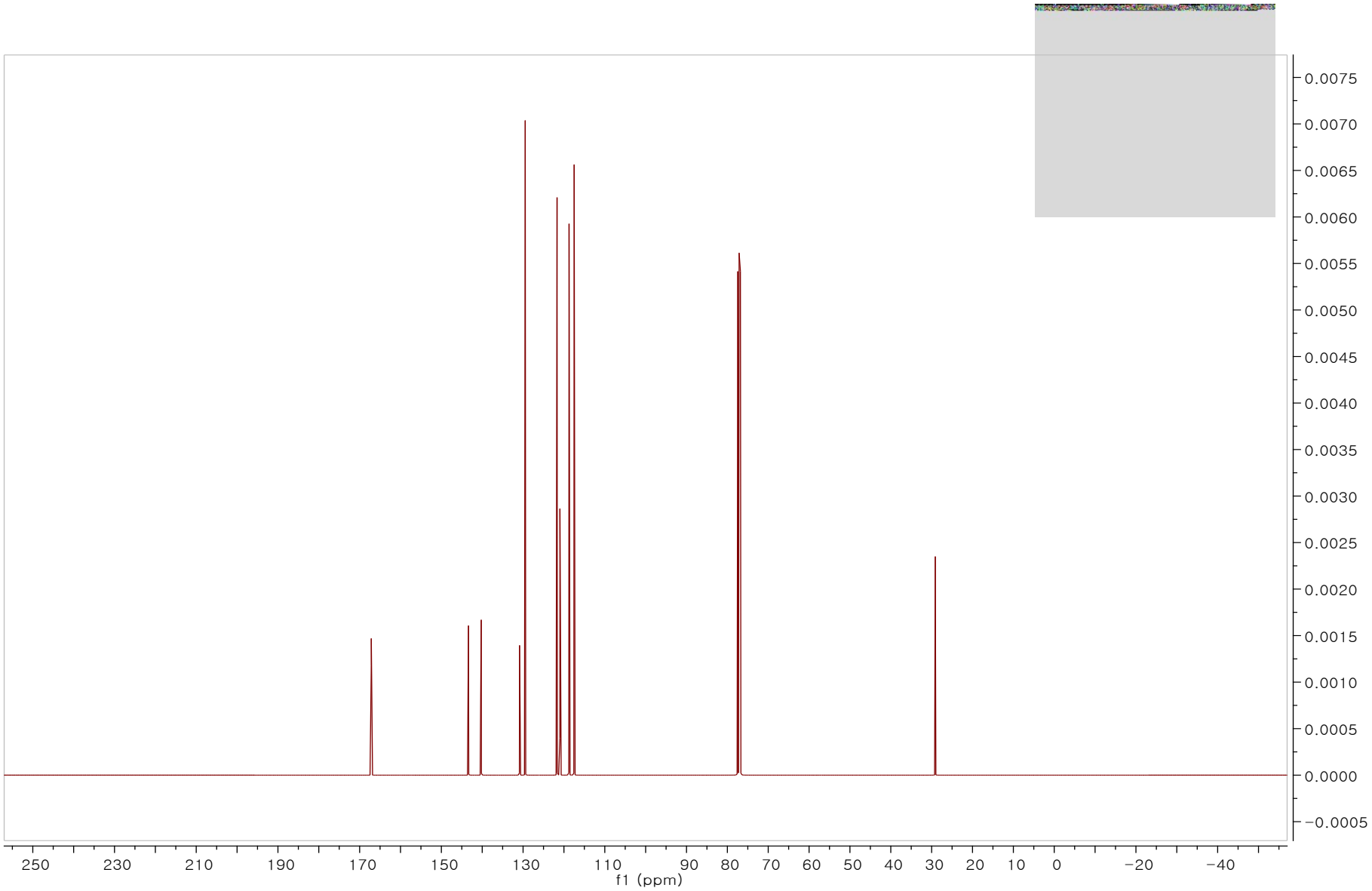
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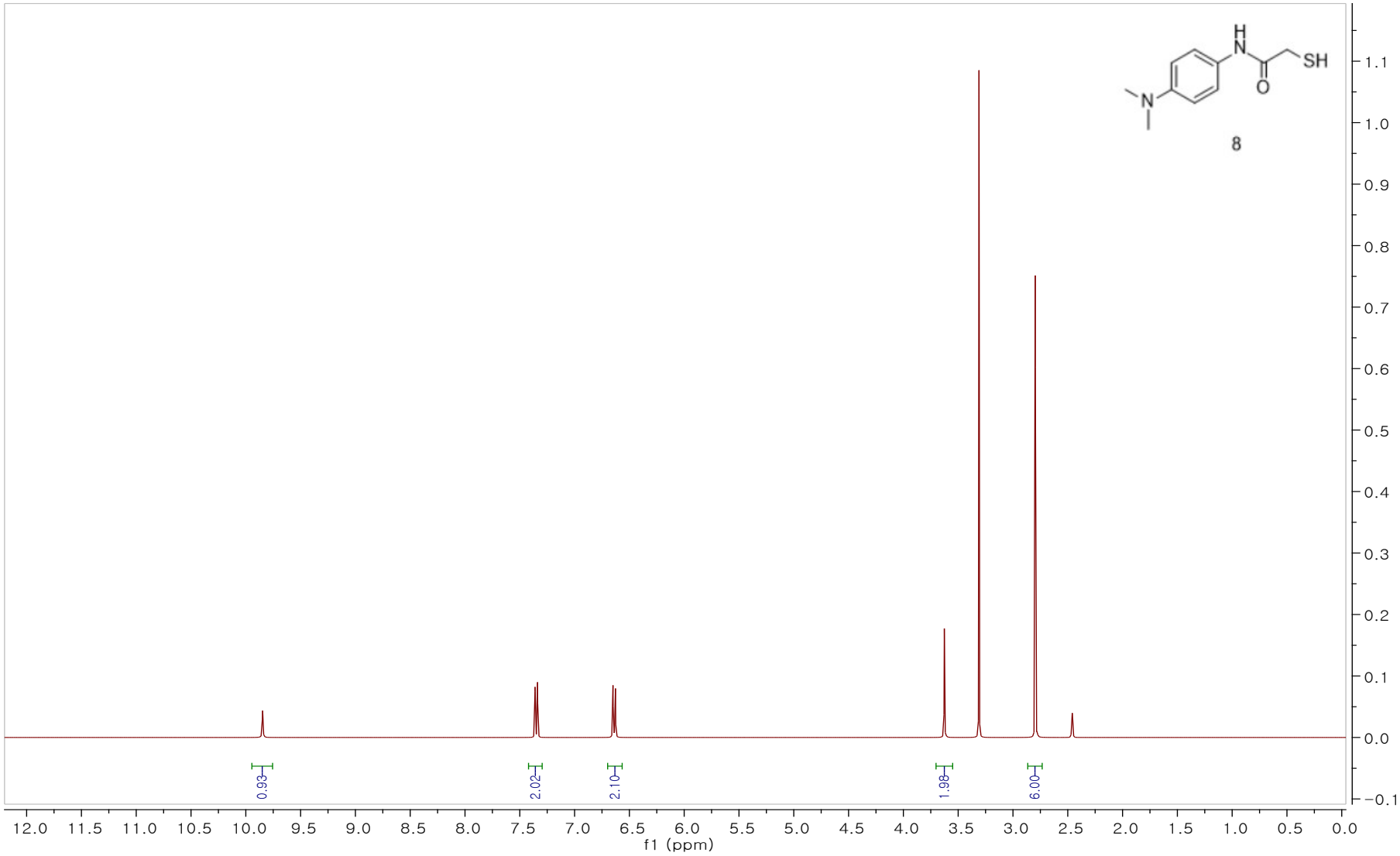
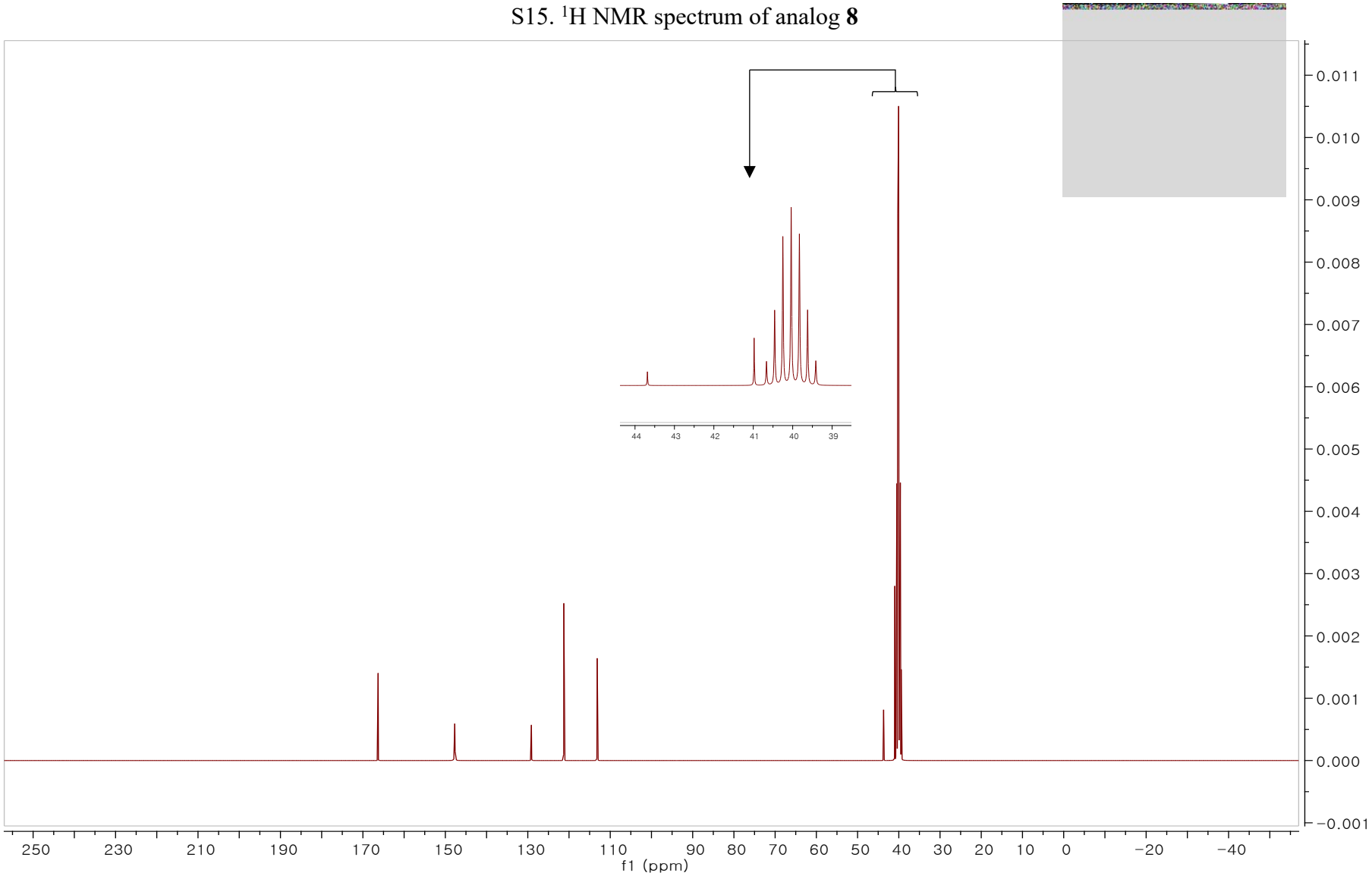
0.0070
0.0065
0.0060
0.0055
0.0050
0.0045
0.0040
0.0035
0.0030
0.0025
0.0020
0.0015
0.0010
0.0005
0.0000
−0.0005
250
230
210
190
170
150
130
110
90
80
70
60
50
40
30
20
10
0
−20
−40
f1 (ppm)

S14. ^{13}C NMR spectrum of analog **7**



S15. ^{1}H NMR spectrum of analog **8**



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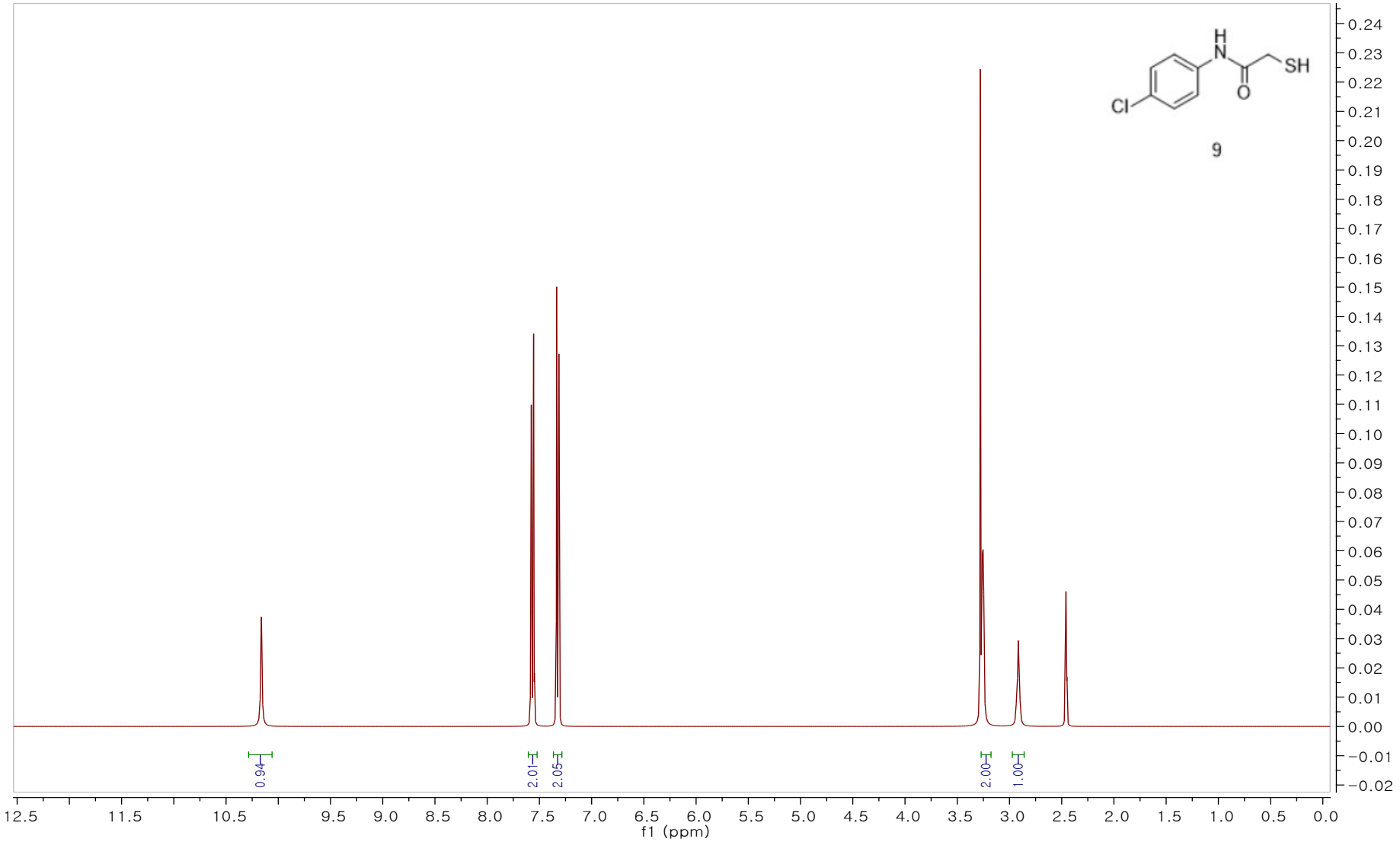

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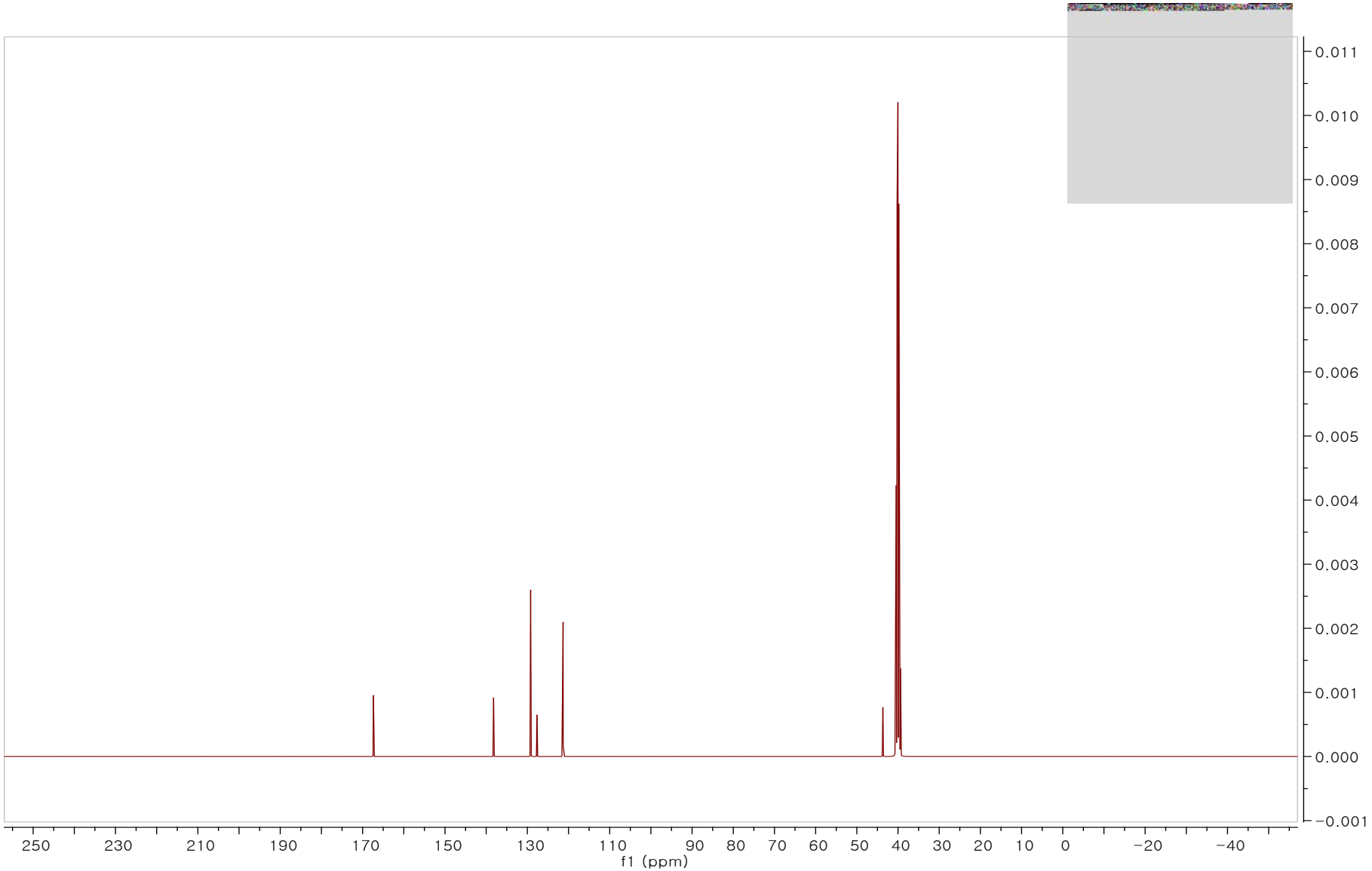

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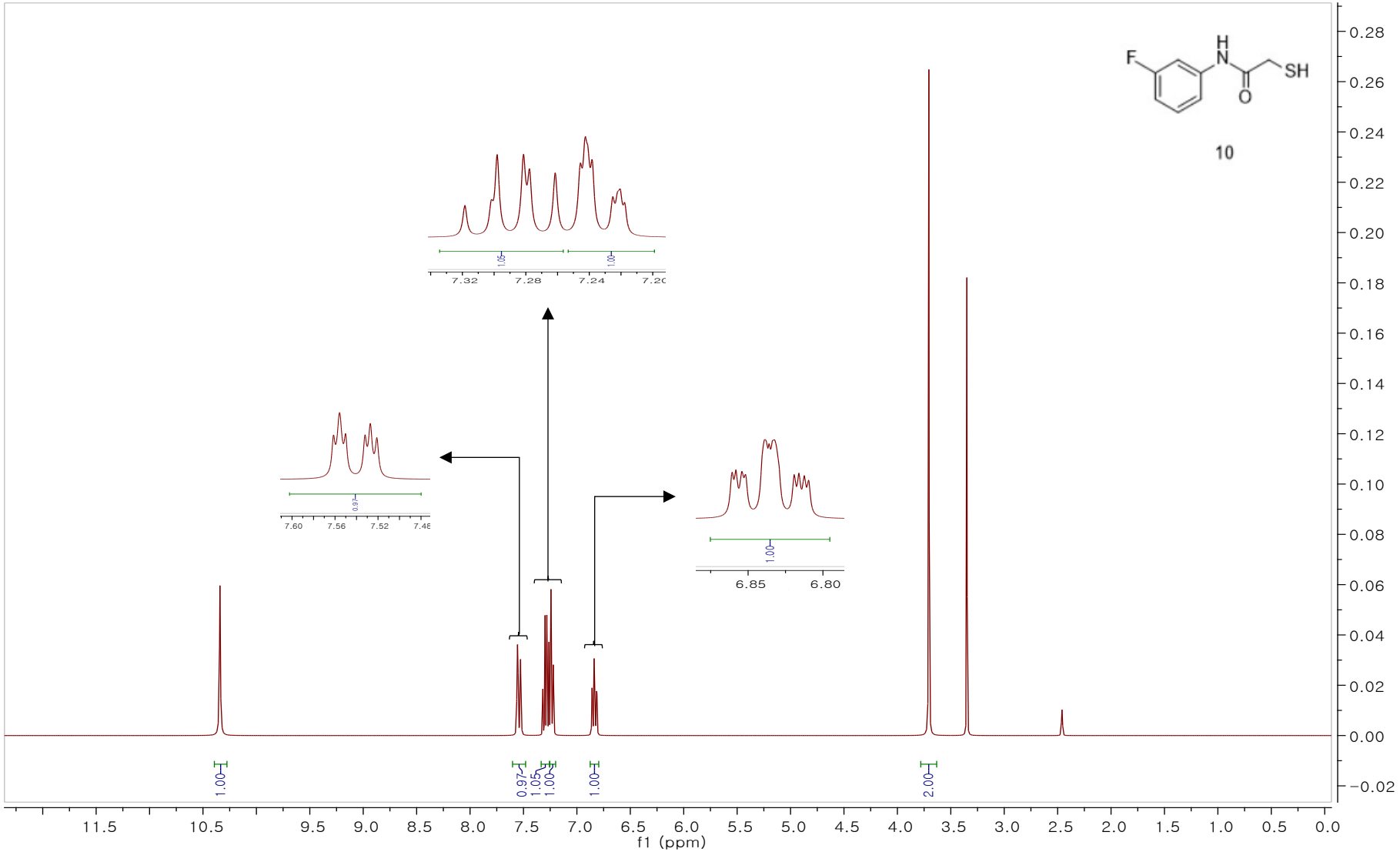
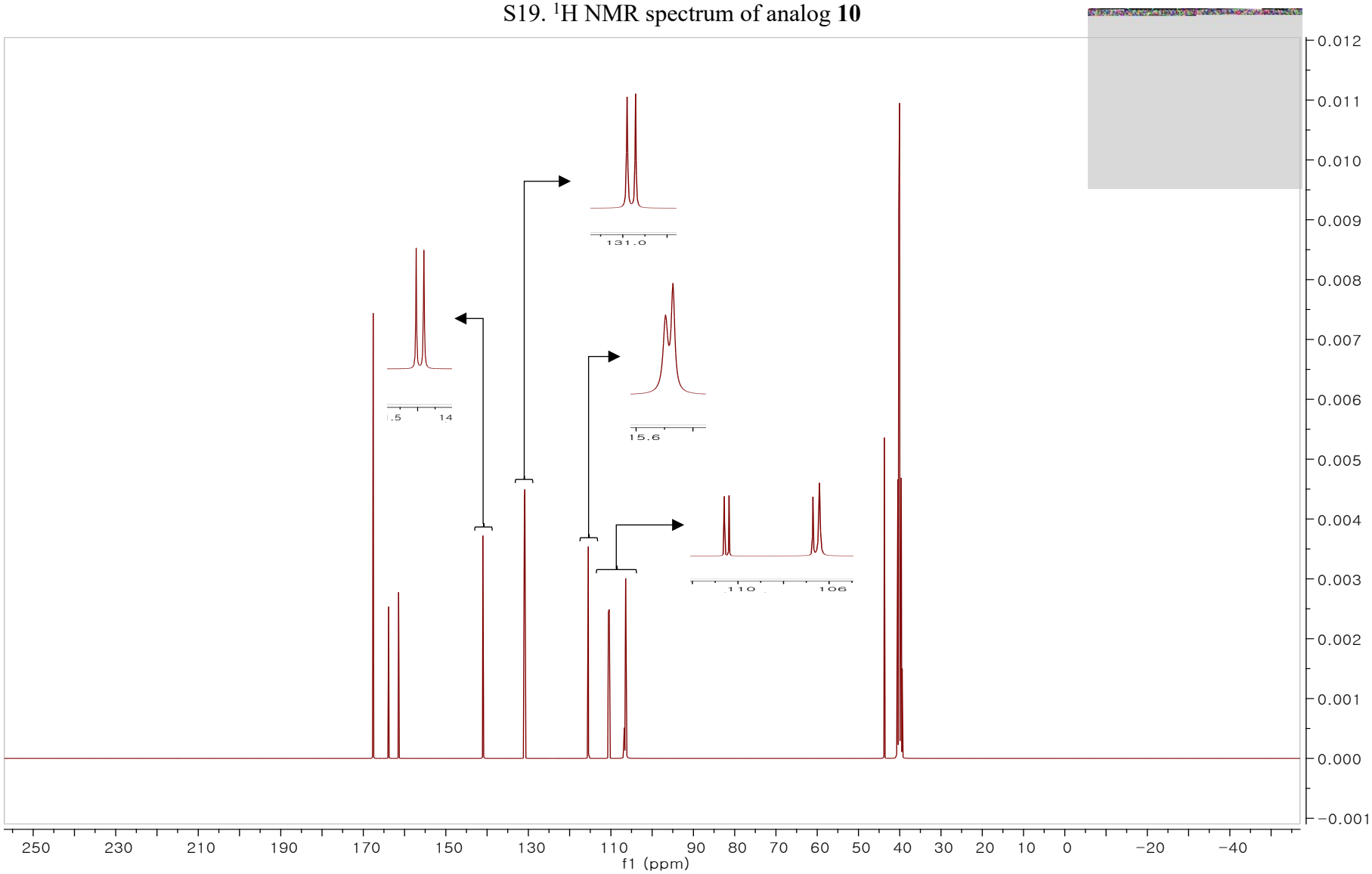

F
H
N
O
SH
10
f1 (ppm)

S19. ^{1}H NMR spectrum of analog **10**



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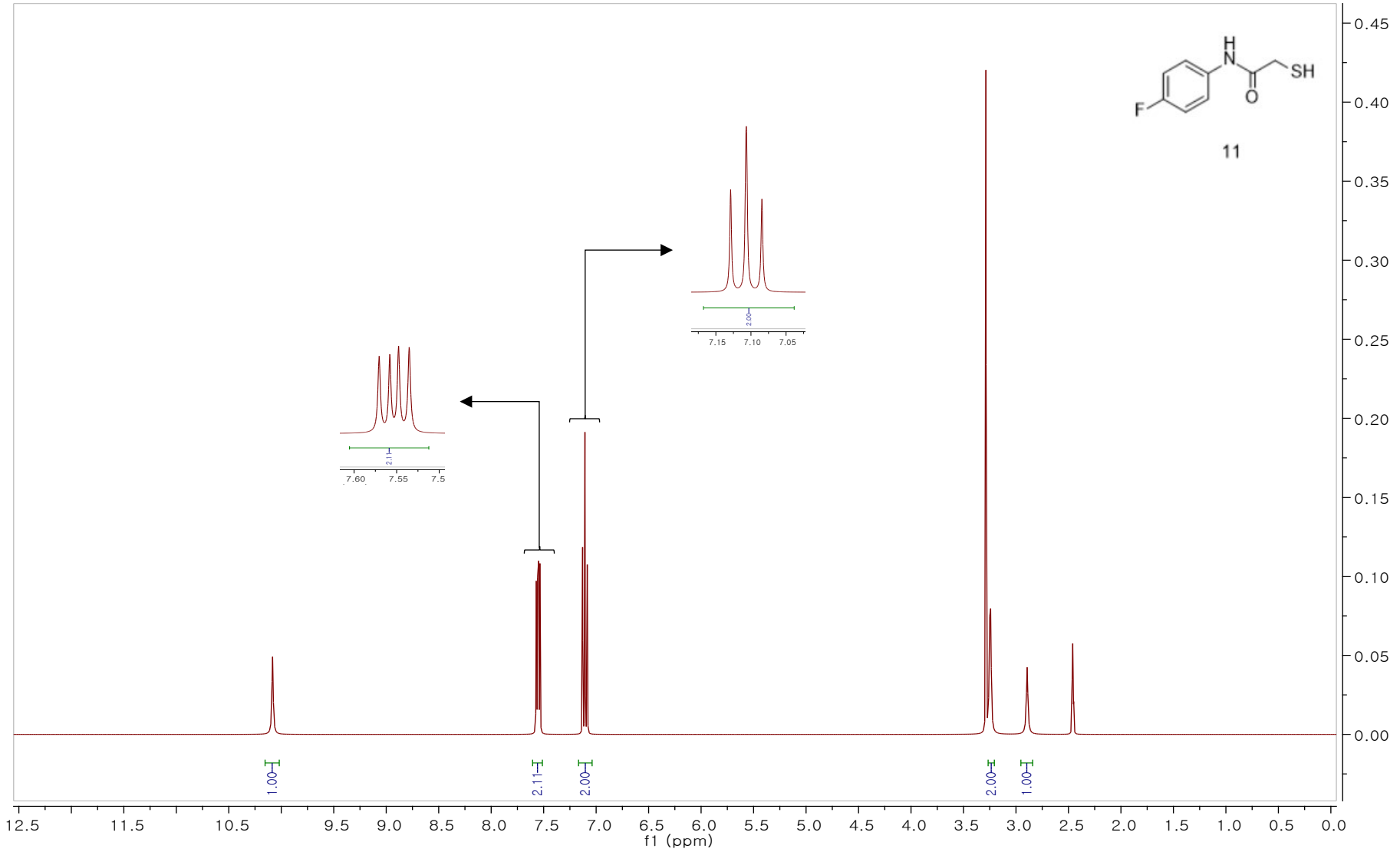
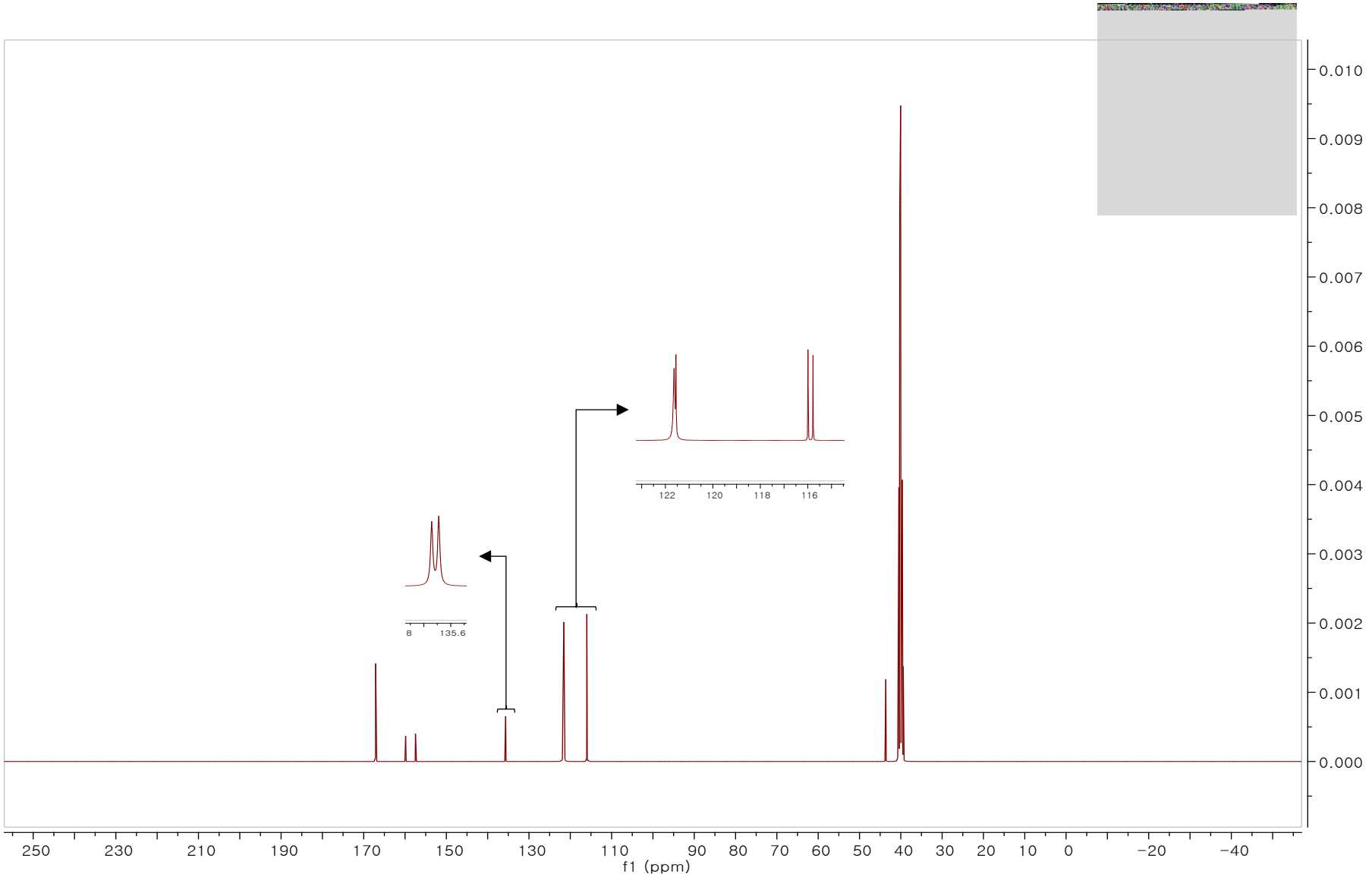


S21. ^{1}H NMR spectrum of analog **11**



S22. ^{13}C NMR spectrum of analog **11**

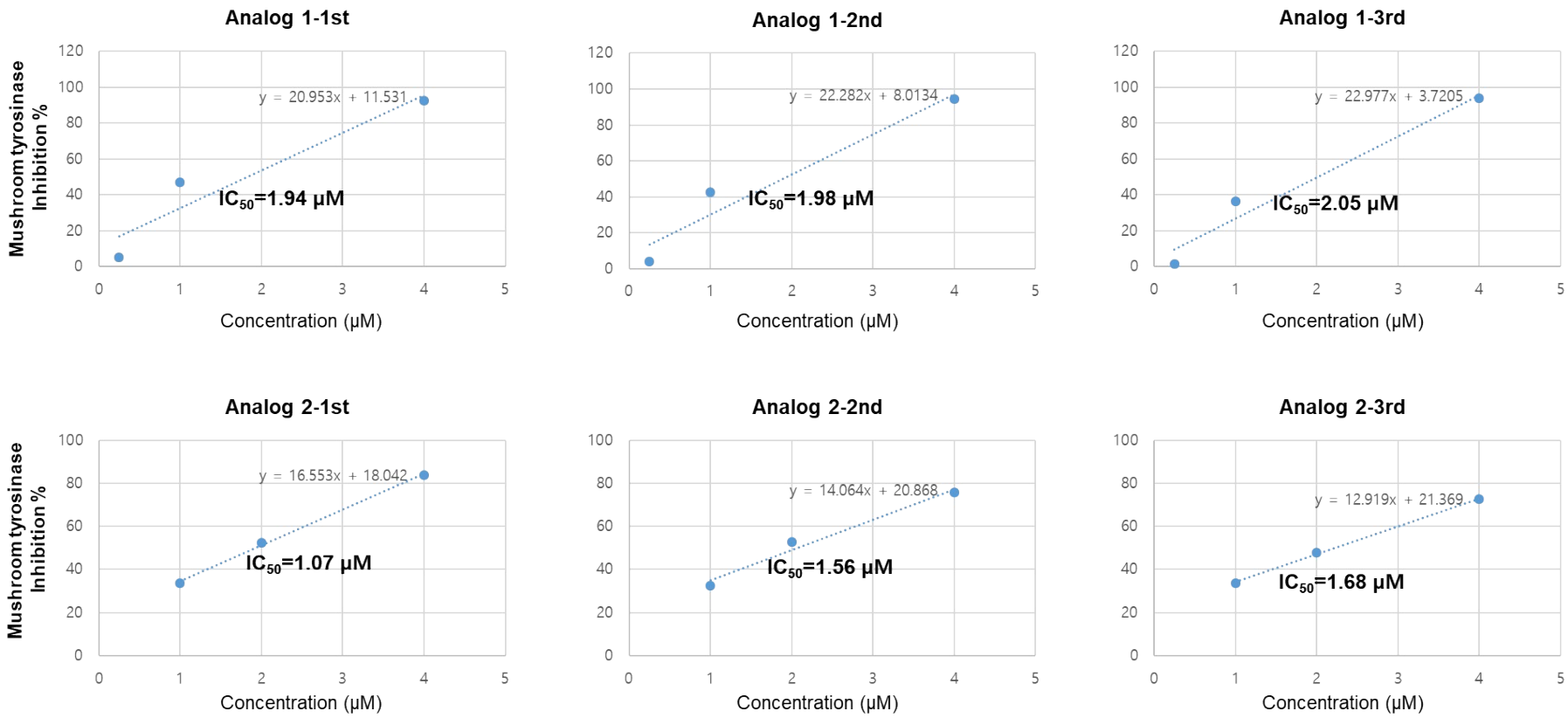
S23. Raw data used for IC_{50} values in the presence of L-tyrosine

Substrate: L-Tyrosine

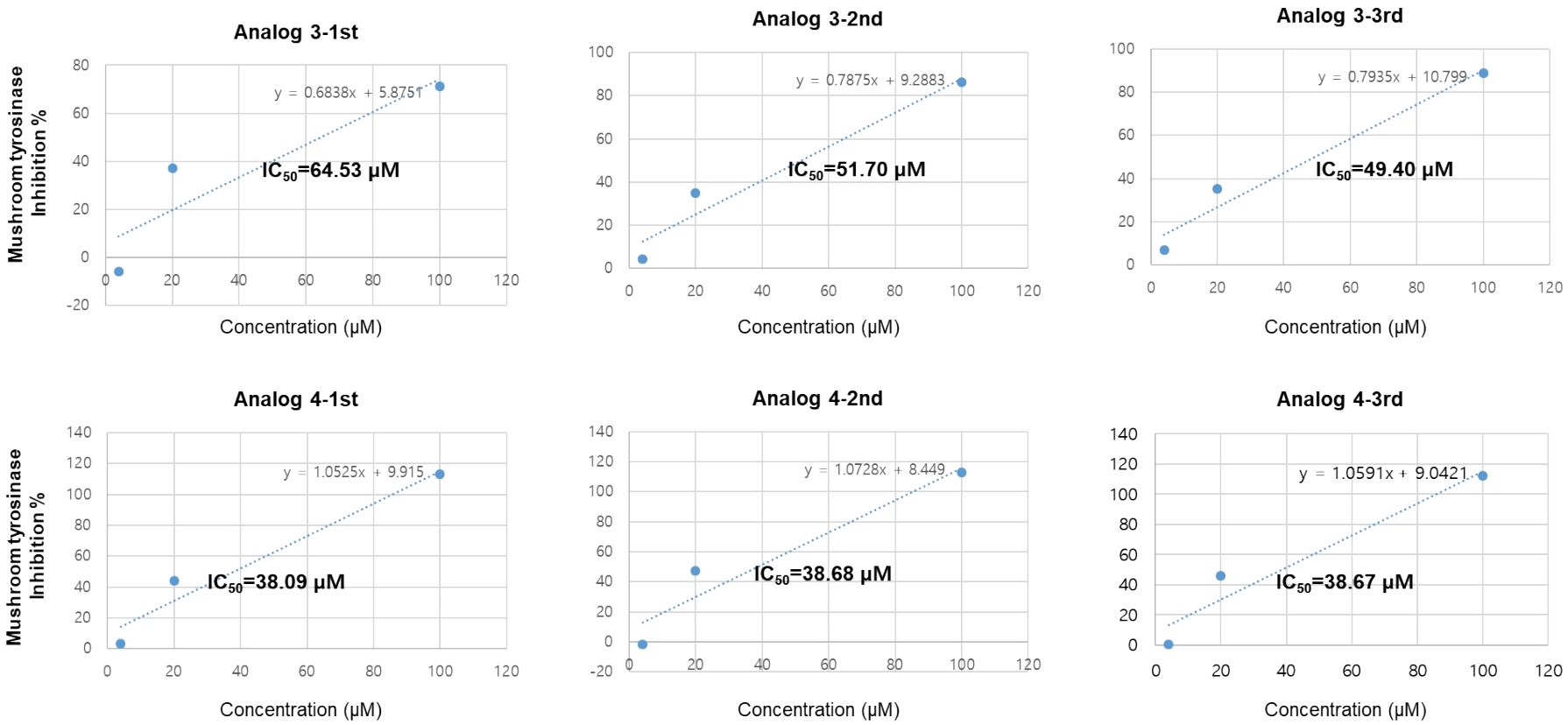
Compd.	Conc.(μM)	OD1	OD2	OD3	color control	Inhibition (%)			IC_{50} (n=3)	IC_{50} average	SEM
1	0.25	0.169	0.1707	0.1736	0.0363	5.06	3.84	1.76	1.94	1.99	0.03
	1	0.1116	0.1181	0.1267	0.0377	47.13	42.48	36.32	1.98		
	4	0.0469	0.0437	0.0451	0.0363	92.42	94.71	93.70	2.05		
2	1	0.1316	0.1334	0.1315	0.039	33.75	32.46	33.82	1.07	1.44	0.23
	2	0.1034	0.103	0.1097	0.0369	52.42	52.71	47.91	1.56		
	4	0.0594	0.0705	0.0748	0.0368	83.83	75.89	72.81	1.68		
3	4	0.1551	0.1438	0.1409	0.0368	-5.91	4.21	6.80	64.53	55.21	4.71
	20	0.1071	0.1094	0.109	0.0367	36.97	34.91	35.27	51.70		
	100	0.0691	0.0523	0.0497	0.0371	71.35	86.39	88.72	49.40		
4	4	0.1439	0.1496	0.1472	0.0358	3.22	-1.88	0.27	38.09	38.48	0.20
	20	0.1297	0.1259	0.1277	0.0672	44.05	47.45	45.84	38.68		
	100	0.0795	0.0797	0.0802	0.094	112.98	112.80	112.35	38.67		
5	4	0.1668	0.1803	0.1808	0.0382	11.62	2.34	1.99		>400	
	100	0.1754	0.2009	0.1929	0.0678	26.05	8.52	14.02			
6	4	0.1467	0.1468	0.1394	0.0394	3.94	3.85	10.47	45.25	44.20	1.69
	20	0.1258	0.1273	0.1239	0.0487	30.98	29.63	32.68	45.91		
	100	0.0949	0.0955	0.0914	0.0987	103.40	102.86	106.54	41.44		
7	0.25	0.1736	0.1747	0.1748	0.0358	1.41	0.62	0.55	2.33	1.91	0.01
	1	0.1066	0.1117	0.105	0.0371	50.27	46.63	51.42	2.37		
	4	0.045	0.0441	0.0456	0.04	96.42	97.07	95.99	2.33		
8	4	0.1793	0.1901	0.1851	0.0369	2.13	-5.29	-1.86	96.36	145.64	16.93
	100	0.1126	0.1384	0.1421	0.0426	51.89	34.16	31.62	138.56		

									152.71		
9	1	0.1205	0.1233	0.1185	0.0367	40.04	38.04	41.47	1.59	0.95	0.06
	2	0.0961	0.1003	0.0945	0.0359	56.93	53.92	58.07	1.73		
	4	0.0508	0.0523	0.049	0.0369	90.05	88.98	91.34	1.51		
10	5	0.1657	0.164	0.1627	0.0367	7.70	8.92	9.85	18.67	17.66	0.66
	10	0.1357	0.1338	0.1391	0.0361	28.74	30.10	26.31	16.67		
	20	0.1036	0.0927	0.1023	0.0373	52.56	60.36	53.49	18.65		
11	4	0.1423	0.1501	0.1484	0.0361	4.92	-2.06	-0.54	63.75	70.28	5.55
	20	0.1255	0.1278	0.1262	0.0371	20.86	18.80	20.23	81.32		
	100	0.0655	0.0827	0.0663	0.0384	75.74	60.34	75.02	65.77		
Kojic acid	6.25	0.0714	0.0731	0.0771	0.0364	19.04	15.11	5.86	23.64	24.26	0.31
	12.5	0.0675	0.0684	0.0648	0.0374	30.38	28.30	36.62	24.48		
	25	0.0517	0.0525	0.0537	0.0367	65.30	63.45	60.68	24.65		
	50	0.0445	0.0435	0.0426	0.0378	84.04	86.35	88.43			

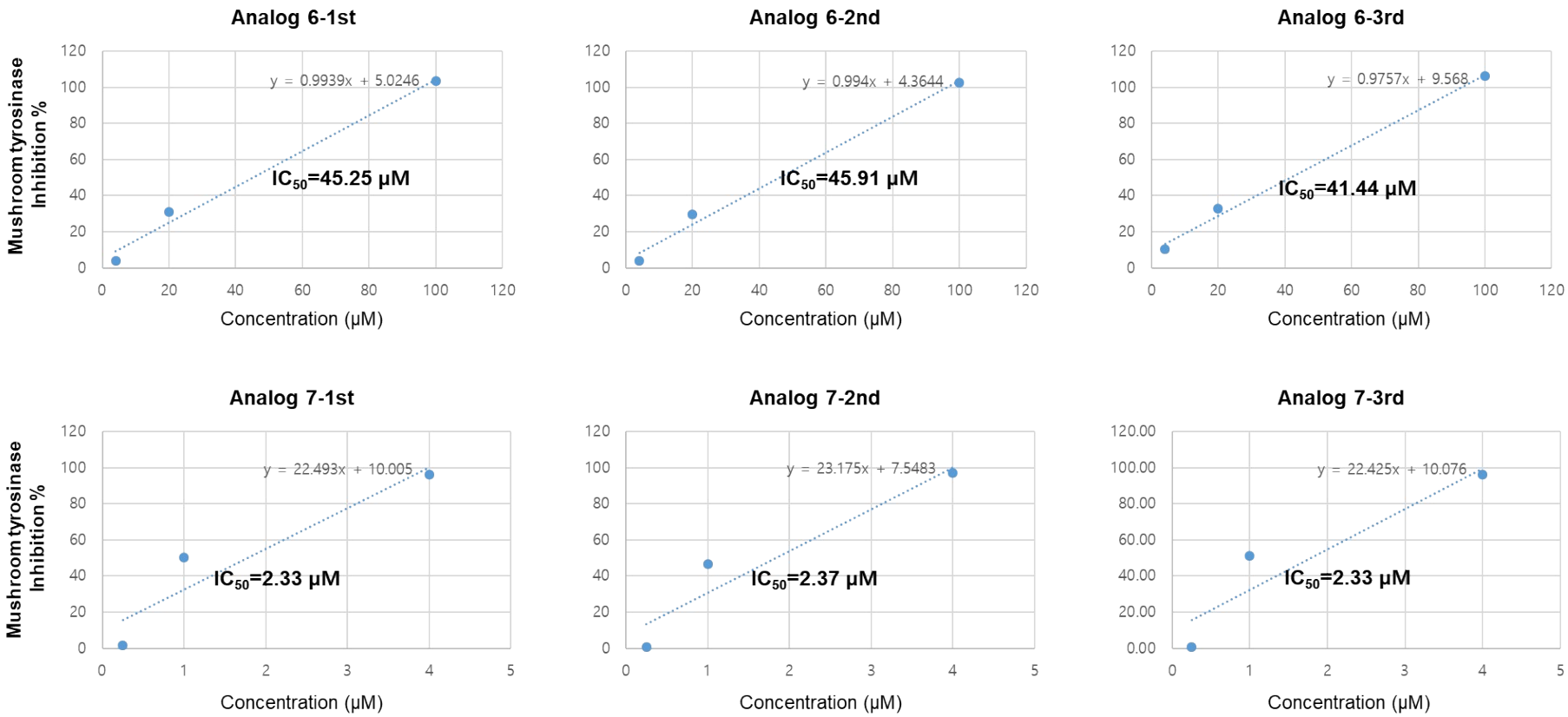
Substrate: L-Tyrosine



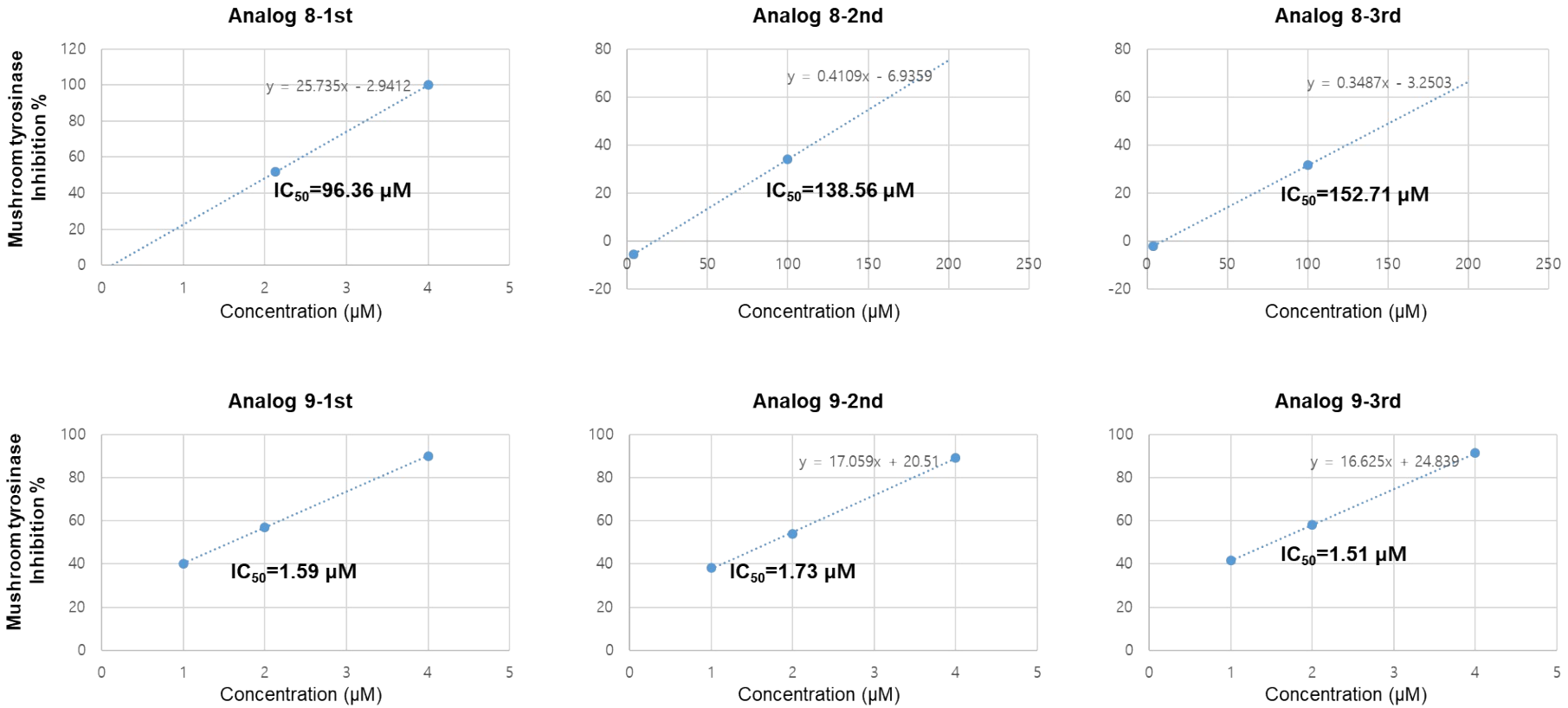
S24. Graphs used to calculate IC_{50} values for **1** and **2** in the presence of L-tyrosine



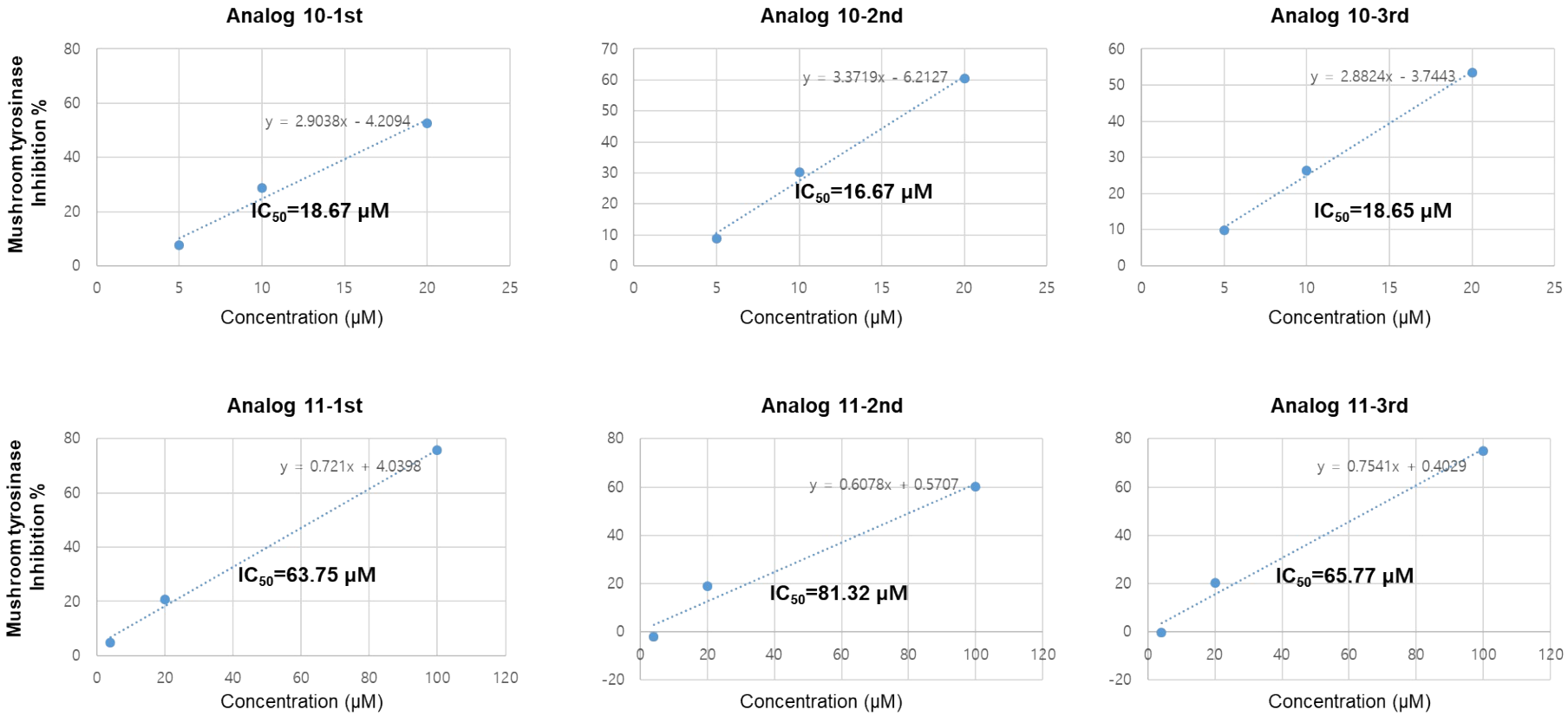
S25. Graphs used to calculate IC_{50} values for **3** and **4** in the presence of L-tyrosine



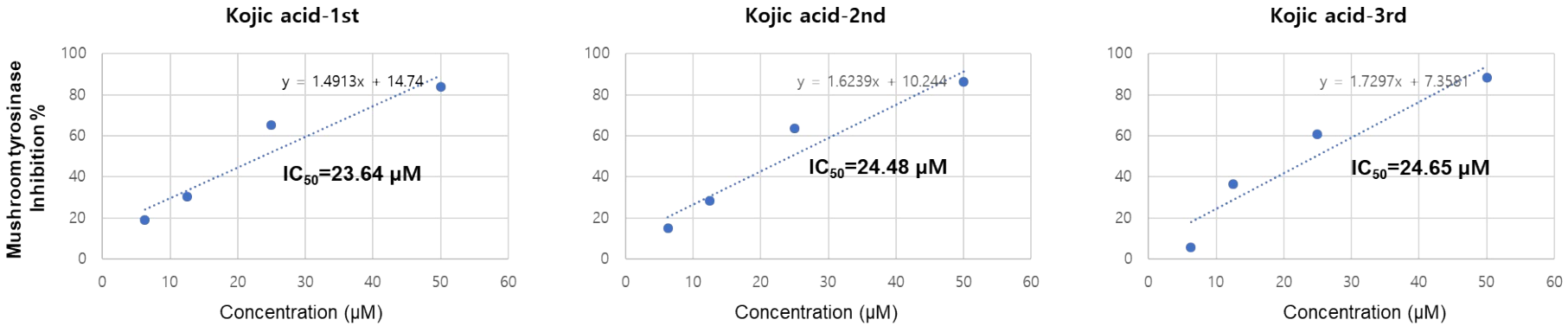
S26. Graphs used to calculate IC_{50} values for **6** and **7** in the presence of L-tyrosine



S27. Graphs used to calculate IC_{50} values for **8** and **9** in the presence of L-tyrosine



S28. Graphs used to calculate IC_{50} values for **10** and 11 in the presence of L-tyrosine



S29. Graphs used to calculate IC_{50} values for kojic acid in the presence of L-tyrosine

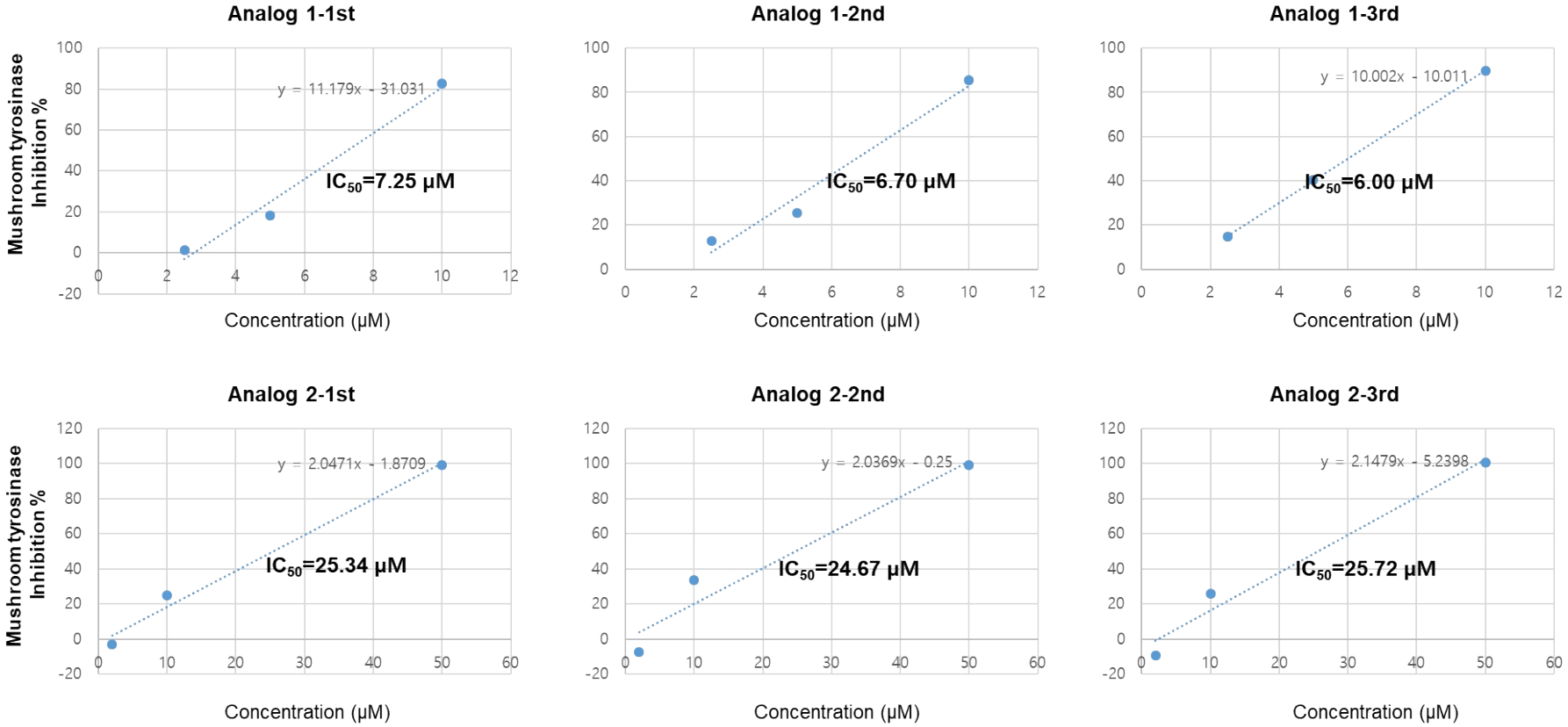
S30. Raw data used for IC_{50} values in the presence of L-dopa

Substrate: L-Dopa

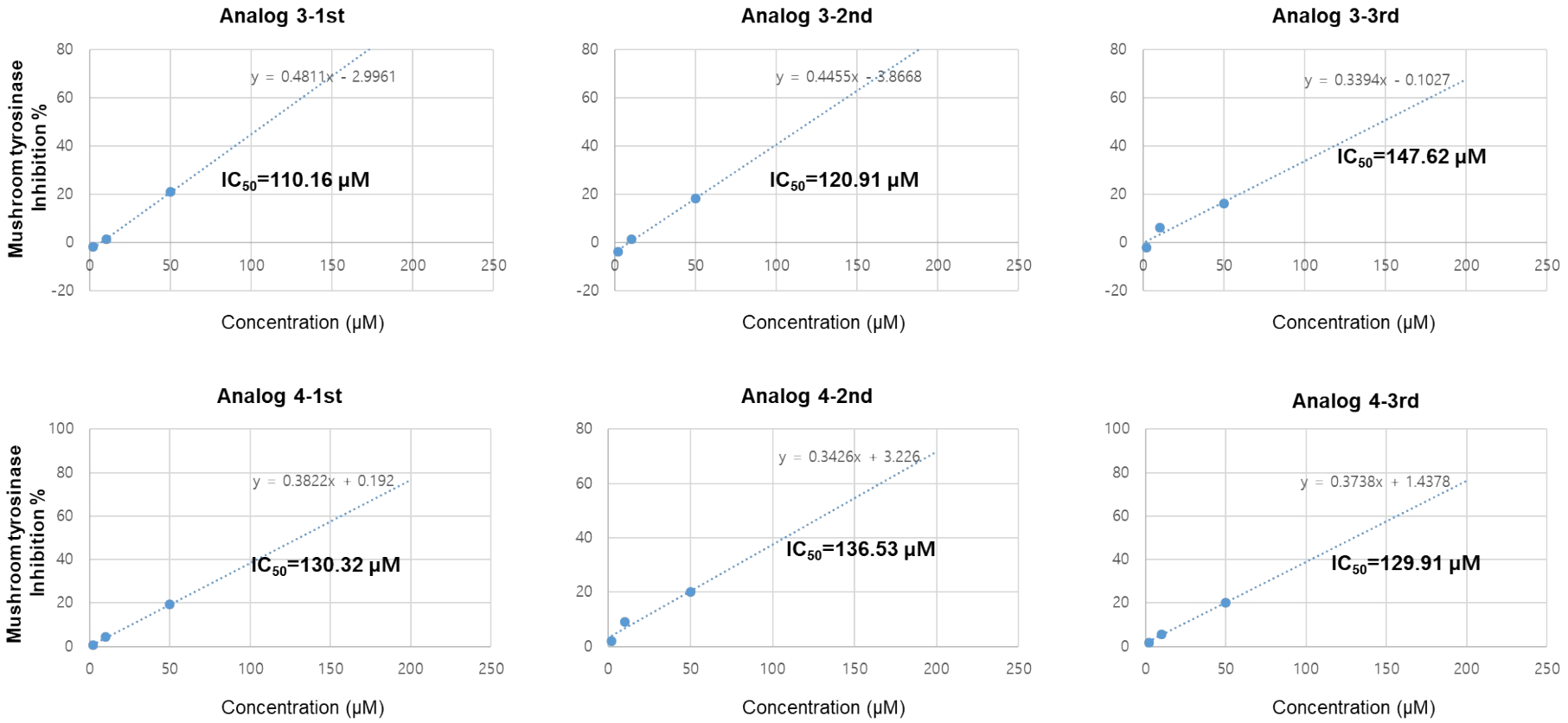
Compd	Conc.(μM)	OD1	OD2	OD3	Color control	inhibition (%)			IC_{50}	IC_{50} average	SEM
1	2.5	0.1578	0.1442	0.1419	0.0422	1.22	12.85	14.81	7.25	6.65	0.36
	5	0.1356	0.1274	0.11	0.0401	18.40	25.41	40.27	6.70		
	10	0.0609	0.0577	0.0527	0.0409	82.91	85.65	89.92	6.00		
2	2	0.2309	0.2388	0.2419	0.0384	-3.14	-7.38	-9.04	25.34	25.24	0.31
	10	0.1778	0.1619	0.1761	0.0379	25.04	33.56	25.95	24.67		
	50	0.0388	0.0389	0.0367	0.0377	99.41	99.36	100.54	25.72		
3	2	0.2282	0.2317	0.2287	0.0384	-1.70	-3.57	-1.96	110.16	126.23	11.14
	10	0.2224	0.2226	0.2132	0.0384	1.41	1.30	6.34	120.91		
	50	0.1855	0.1908	0.1944	0.0383	21.13	18.29	16.36	147.62		
4	2	0.2245	0.2223	0.2224	0.0392	0.71	1.89	1.84	130.32	132.25	2.14
	10	0.2194	0.2105	0.217	0.0408	4.30	9.07	5.59	136.53		
	50	0.2183	0.217	0.2168	0.0676	19.25	19.95	20.06	129.91		
5	4	0.1569	0.1594	0.1661	0.0416	1.48	-0.66	-6.38	110.13	117.86	3.89
	20	0.148	0.1481	0.1389	0.0484	14.90	14.81	22.67	120.86		
	100	0.127	0.1319	0.1339	0.0627	45.06	40.87	39.16	122.58		
6	4	0.1669	0.1664	0.1578	0.0467	-2.71	-2.28	5.07	89.16	95.31	2.51
	20	0.1443	0.1535	0.1333	0.0525	21.56	13.70	30.96	105.17		
	100	0.1378	0.1467	0.1409	0.0844	54.37	46.77	51.72	91.59		
7	2.5	0.1504	0.1456	0.1396	0.0413	6.78	10.88	16.01	7.05	6.49	0.28
	5	0.1254	0.1192	0.123	0.041	27.88	33.18	29.93	6.21		
	10	0.0673	0.0513	0.0526	0.0434	79.58	93.25	92.14	6.21		
8	4	0.1648	0.1628	0.1753	0.0409	-5.87	-4.16	-14.84	268.56	309.49	52.67

	20	0.149	0.1524	0.1644	0.0397	6.61	3.70	-6.55	414.02		
	100	0.1395	0.1468	0.1449	0.0415	16.26	10.03	11.65	245.89		
9	2	0.2134	0.2193	0.1783	0.0376	2.77	-0.50	22.18	18.83	18.03	1.35
	10	0.1138	0.12	0.1264	0.038	58.08	54.65	51.11	19.86		
	50	0.0856	0.0824	0.0818	0.0827	98.40	100.17	100.50	15.39		
10	4	0.1572	0.189	0.1602	0.0413	0.97	-26.20	-1.59	108.04	98.18	9.34
	20	0.1498	0.1425	0.1321	0.0402	6.35	12.59	21.48	106.98		
	100	0.1074	0.1112	0.0901	0.0446	46.34	43.09	61.12	79.51		
11	4	0.1647	0.1565	0.159	0.041	-5.70	1.31	-0.83	195.01	257.61	32.48
	20	0.1546	0.1455	0.1465	0.0409	2.85	10.62	9.77	303.94		
	100	0.1335	0.1387	0.1382	0.0432	22.84	18.40	18.83	273.89		
Kojic acid	6.25	0.0981	0.0956	0.0977	0.0357	3.70	7.56	4.32	31.39	30.10	0.75
	12.5	0.0877	0.0825	0.0873	0.0381	23.46	31.48	24.07	30.14		
	25	0.0757	0.0764	0.0727	0.0372	40.59	39.51	45.22	28.78		
	50	0.0517	0.051	0.0467	0.0385	79.63	80.71	87.35			

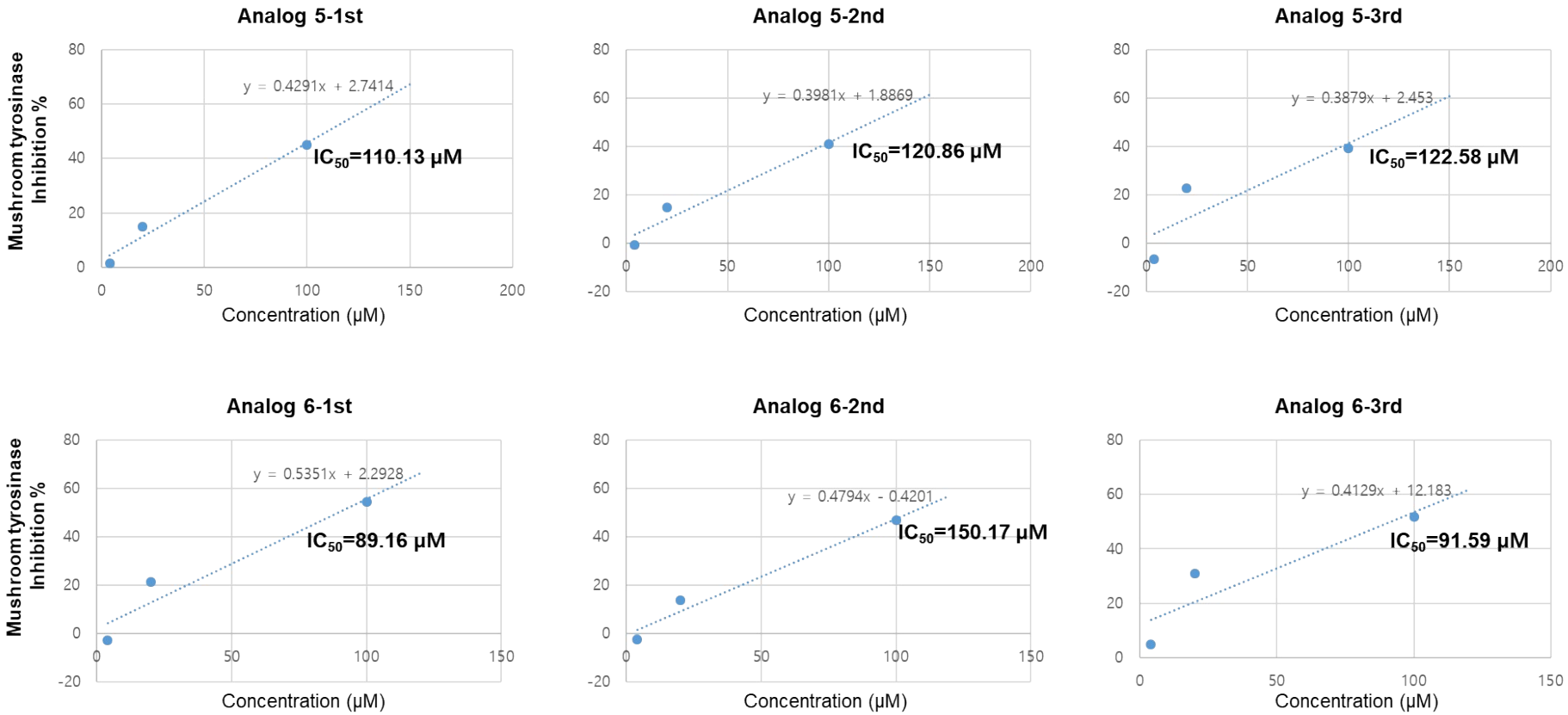
Substrate: L-Dopa



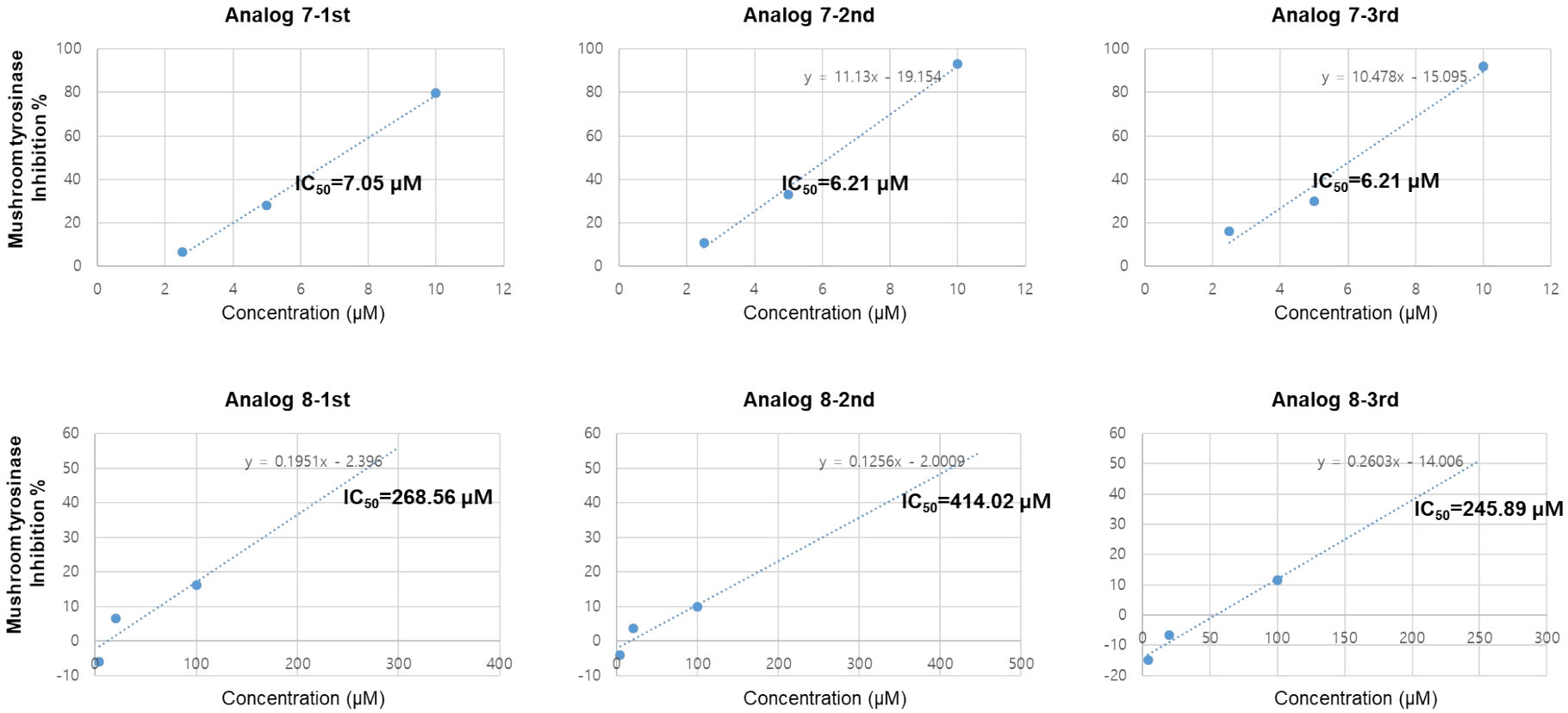
S31. Graphs used to calculate IC_{50} values for **1** and **2** in the presence of L-dopa



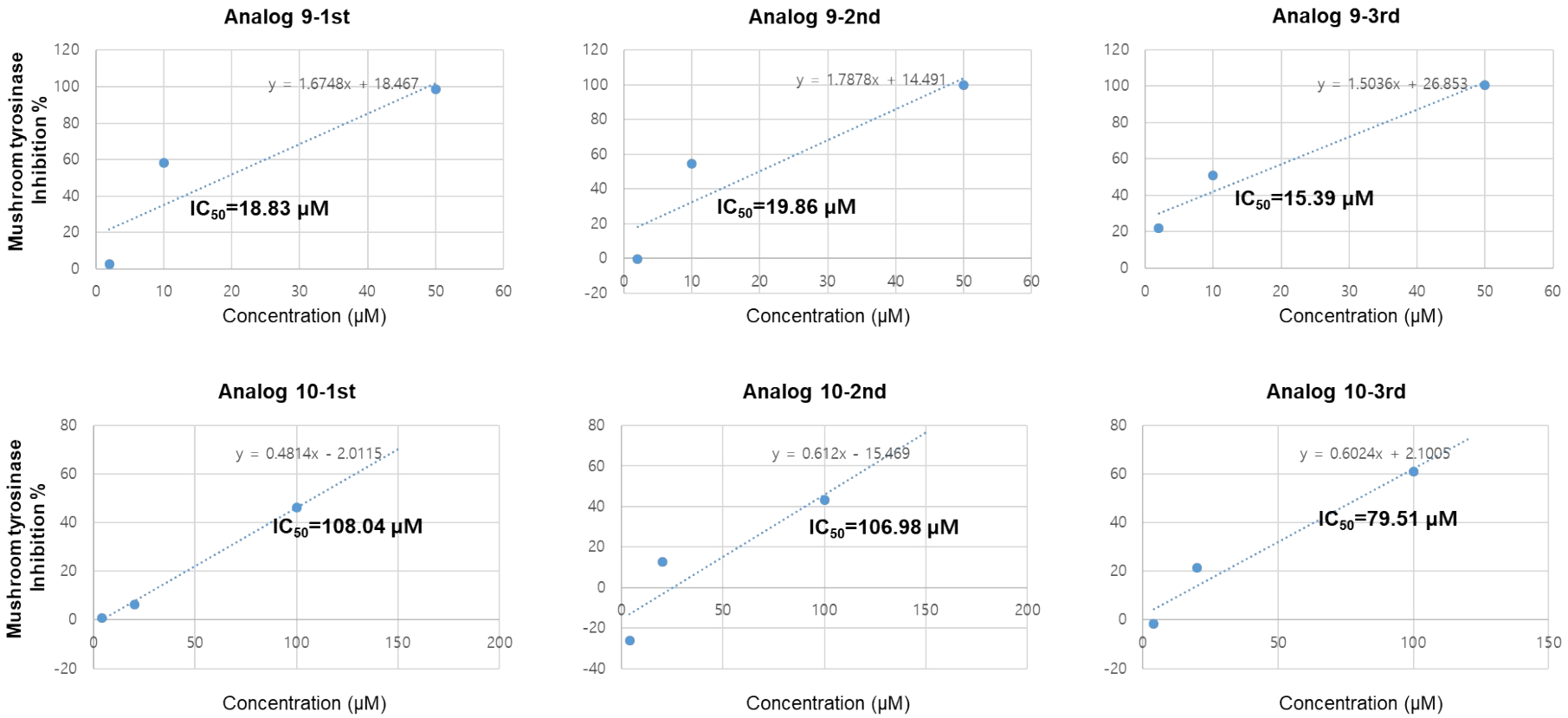
S32. Graphs used to calculate IC_{50} values for **3** and **4** in the presence of L-dopa



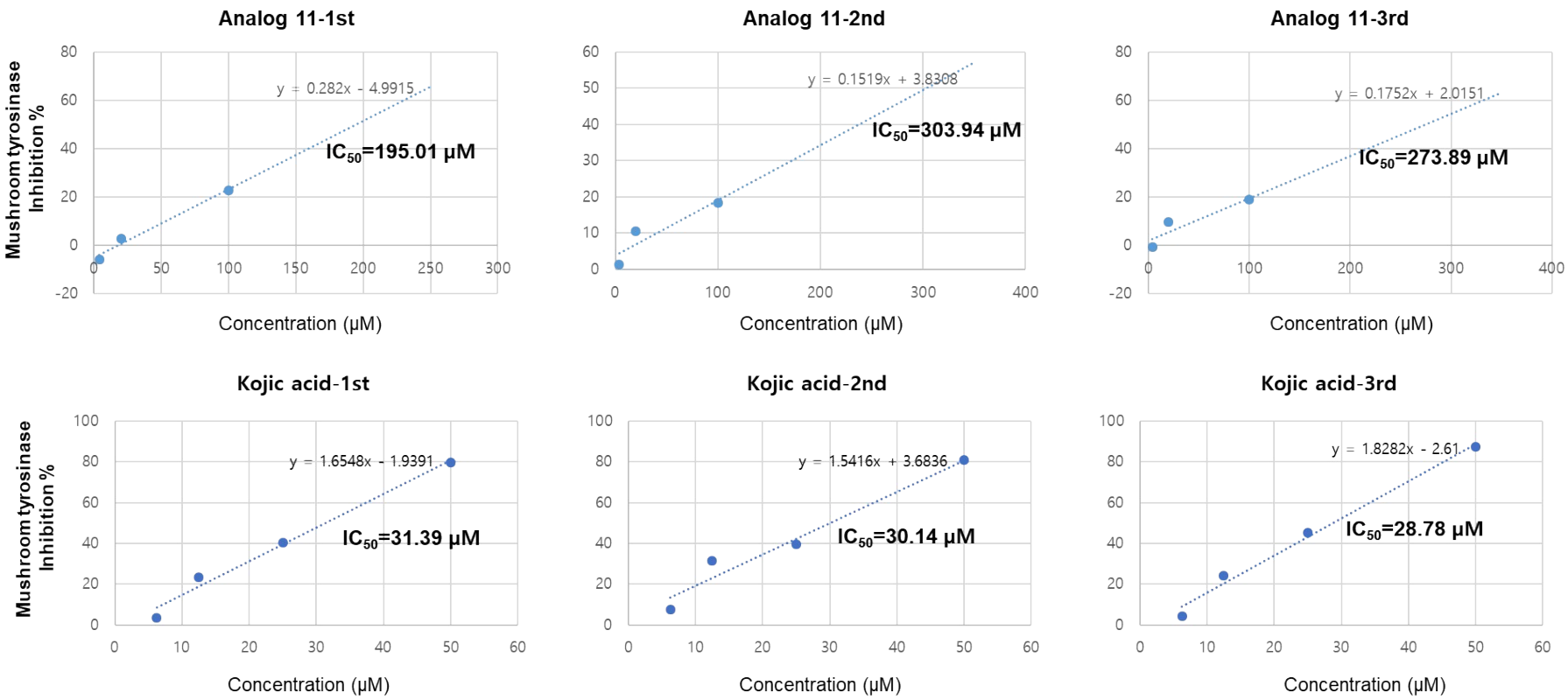
S33. Graphs used to calculate IC_{50} values for **5** and **6** in the presence of L-dopa



S34. Graphs used to calculate IC_{50} values for **7** and **8** in the presence of L-dopa

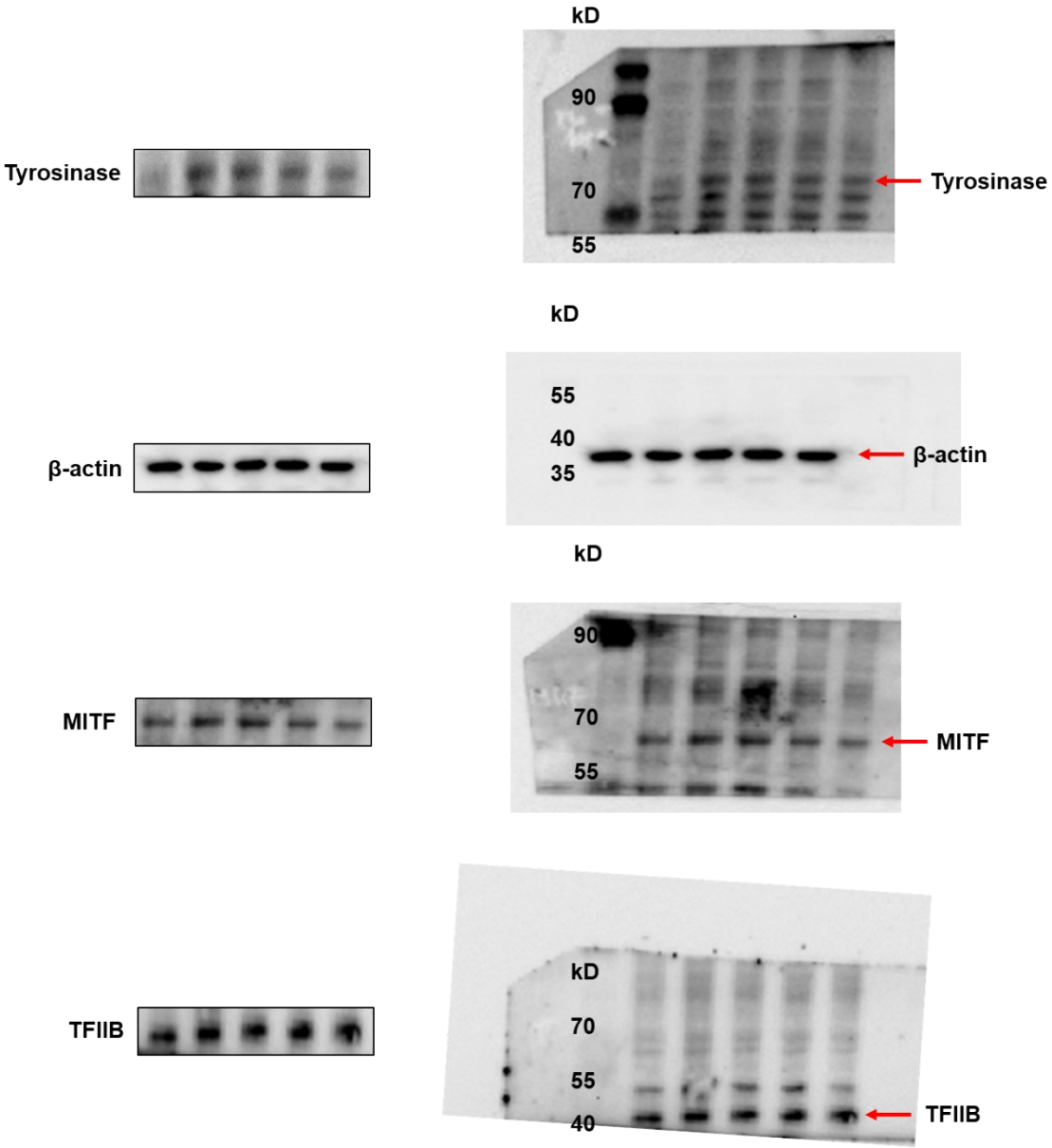


S35. Graphs used to calculate IC_{50} values for **9** and **10** in the presence of L-dopa



S36. Graphs used to calculate IC_{50} values for **11** and kojic acid in the presence of L-dopa

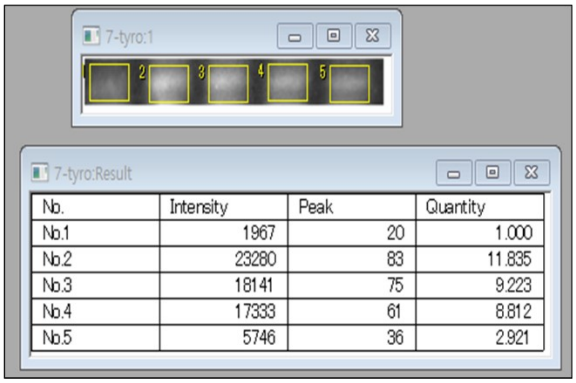
Original blot (1st set)



S37. Original western blot for analog **7** (1st set)

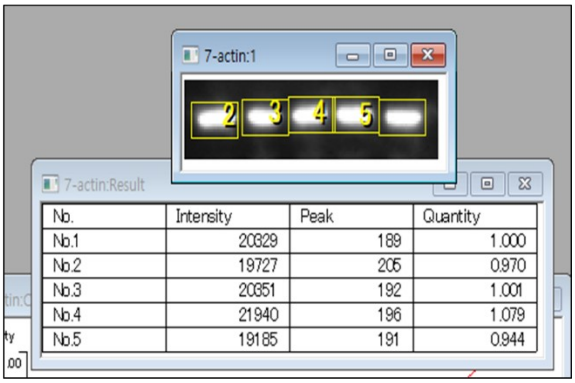
Quantification of band

(A) Tyrosinase



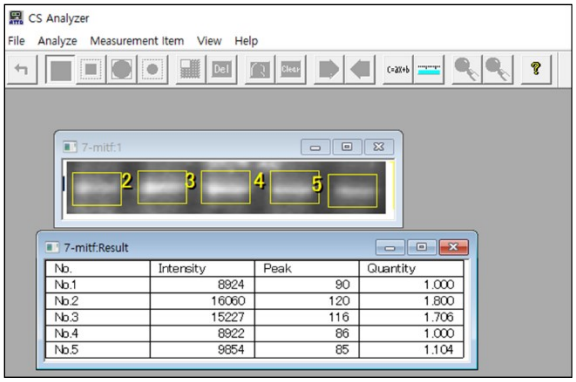
No.	Intensity	Peak	Quantity
No.1	1967	20	1.000
No.2	23280	83	11.835
No.3	18141	75	9.223
No.4	17333	61	8.812
No.5	5746	36	2.921

(B) β-actin



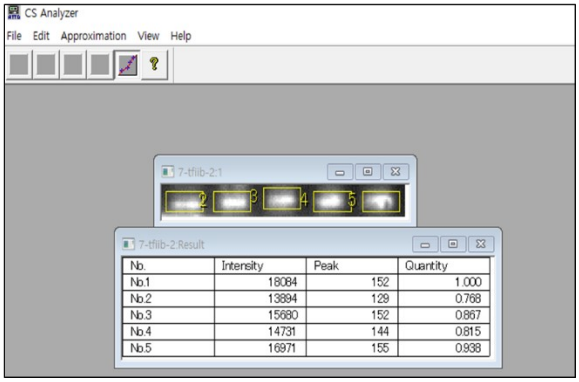
No.	Intensity	Peak	Quantity
No.1	20329	189	1.000
No.2	19727	205	0.970
No.3	20351	192	1.001
No.4	21940	196	1.079
No.5	19185	191	0.944

(C) MITF



No.	Intensity	Peak	Quantity
No.1	8924	90	1.000
No.2	16060	120	1.800
No.3	15227	116	1.706
No.4	8922	86	1.000
No.5	9854	85	1.104

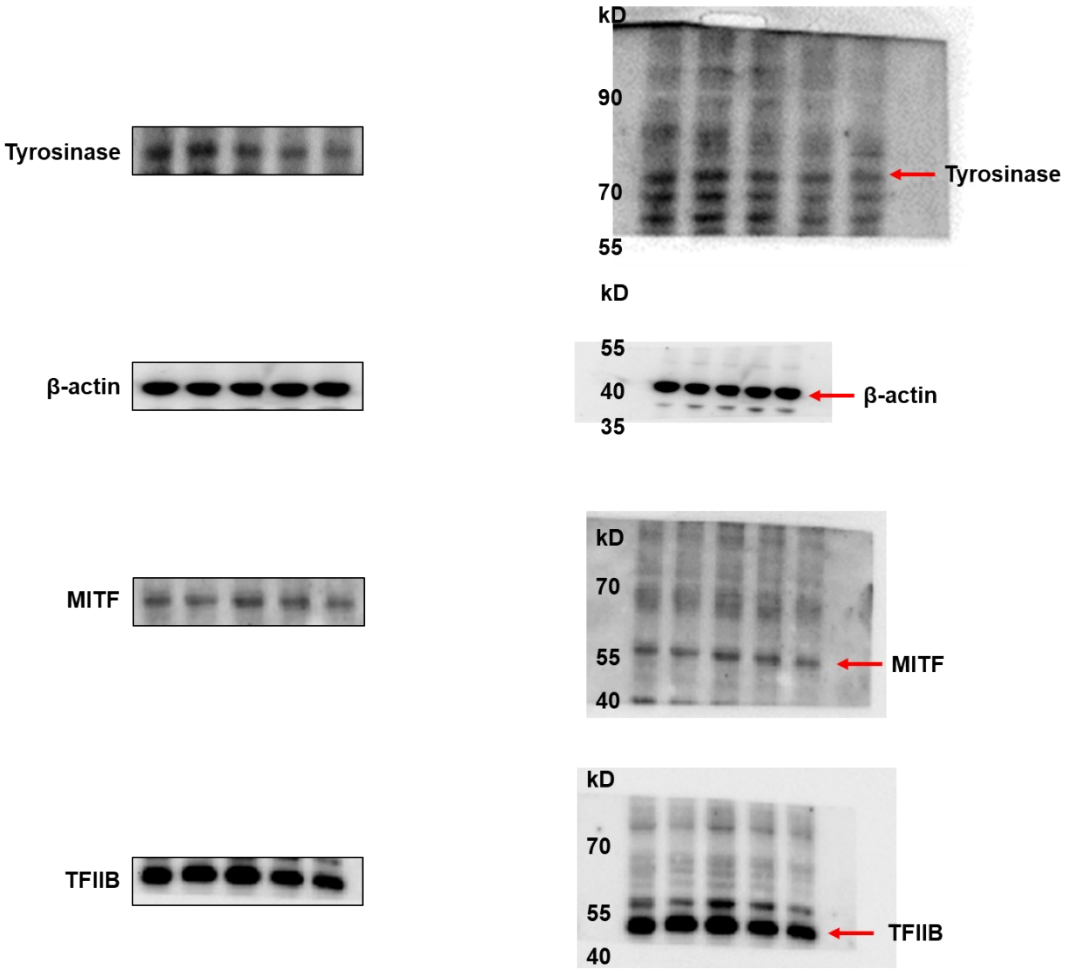
(D) TFIIB



No.	Intensity	Peak	Quantity
No.1	18084	152	1.000
No.2	13894	129	0.768
No.3	15680	152	0.867
No.4	14731	144	0.815
No.5	16971	155	0.938

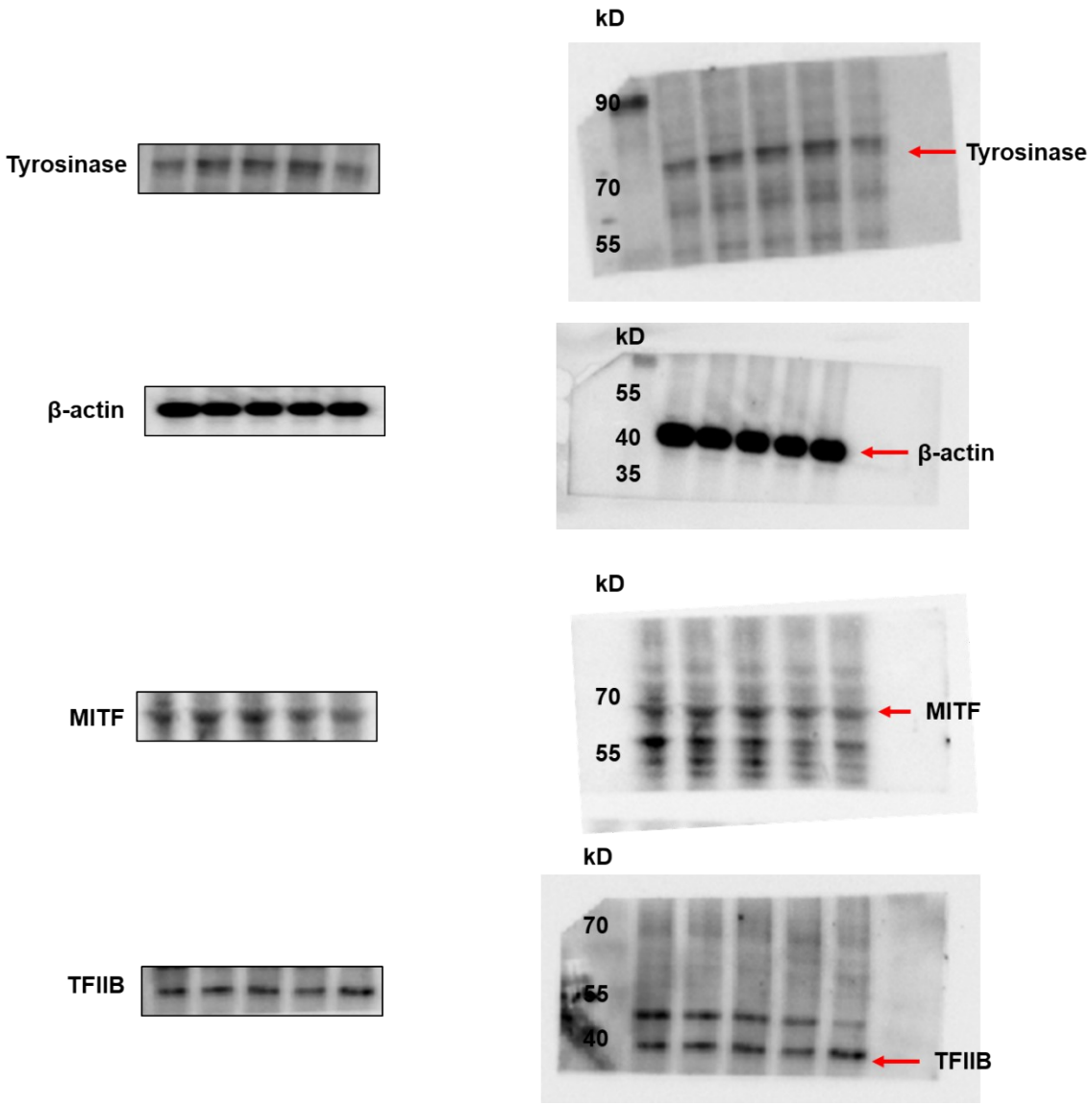
S38. Quantification analysis of protein expressions (A) tyrosinase, (B) β-actin, (c) MITF, and (D) TFIIB for analog **7** by CS analyzer software program

Original blot (2nd set)

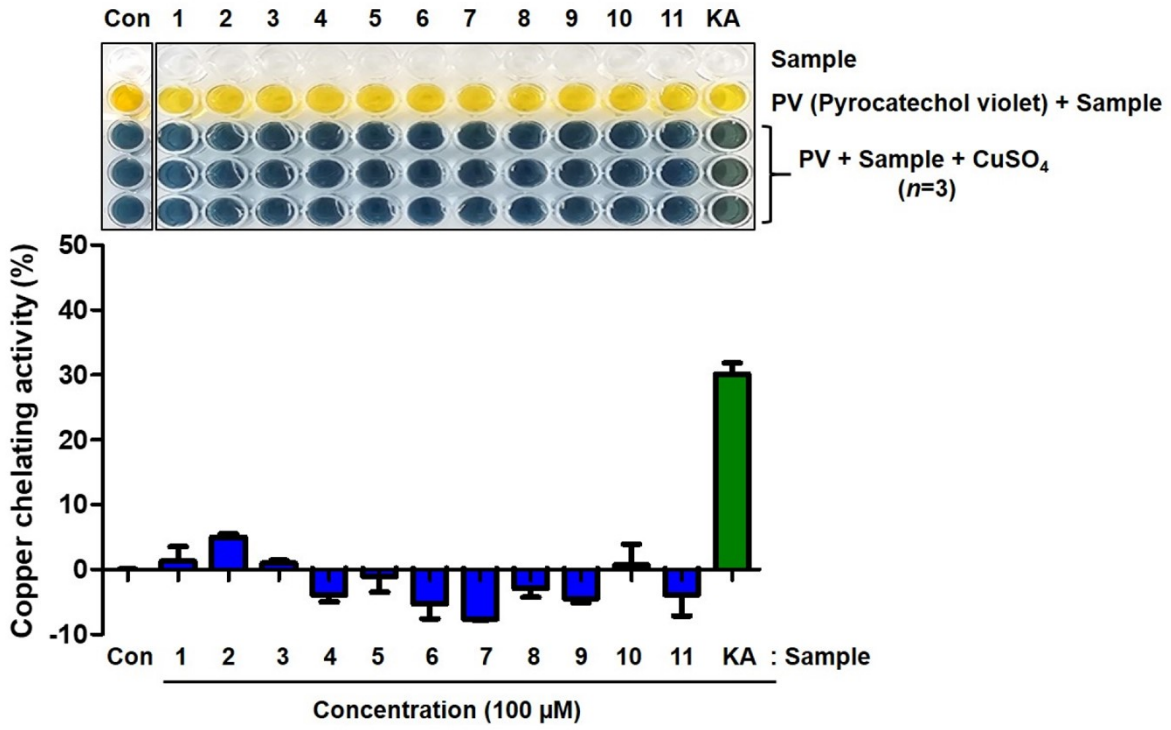


S39. Original western blot for analog **7** (2nd set)

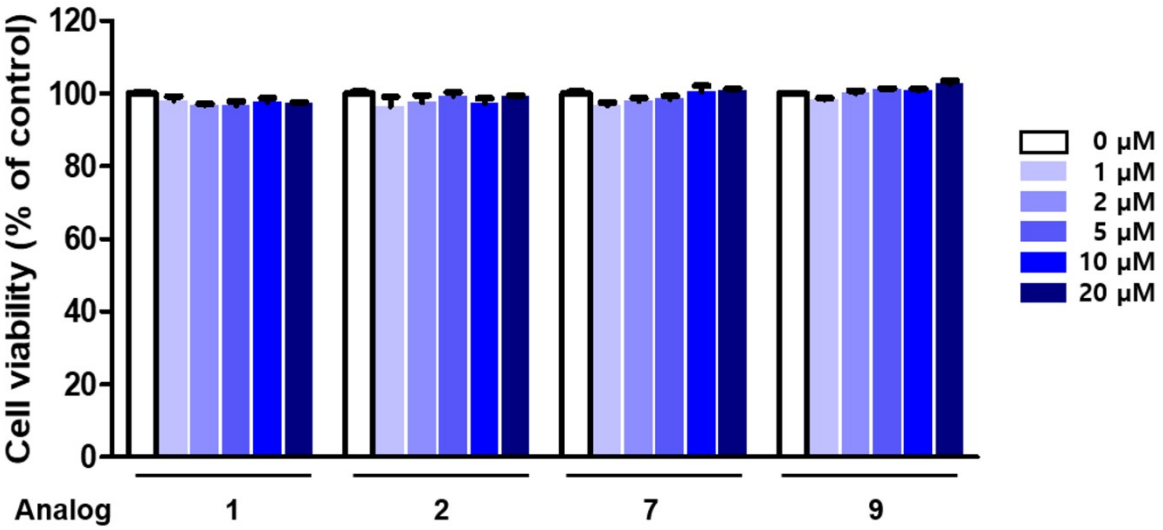
Original blot (3rd set)



S40. Original western blot for analog **7** (3rd set)



S41. Copper chelation activity of kojic acid and 2-MAA analogs **1**–**11** using a pyrocatechol violet reagent. KA, kojic acid (a positive control). All samples were exposed at a concentration of 100 μM in the presence of 80 μM pyrocatechol violet (PV) and 209 μM $CuSO_4$. When PV chelates with copper ions, the color of PV changes to bluish-violet.



S42. Effect of analogs **1**, **2**, **7**, and **9** on HaCaT cell viability. Each analog was exposed at concentrations of 0, 1, 2, 5, 10, and 20 μM for 24 h. HaCaT, human immortal keratinocyte.