

SUPPLEMENTARY INFORMATION

Effective synthesis of novel dihydrobenzoxazoles bearing the 2-aminothiazole moiety and evaluation of the antiproliferative activity of their acylated derivatives

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^aInstitute of Bioorganic Chemistry, National Academy of Sciences of Belarus, Akad. Kuprevicha st. 5/2, Minsk 220141, Belarus

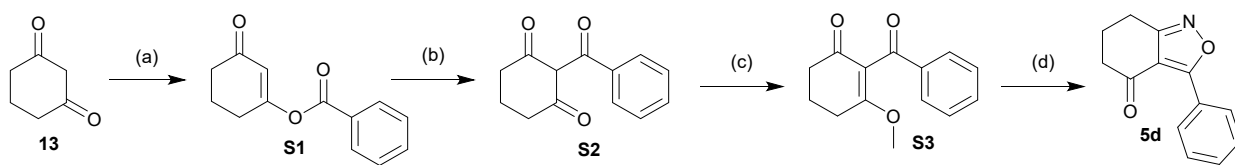
^bDepartment of Experimental Tumor Biology, Blokhin N.N. National Medical Research Center of Oncology, Kashirskoye sh. 24, Moscow 115522, Russian Federation

^cInstitute of Gene Biology, Russian Academy of Sciences, Vavilova st. 34/5, Moscow 119334, Russian Federation

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Synthesis of compound 5d



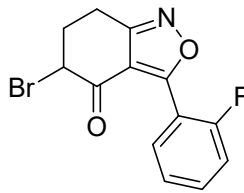
Scheme S1. Synthesis of compound 5d. Reagents and conditions: (a) benzoyl chloride, Py, CHCl₃, r.t., 1 h, 94%; (b) acetone cyanohydrin, Et₃N, CH₃CN, r.t., 3 h, 91%; (c) (CH₃)₂SO₄, K₂CO₃, CH₃CN, reflux, 0.5 h, 80%; (d) NH₂OH·HCl, NaOH, MeOH, r.t., 3 h, 89%.

3-Oxocyclohex-1-en-1-yl benzoate S1 was prepared according to the method described in the work¹. Yield 94%. Light yellow oil (lit.¹ oil). ¹H NMR(Chloroform-*d*, 500 MHz): δ (ppm) 8.07 (dd, *J*=8.3, 1.4 Hz, 2H), 7.67 – 7.60 (m, 1H), 7.53 – 7.46 (m, 2H), 6.04 (t, *J*=1.3 Hz, 1H), 2.71 – 2.63 (m, 2H), 2.49 – 2.42 (m, 2H), 2.12 (p, *J*=6.4 Hz, 2H). ¹³C NMR(Chloroform-*d*, 126 MHz): δ (ppm) 199.60, 170.30, 163.29, 134.22, 130.33, 128.82, 128.74, 117.98, 36.90, 28.54, 21.46.

2-Benzoylcyclohexane-1,3-dione S2 was prepared according to the method described for the synthesis of analogous compounds in the work². Yield 87%. Light yellow oil (lit. m.p.¹ 55-58 °C). ¹H NMR(Chloroform-*d*, 500 MHz): δ (ppm) 16.96 (s, 1H), 7.55 – 7.47 (m, 3H), 7.43 – 7.36 (m, 2H), 2.75 (t, *J*=6.3 Hz, 2H), 2.50 (t, *J*=6.6 Hz, 2H), 2.07 (p, *J*=6.5 Hz, 2H). ¹³C NMR(Chloroform-*d*, 126 MHz): δ (ppm) 199.14, 196.39, 194.34, 138.30, 132.04, 128.40, 127.84, 113.39, 38.27, 32.57, 19.27.

2-Benzoyl-3-methoxycyclohex-2-en-1-one S3 was prepared according to the method described for the synthesis of compound 16 (see experimental part of this article). Yield 80%. M.p. 160-164 °C. ¹H NMR(Chloroform-*d*, 500 MHz): δ (ppm) 7.91 – 7.80 (m, 2H), 7.55 – 7.50 (m, 1H), 7.45 – 7.40 (m, 2H), 3.72 (s, 3H), 2.68 (t, *J*=6.2 Hz, 2H), 2.46 (t, *J*=6.7 Hz, 2H), 2.14 (p, *J*=6.5 Hz, 2H). ¹³C NMR(Chloroform-*d*, 126 MHz): δ (ppm) 196.58, 195.43, 174.87, 137.49, 133.40, 129.26, 128.64, 119.94, 56.72, 36.53, 26.01, 20.70.

3-Phenyl-6,7-dihydrobenzo[*c*]isoxazol-4(5*H*)-one 5d was prepared according to the method described for the synthesis of compound 5e (see experimental part of this article). Yield 92%, mp 58-62 °C (lit.³ 65-67.5 °C). ¹H NMR(Chloroform-*d*, 500 MHz): δ (ppm) 8.50 – 8.36 (m, 2H), 7.59 – 7.46 (m, 3H), 3.03 – 2.95 (m, 2H), 2.67 – 2.58 (m, 2H), 2.17 (p, *J*=6.5 Hz, 2H). ¹³C NMR(Chloroform-*d*, 126 MHz): δ (ppm) 192.59, 170.82, 165.02, 132.44, 128.83, 126.47, 112.49, 40.20, 22.38, 21.98.



5-bromo-3-(2-fluorophenyl)-6,7-dihydrobenzo[c]isoxazol-4(5H)-one

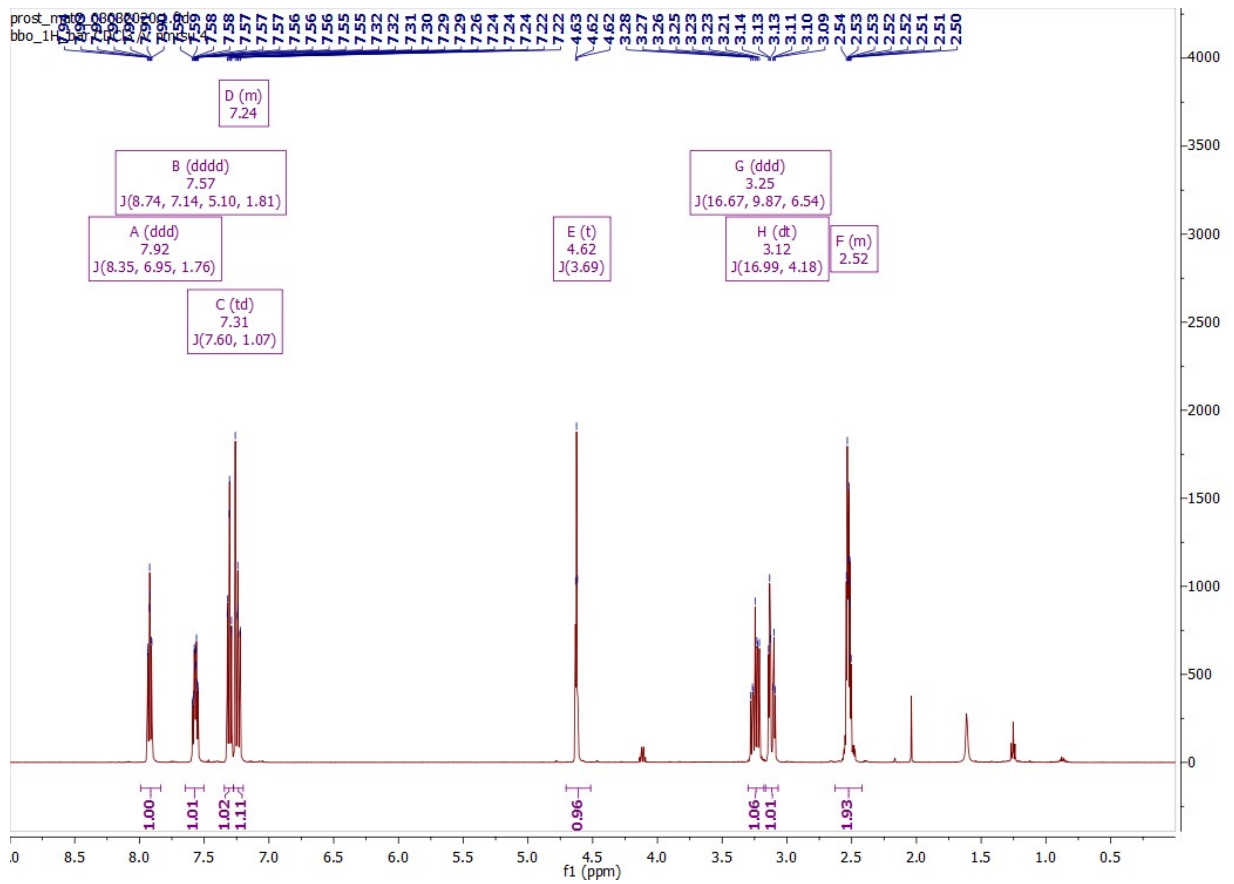
Chemical Formula: C₁₃H₉BrFNO₂

Exact Mass: 308,9801

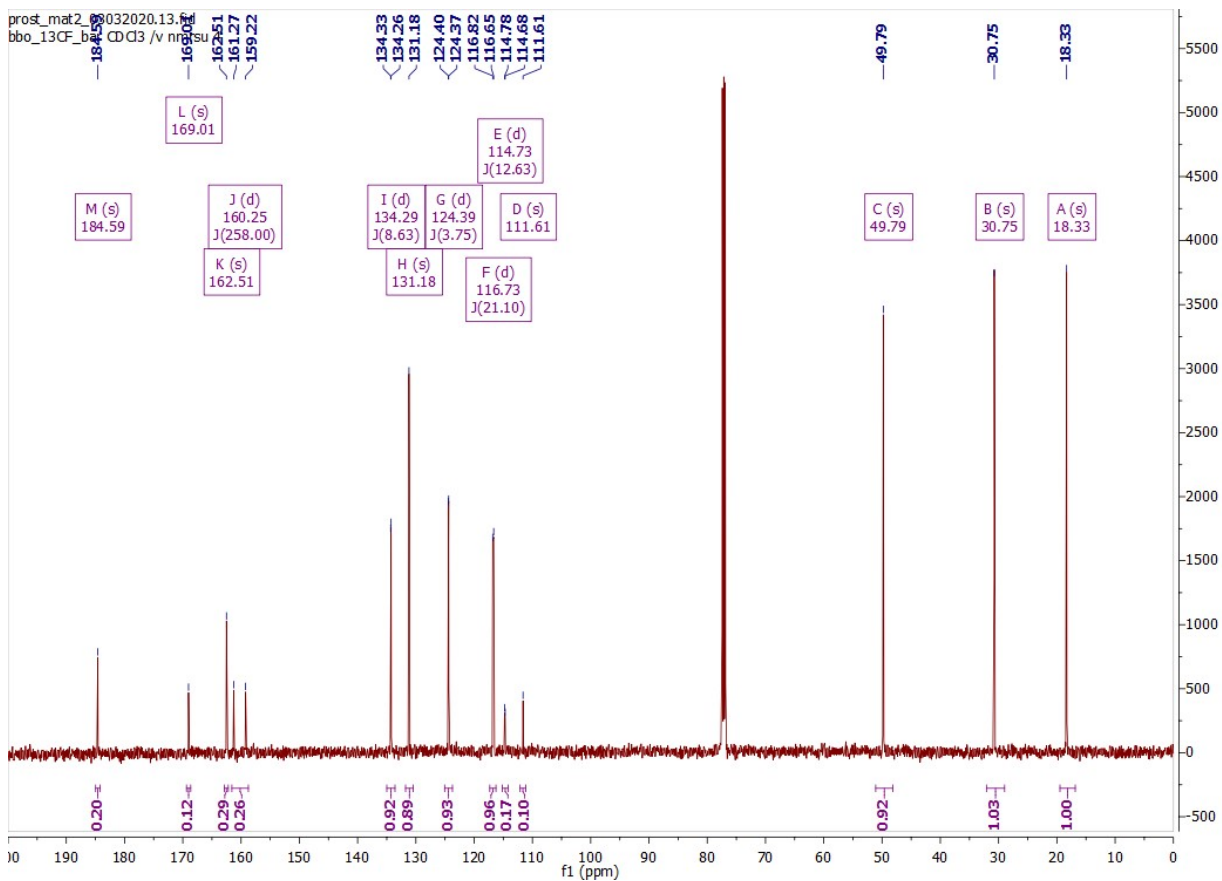
Molecular Weight: 310,1224

6a

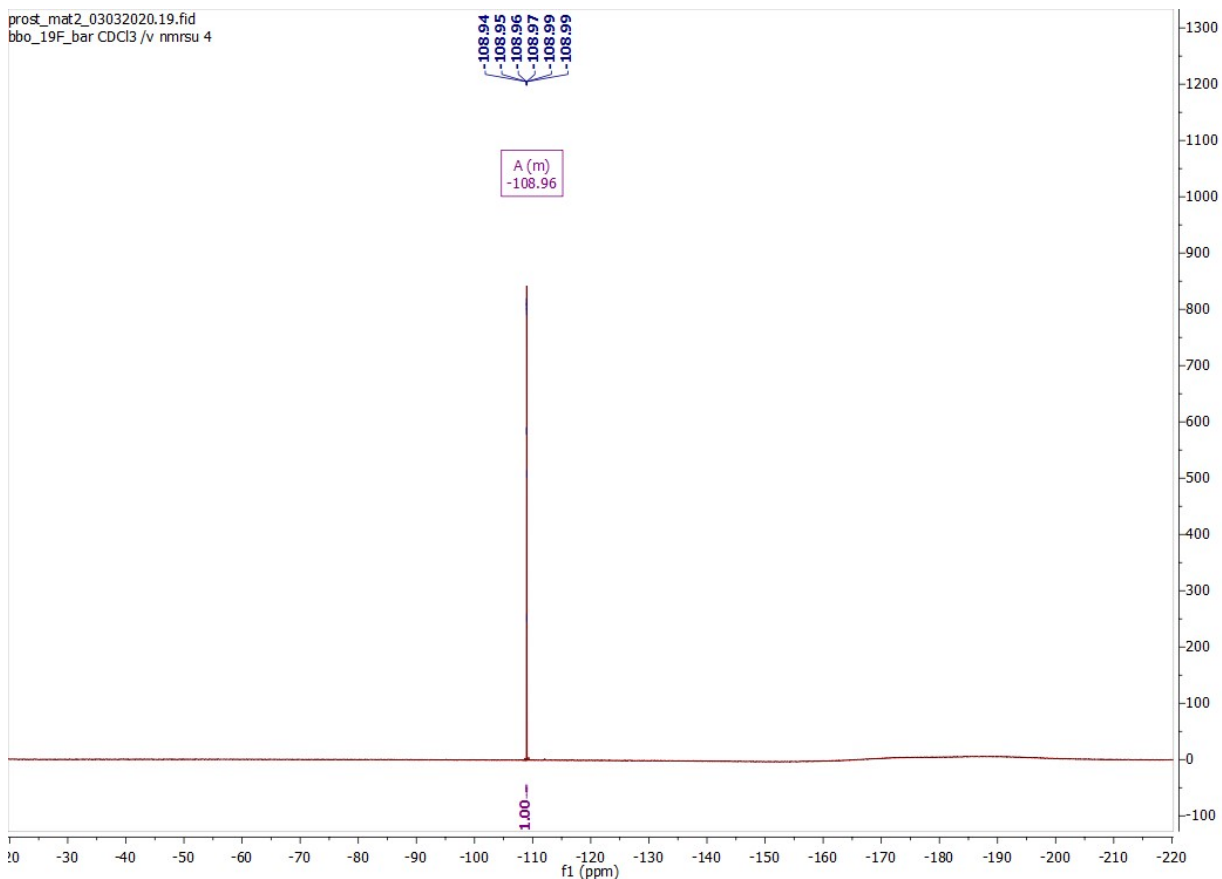
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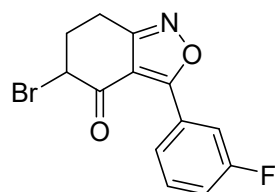


^{13}C spectra of **6a**:



^{19}F spectra of **6a**:





5-bromo-3-(3-fluorophenyl)-6,7-dihydrobenzo[c]isoxazol-4(5H)-one

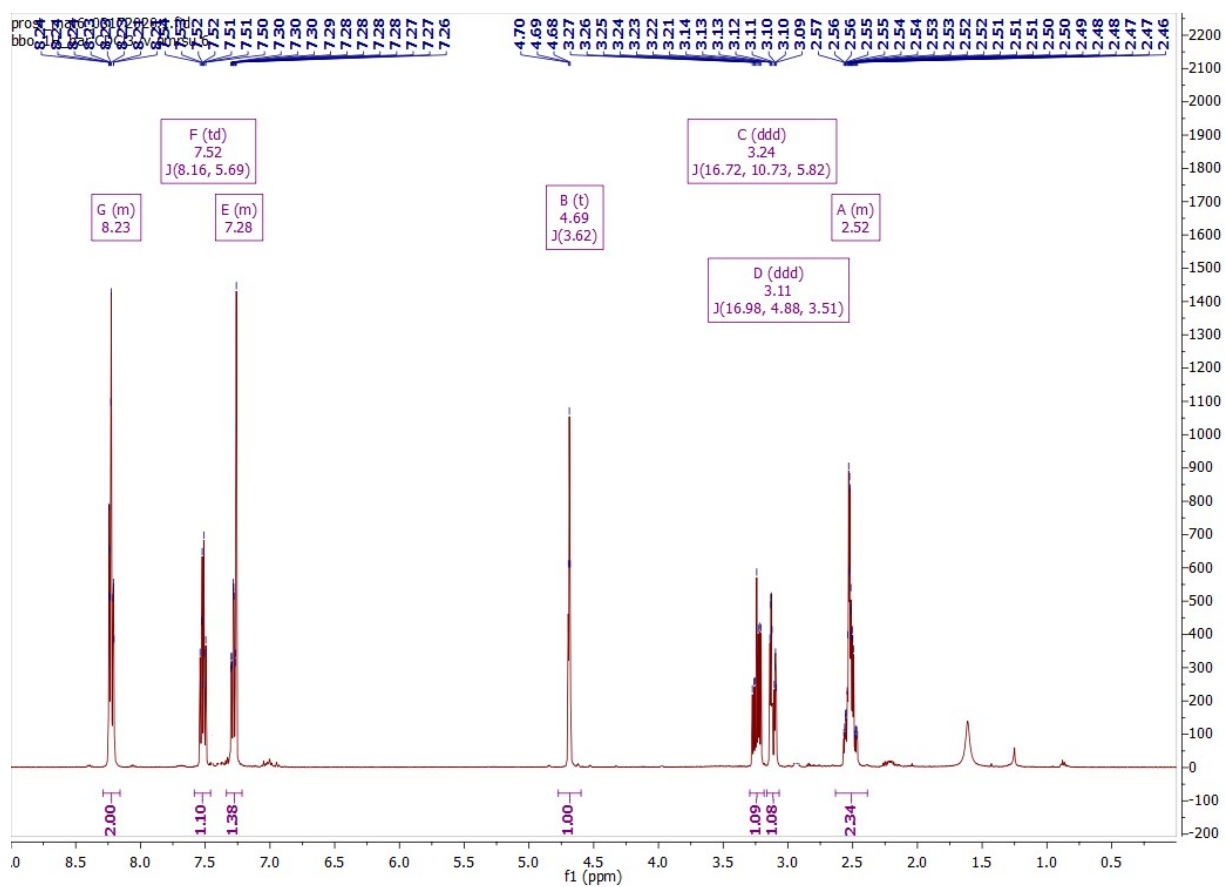
Chemical Formula: C₁₃H₉BrFNO₂

Exact Mass: 308,9801

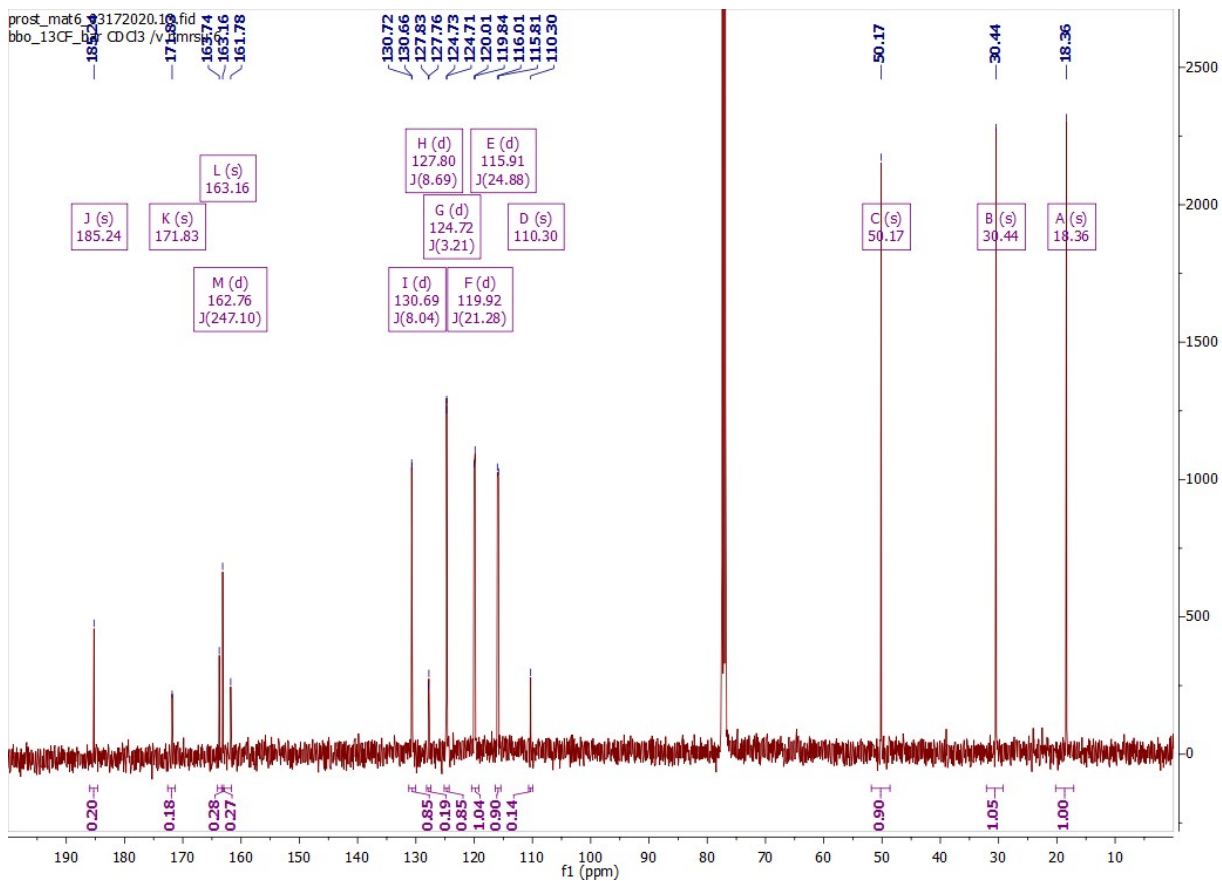
Molecular Weight: 310,1224

6b

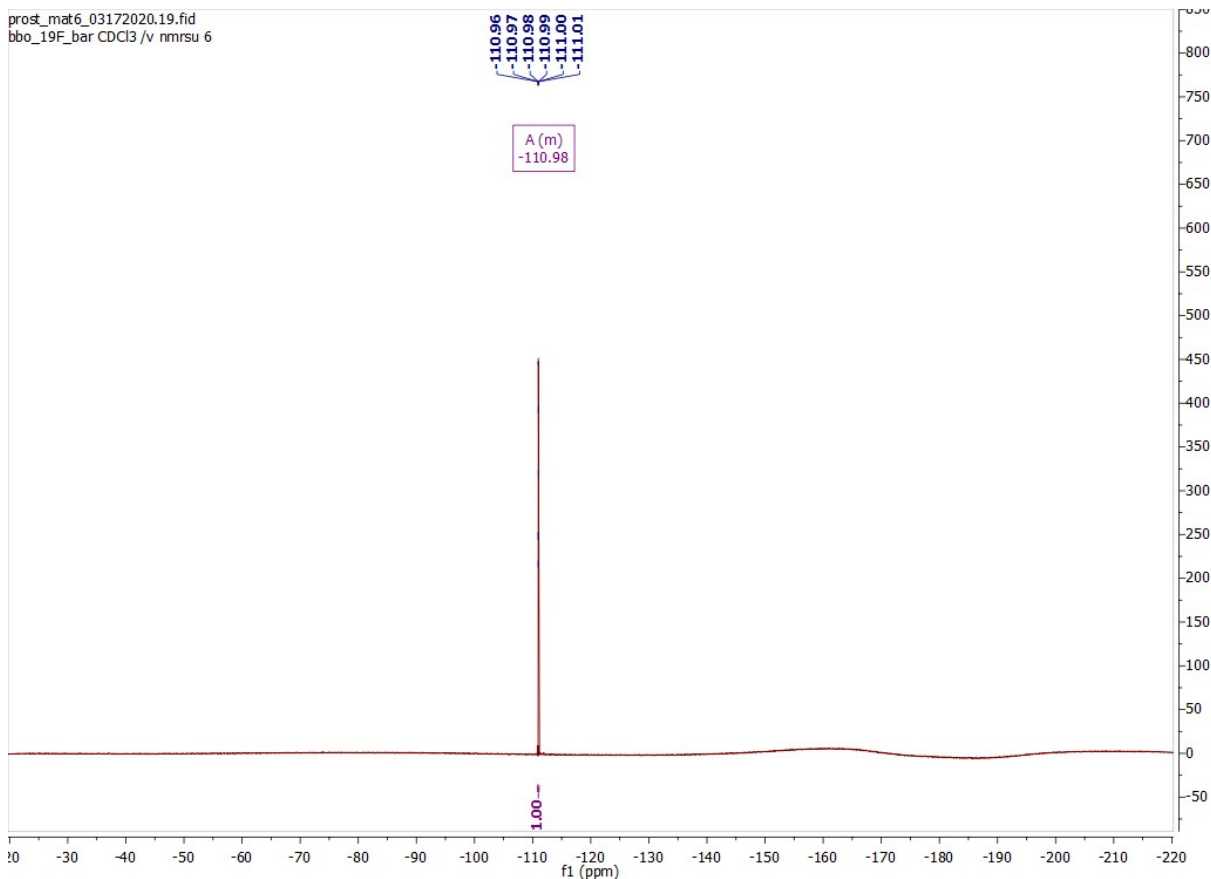
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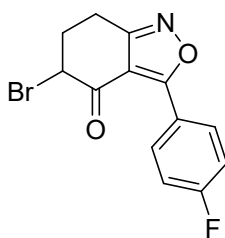


¹³C spectra of **6b**:



¹⁹F spectra of **6b**:





5-bromo-3-(4-fluorophenyl)-6,7-dihydrobenzo[c]isoxazol-4(5H)-one

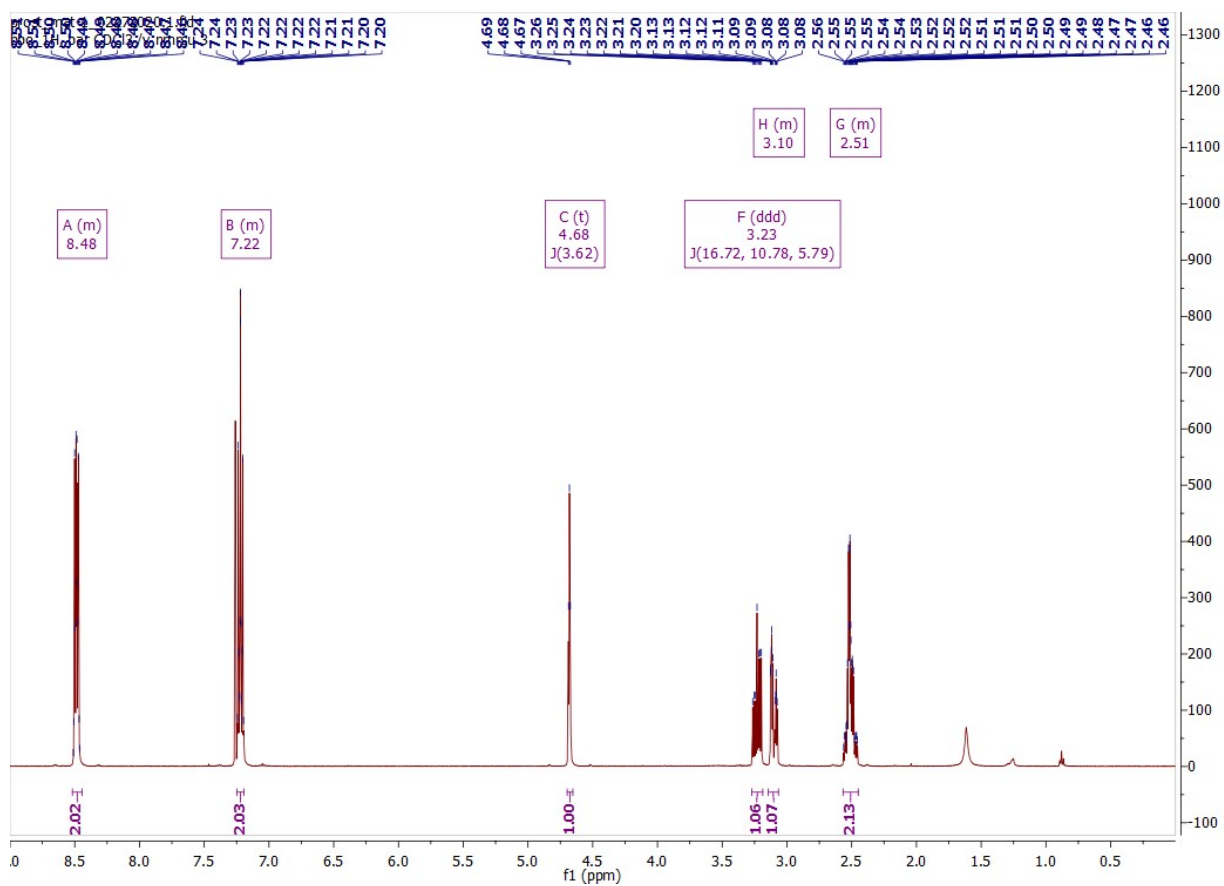
Chemical Formula: C₁₃H₉BrFNO₂

Exact Mass: 308,9801

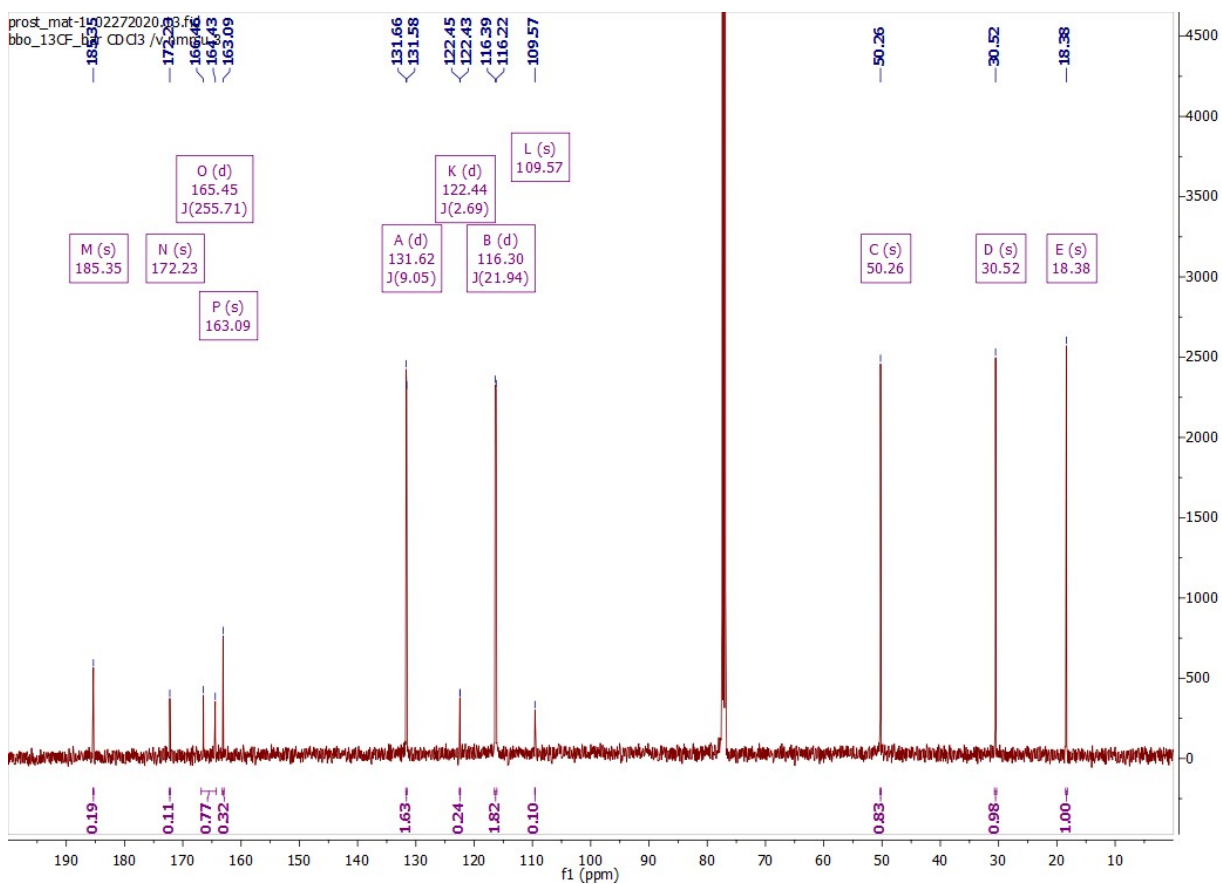
Molecular Weight: 310,1224

6c

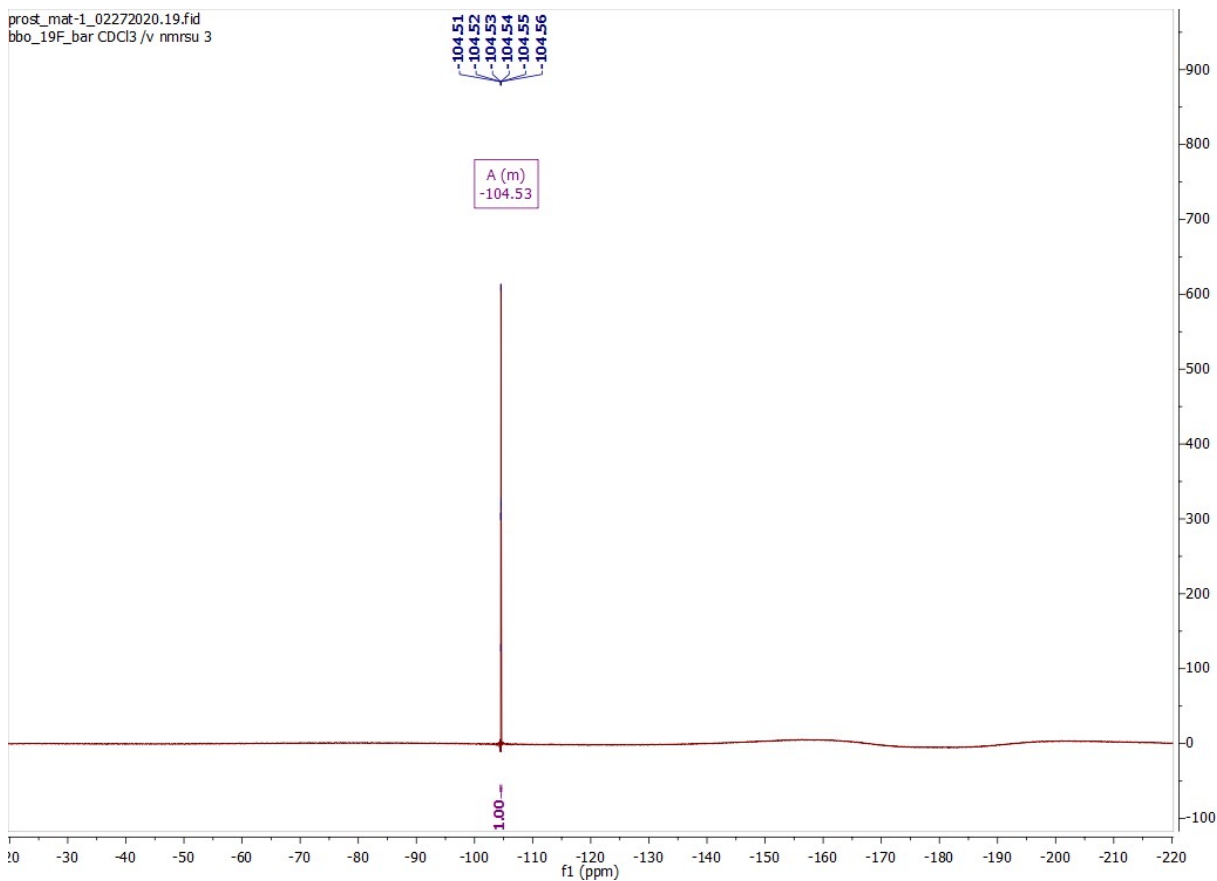
¹H spectra of **6c**:

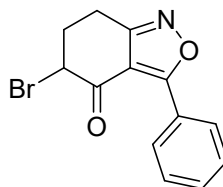


¹³C spectra of **6c**:



¹⁹F spectra of **6c**:





5-bromo-3-phenyl-6,7-dihydrobenzo[*c*]isoxazol-4(5*H*)-one

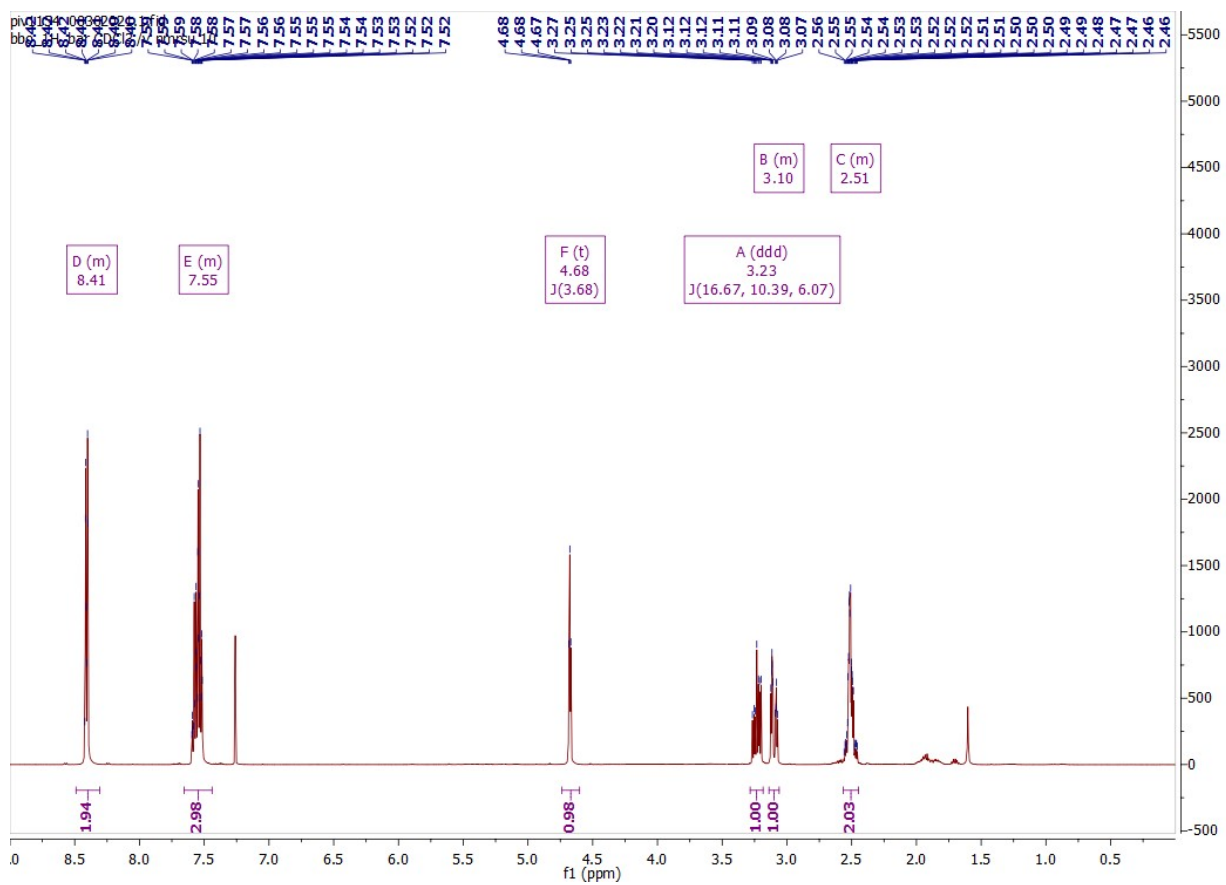
Chemical Formula: $C_{13}H_{10}BrNO_2$

Exact Mass: 290,9895

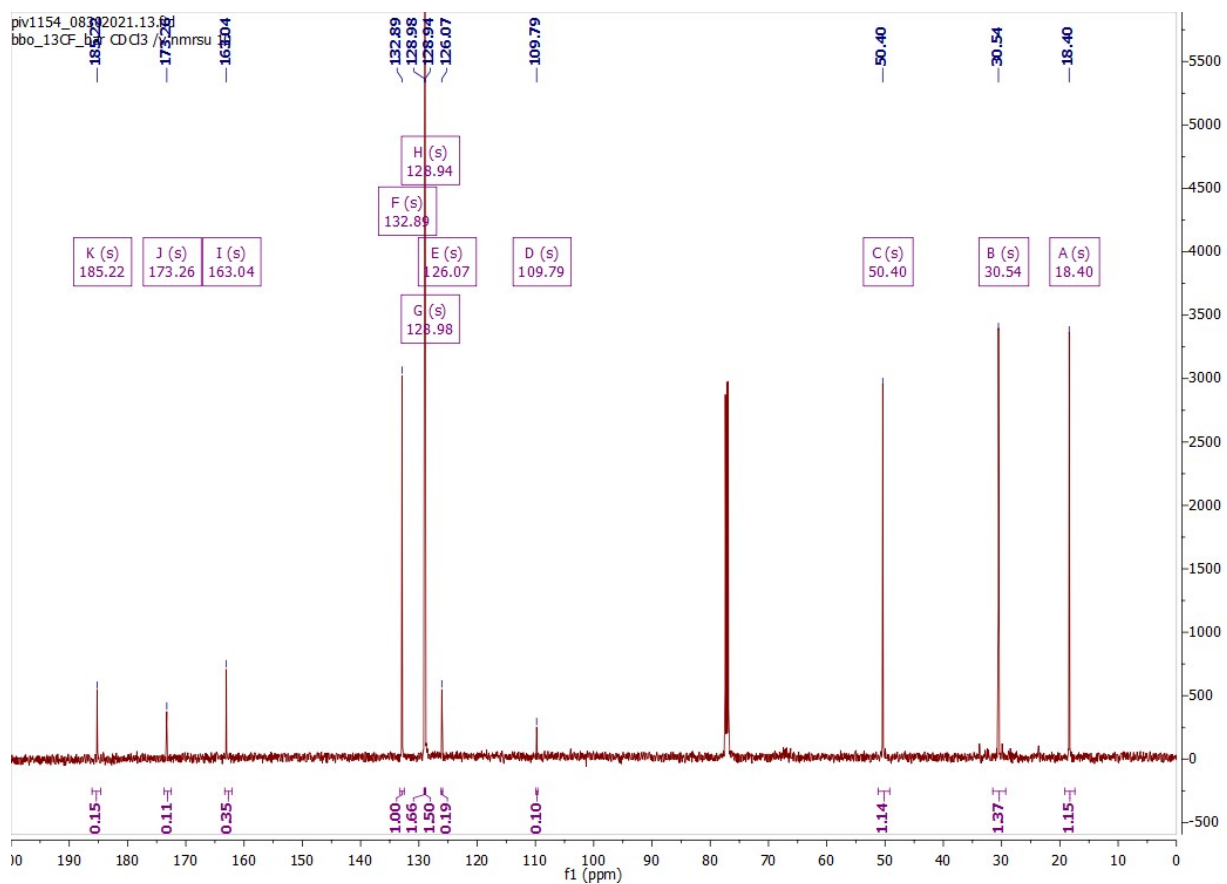
Molecular Weight: 292,1320

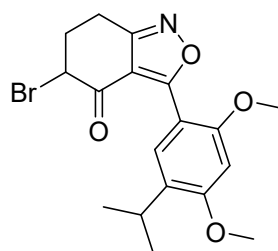
6d

1H spectra of **6d**:



^{13}C spectra of **6d**:





5-bromo-3-(5-isopropyl-2,4-dimethoxyphenyl)-6,7-dihydrobenzo[*c*]isoxazol-4(5*H*)-one

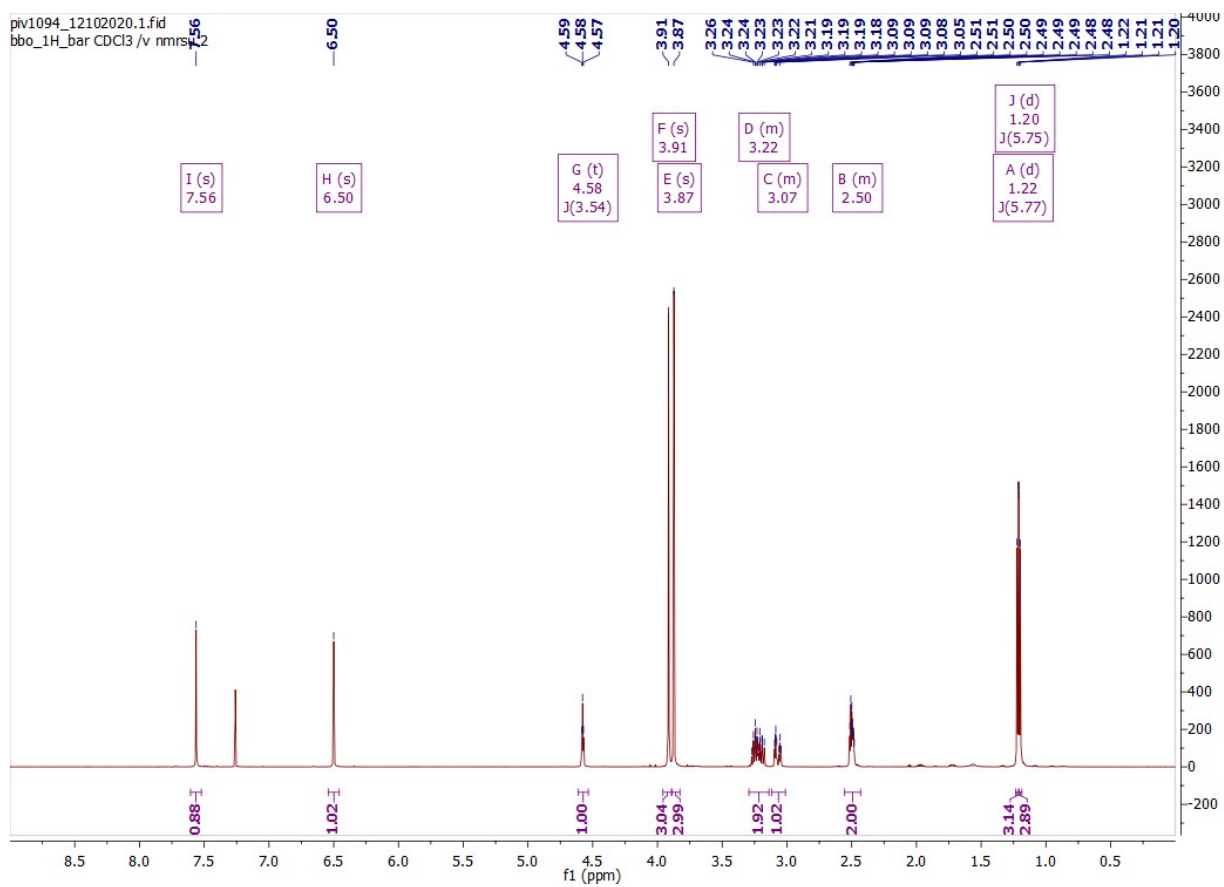
Chemical Formula: C₁₈H₂₀BrNO₄

Exact Mass: 393,0576

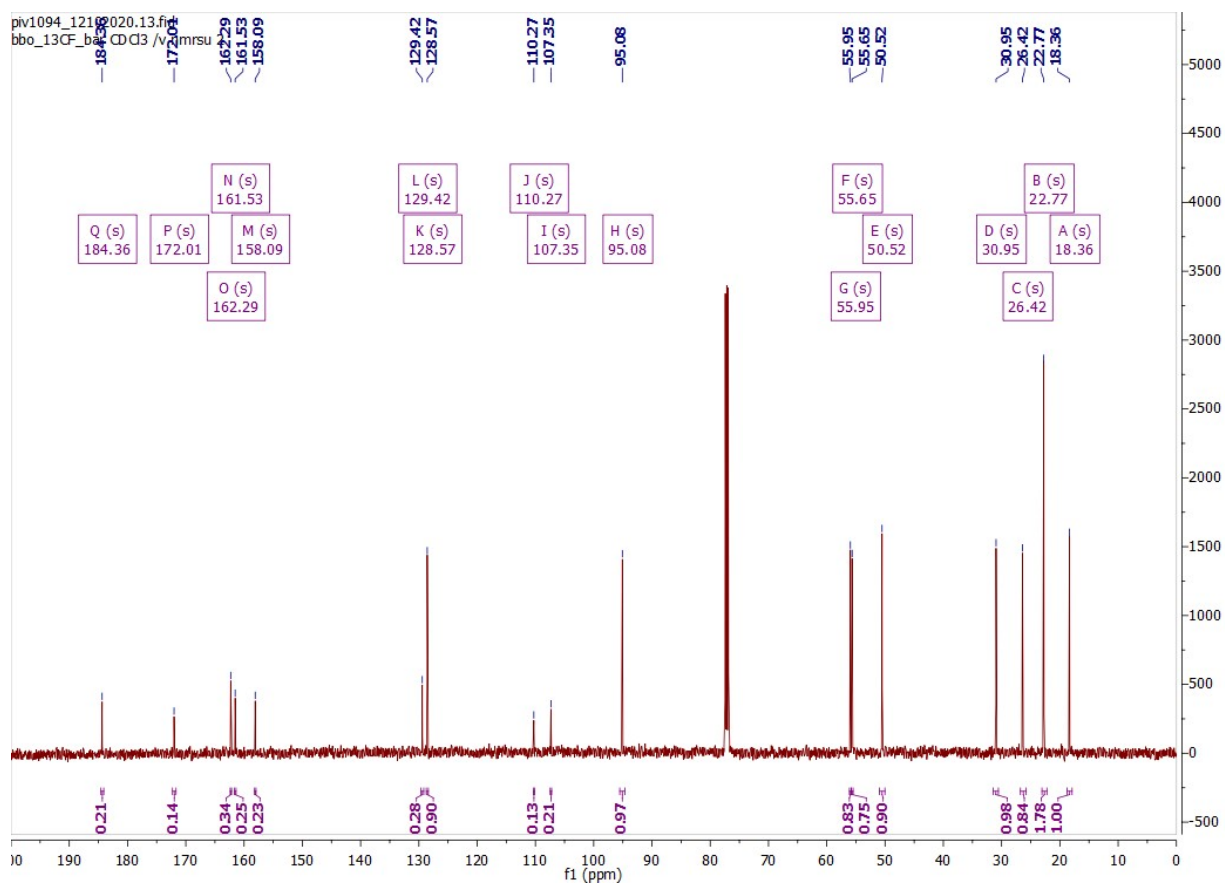
Molecular Weight: 394,2650

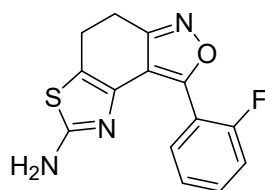
6e

¹H spectra of **6e**:



¹³C spectra of 6e:





8-(2-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-c]isoxazol-2-amine

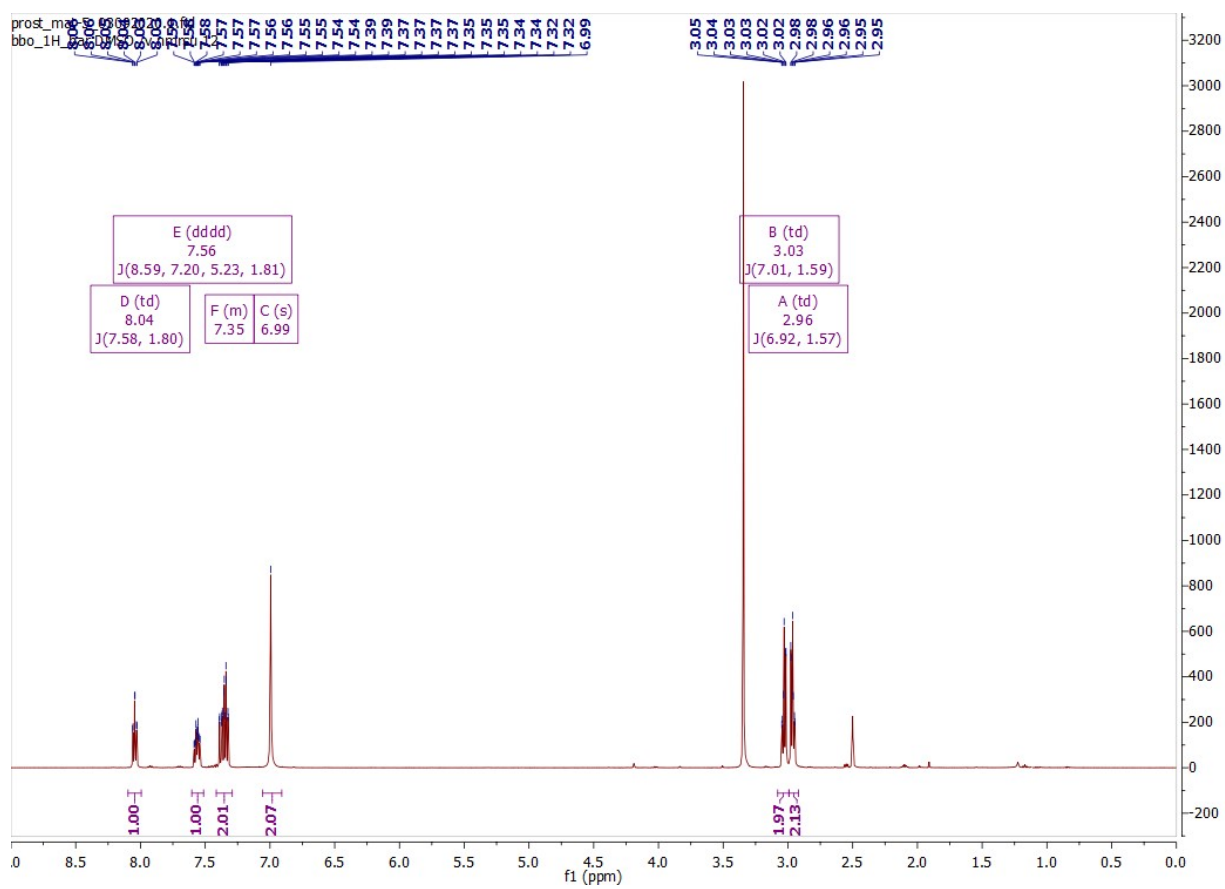
Chemical Formula: C₁₄H₁₀FN₃OS

Exact Mass: 287,0529

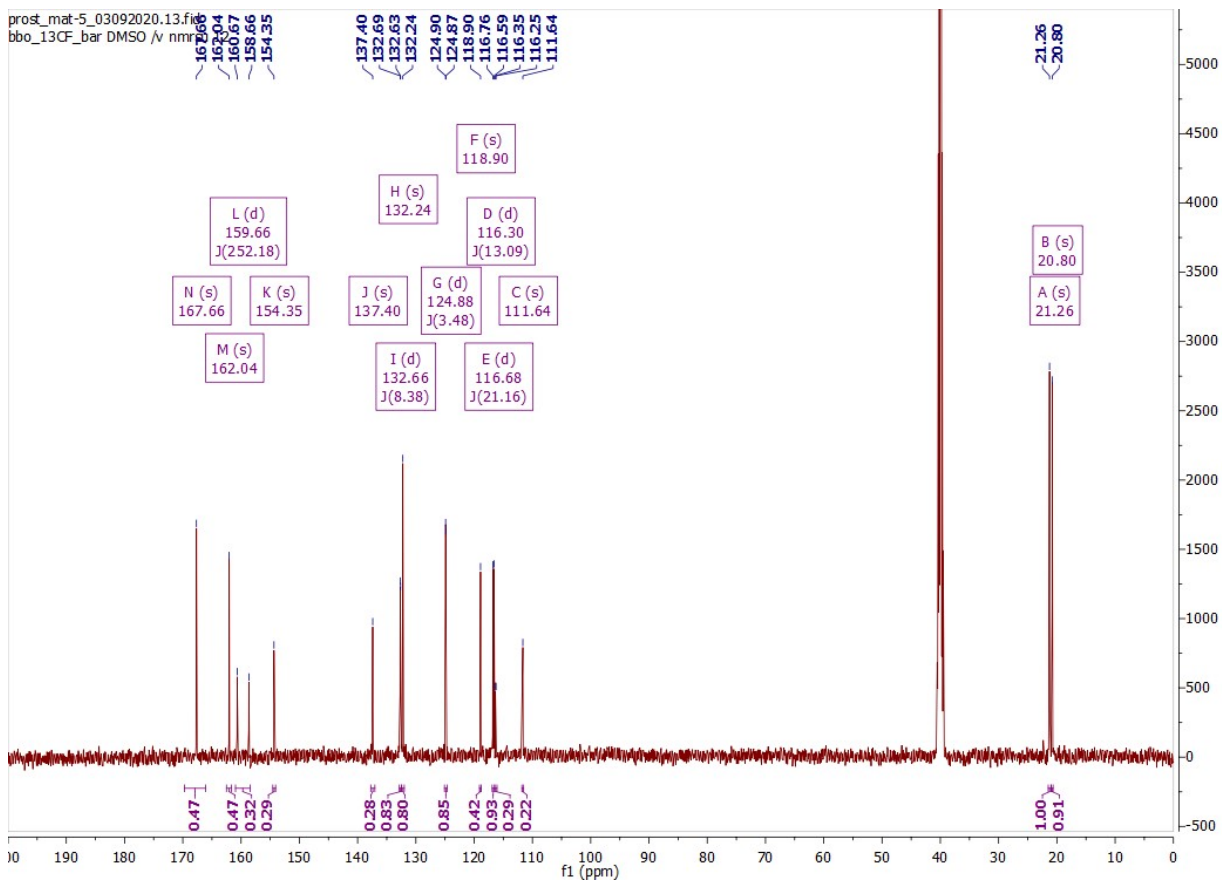
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7a

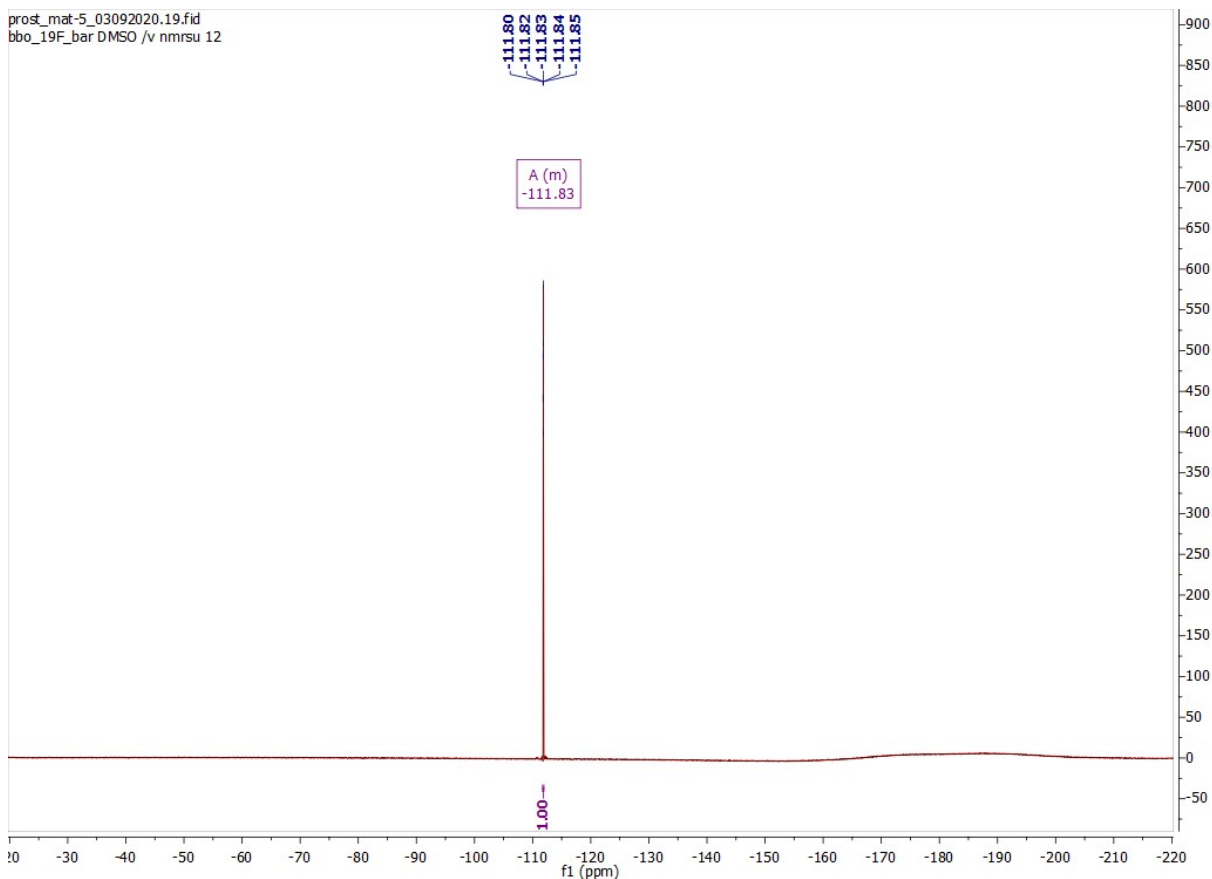
¹H spectra of **7a**:

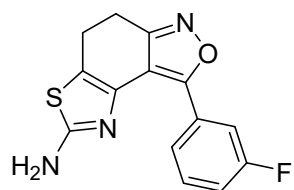


¹³C spectra of 7a:



¹⁹F spectra of 7a:





8-(3-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-c]isoxazol-2-amine

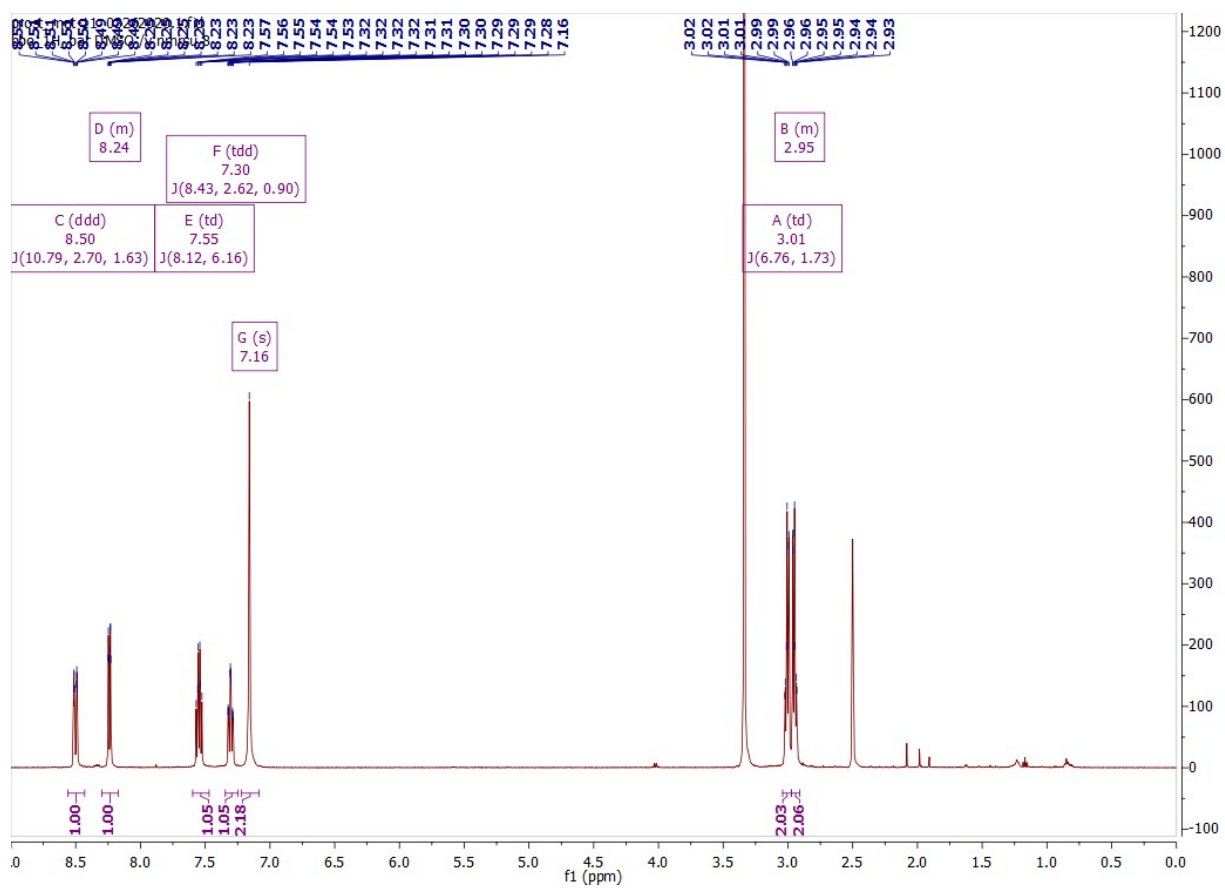
Chemical Formula: C₁₄H₁₀FN₃OS

Exact Mass: 287,0529

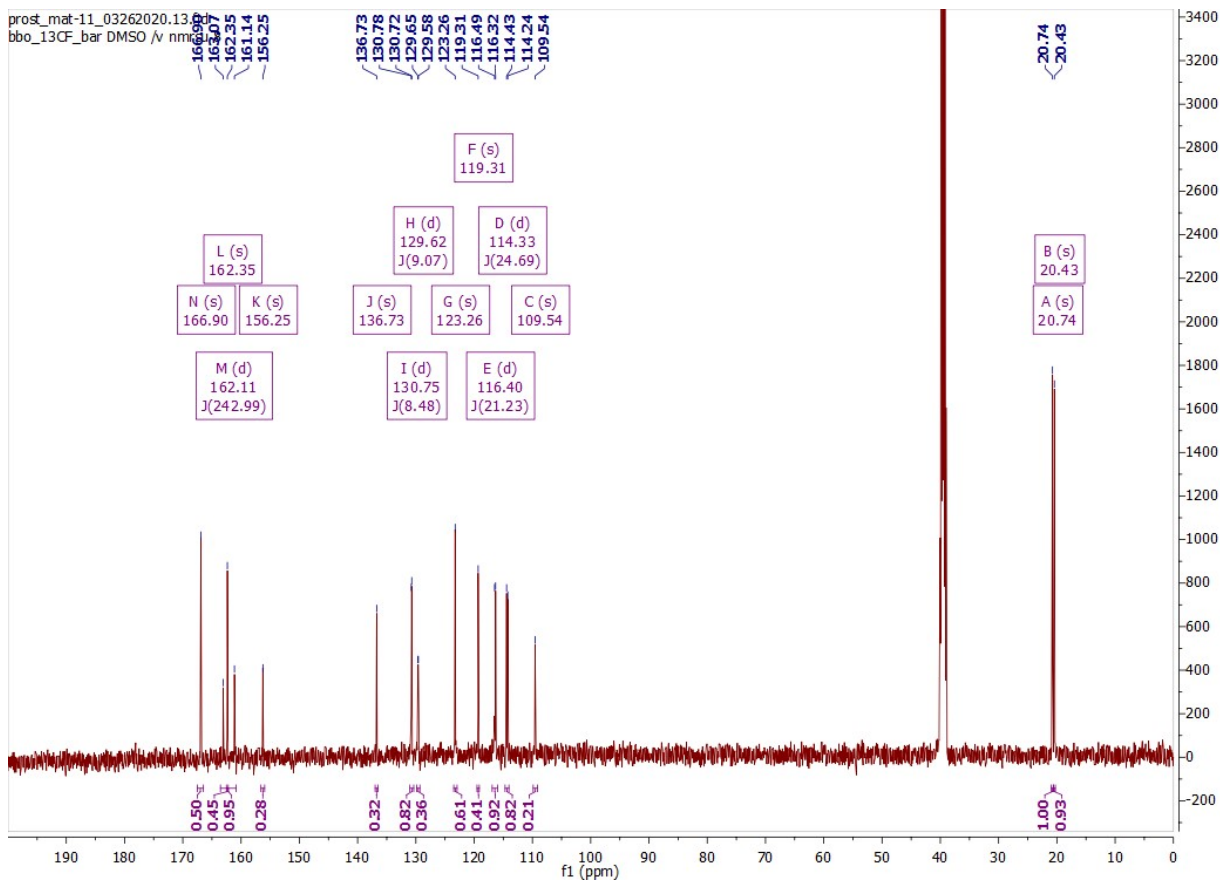
Molecular Weight: 287,3124

7b

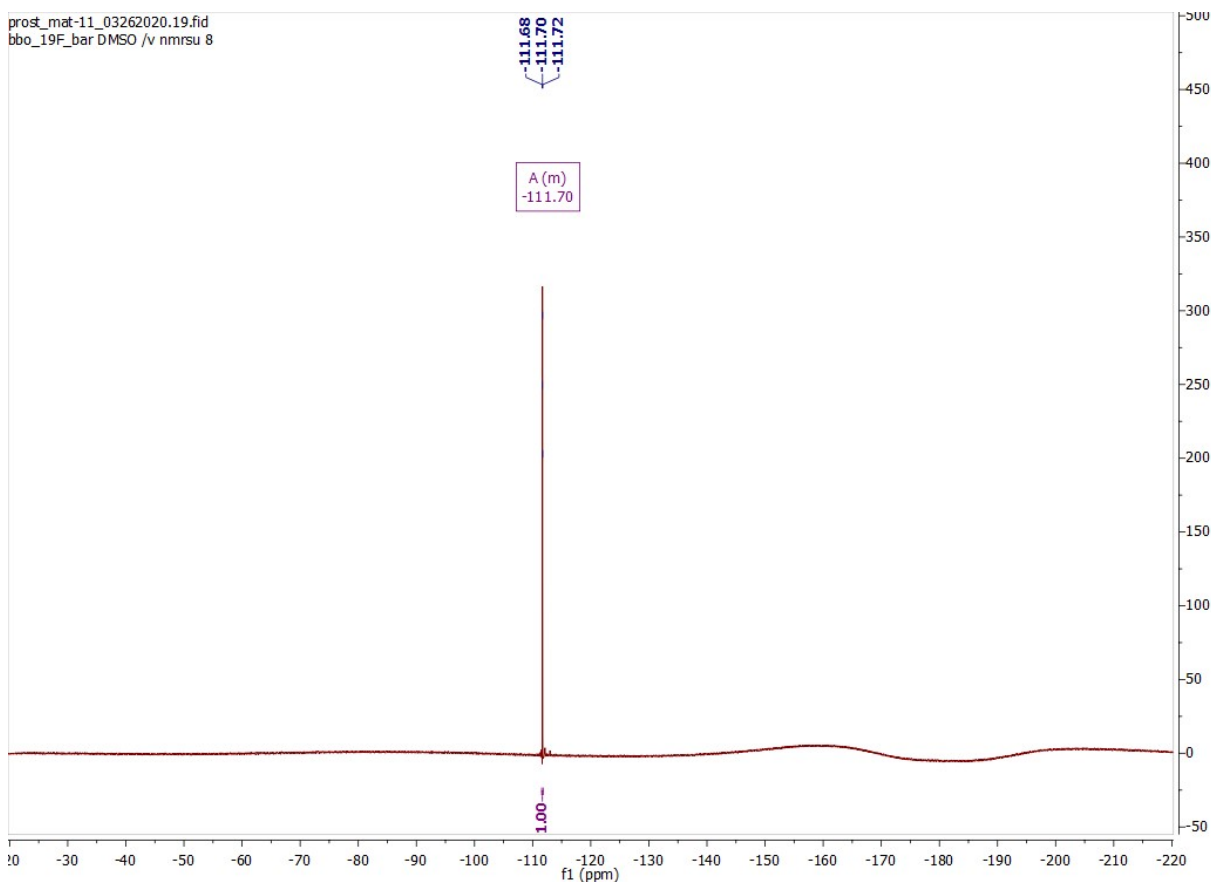
¹H spectra of **7b**:

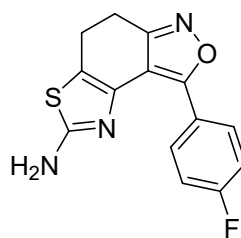


¹³C spectra of **7b**:



¹⁹F spectra of **7b**:





8-(4-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-c]isoxazol-2-amine

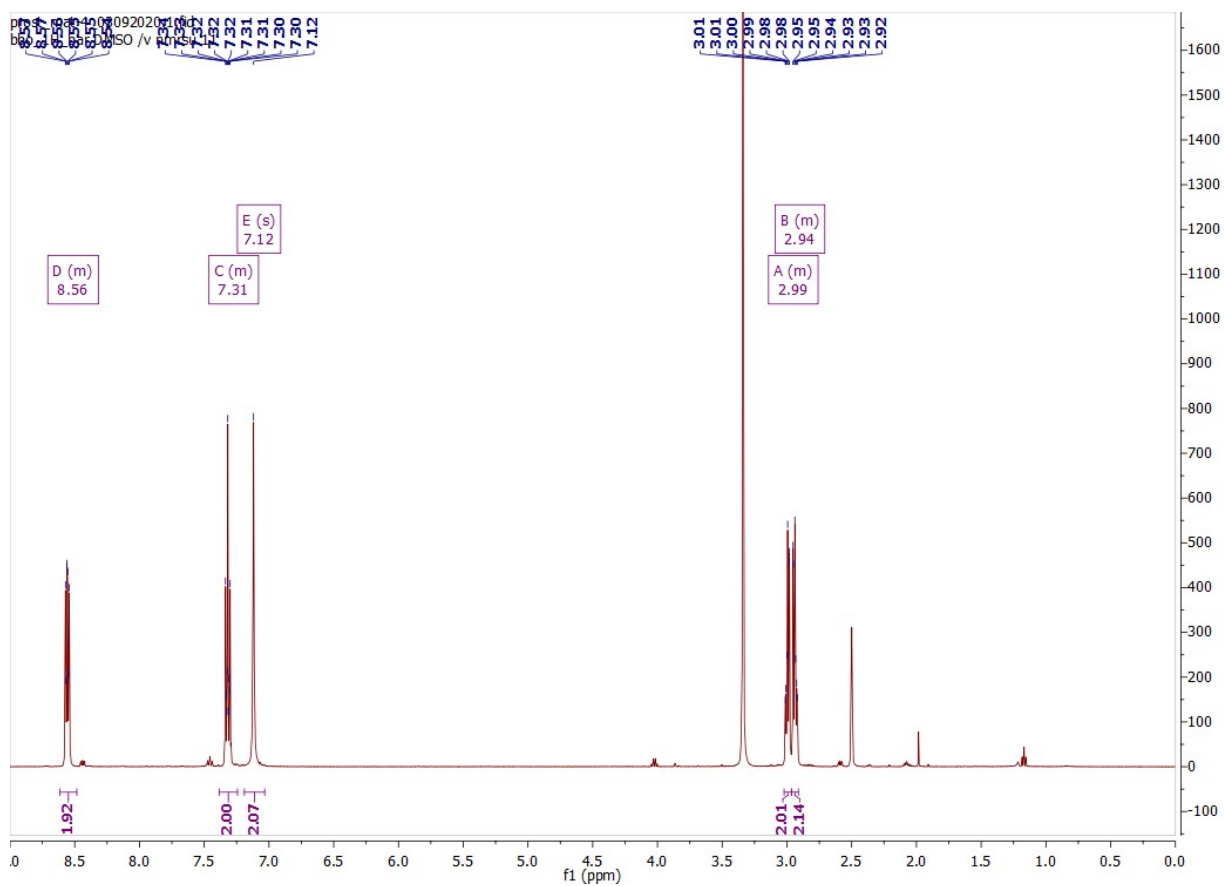
Chemical Formula: C₁₄H₁₀FN₃OS

Exact Mass: 287,0529

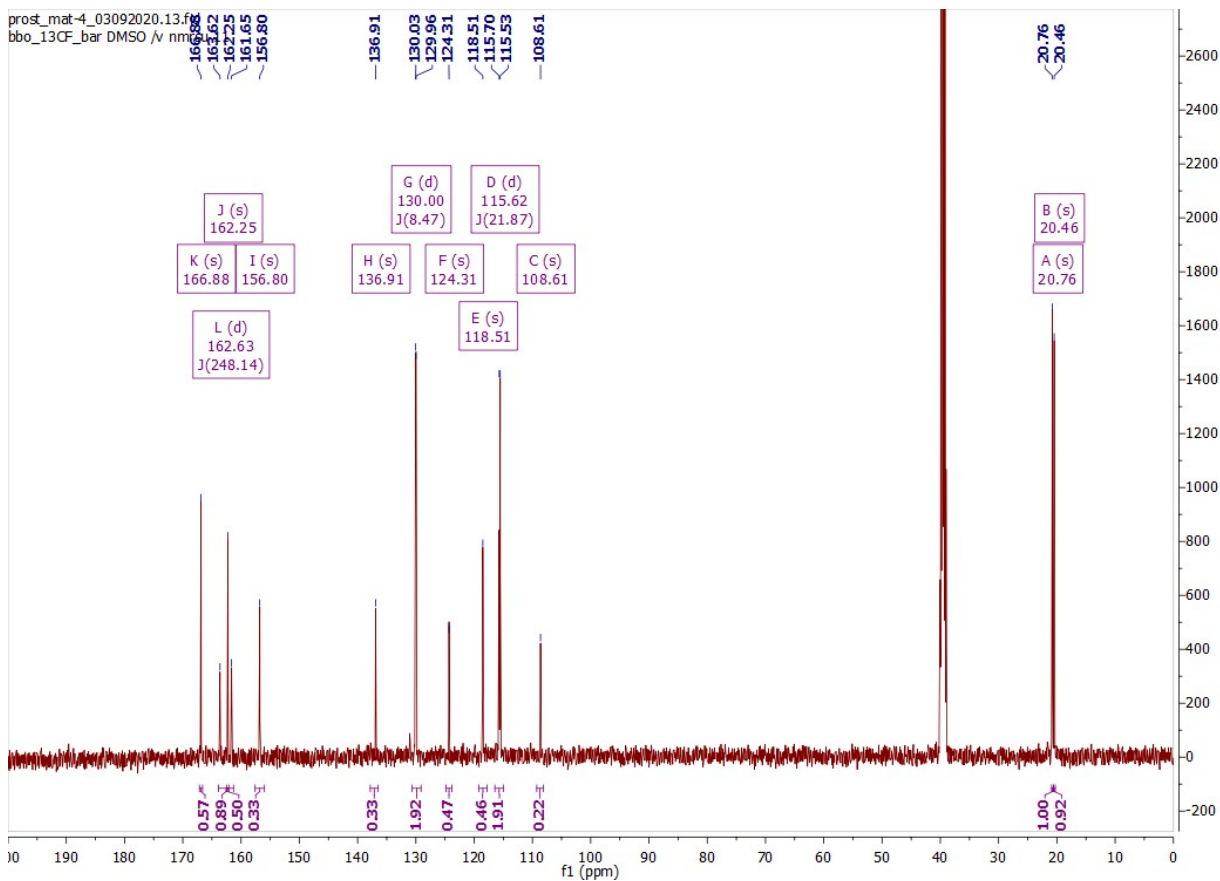
Molecular Weight: 287,3124

7c

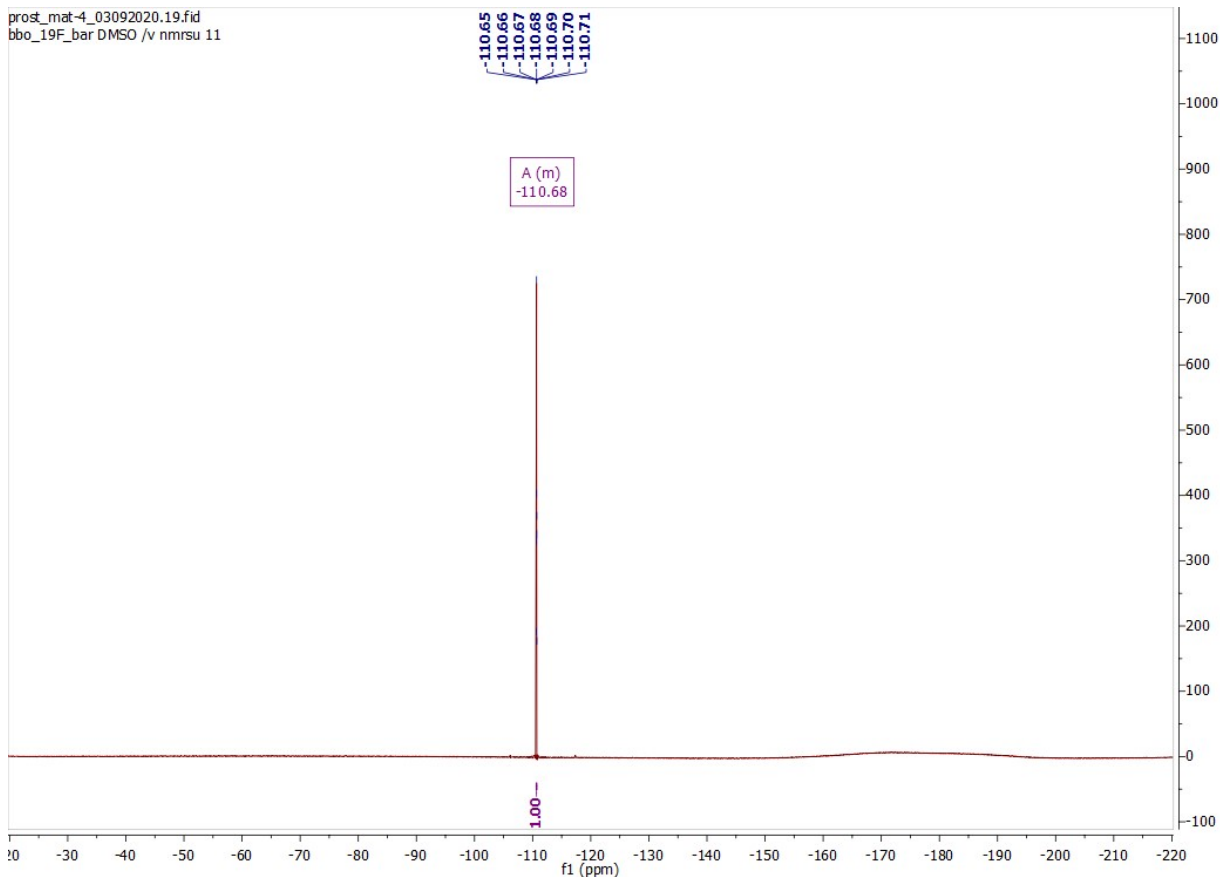
¹H spectra of 7c:

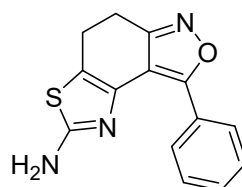


¹³C spectra of 7c:



¹⁹F spectra of 7c:





8-phenyl-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-c]isoxazol-2-amine

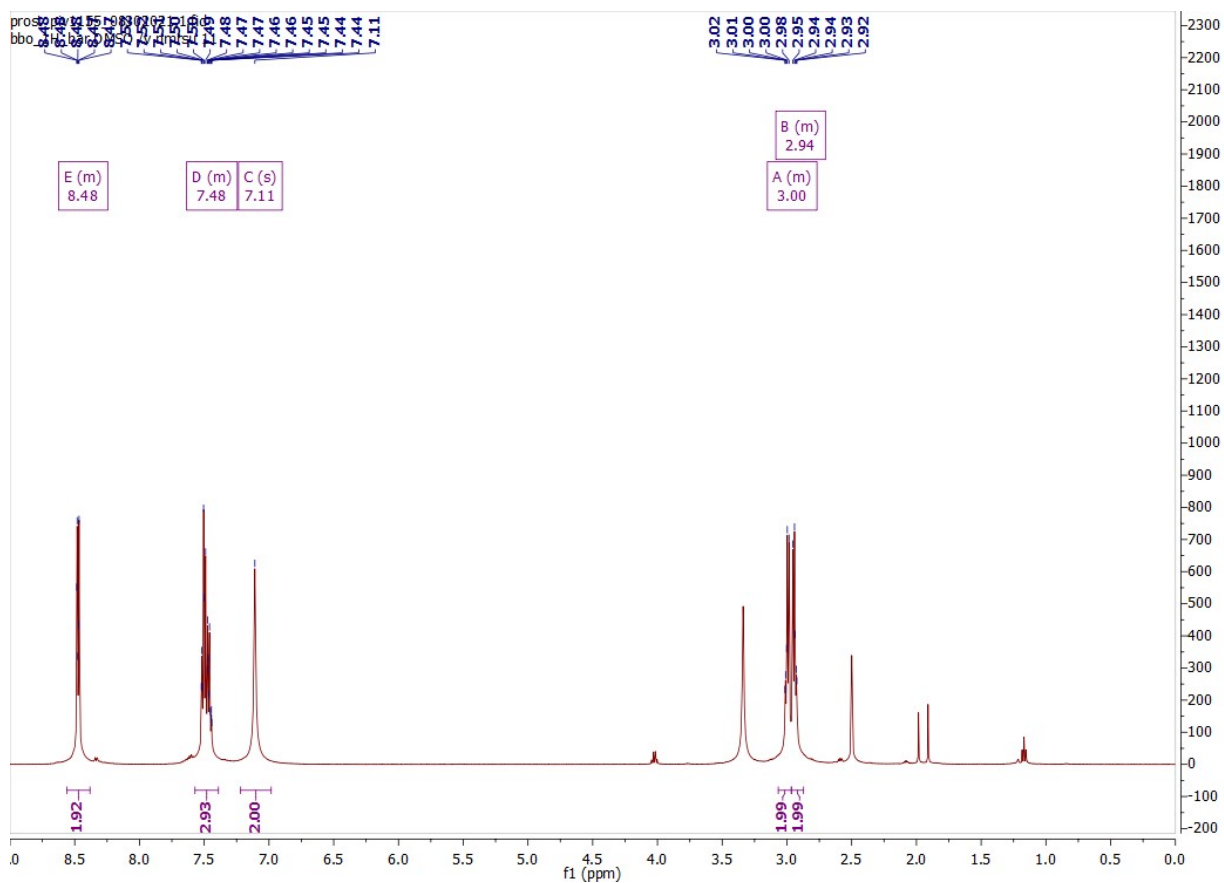
Chemical Formula: C₁₄H₁₁N₃OS

Exact Mass: 269,0623

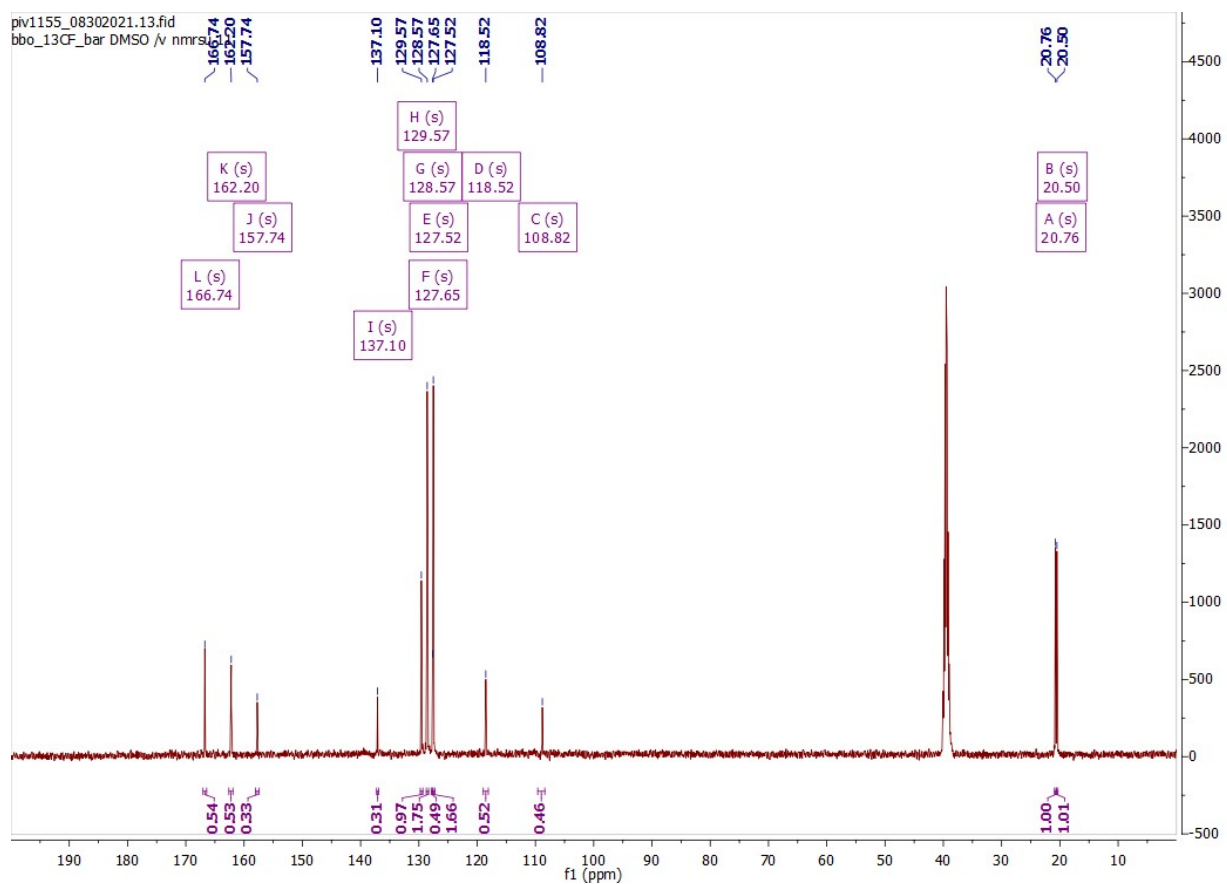
Molecular Weight: 269,3220

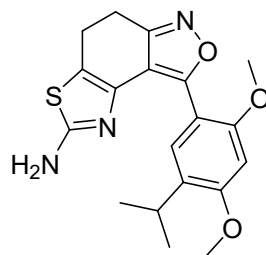
7d

¹H spectra of **7d**:



¹³C spectra of 7d:





8-(5-isopropyl-2,4-dimethoxyphenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-c]isoxazol-2-amine

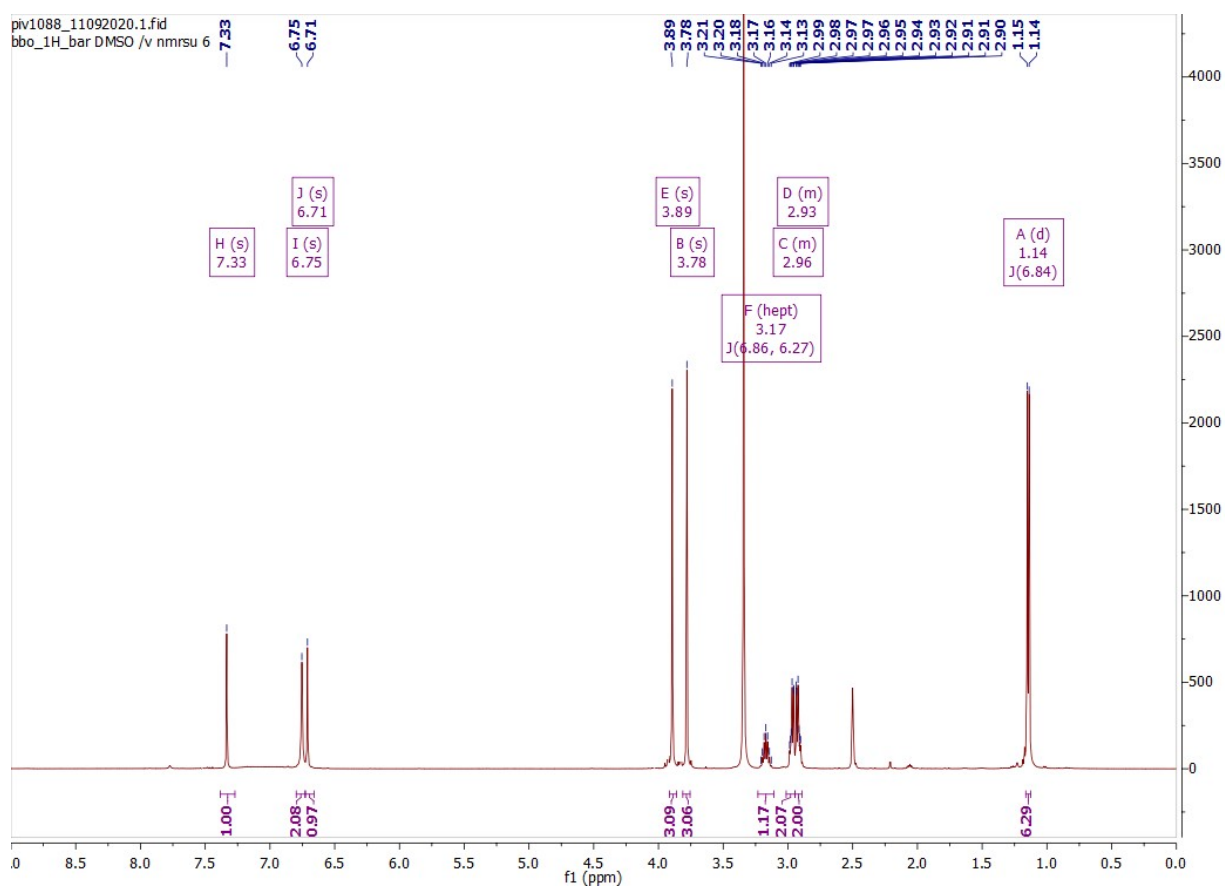
Chemical Formula: C₁₉H₂₁N₃O₃S

Exact Mass: 371,1304

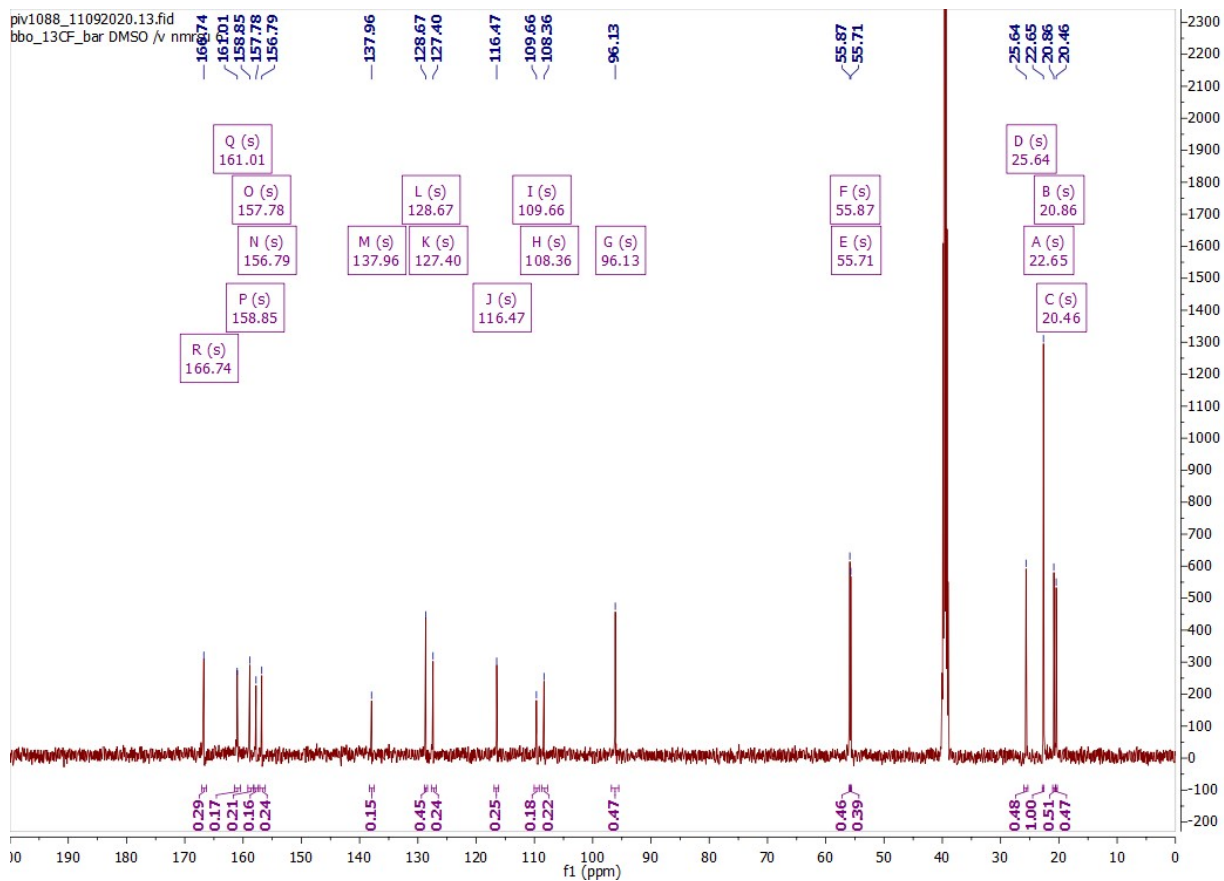
Molecular Weight: 371,4550

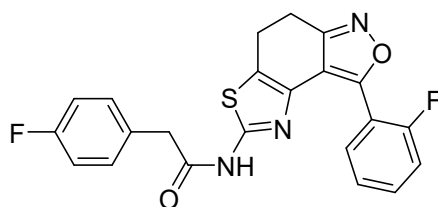
7e

¹H spectra of 7d:



¹³C spectra of 7e:





2-(4-fluorophenyl)-N-(8-(2-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-c]isoxazol-2-yl)acetamide

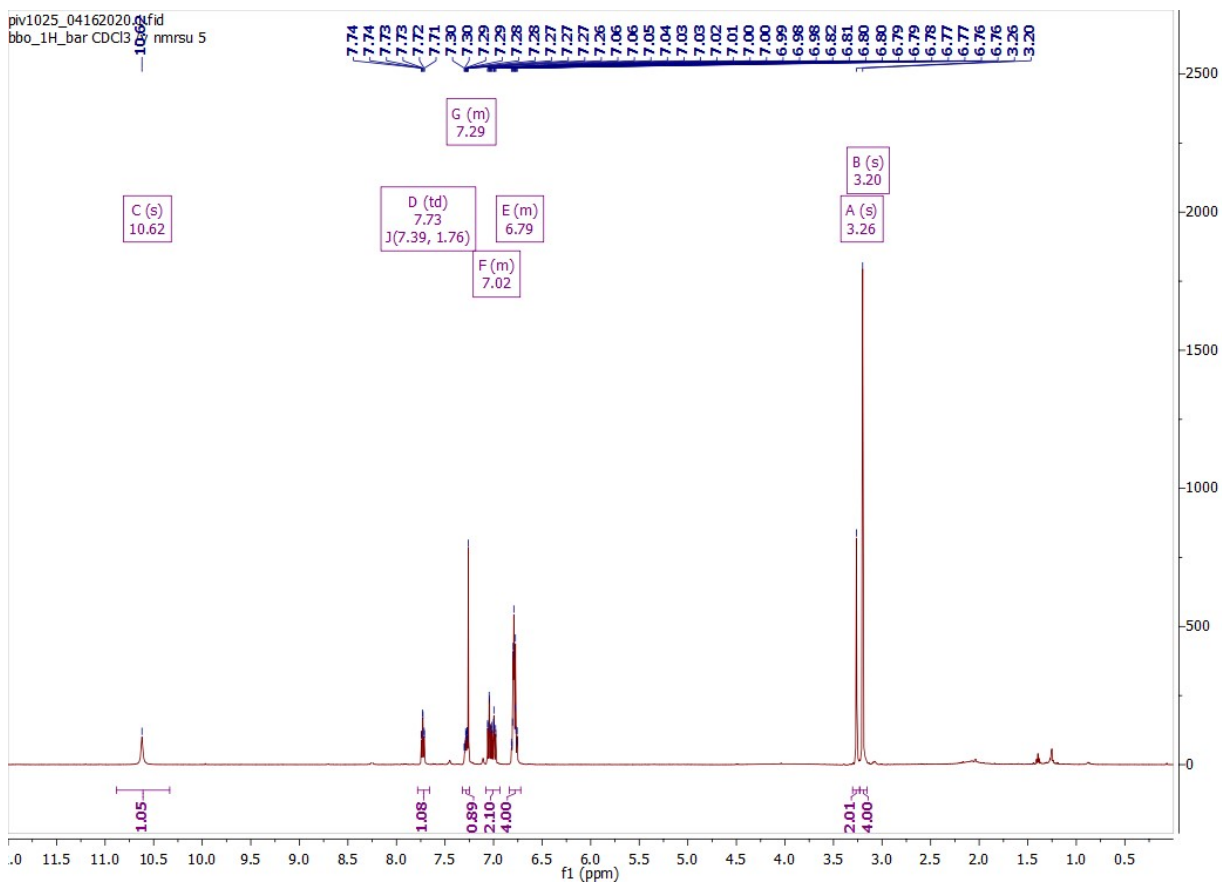
Chemical Formula: $C_{22}H_{15}F_2N_3O_2S$

Exact Mass: 423,0853

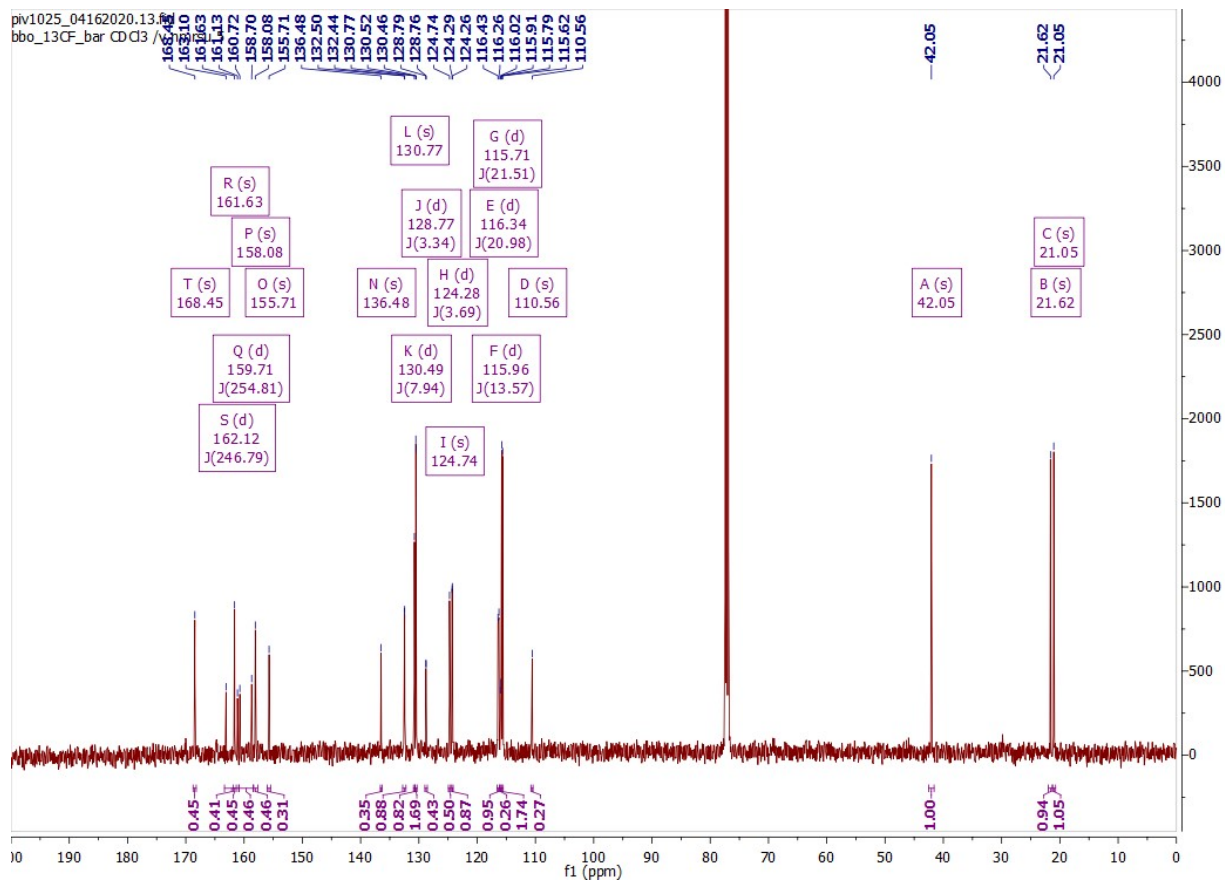
Molecular Weight: 423,4378

8a

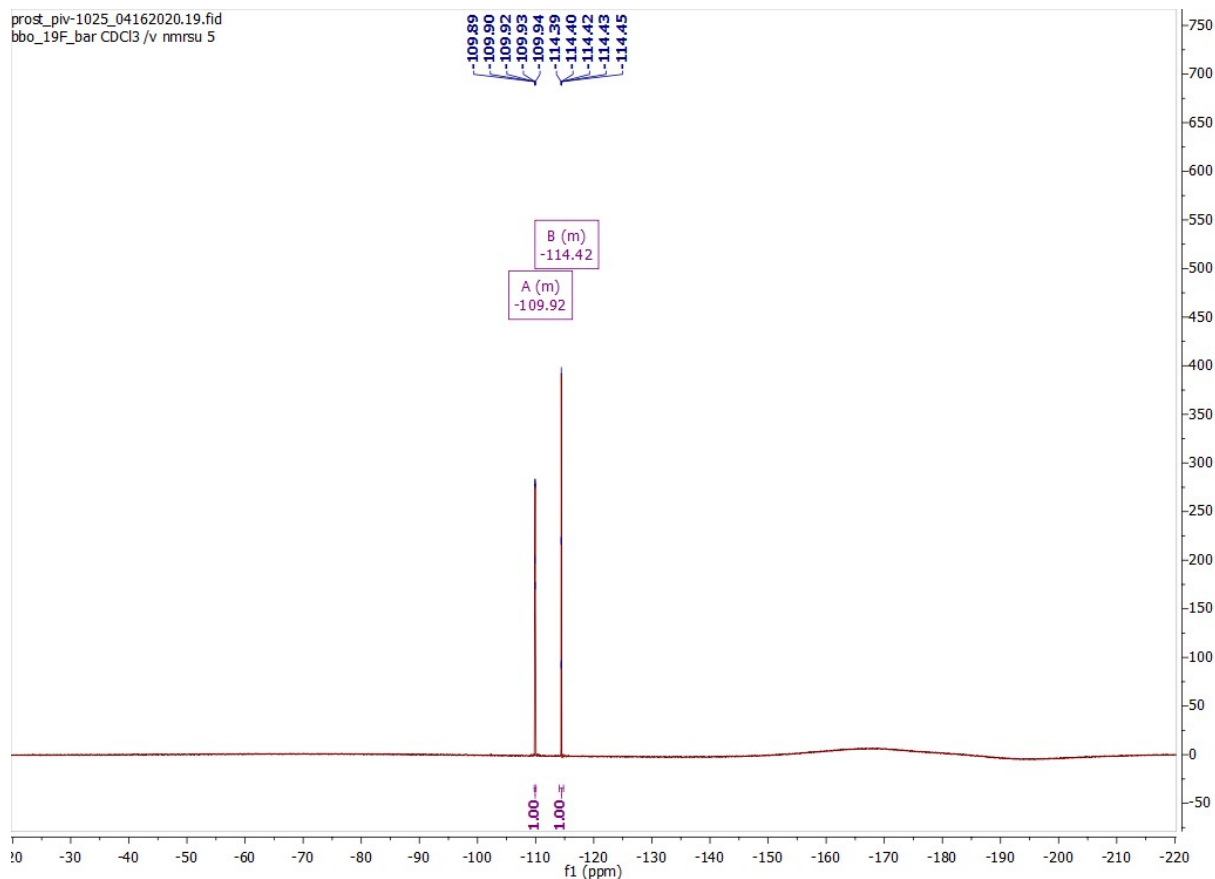
1H spectra of **8a**:



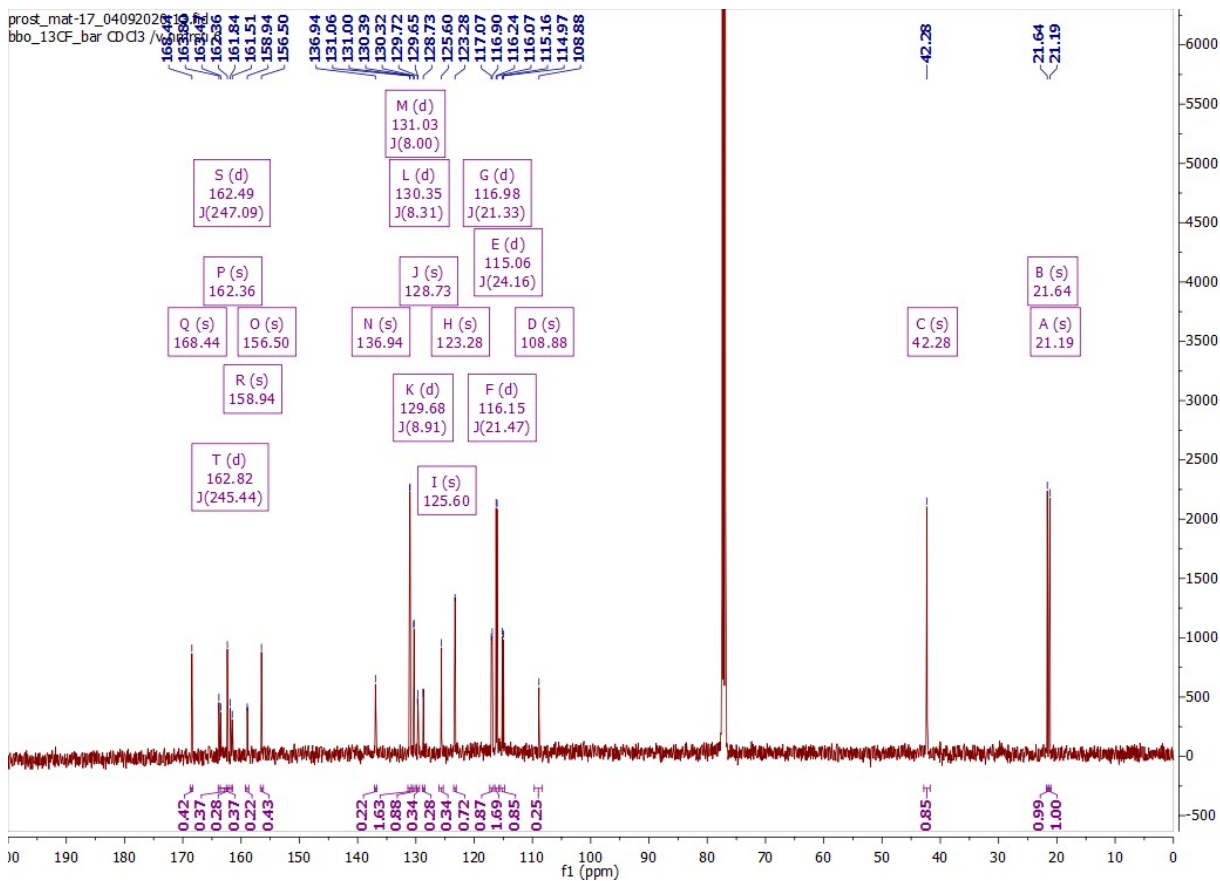
¹³C spectra of **8a**:



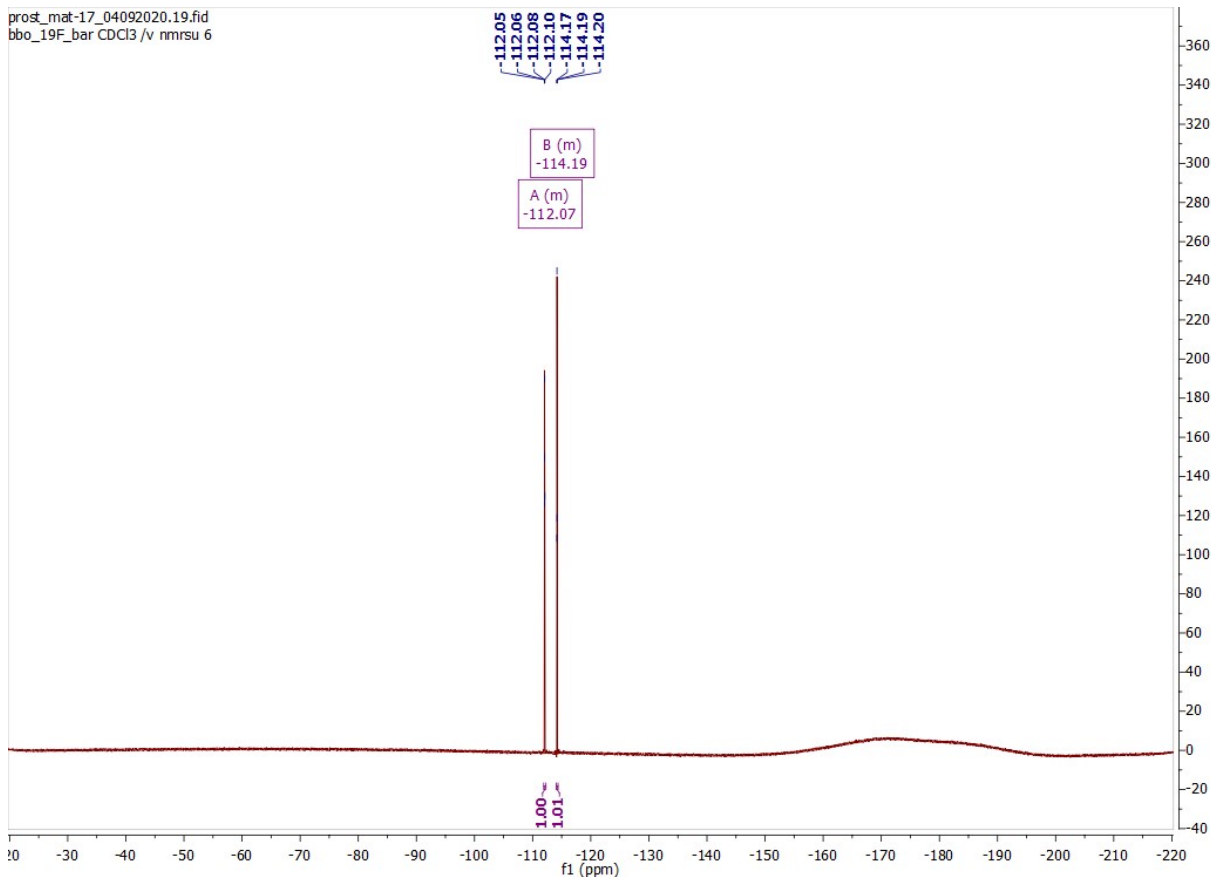
¹⁹F spectra of **8a**:

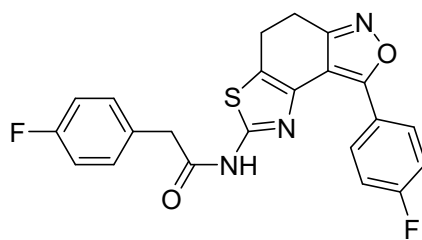


¹³C spectra of **8b**:



¹⁹F spectra of **8b**:





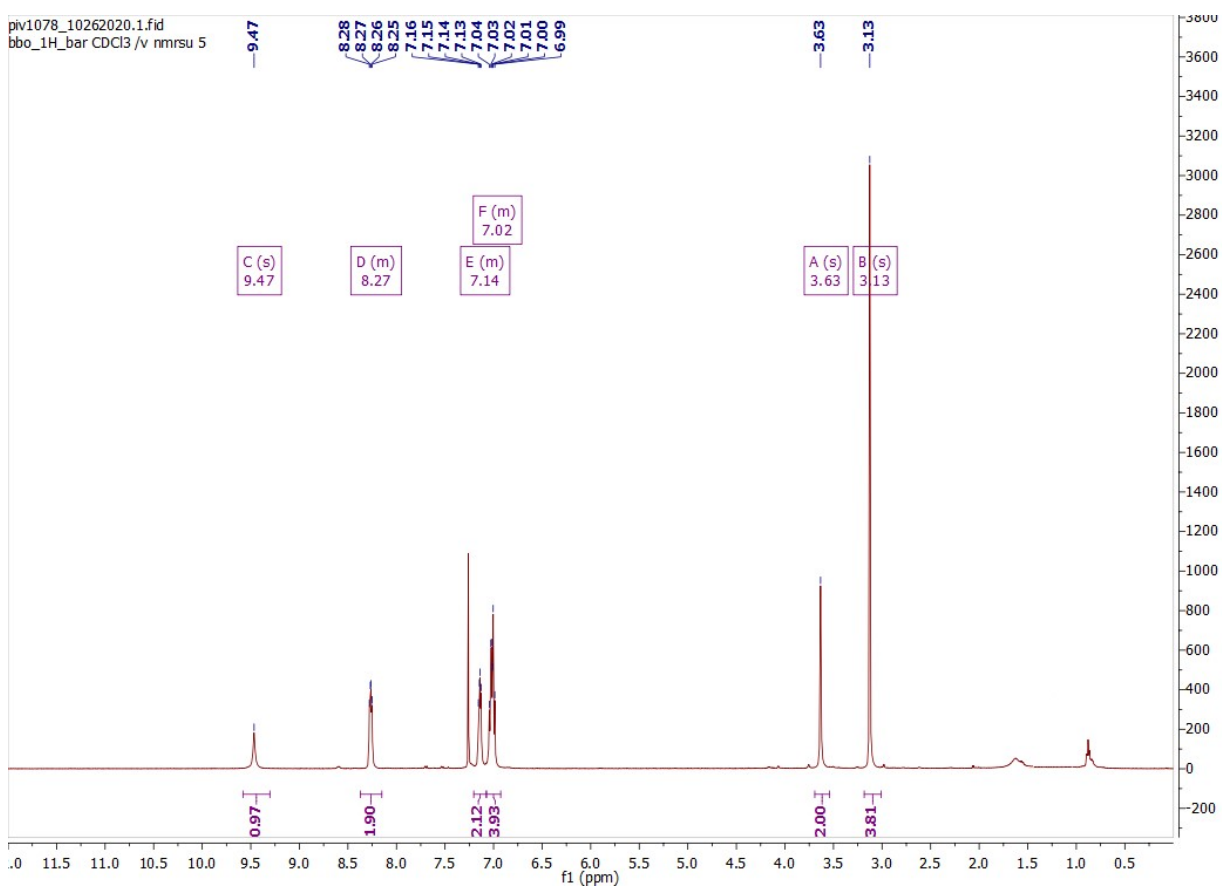
2-(4-fluorophenyl)-N-(8-(4-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-c]isoxazol-2-yl)acetamide

Chemical Formula: C₂₂H₁₅F₂N₃O₂S

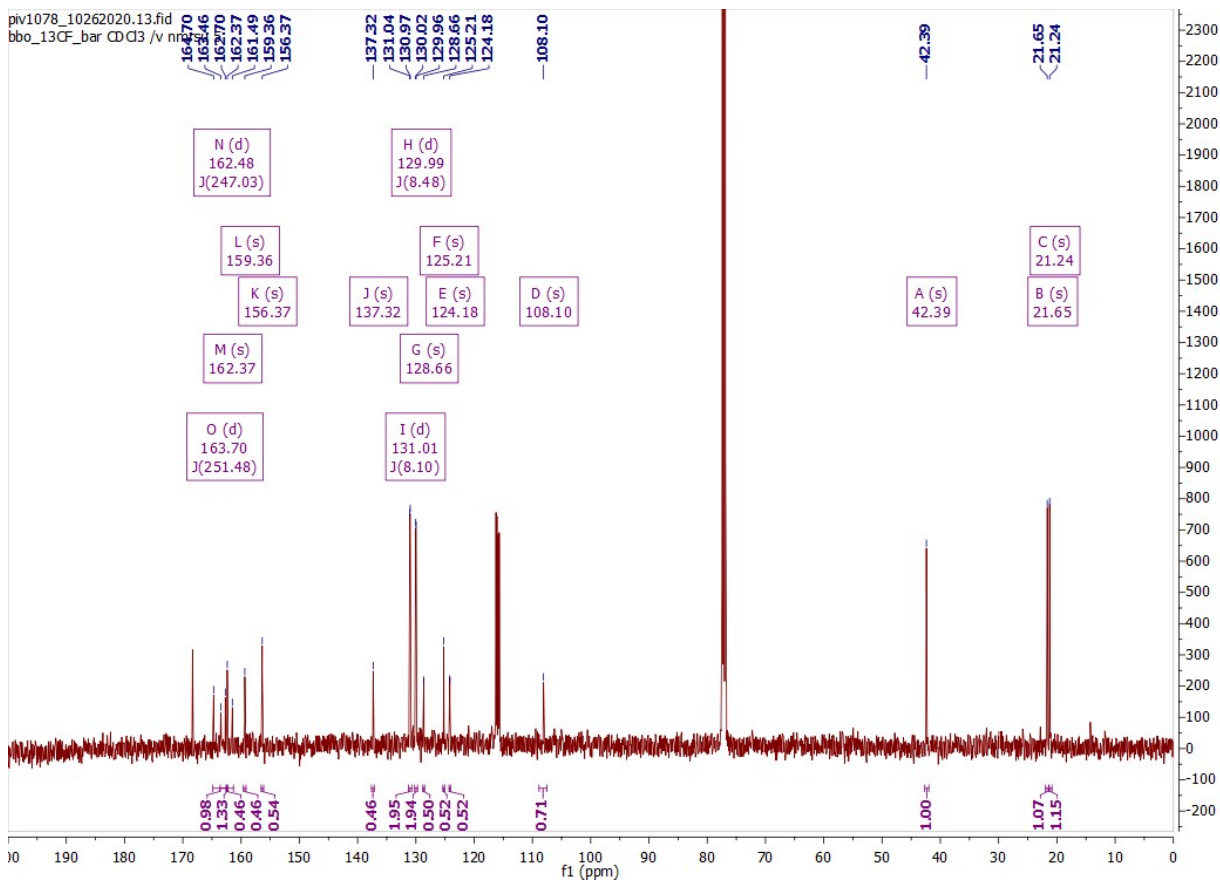
Molecular Weight: 423.44

8c

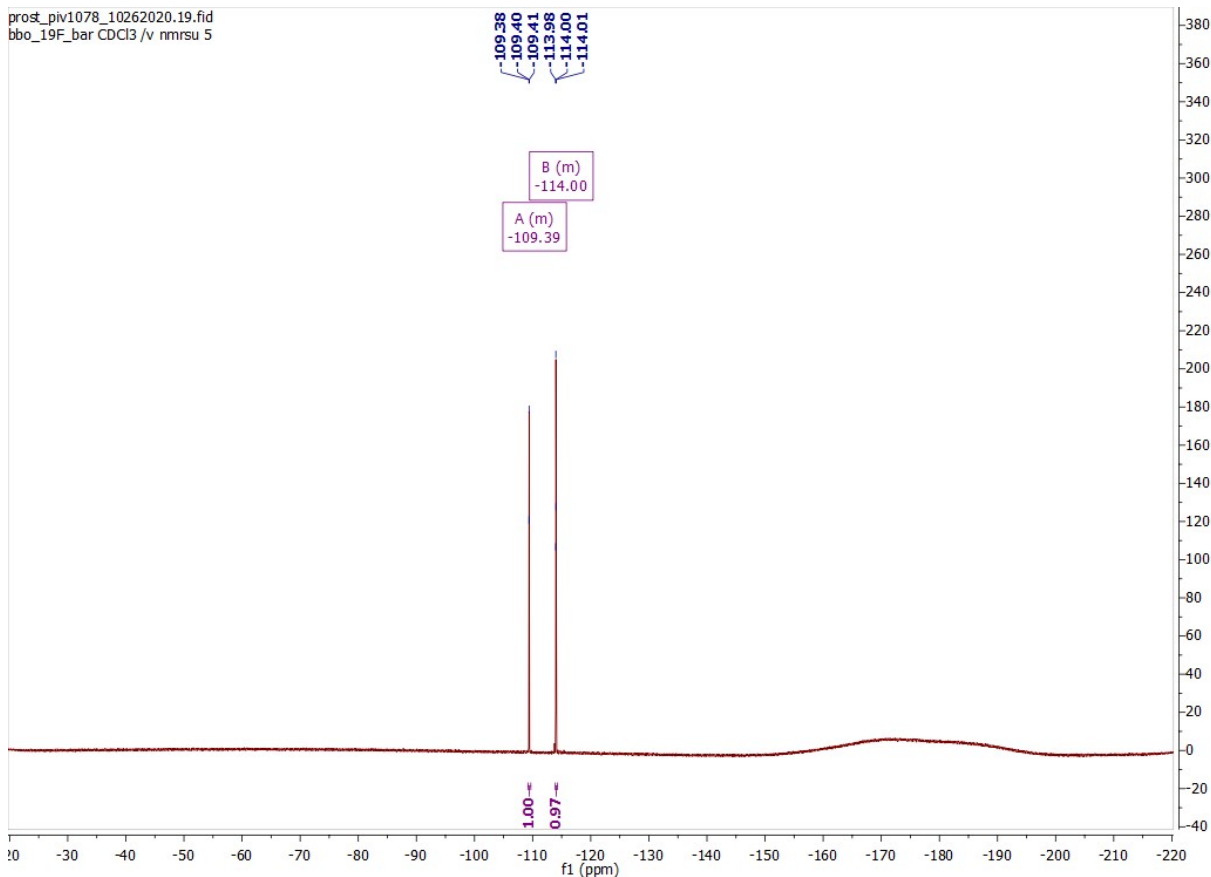
¹H spectra of **8c**:

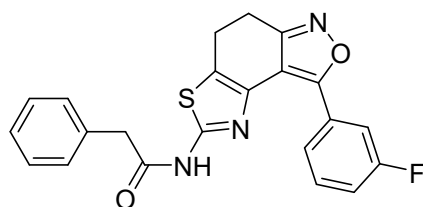


¹³C spectra of **8c**:



¹⁹F spectra of **8c**:

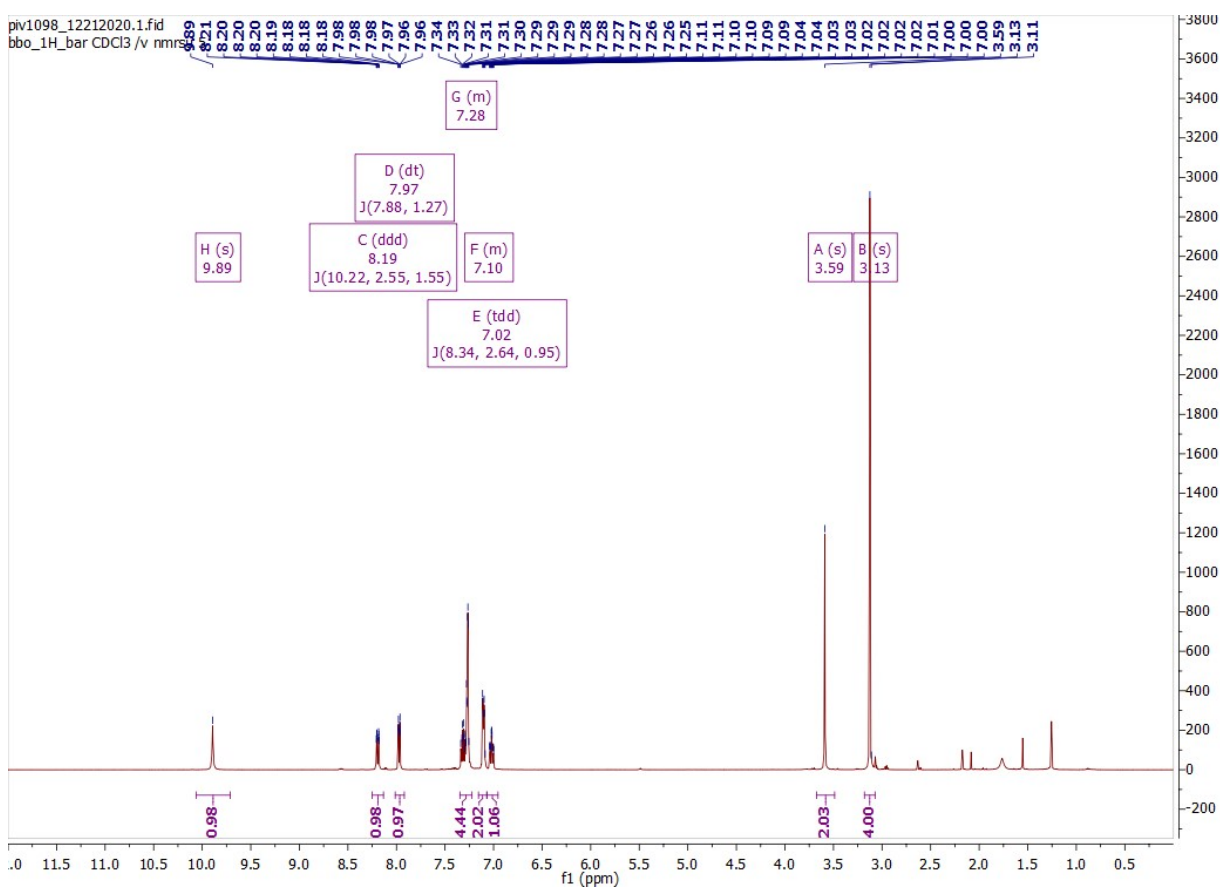




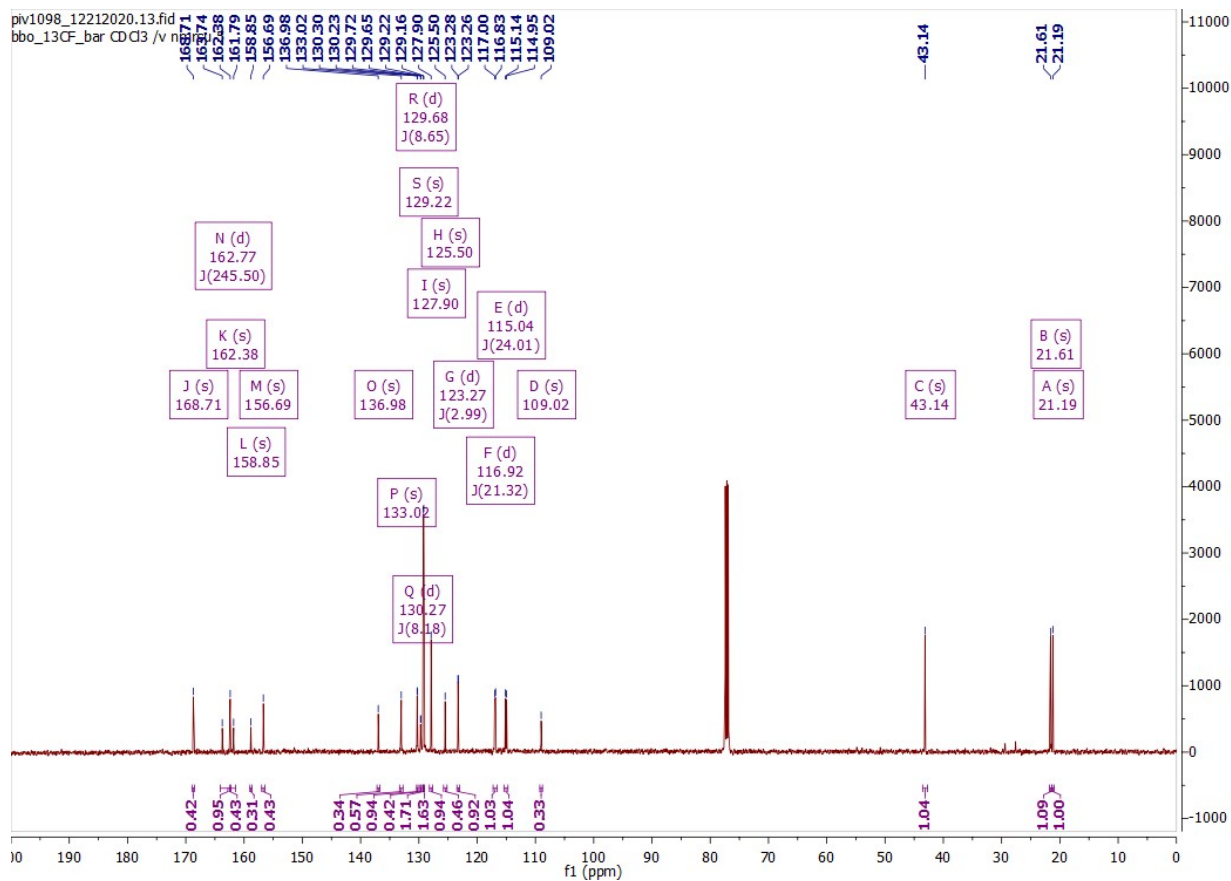
N-(8-(3-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-c]isoxazol-2-yl)-2-phenylacetamide
 Chemical Formula: C₂₂H₁₆FN₃O₂S
 Exact Mass: 405,0947
 Molecular Weight: 405,4474

8d

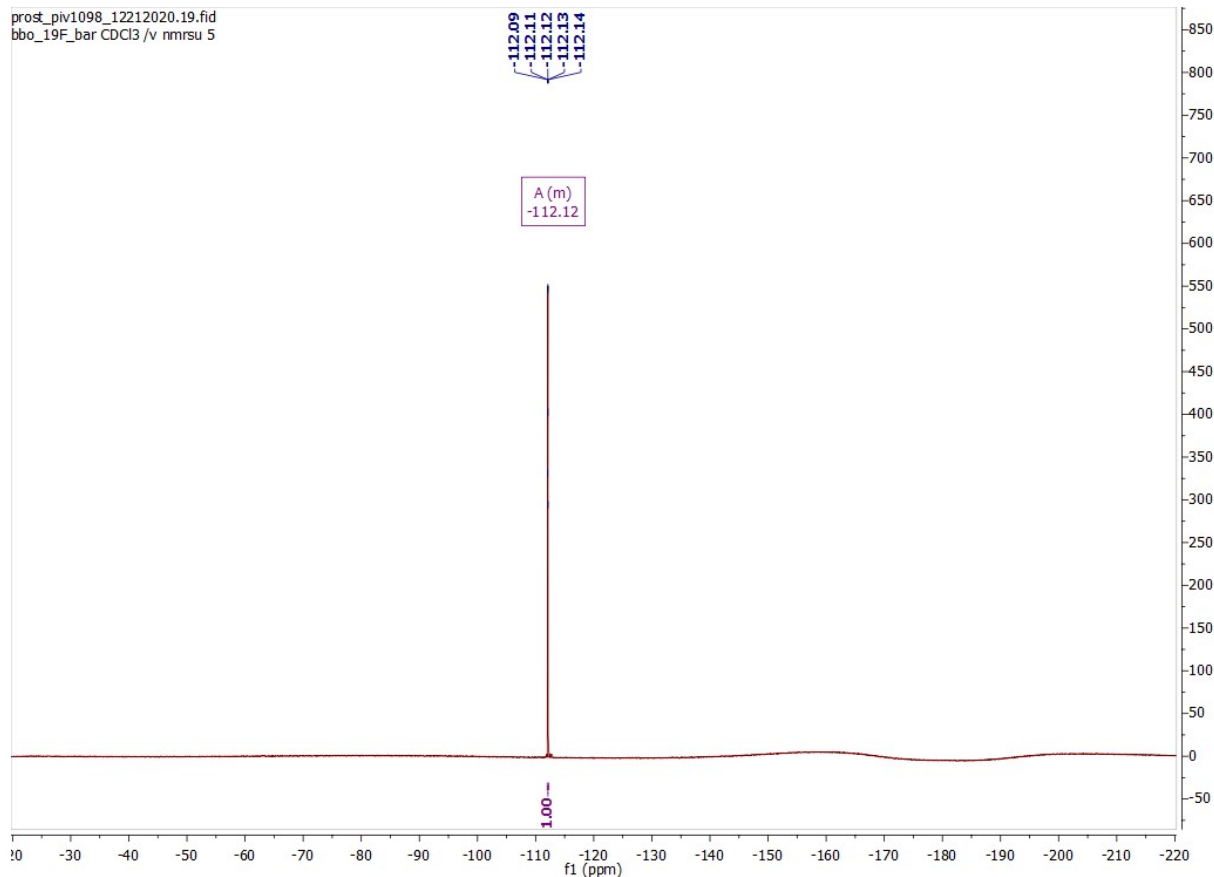
¹H spectra of **8d**:

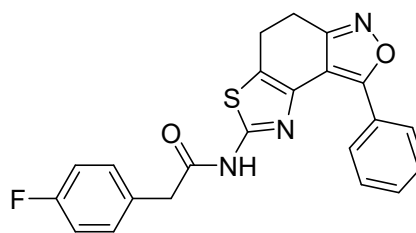


¹³C spectra of **8d**:



¹⁹F NMR spectra of **8d**:





2-(4-fluorophenyl)-*N*-(8-phenyl-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-c]isoxazol-2-yl)acetamide

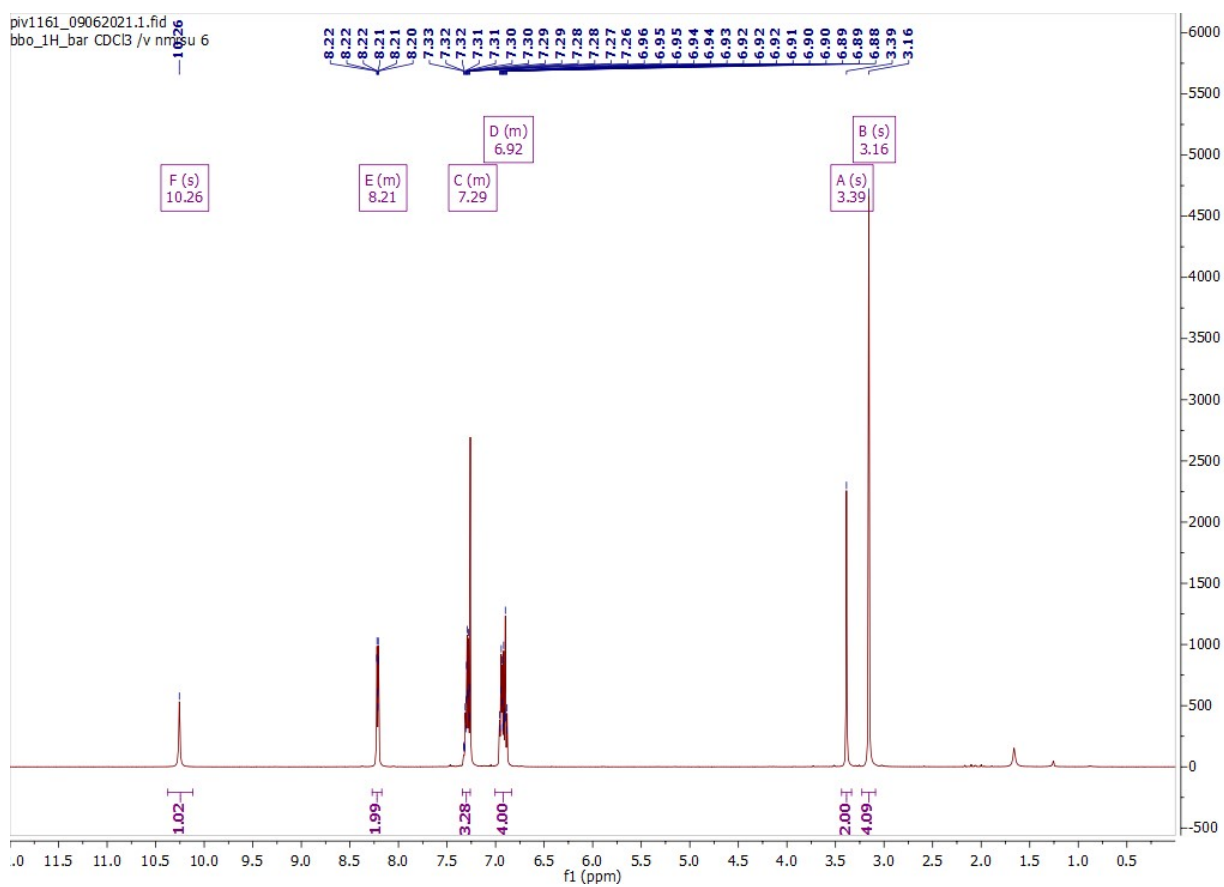
Chemical Formula: C₂₂H₁₆FN₃O₂S

Exact Mass: 405,0947

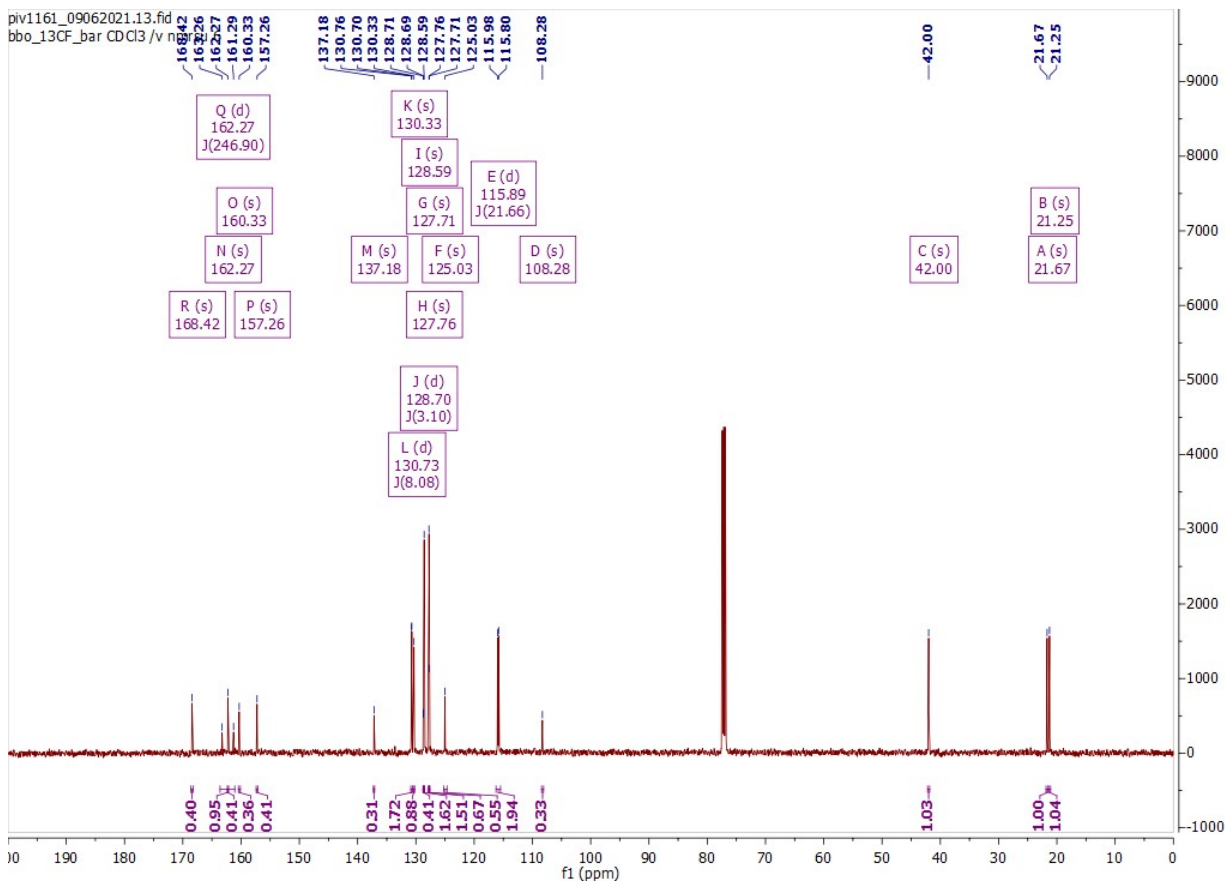
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8e

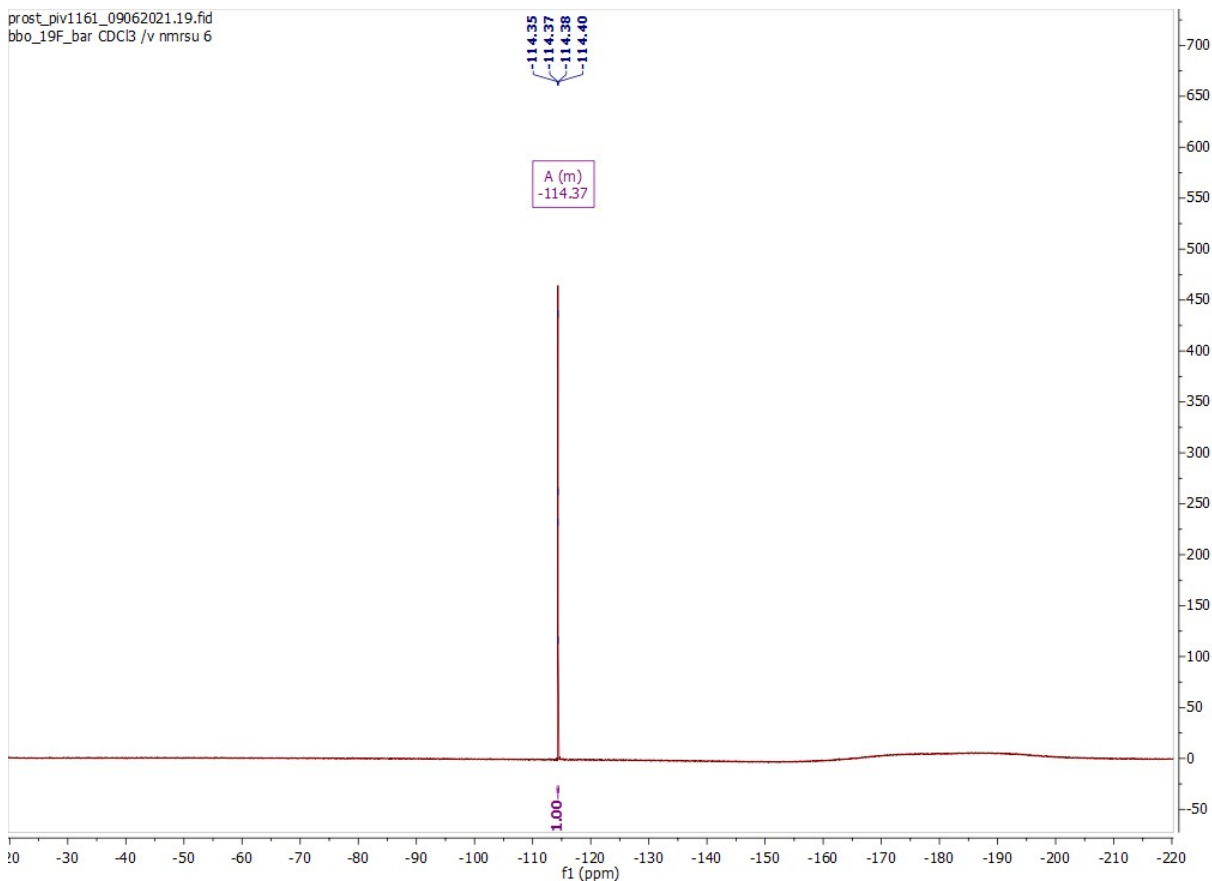
¹H spectra of **8e**:

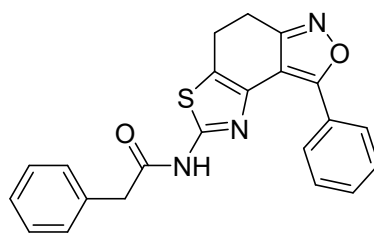


¹³C spectra of **8e**:



¹⁹F NMR spectra of **8e**:





2-phenyl-*N*-(8-phenyl-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-*c*]isoxazol-2-yl)acetamide

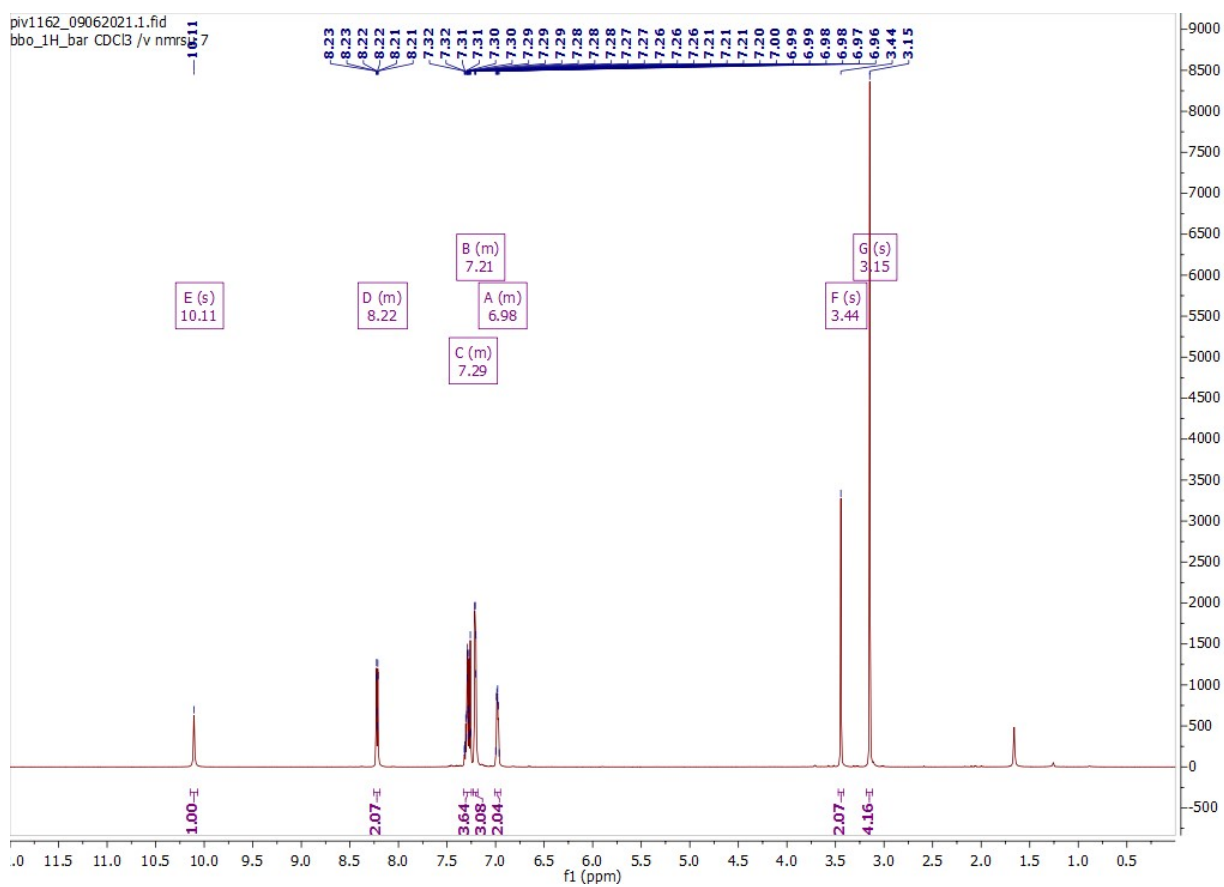
Chemical Formula: C₂₂H₁₇N₃O₂S

Exact Mass: 387,1041

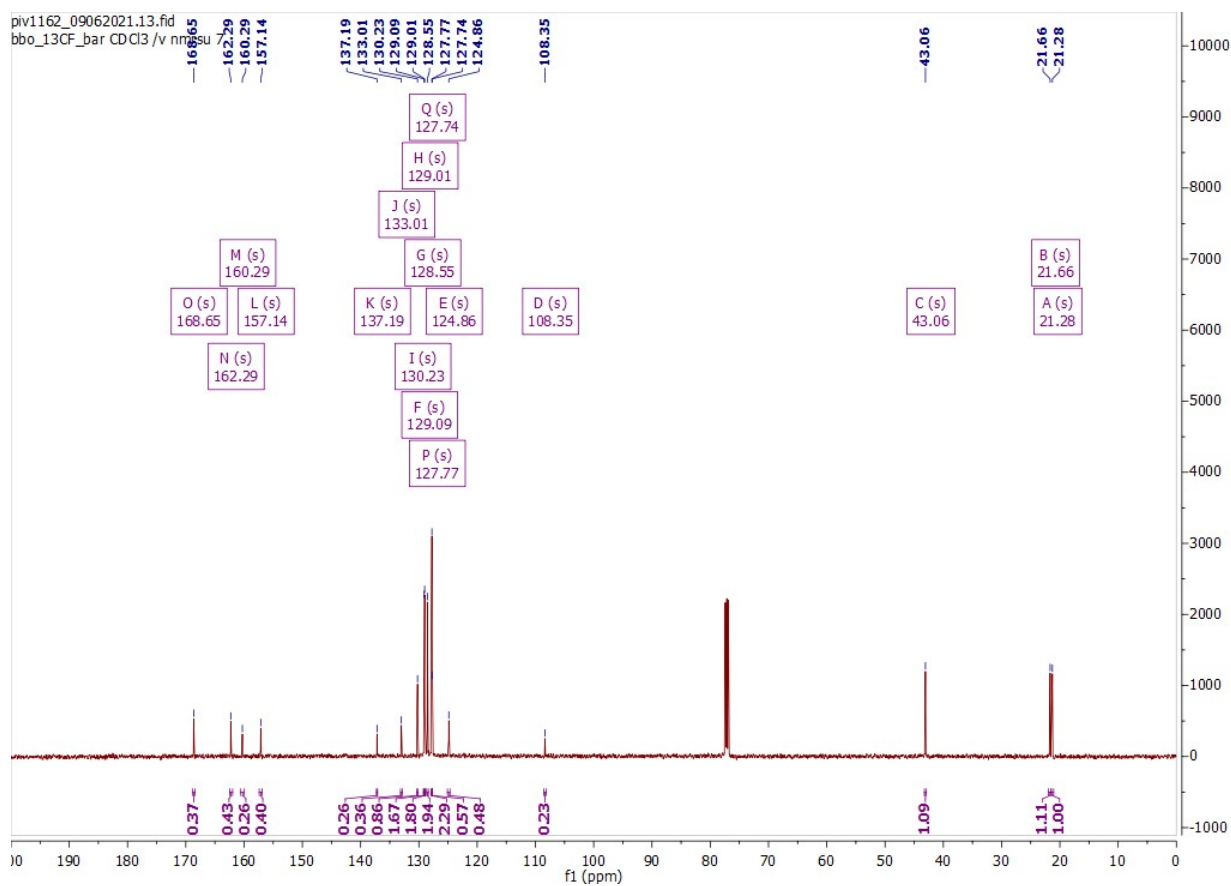
Molecular Weight: 387,4570

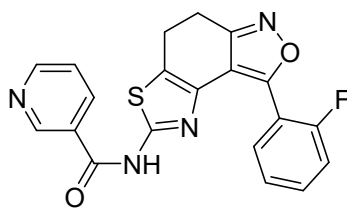
8f

¹H spectra of **8f**:



¹H spectra of **8f**:





N-(8-(2-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-*c*]isoxazol-2-yl)nicotinamide

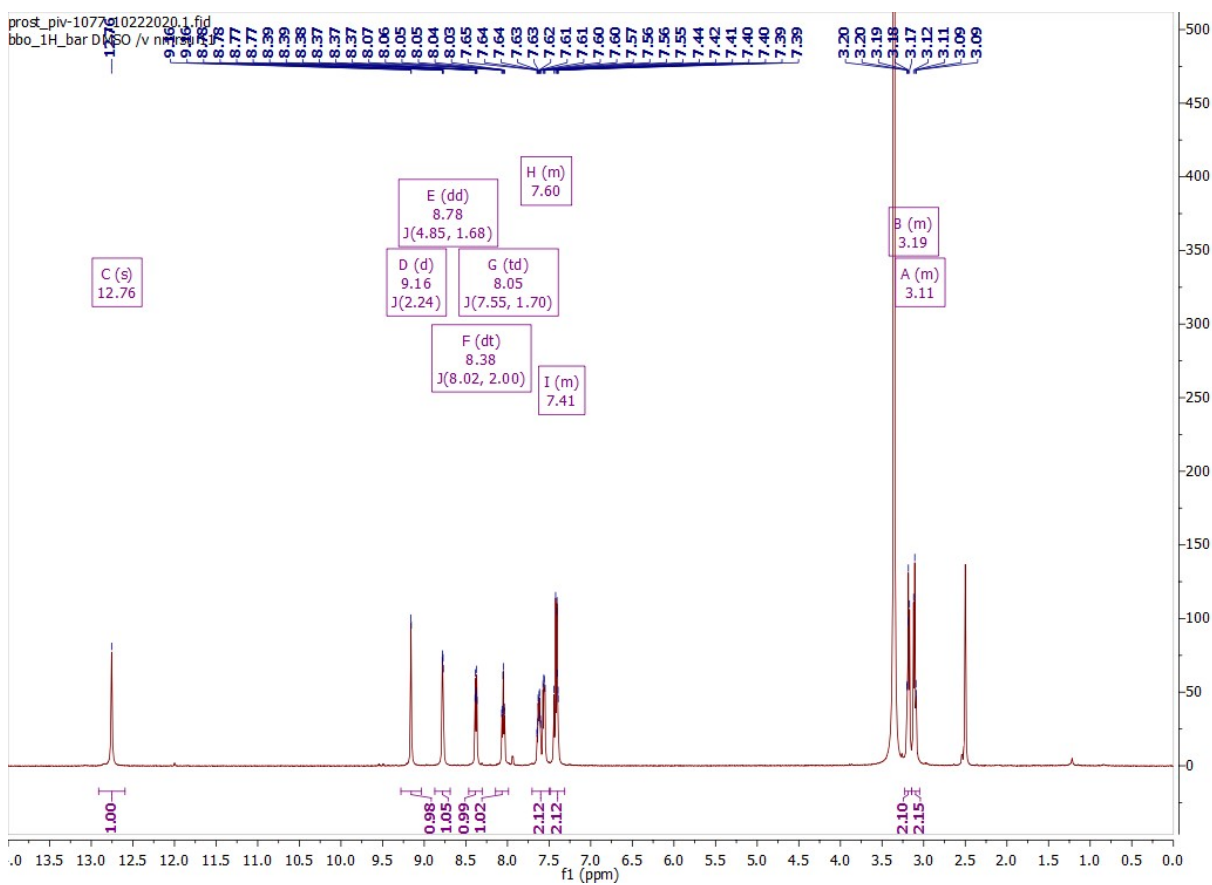
Chemical Formula: C₂₀H₁₃FN₄O₂S

Exact Mass: 392,0743

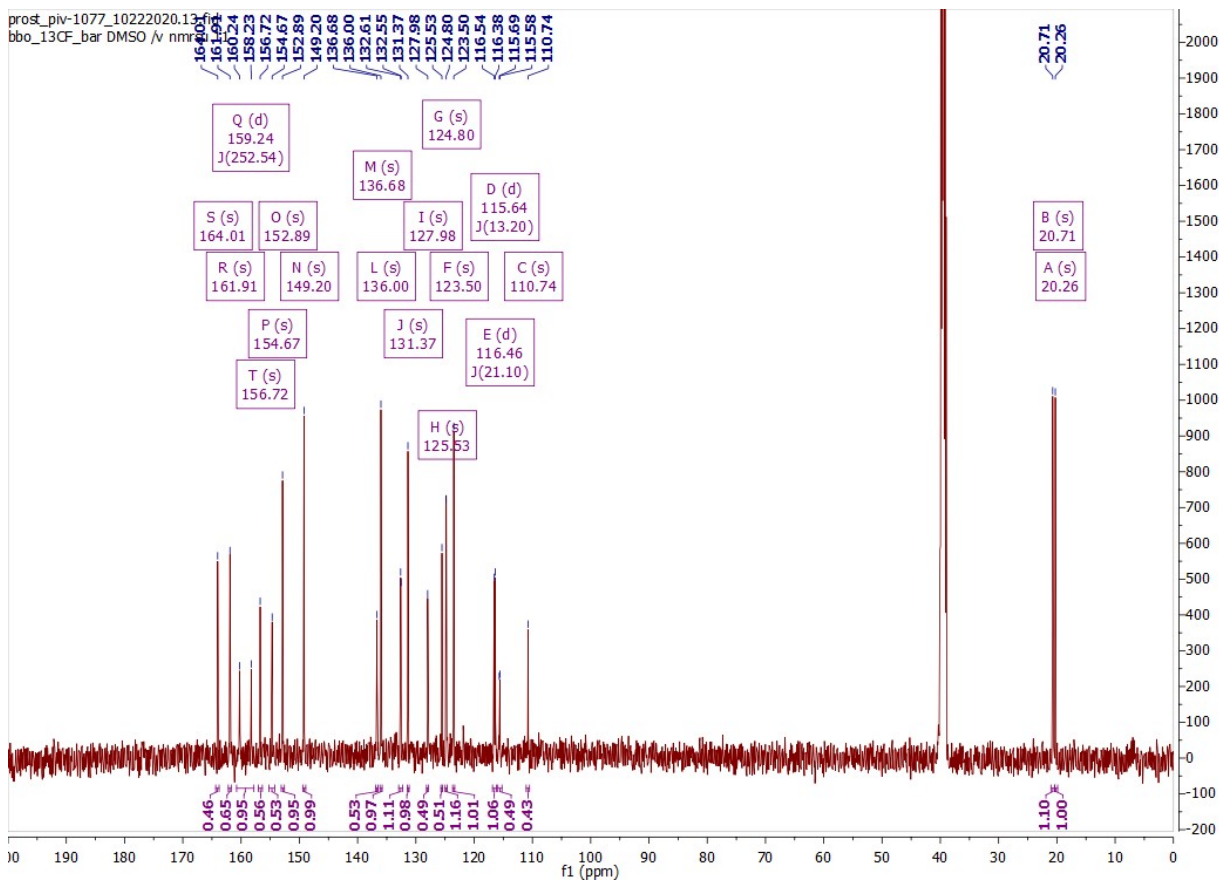
Molecular Weight: 392,4084

8g

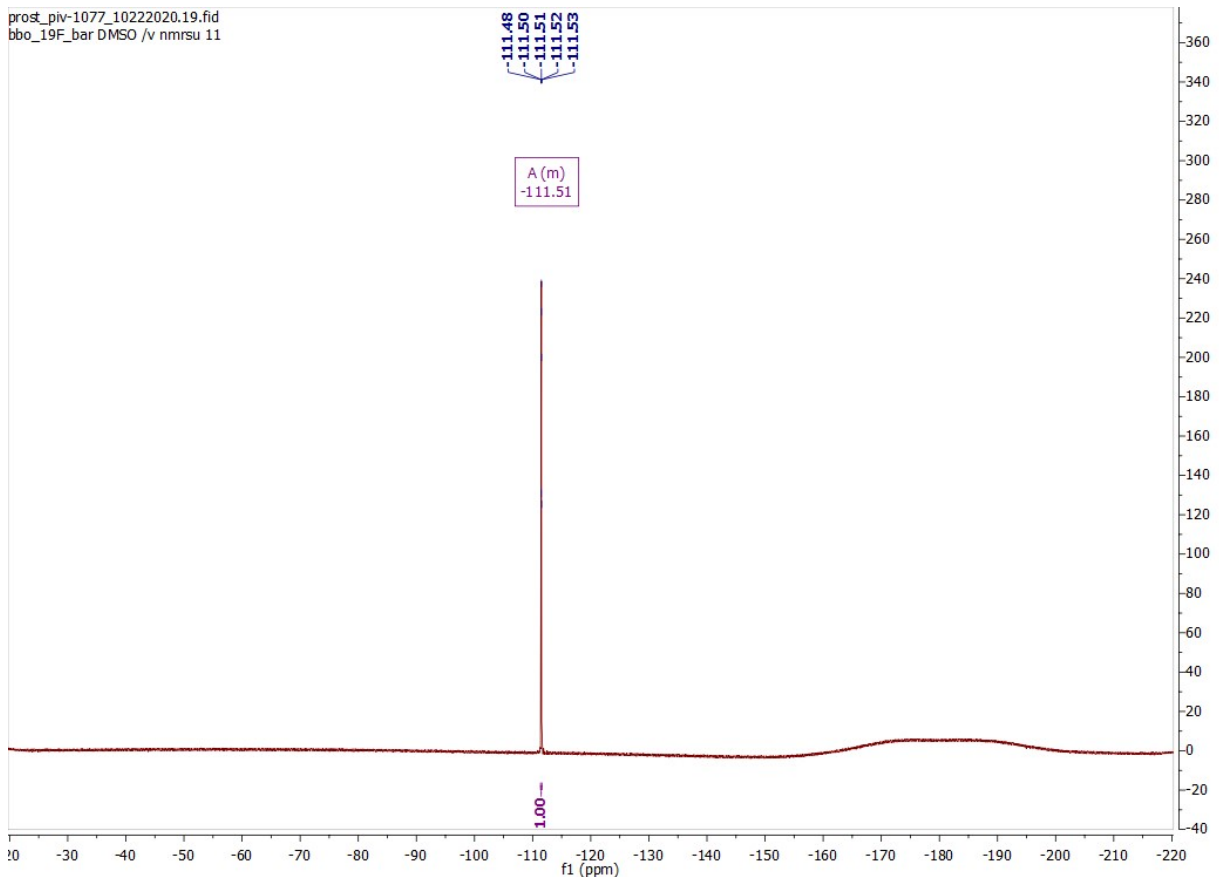
¹H spectra of **8g**:

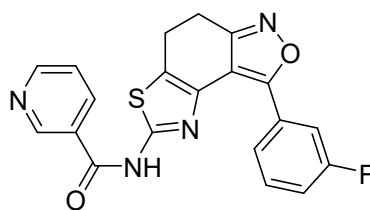


¹³C spectra of **8g**:



¹⁹F NMR spectra of **8g**:





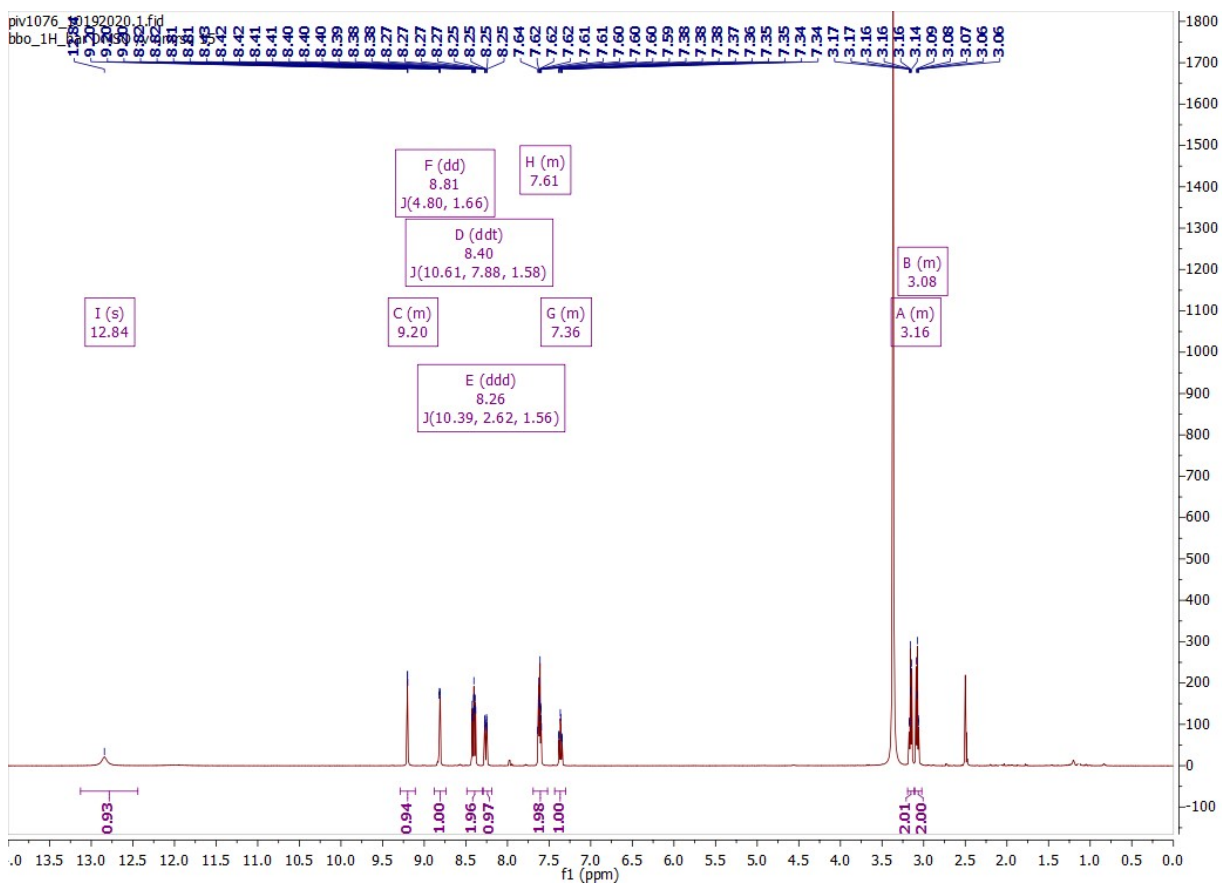
N-(8-(3-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-*c*]isoxazol-2-yl)nicotinamide

Chemical Formula: C₂₀H₁₃FN₄O₂S

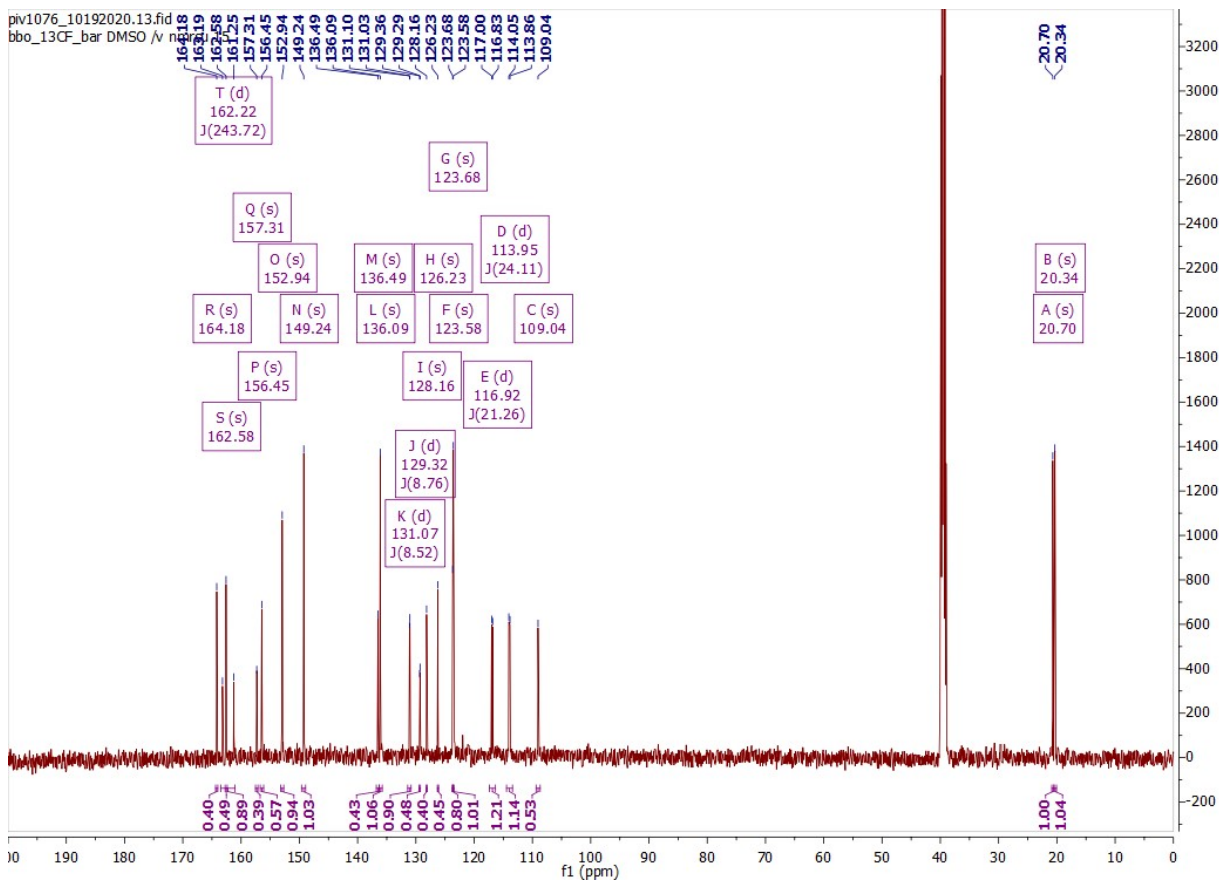
Molecular Weight: 392,41

8h

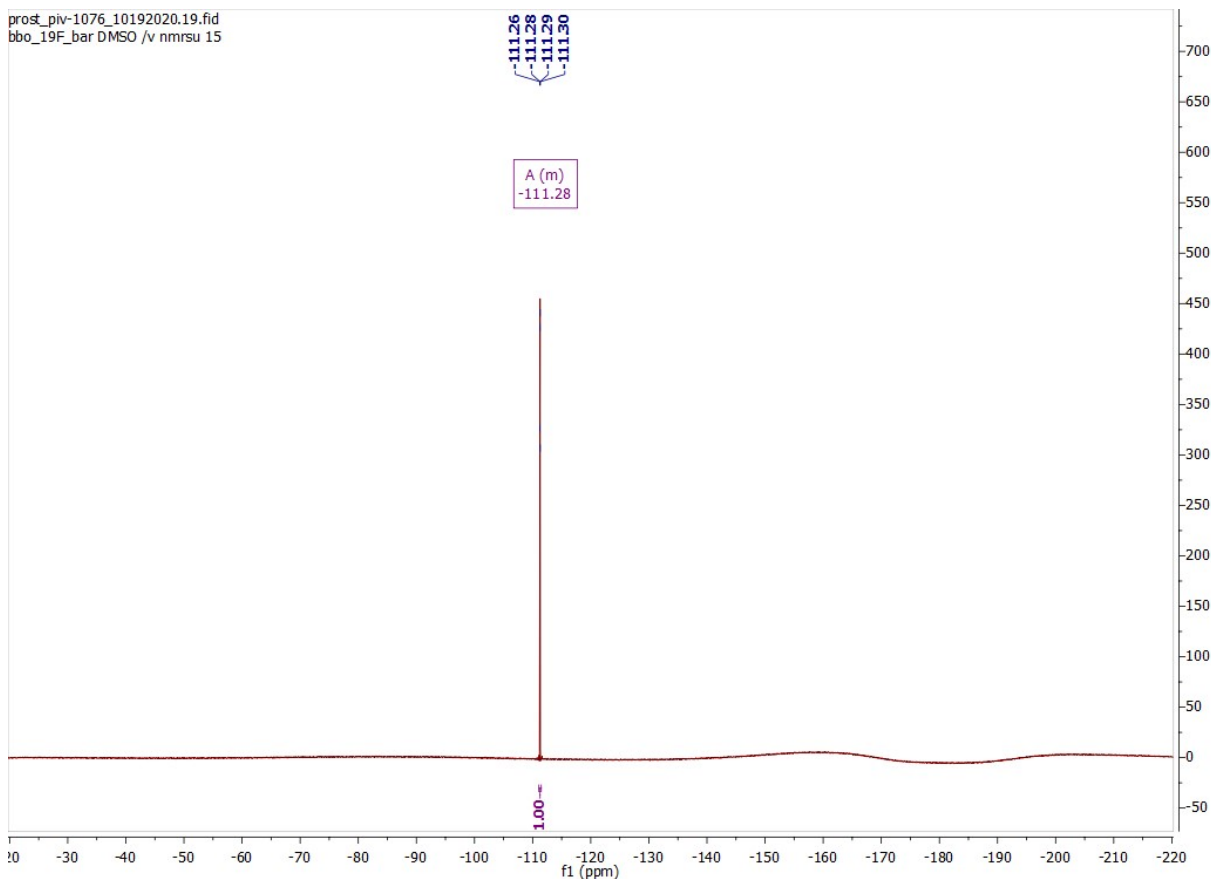
¹H spectra of 8h:

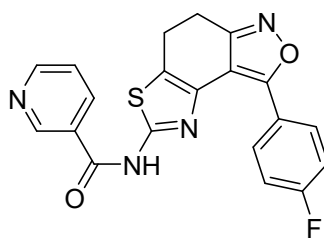


¹³C spectra of **8h**:



¹⁹F spectra of **8h**:





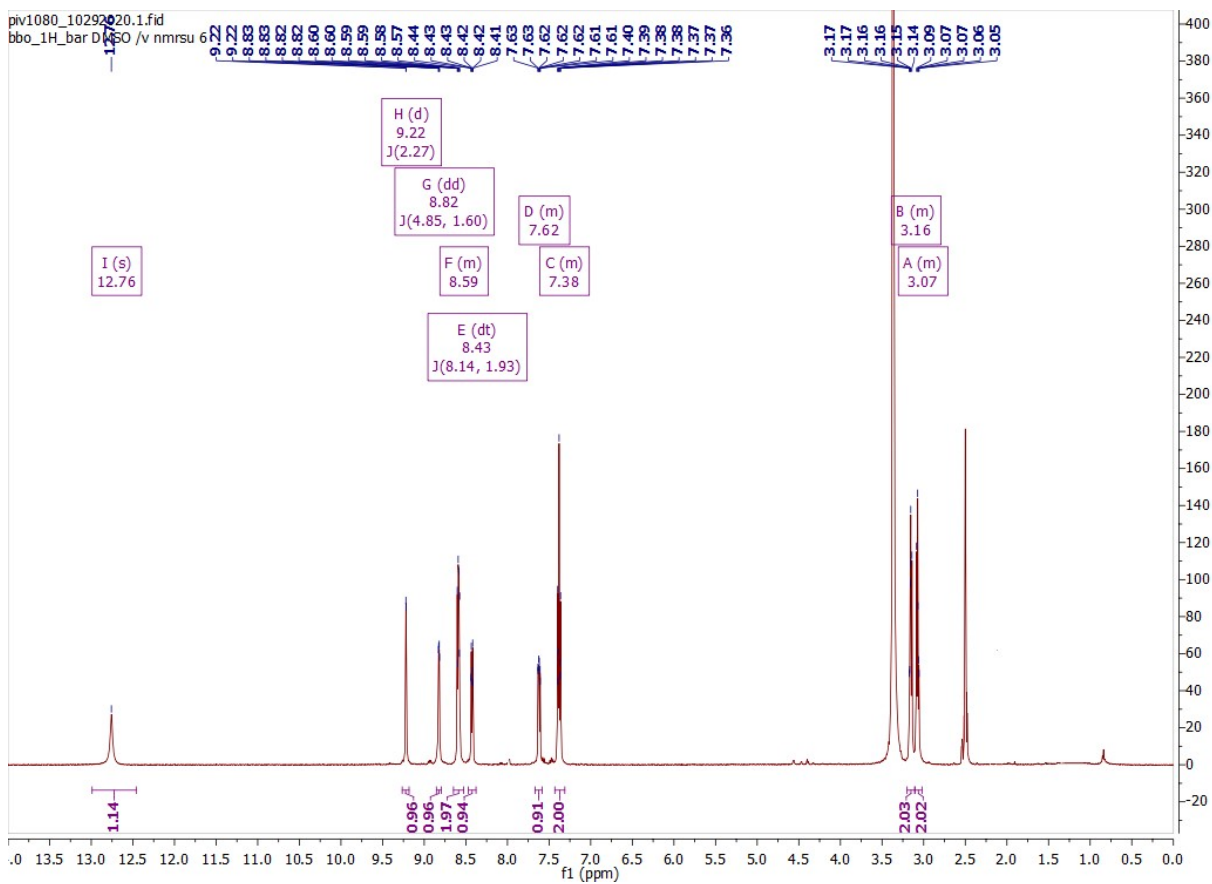
N-(8-(4-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-*c*]isoxazol-2-yl)nicotinamide

Chemical Formula: C₂₀H₁₃FN₄O₂S

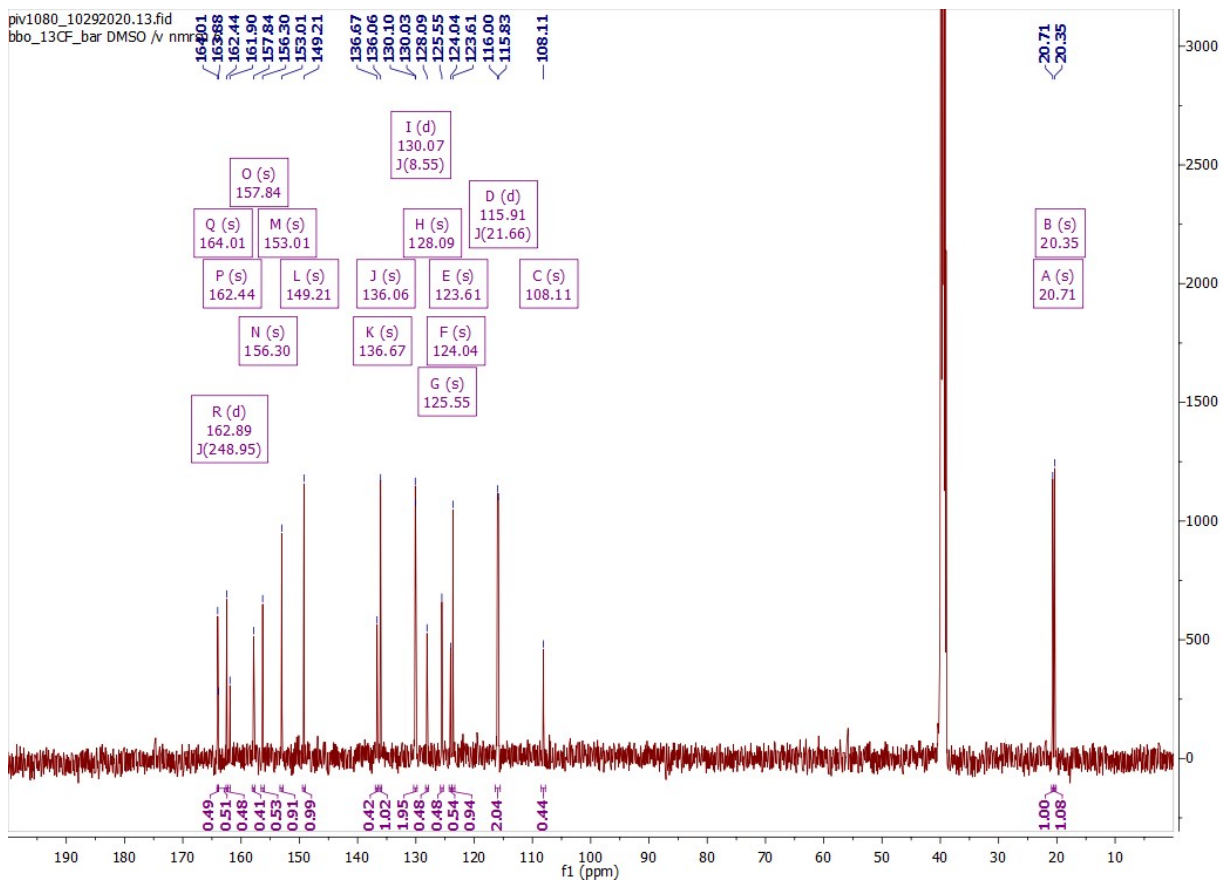
Molecular Weight: 392,41

8i

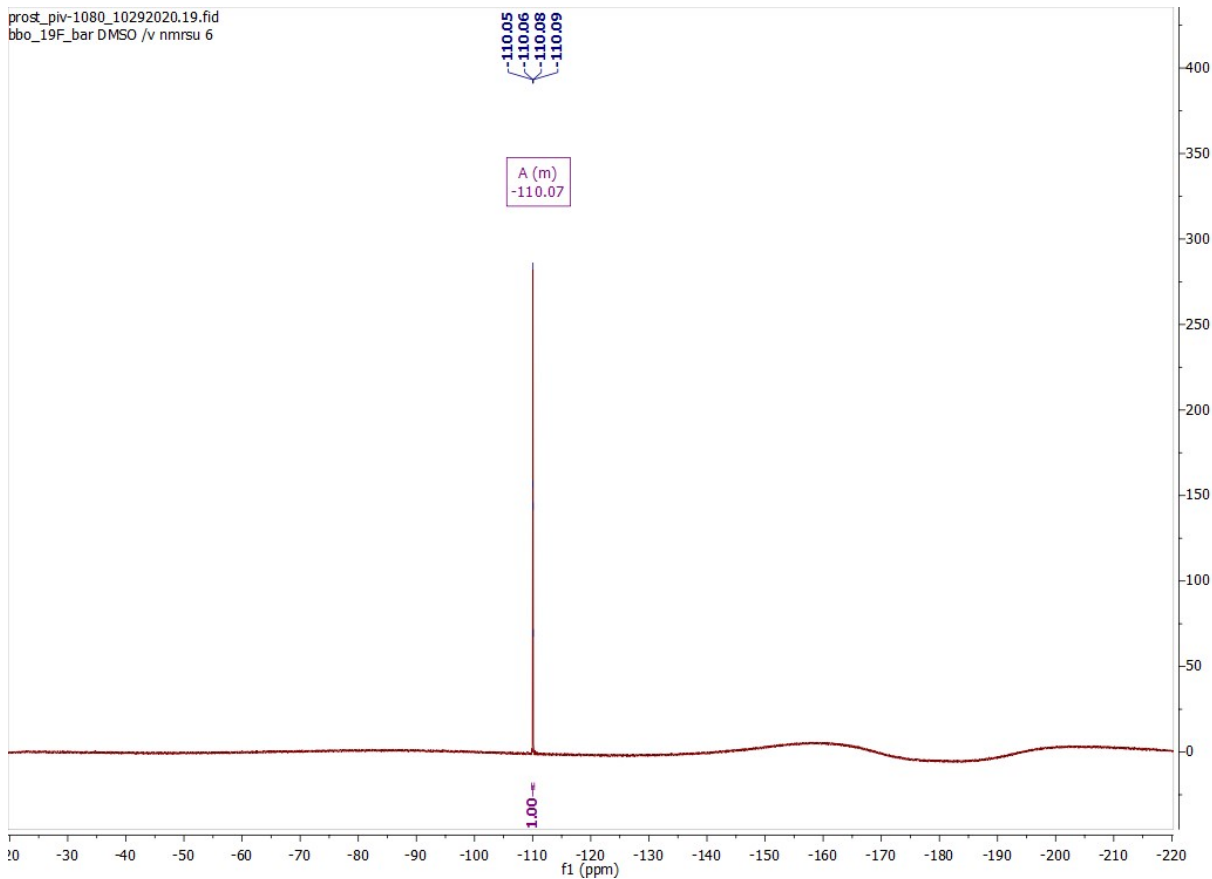
¹H spectra of **8i**:

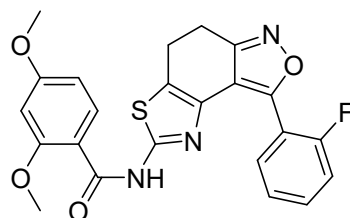


¹³C spectra of **8i**:



¹⁹F spectra of **8i**:





N-(8-(2-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-*c*]isoxazol-2-yl)-2,4-dimethoxybenzamide

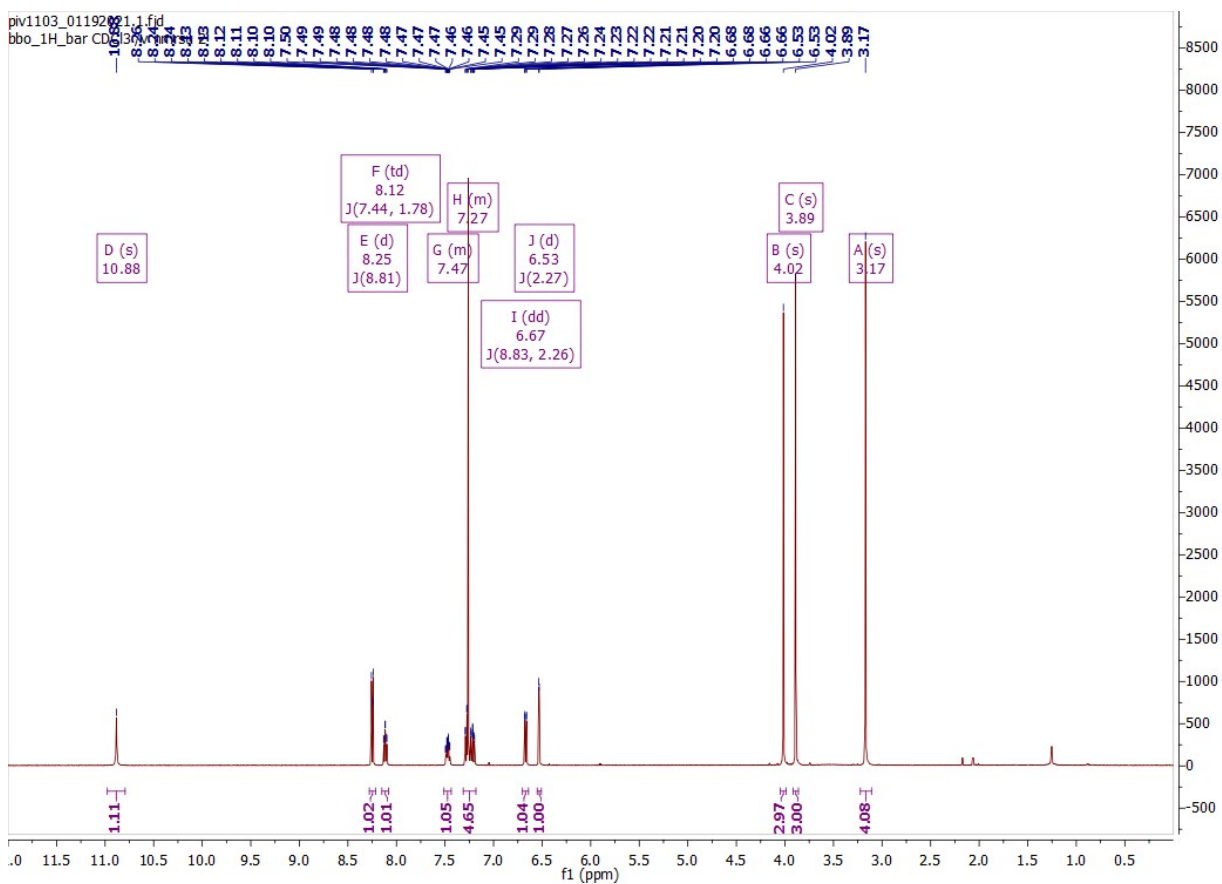
Chemical Formula: C₂₃H₁₈FN₃O₄S

Exact Mass: 451,1002

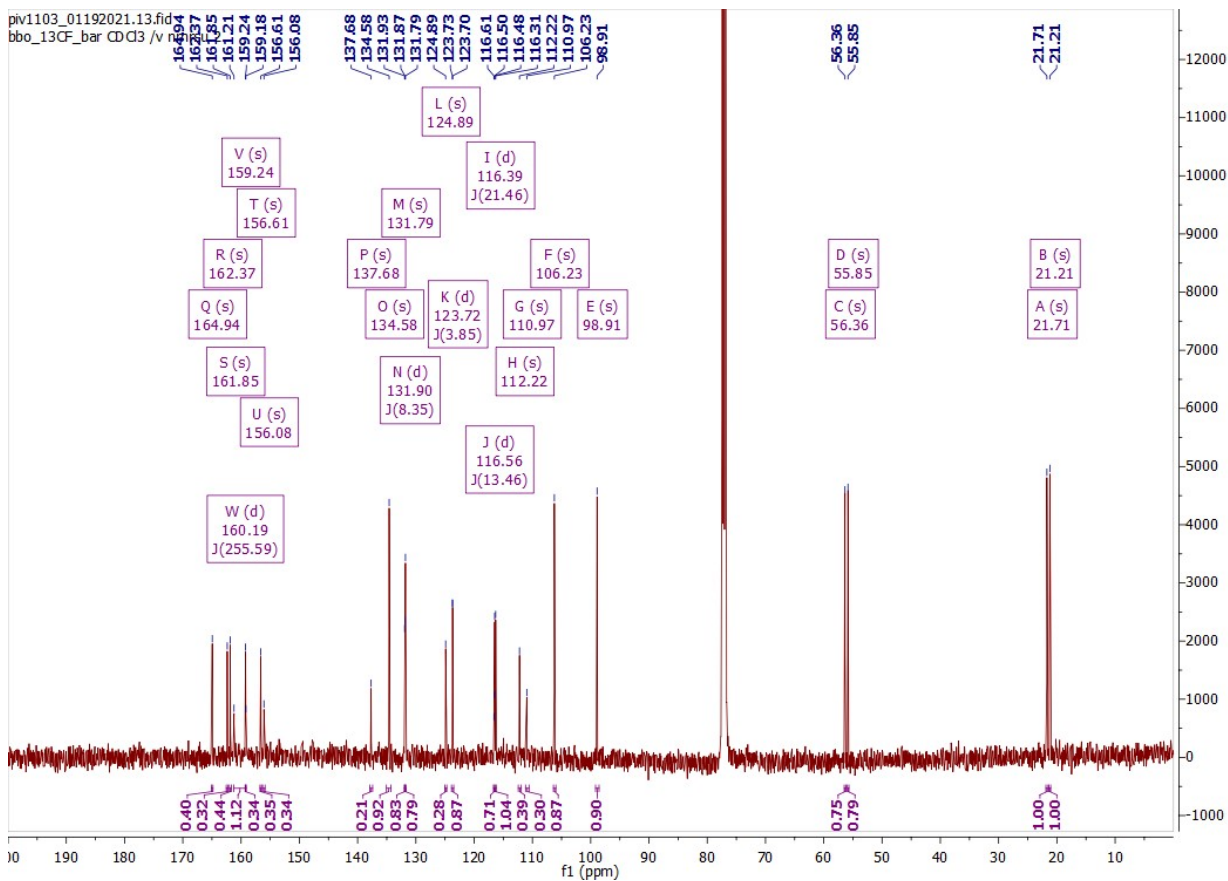
Molecular Weight: 451,4724

8j

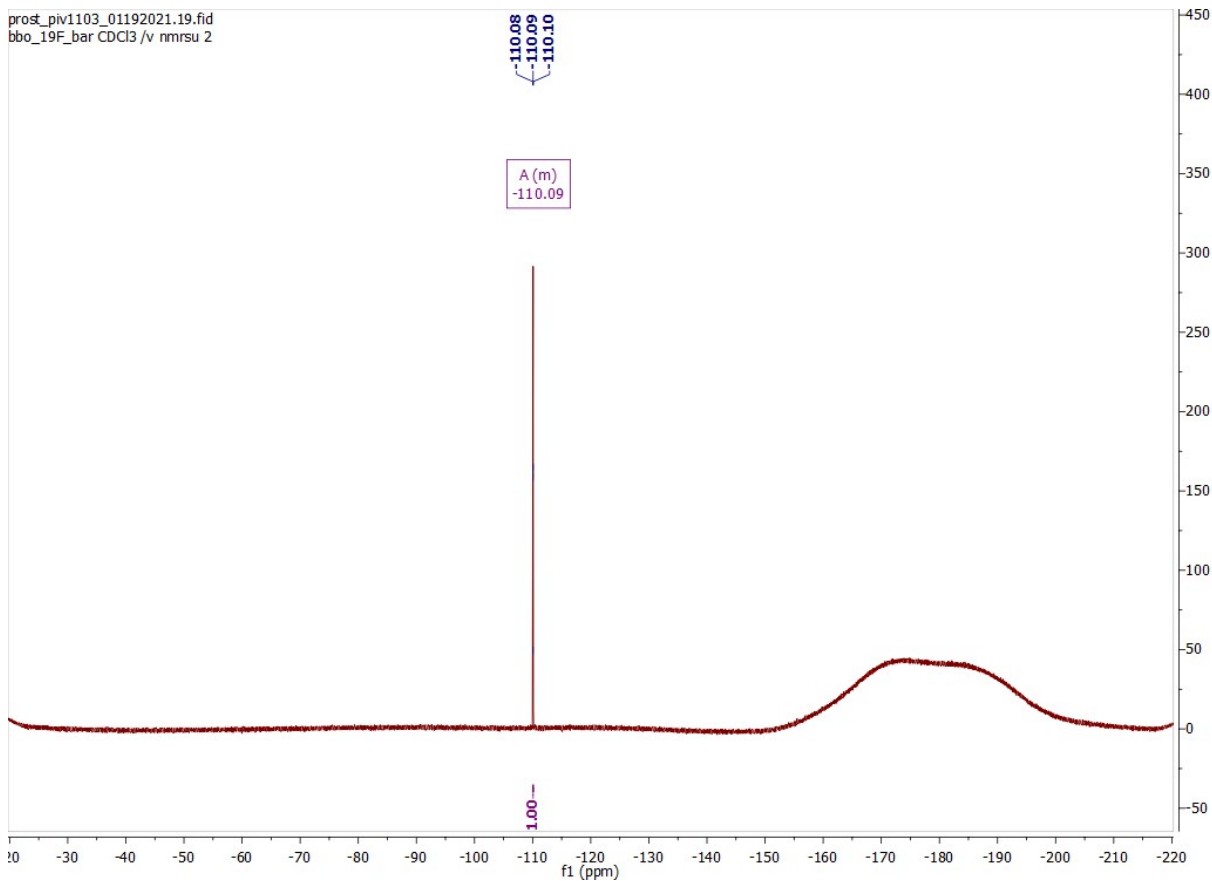
¹H spectra of **8j**:

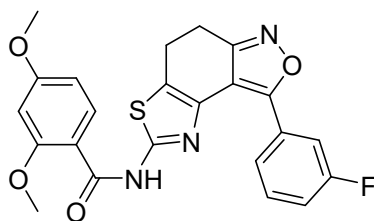


¹³C spectra of **8j**:



¹⁹F NMR spectra of **8j**:





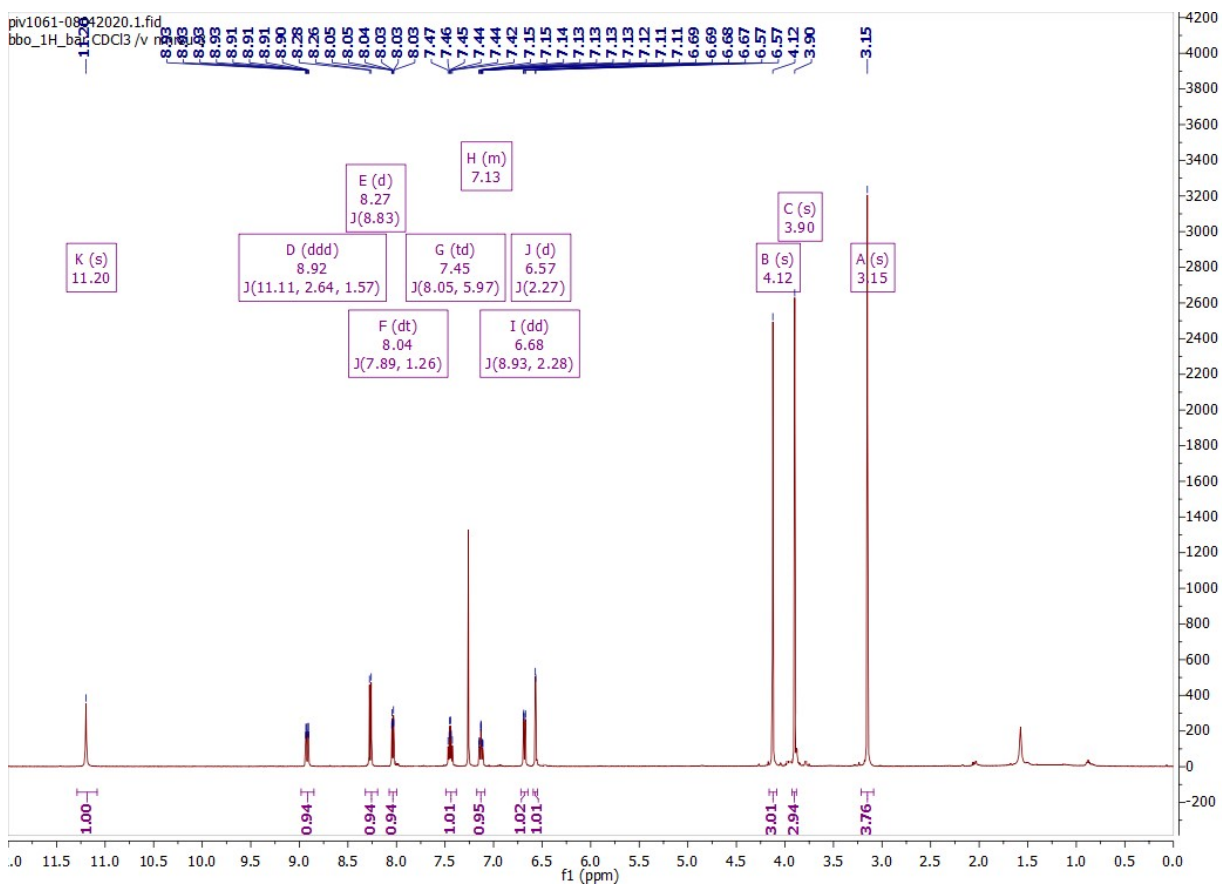
N-(8-(3-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-*c*]isoxazol-2-yl)-2,4-dimethoxybenzamide

Chemical Formula: C₂₃H₁₈FN₃O₄S

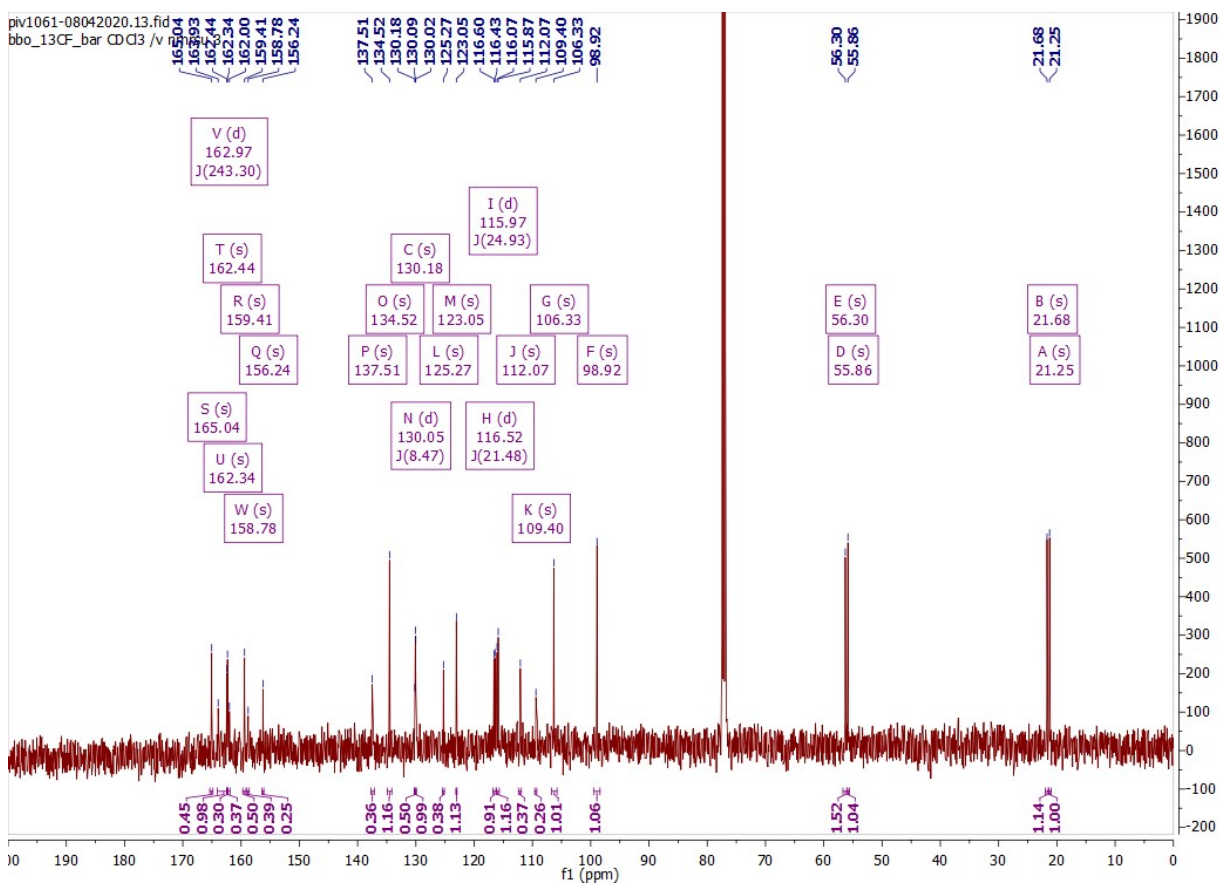
Molecular Weight: 451,47

8k

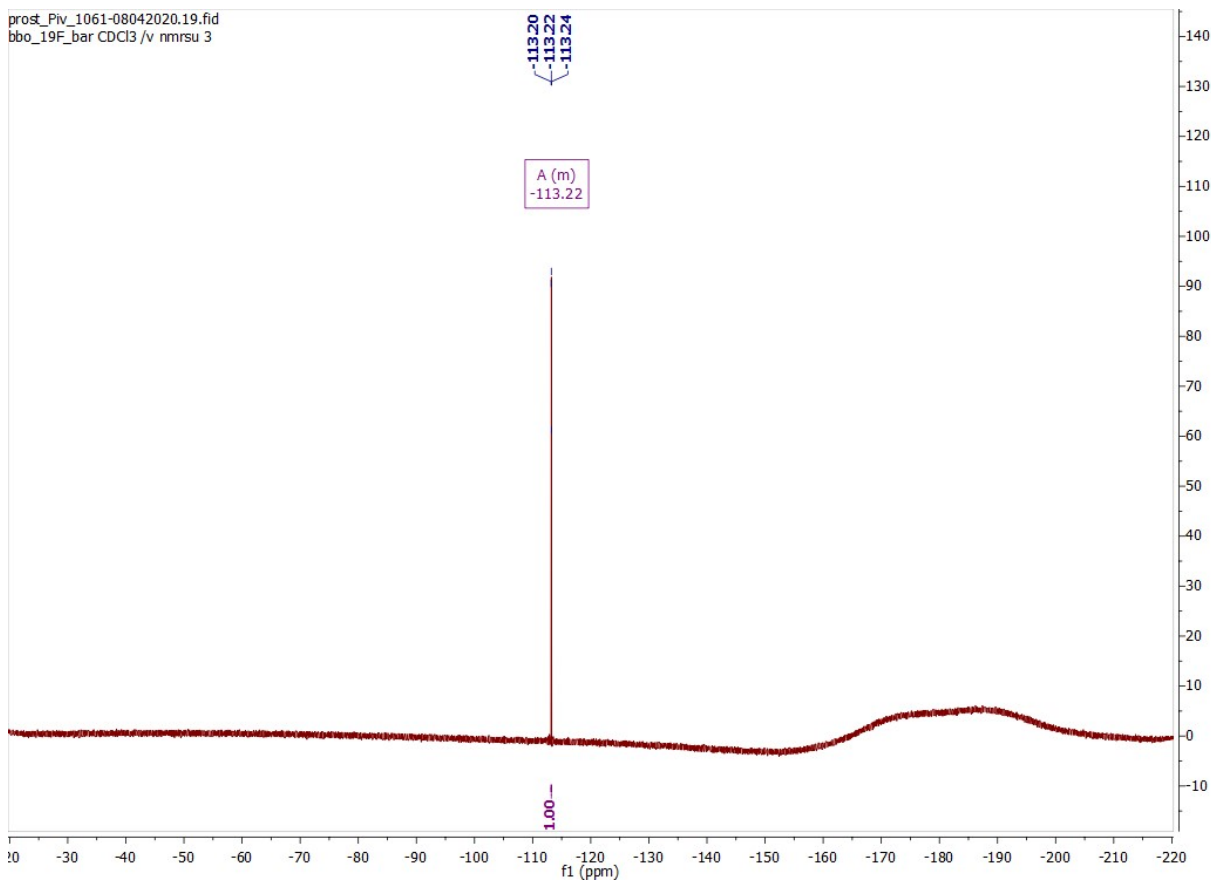
¹H spectra of **8k**:

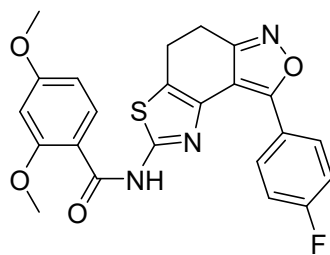


¹³C spectra of **8k**:



¹⁹F spectra of **8k**:

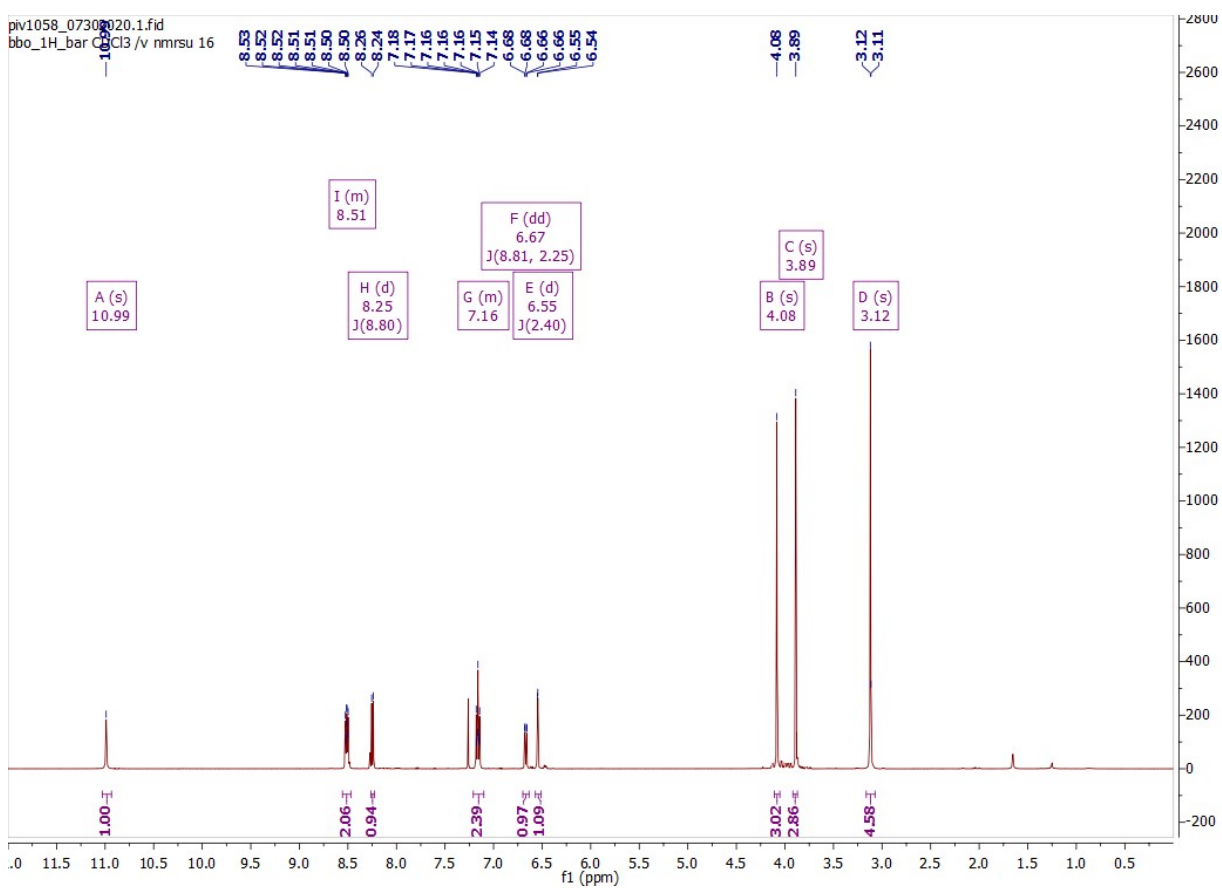




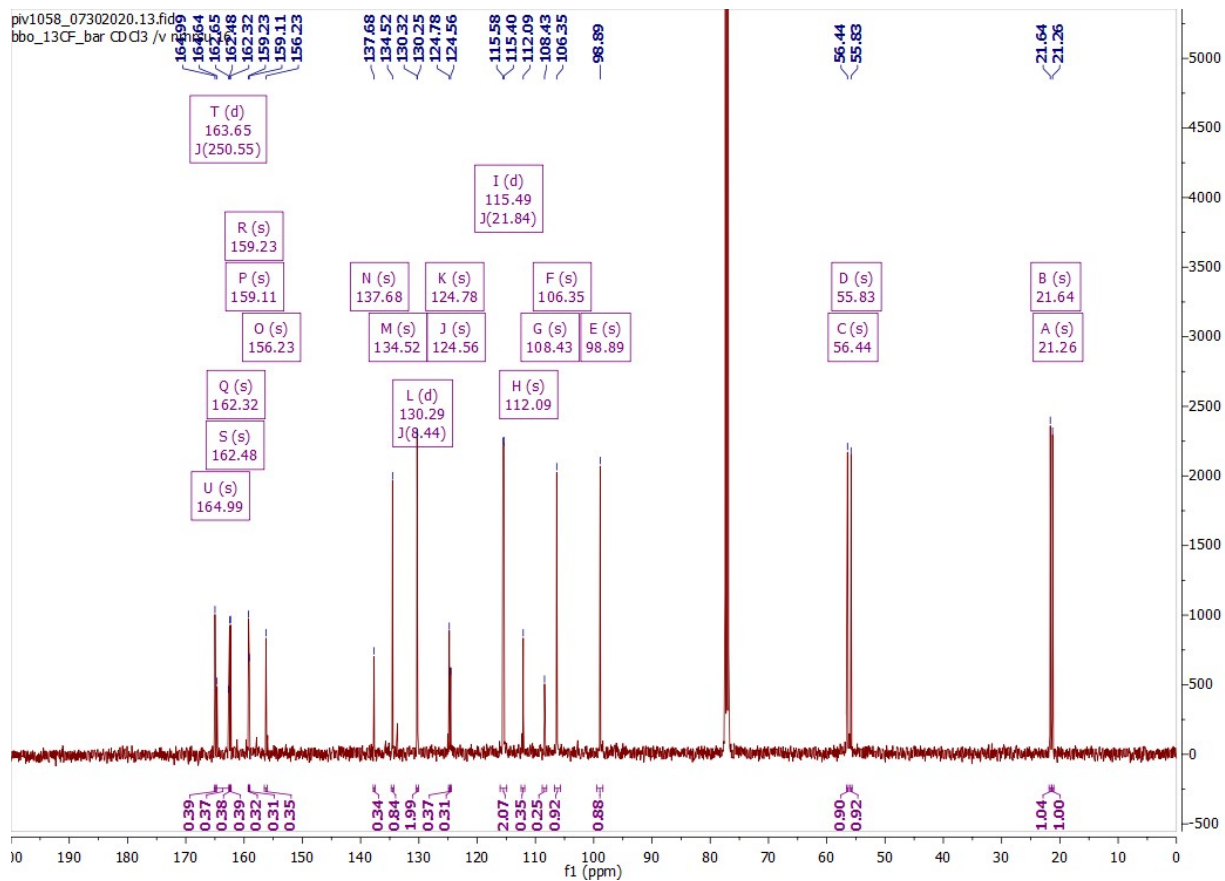
N-(8-(4-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-*c*]isoxazol-2-yl)-2,4-dimethoxybenzamide
 Chemical Formula: C₂₃H₁₈FN₃O₄S
 Molecular Weight: 451,4724

81

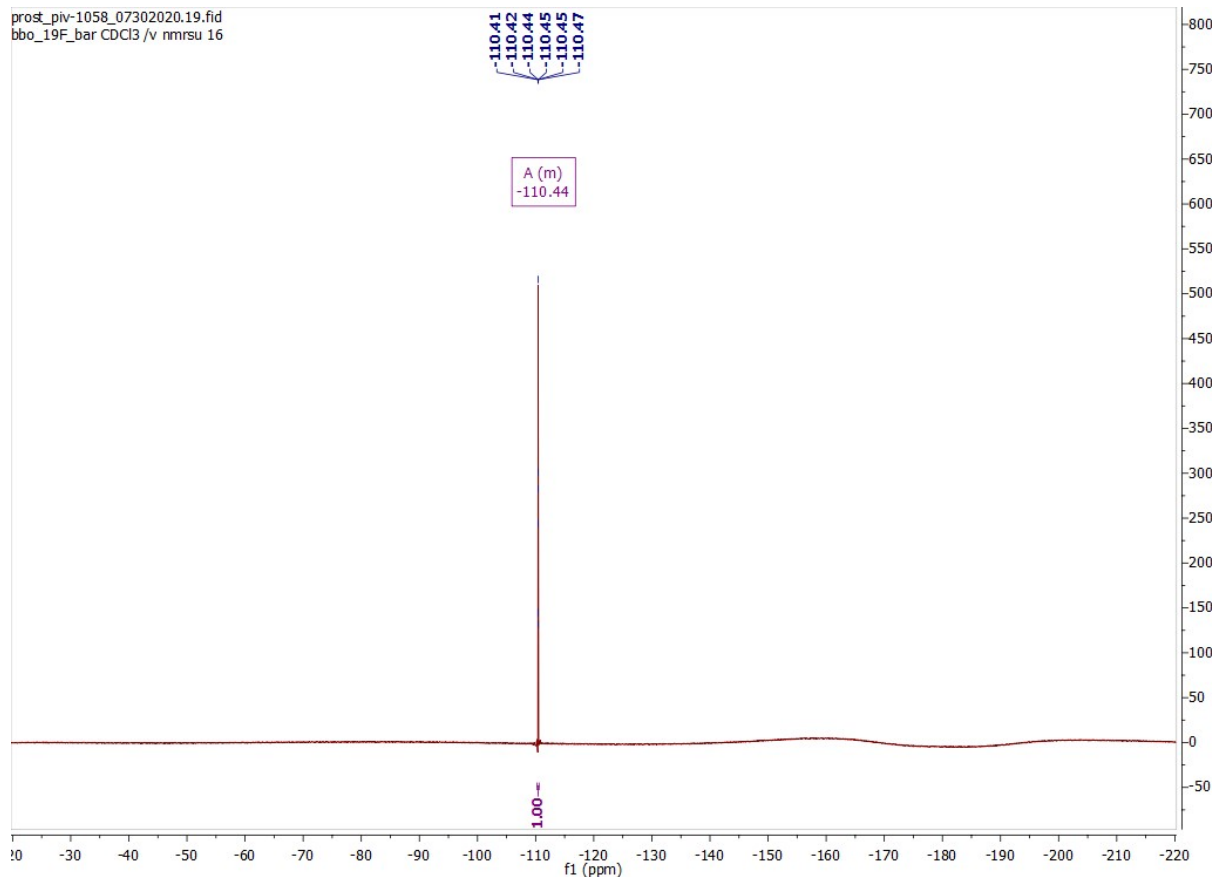
¹H spectra of **81**:

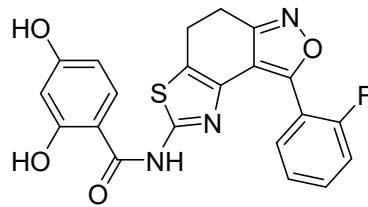


¹³C spectra of **8l**:



¹⁹F spectra of **8l**:

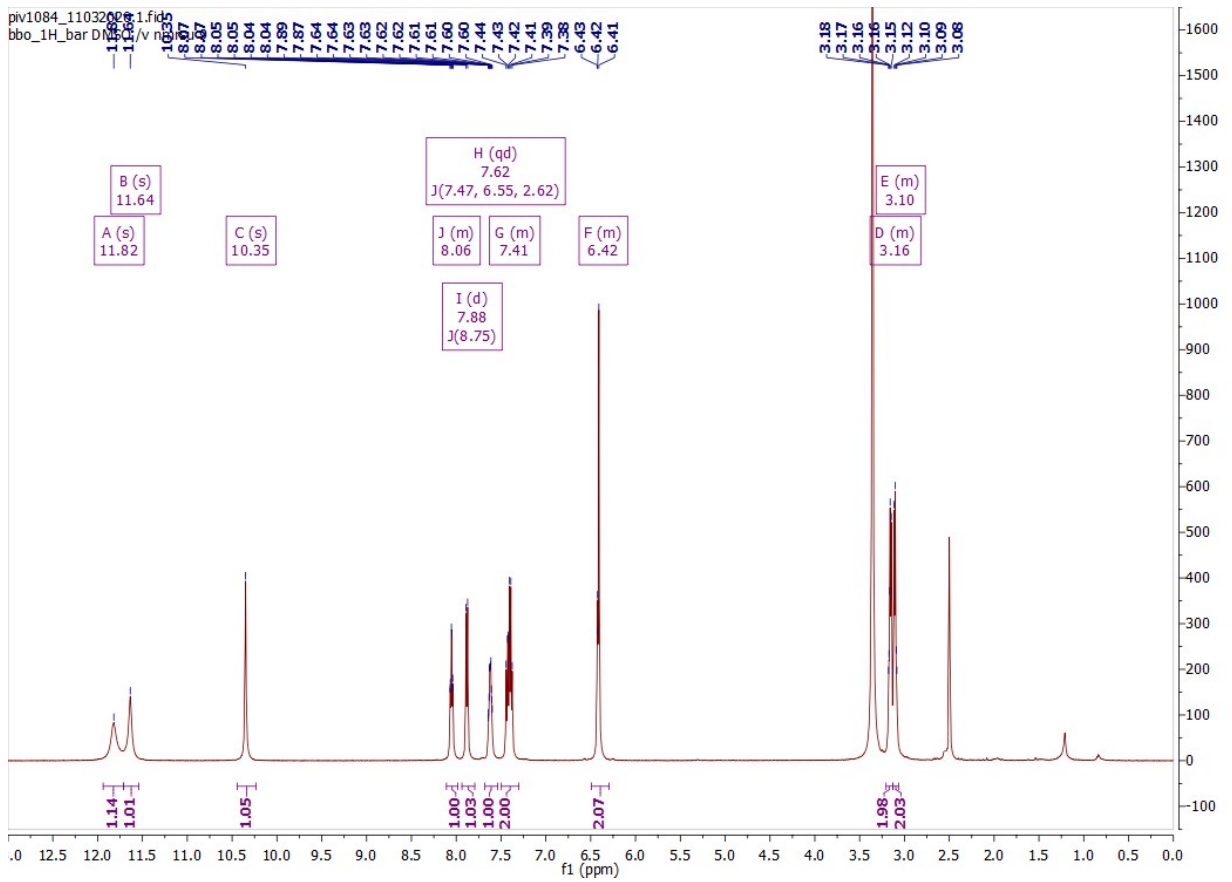




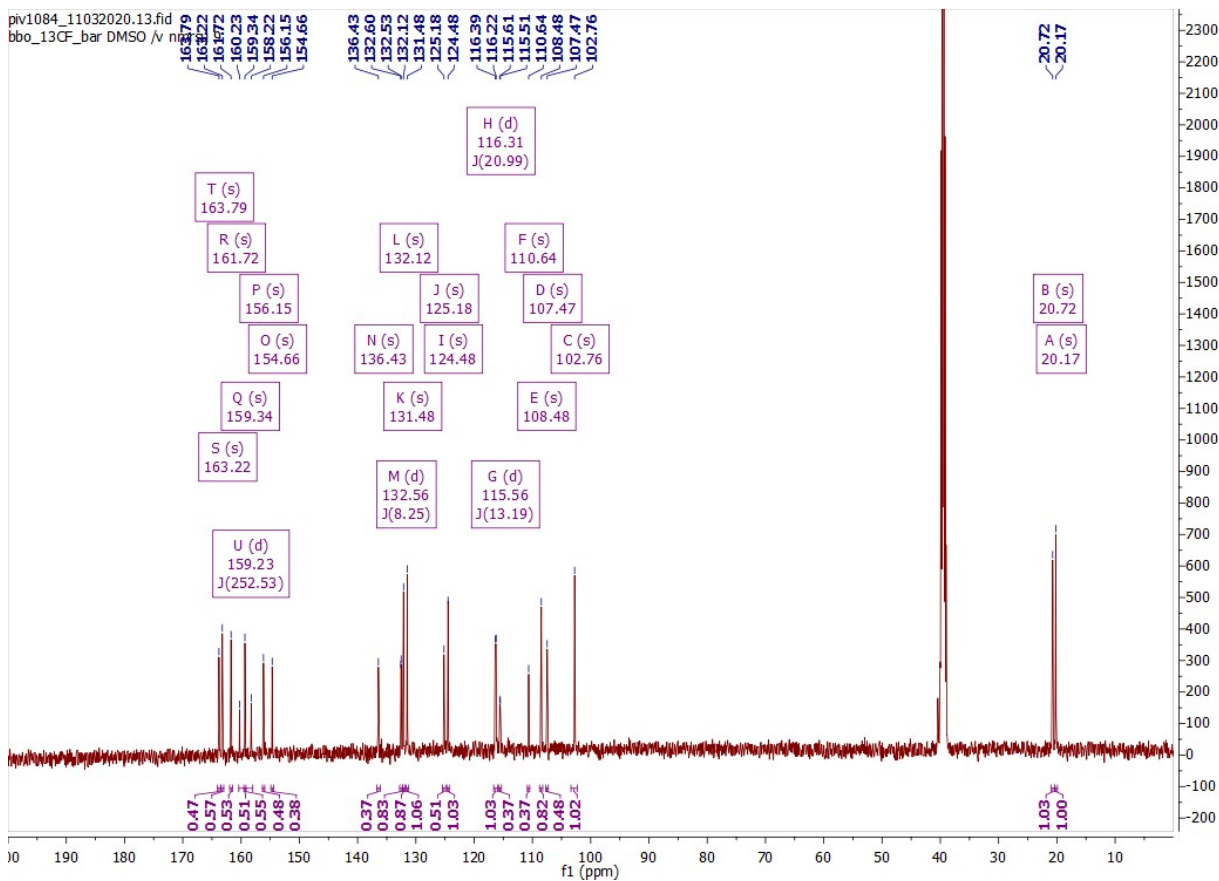
N-(8-(2-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-*c*]isoxazol-2-yl)-2,4-dihydroxybenzamide
 Chemical Formula: C₂₁H₁₄FN₃O₄S
 Exact Mass: 423,0689
 Molecular Weight: 423,4184

8m

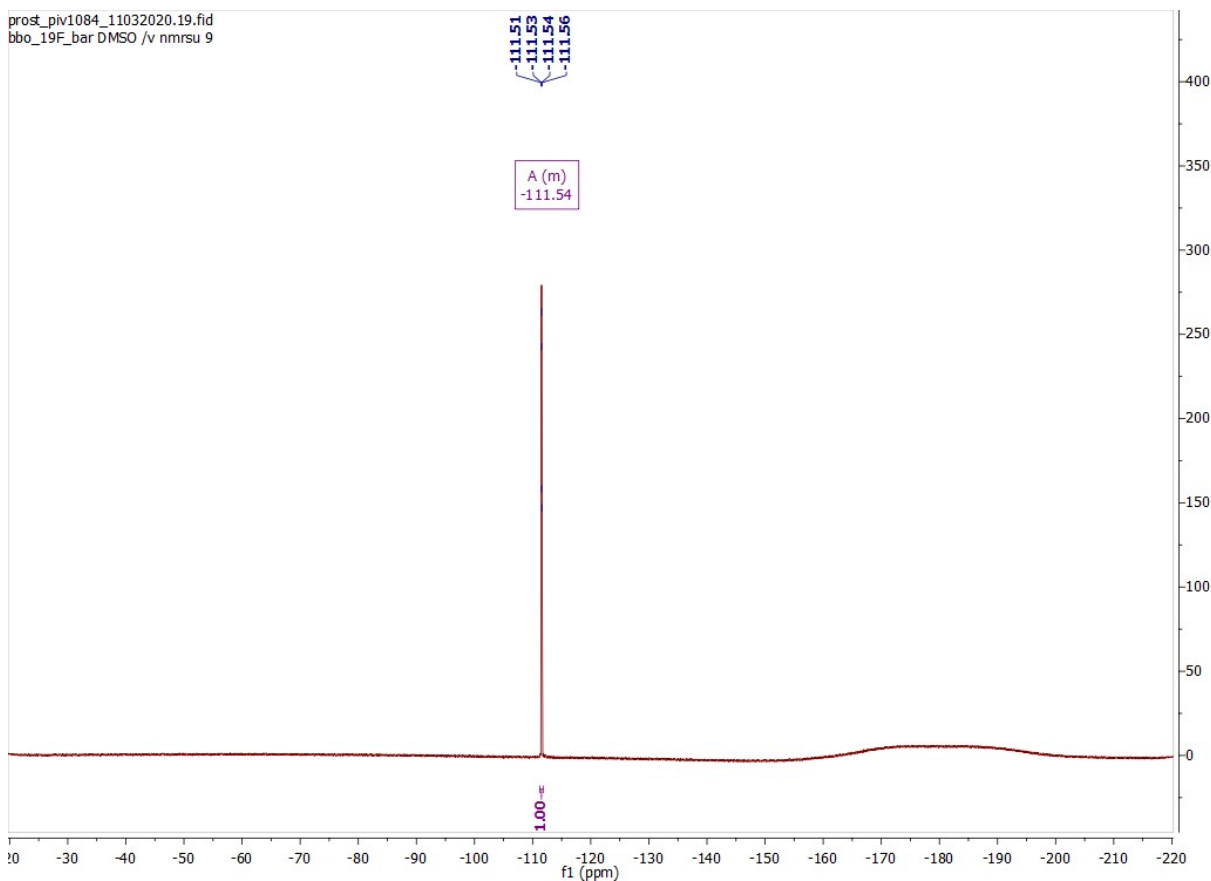
¹H spectra of **8m**:

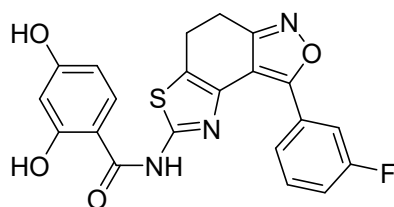


¹³C spectra of **8m**:



¹⁹F spectra of **8m**:





N-(8-(3-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-*c*]isoxazol-2-yl)-2,4-dihydroxybenzamide

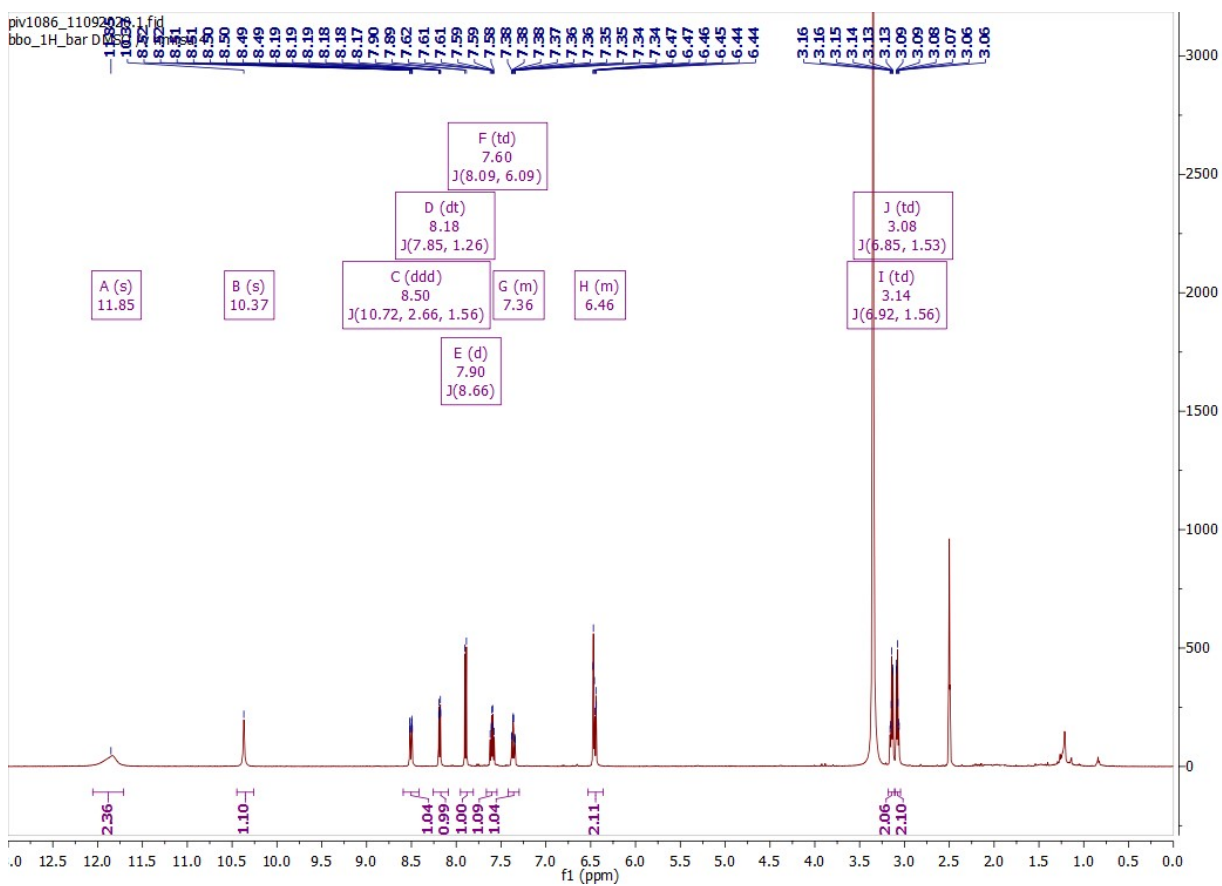
Chemical Formula: C₂₁H₁₄FN₃O₄S

Exact Mass: 423,0689

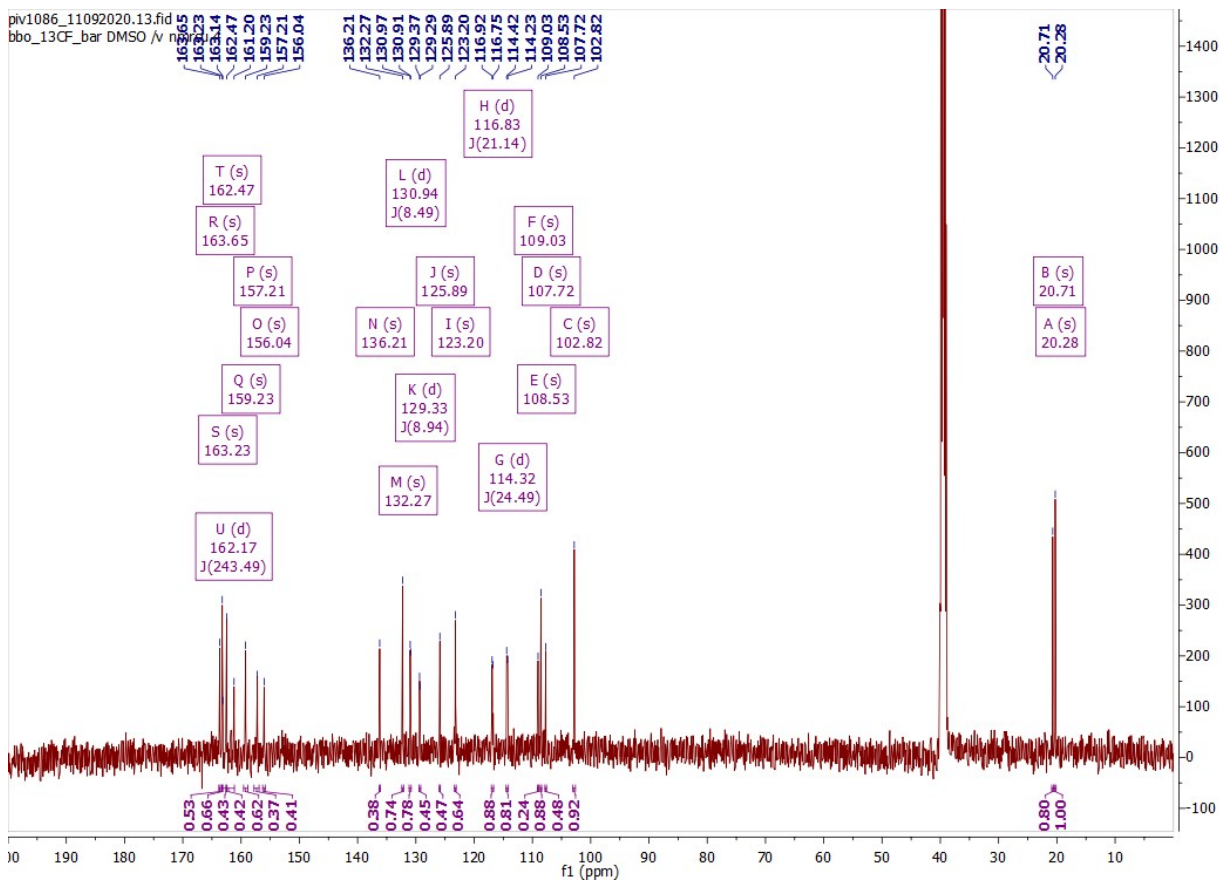
Molecular Weight: 423,4184

8n

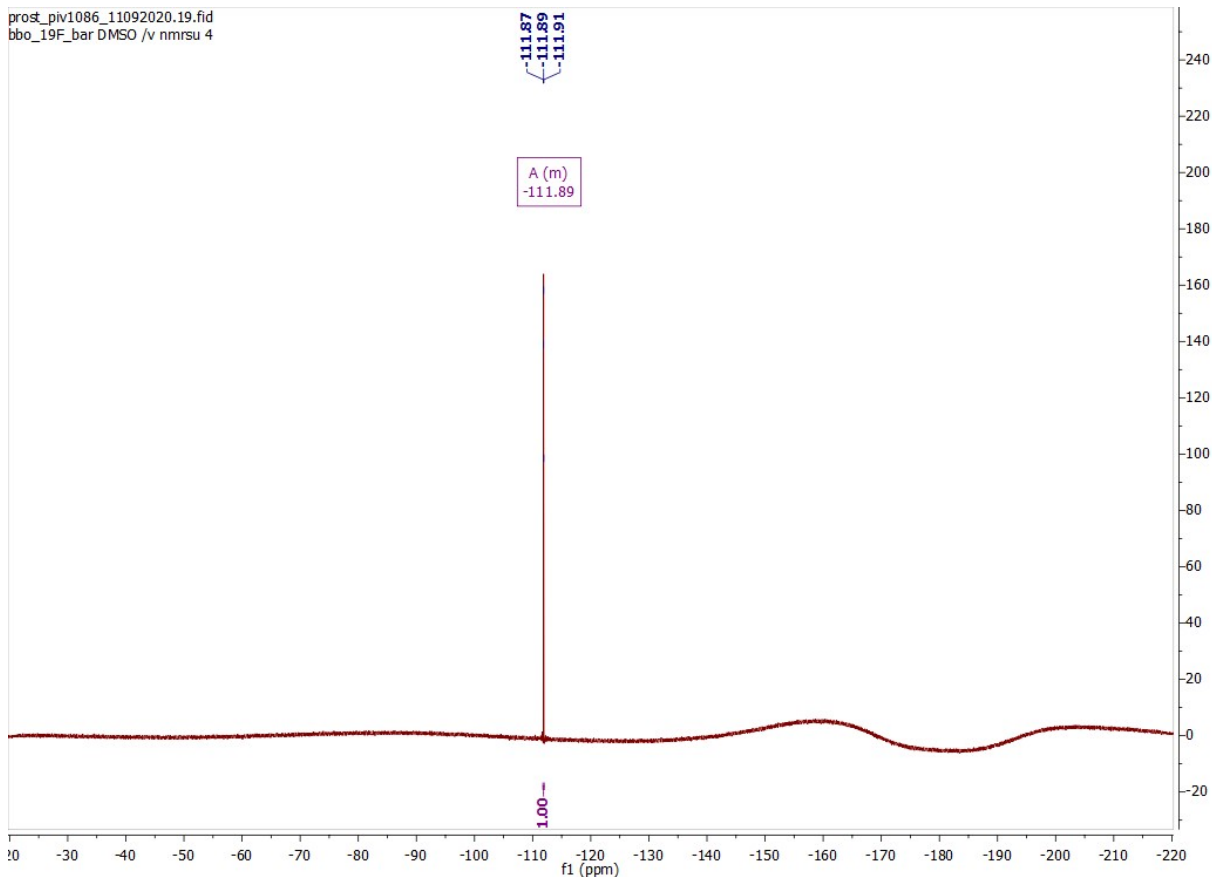
¹H spectra of **8n**:

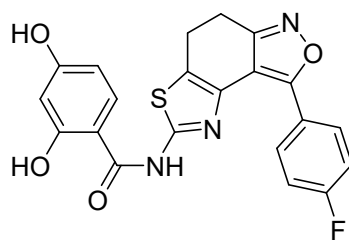


¹³C spectra of **8n**:



¹⁹F NMR spectra of **8n**:

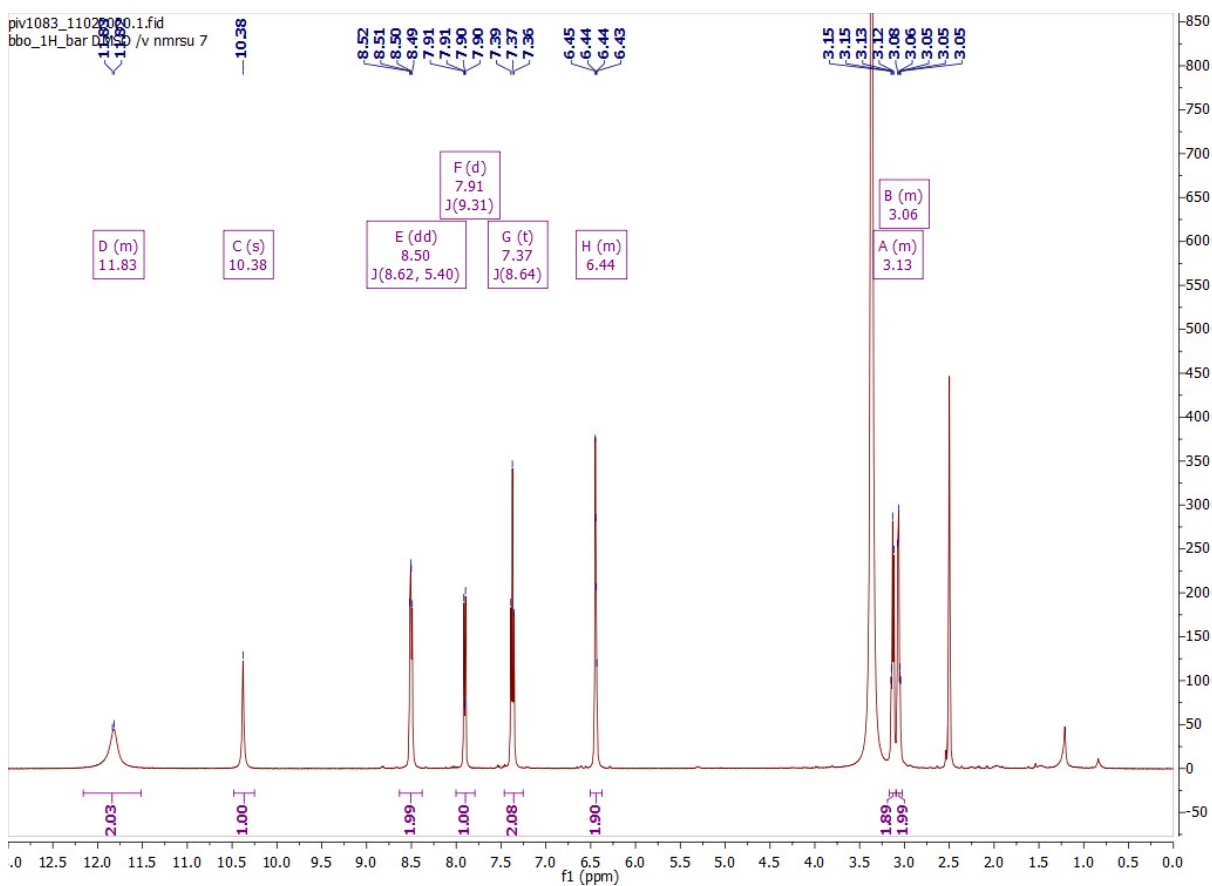




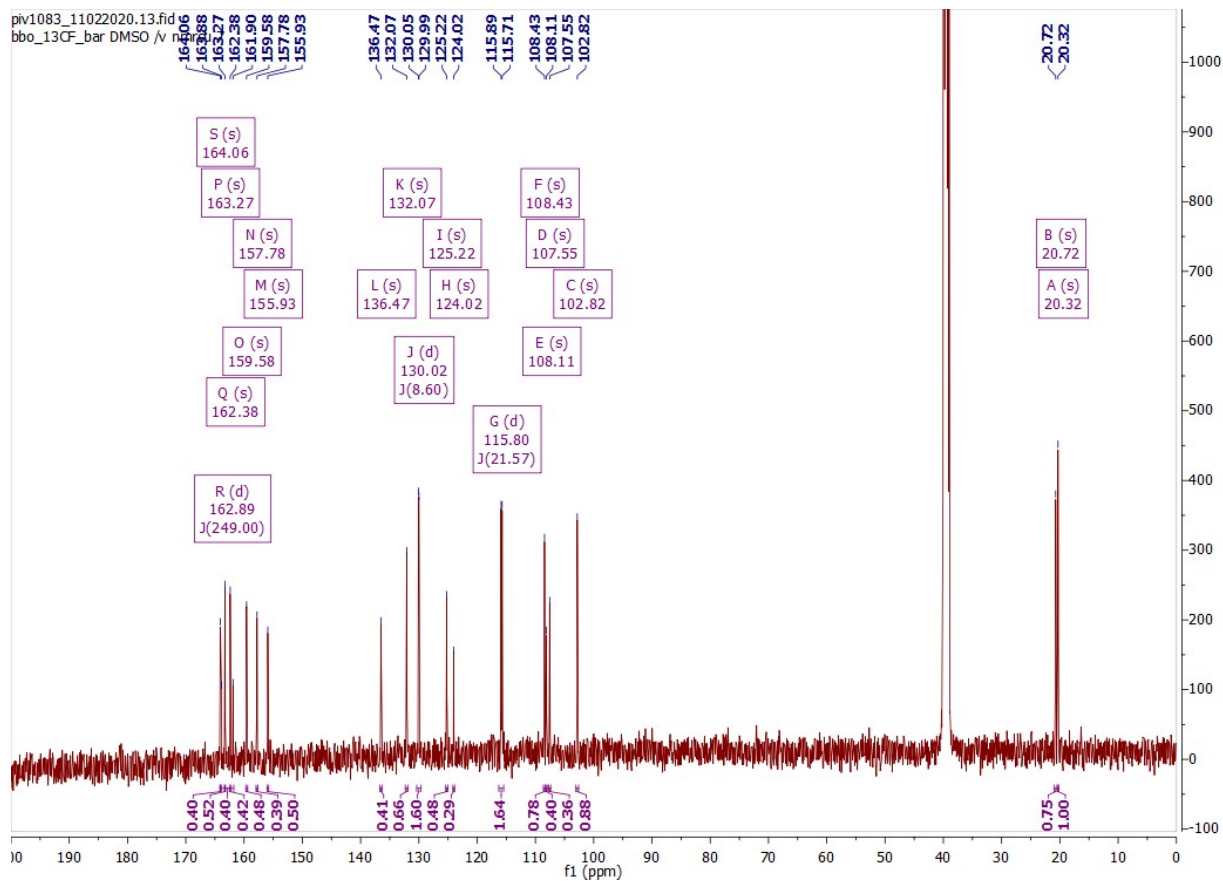
N-(8-(4-fluorophenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-c]isoxazol-2-yl)-2,4-dihydroxybenzamide
 Chemical Formula: C₂₁H₁₄FN₃O₄S
 Exact Mass: 423,0689
 Molecular Weight: 423,4184

80

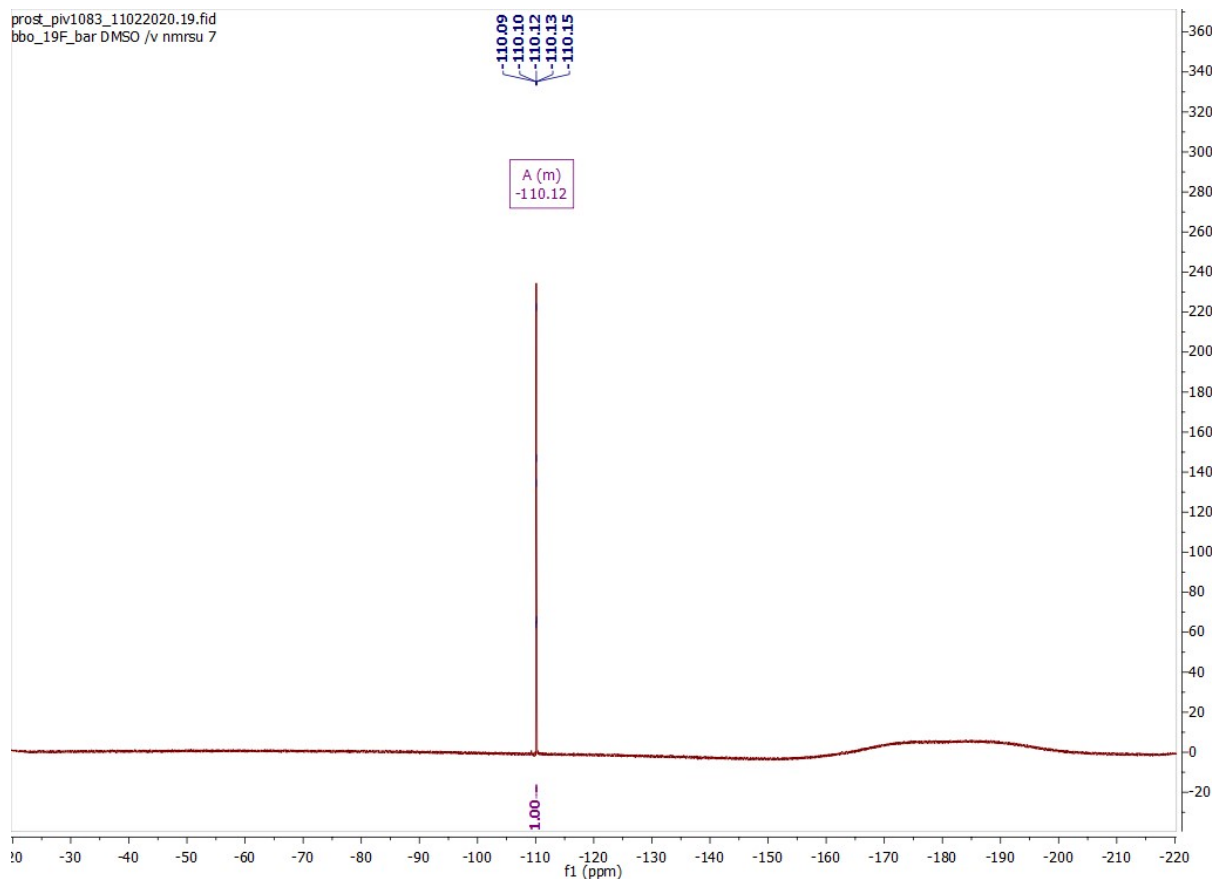
¹H spectra of **80**:

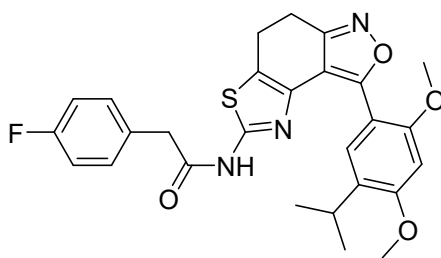


¹³C spectra of **8o**:



¹⁹F spectra of **8o**:

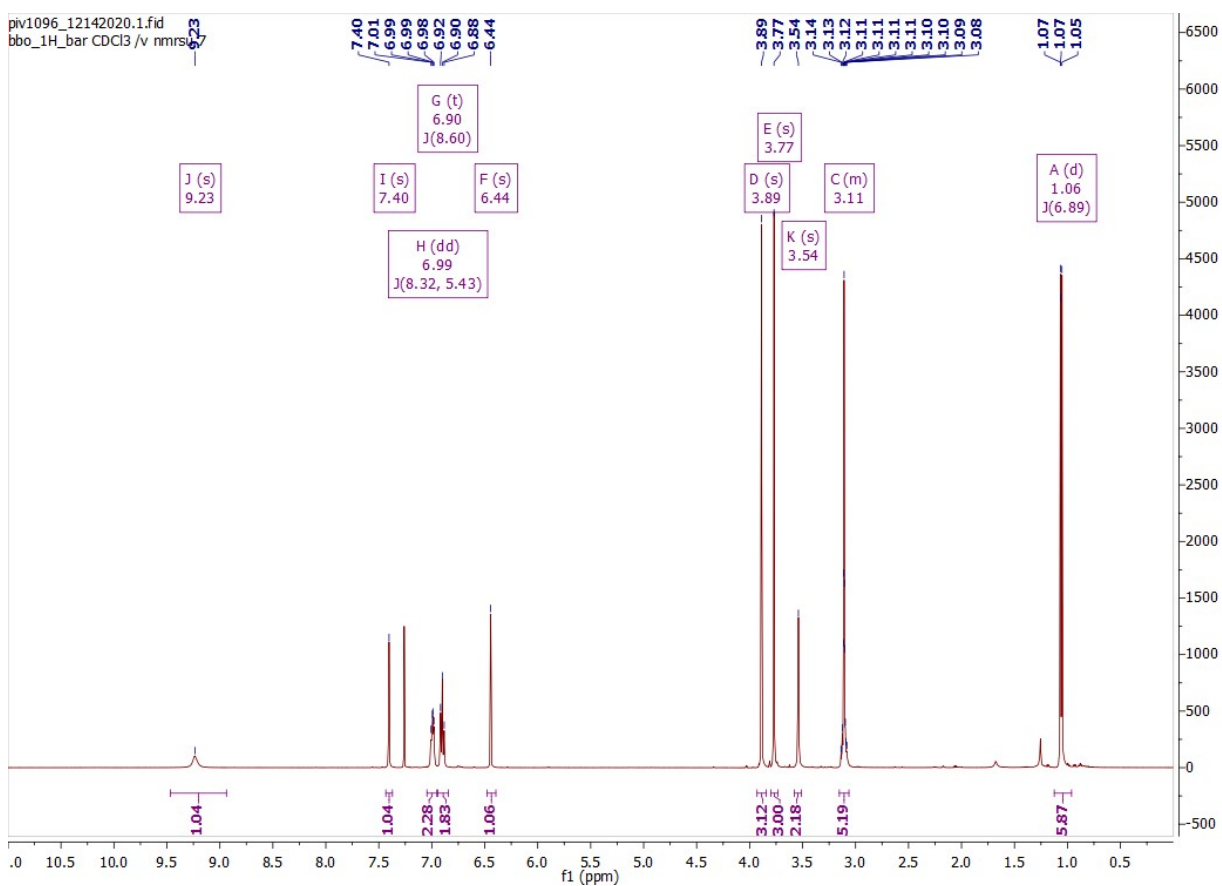


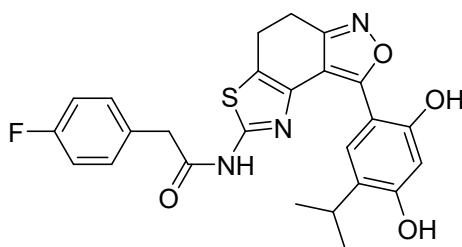


2-(4-fluorophenyl)-*N*-(8-(5-isopropyl-2,4-dimethoxyphenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-*c*]isoxazol-2-yl)acetamide
 Chemical Formula: C₂₇H₂₆FN₃O₄S
 Exact Mass: 507,1628
 Molecular Weight: 507,5804

8p

¹H spectra of **8p**:

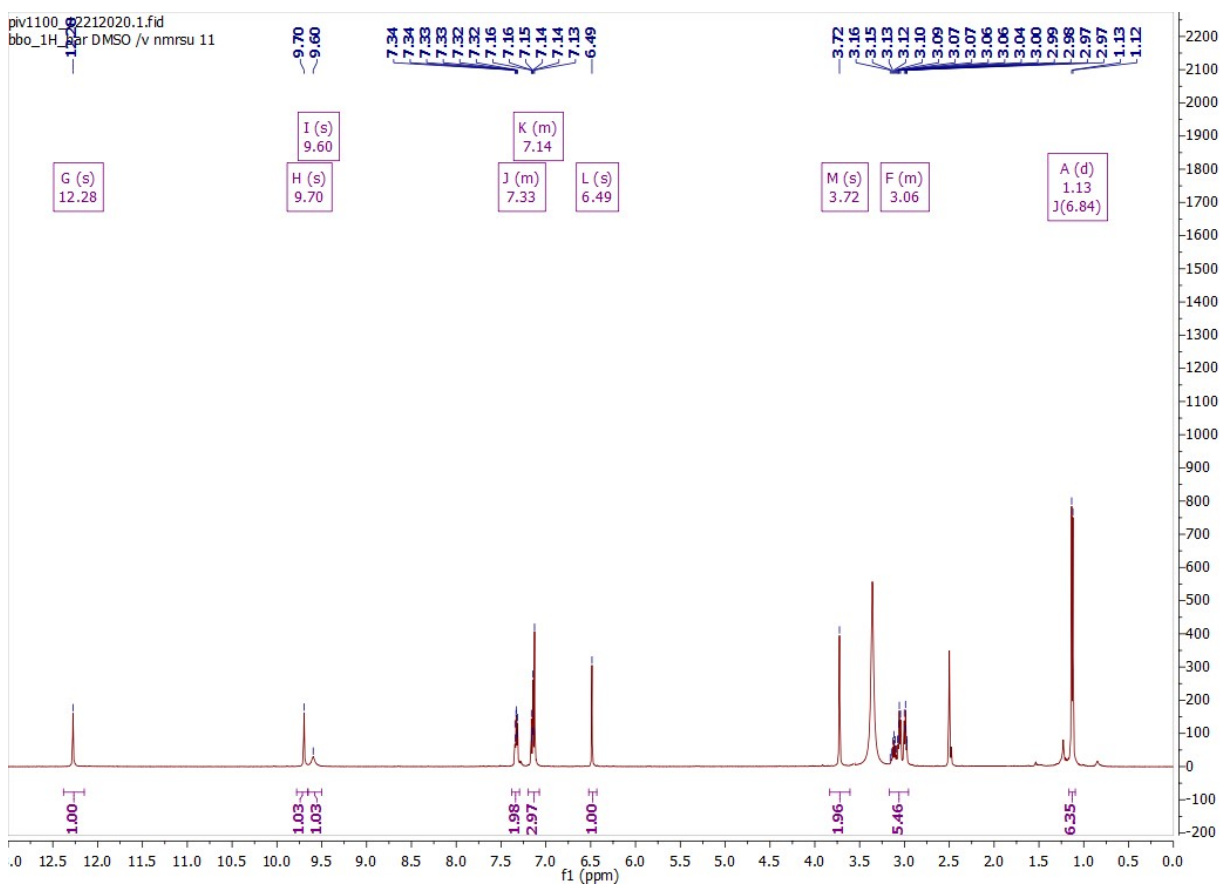




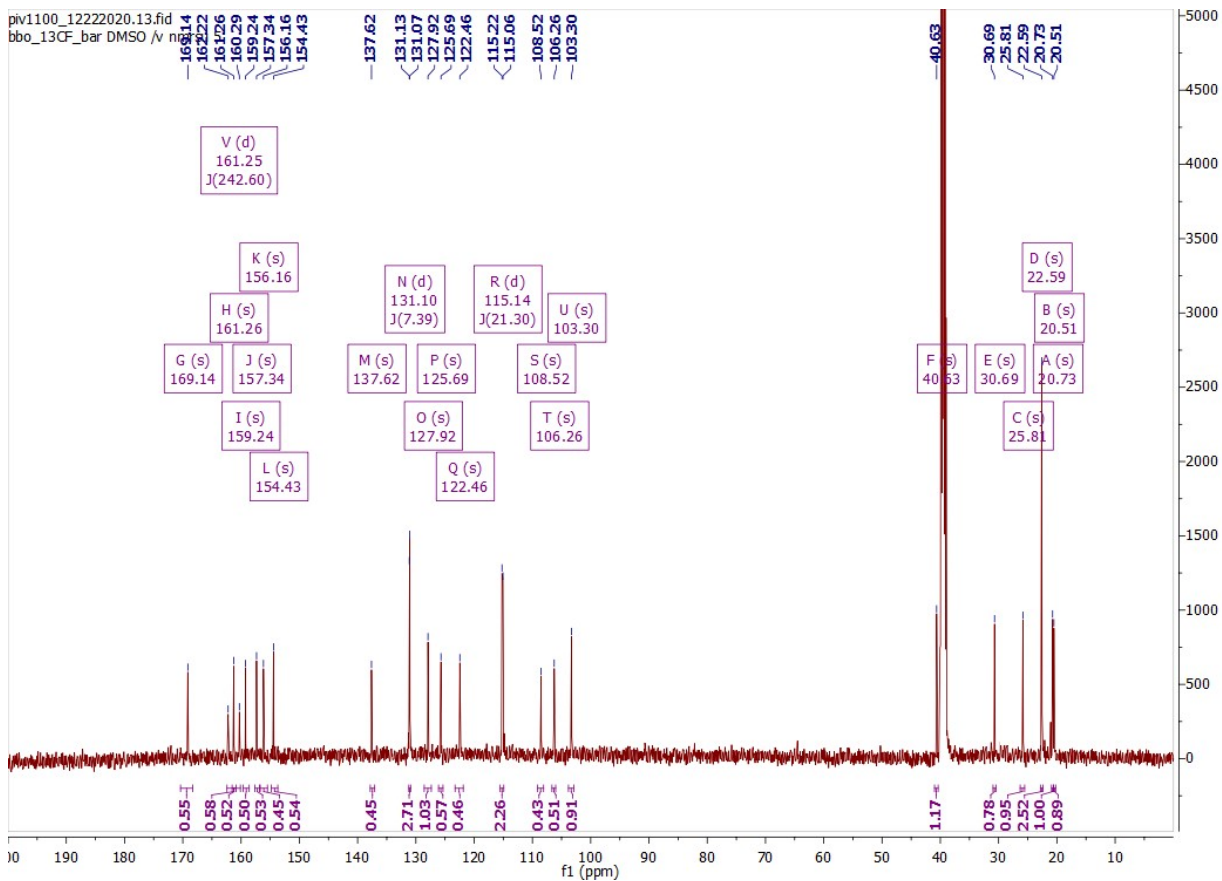
N-(8-(2,4-dihydroxy-5-isopropylphenyl)-4,5-dihydrothiazolo[4',5':3,4]benzo[1,2-c]isoxazol-2-yl)-2-(4-fluorophenyl)acetamide
 Chemical Formula: C₂₅H₂₂FN₃O₄S
 Exact Mass: 479,1315
 Molecular Weight: 479,5264

8q

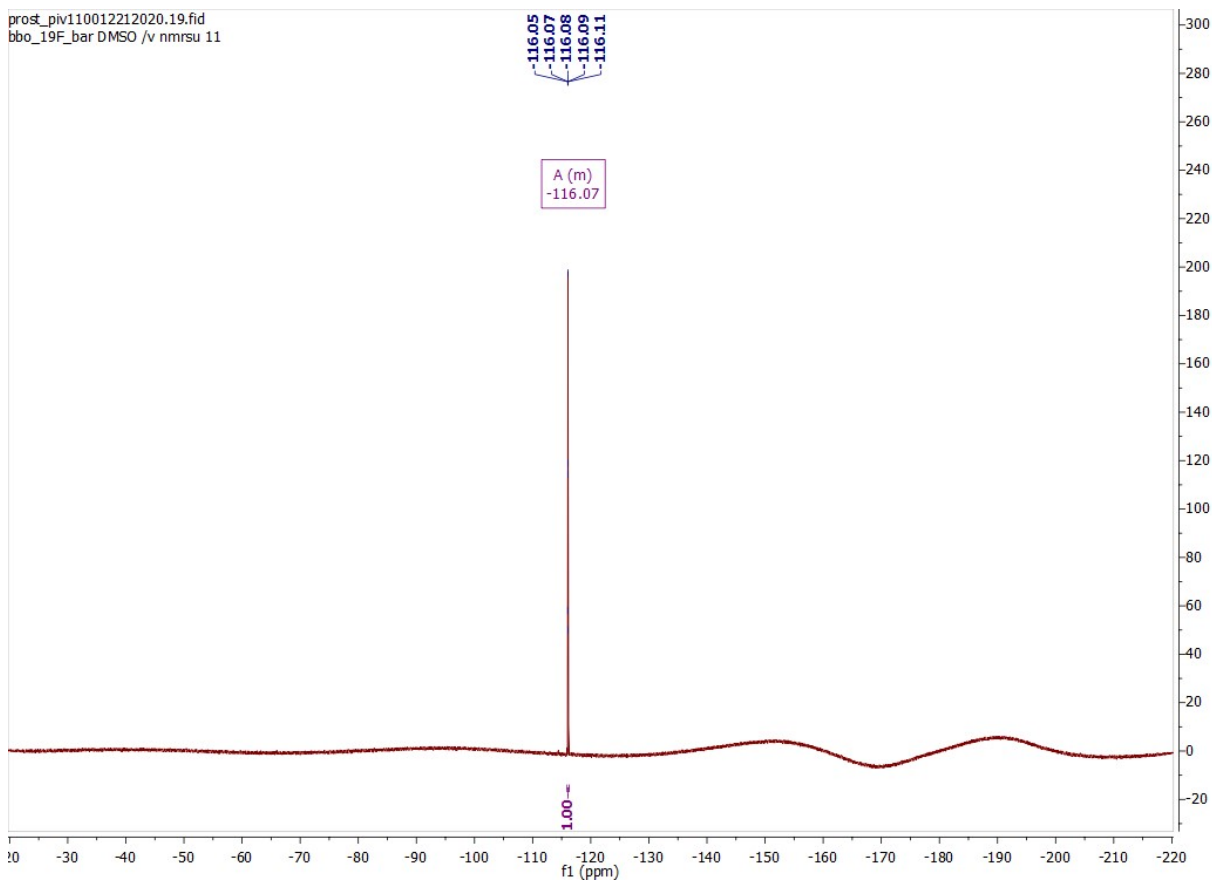
¹H spectra of 8q:

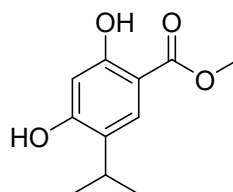


¹³C spectra of **8q**:



¹⁹F spectra of **8q**:





methyl 2,4-dihydroxy-5-isopropylbenzoate

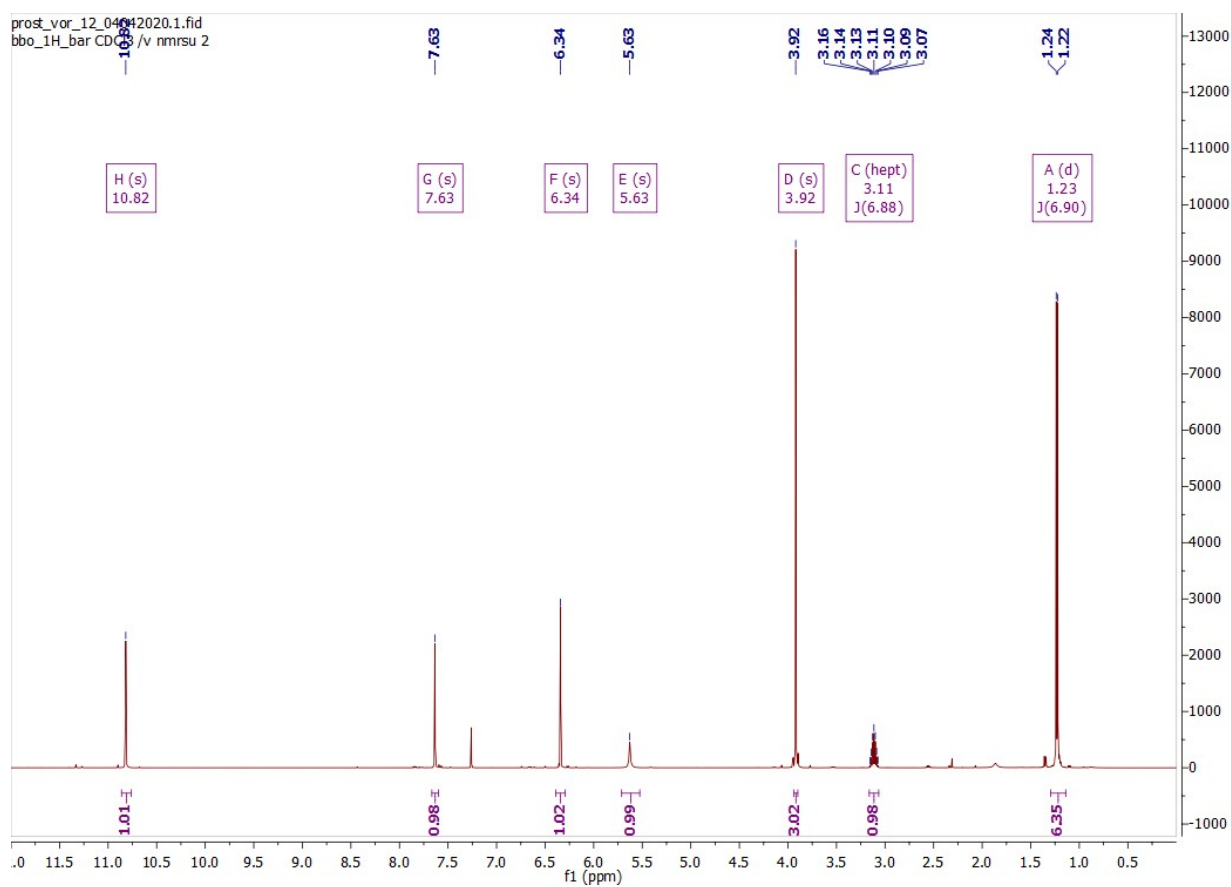
Chemical Formula: $C_{11}H_{14}O_4$

Exact Mass: 210,0892

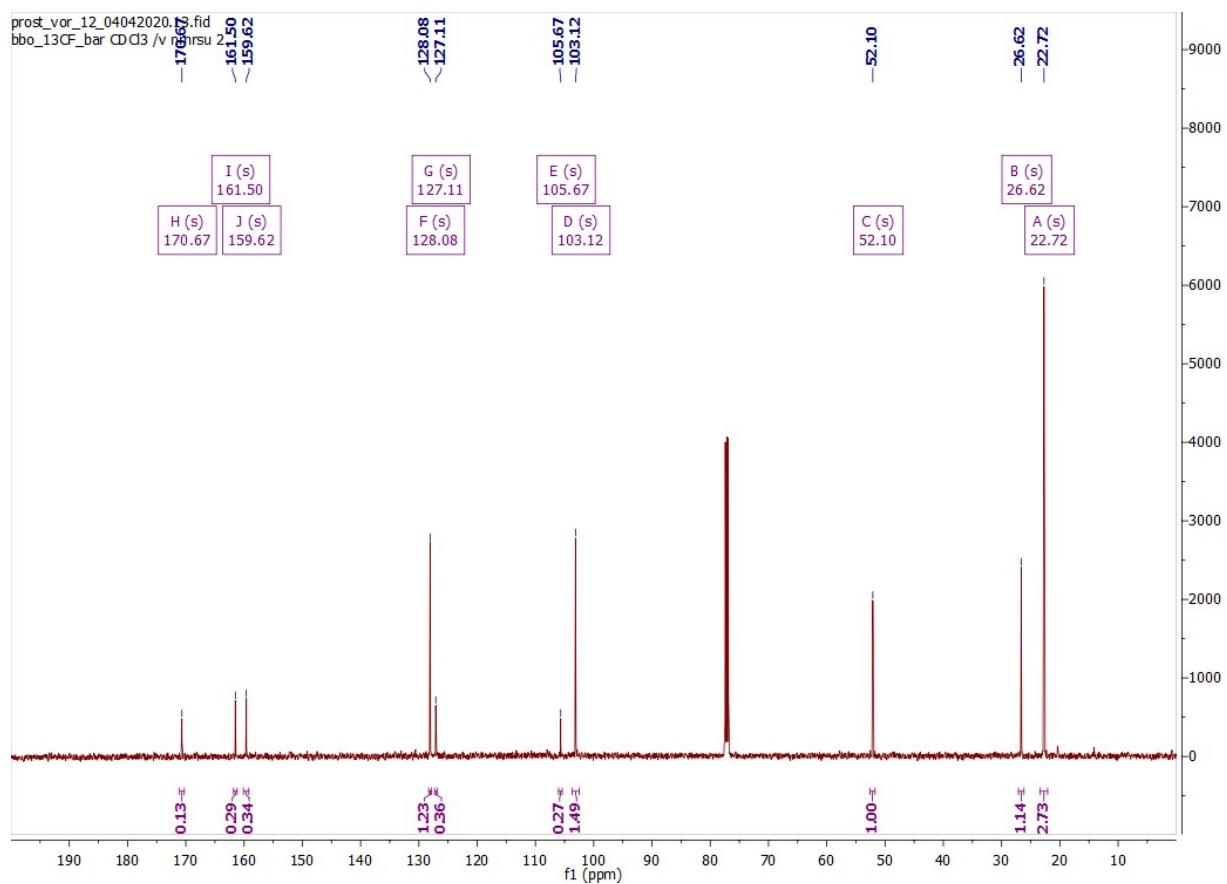
Molecular Weight: 210,2290

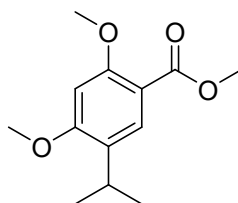
10

1H spectra of **10**:



¹³C spectra of 10:





methyl 5-isopropyl-2,4-dimethoxybenzoate

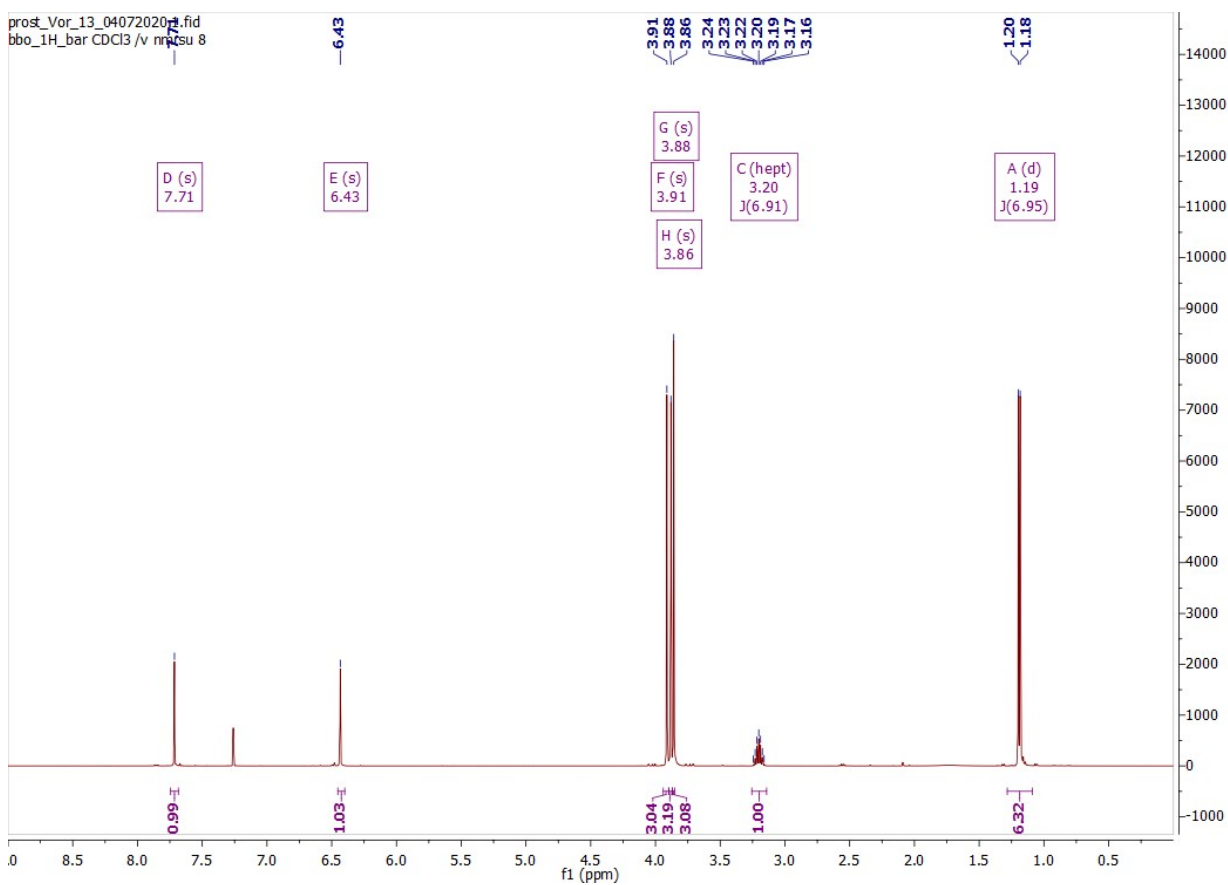
Chemical Formula: C₁₃H₁₈O₄

Exact Mass: 238,1205

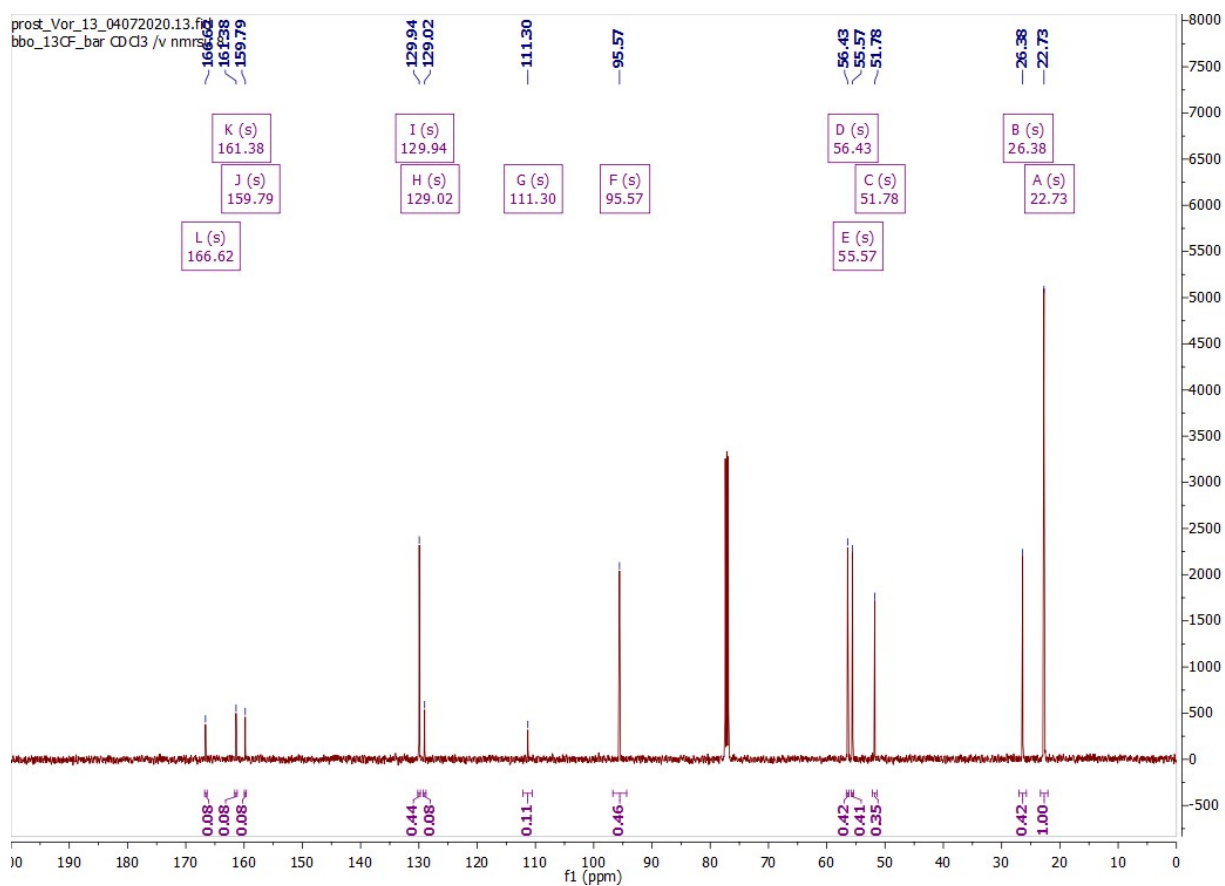
Molecular Weight: 238,2830

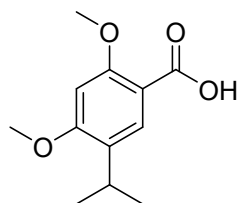
11

¹H spectra of **11**:



¹³C spectra of 11:





5-isopropyl-2,4-dimethoxybenzoic acid

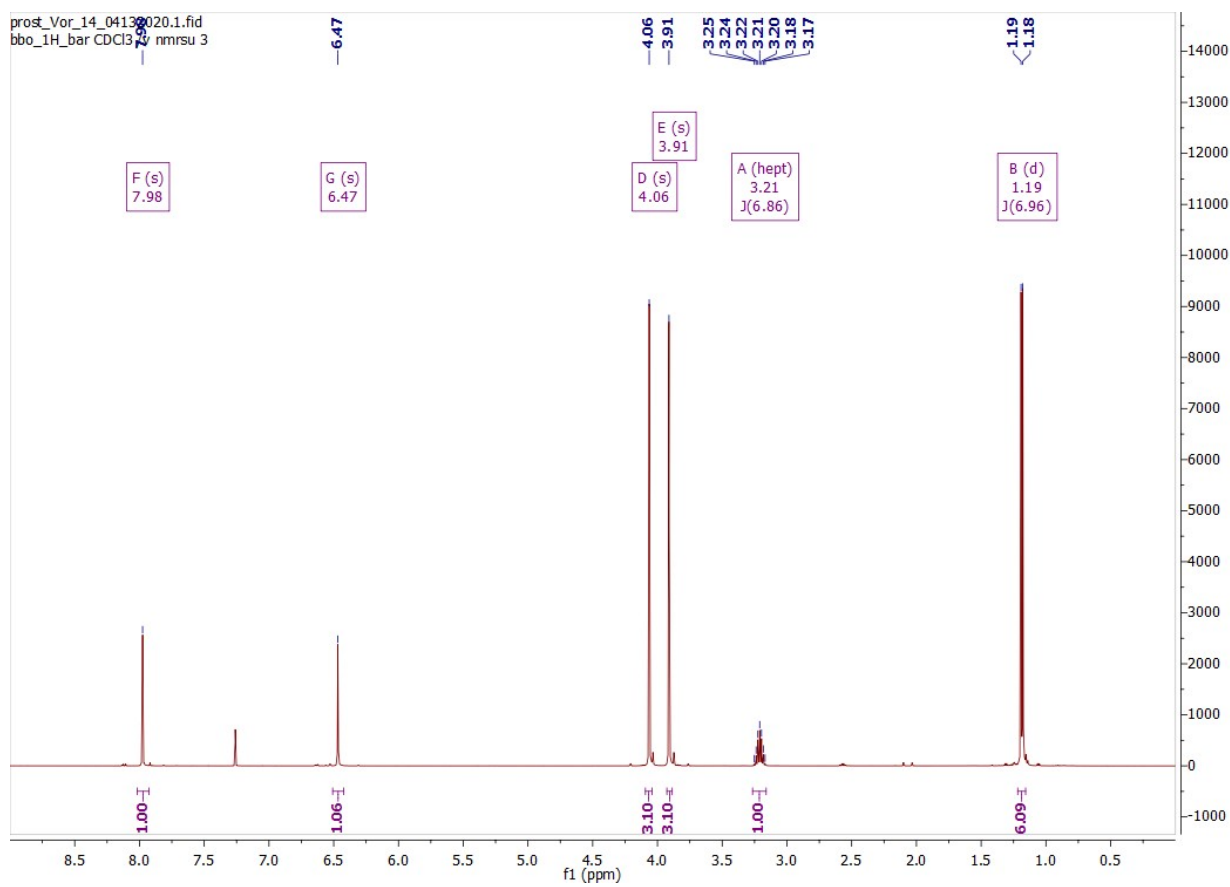
Chemical Formula: C₁₂H₁₆O₄

Exact Mass: 224,1049

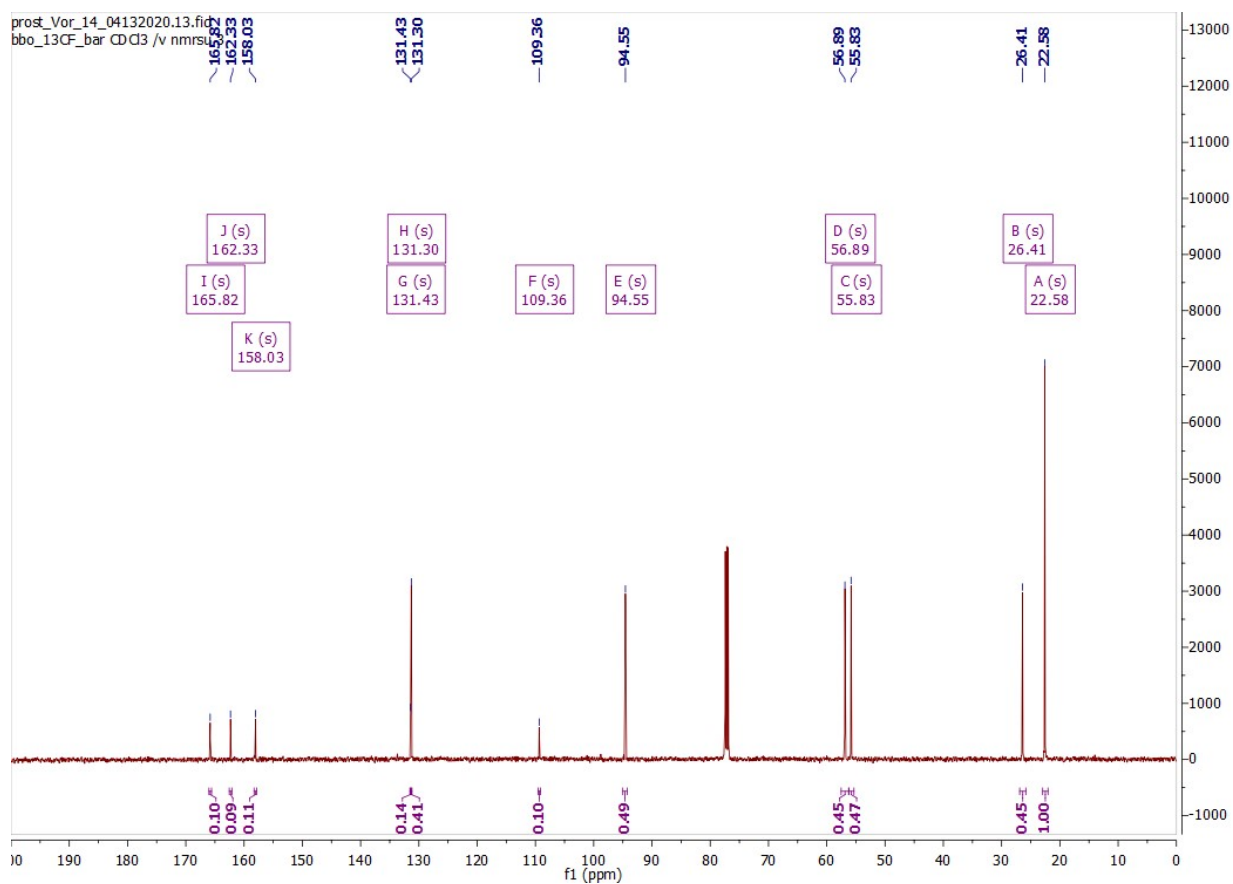
Molecular Weight: 224,2560

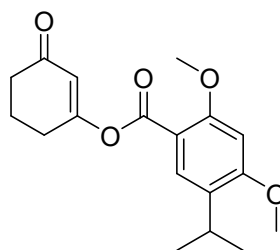
12

¹H spectra of 12:



¹³C spectra of 12:





3-oxocyclohex-1-en-1-yl 5-isopropyl-2,4-dimethoxybenzoate

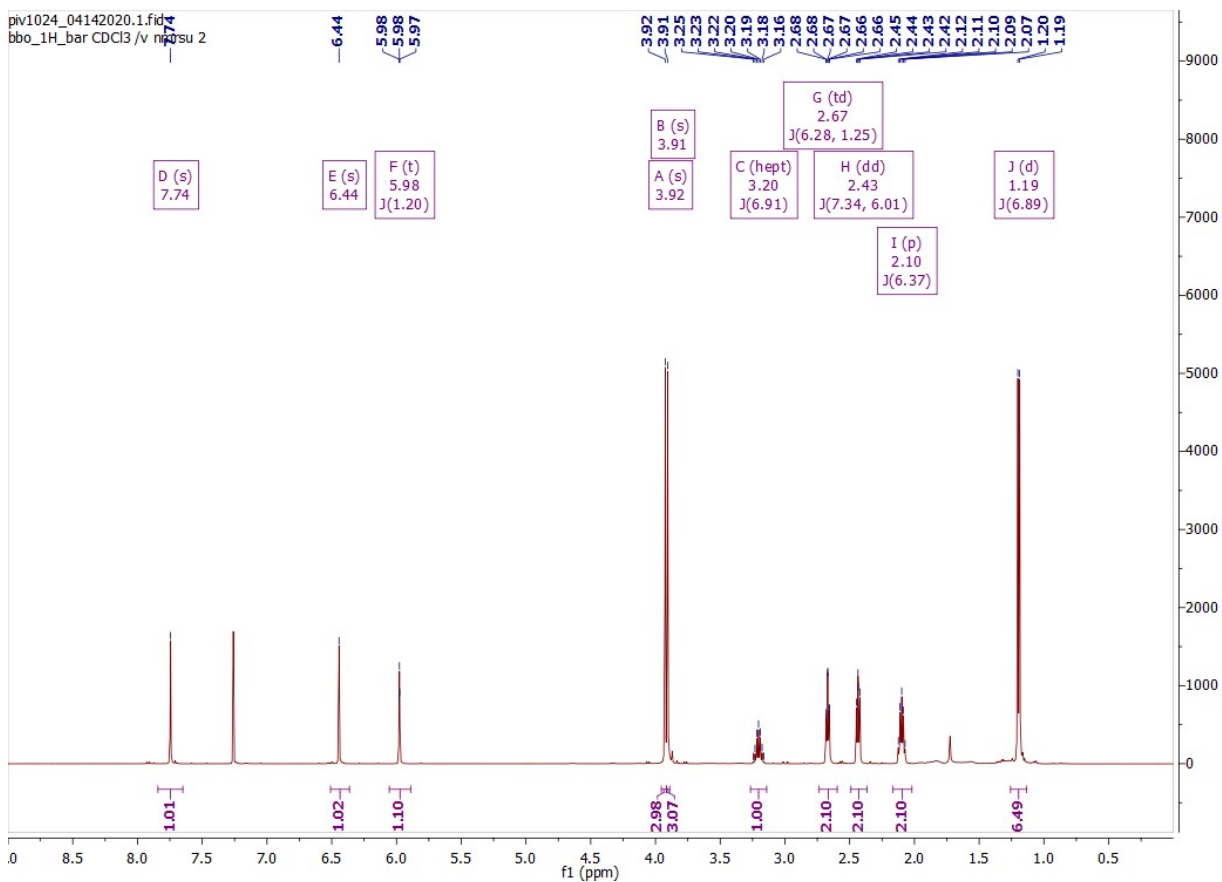
Chemical Formula: $C_{18}H_{22}O_5$

Exact Mass: 318,1467

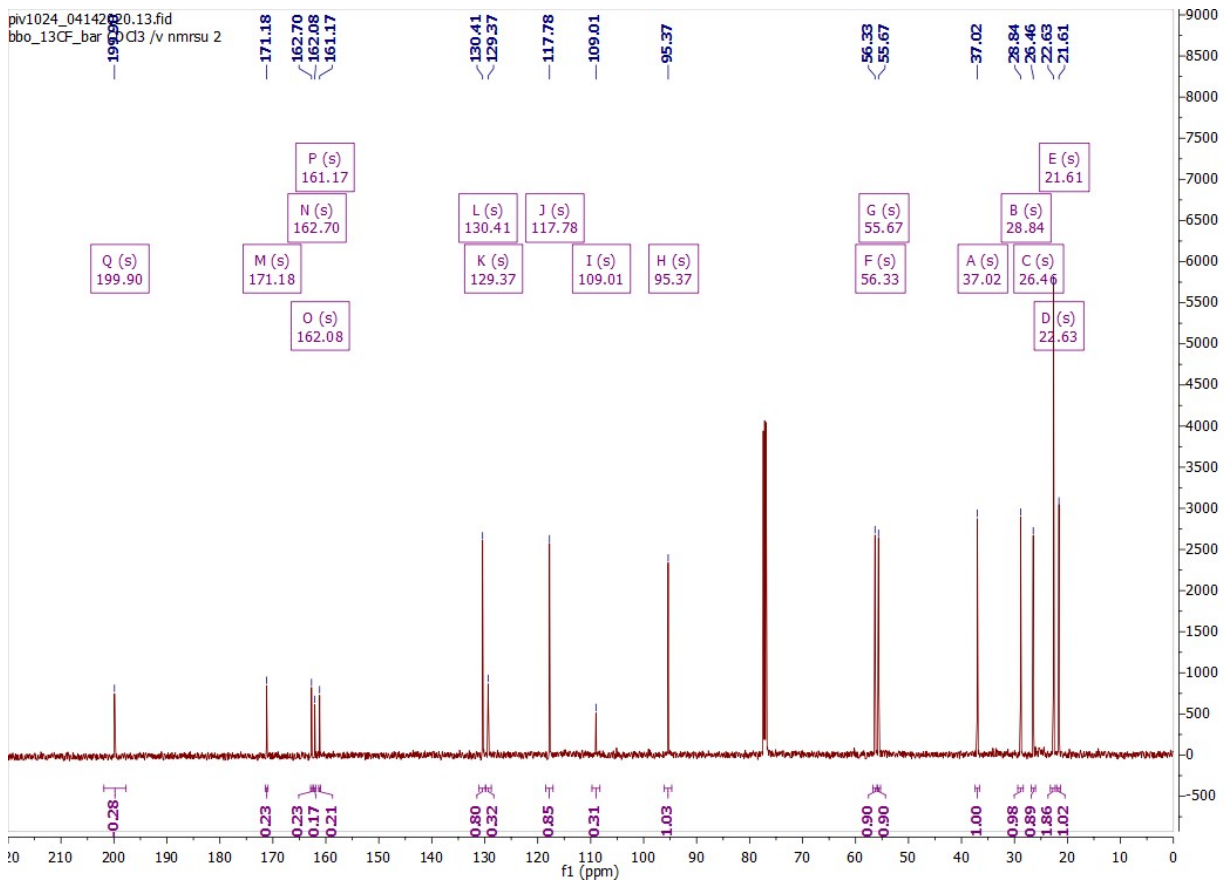
Molecular Weight: 318,3690

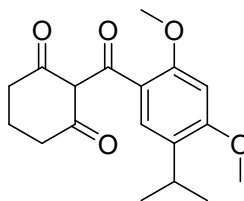
14

1H spectra of 13:



¹³C spectra of 14:





2-(5-isopropyl-2,4-dimethoxybenzoyl)cyclohexane-1,3-dione

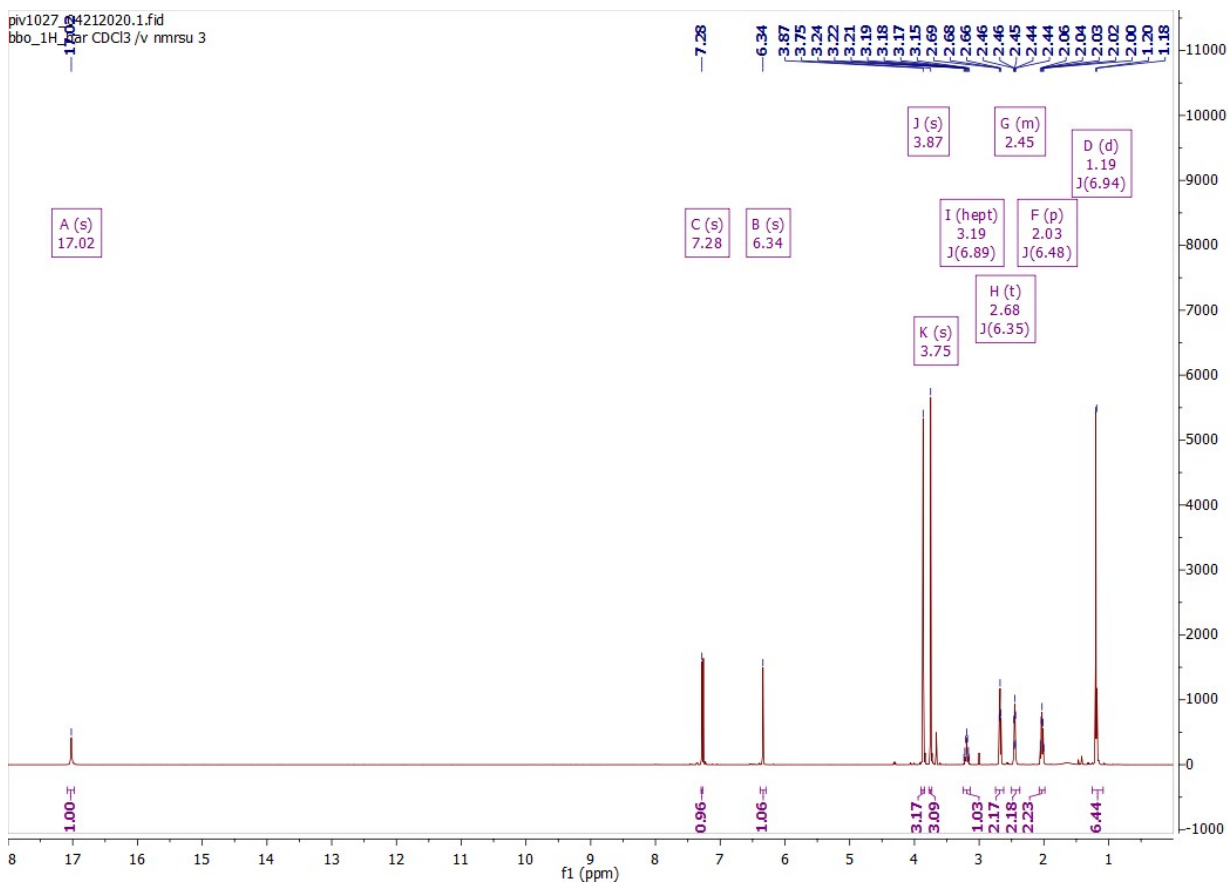
Chemical Formula: $C_{18}H_{22}O_5$

Exact Mass: 318,1467

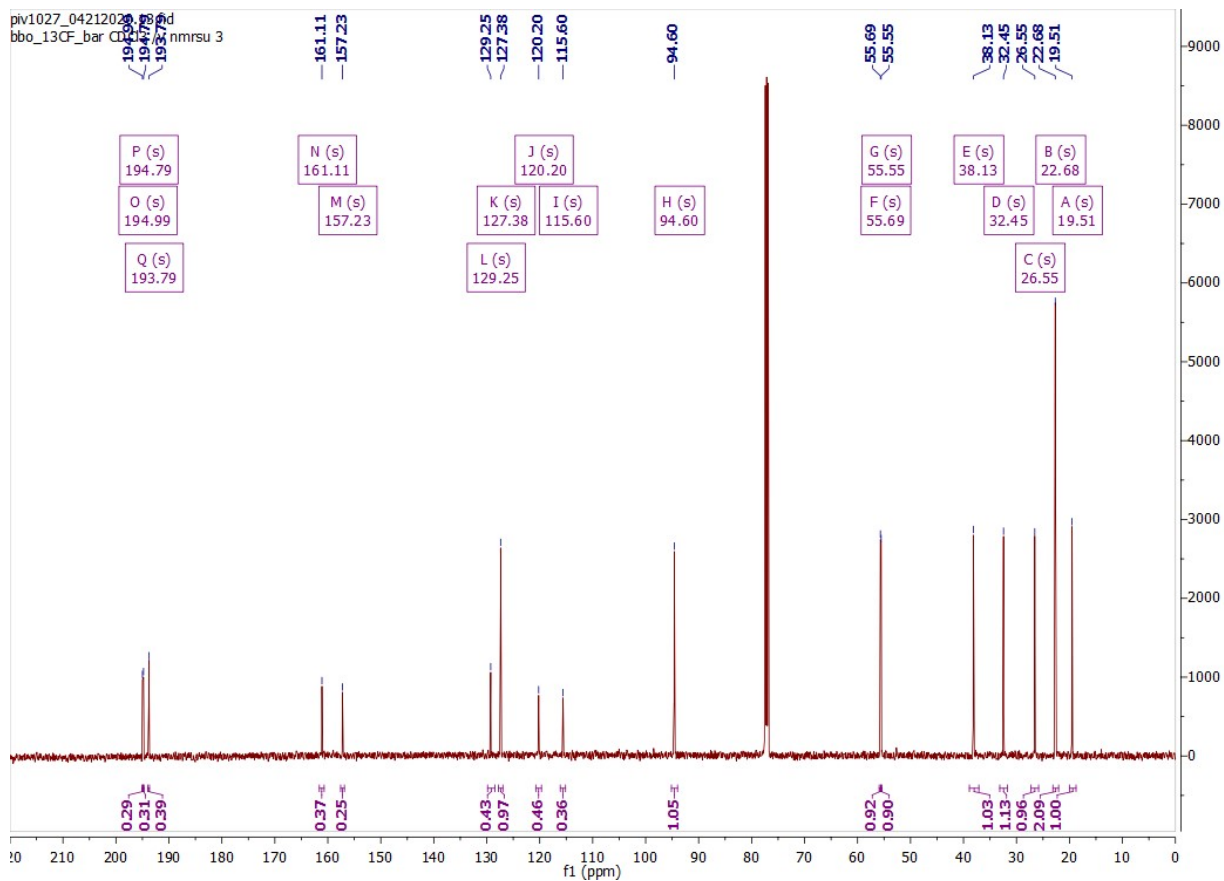
Molecular Weight: 318,3690

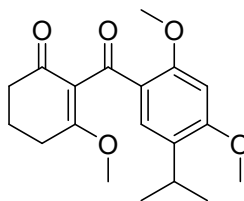
15

1H spectra of 15:



¹³C spectra of 15:





2-(5-isopropyl-2,4-dimethoxybenzoyl)-3-methoxycyclohex-2-en-1-one

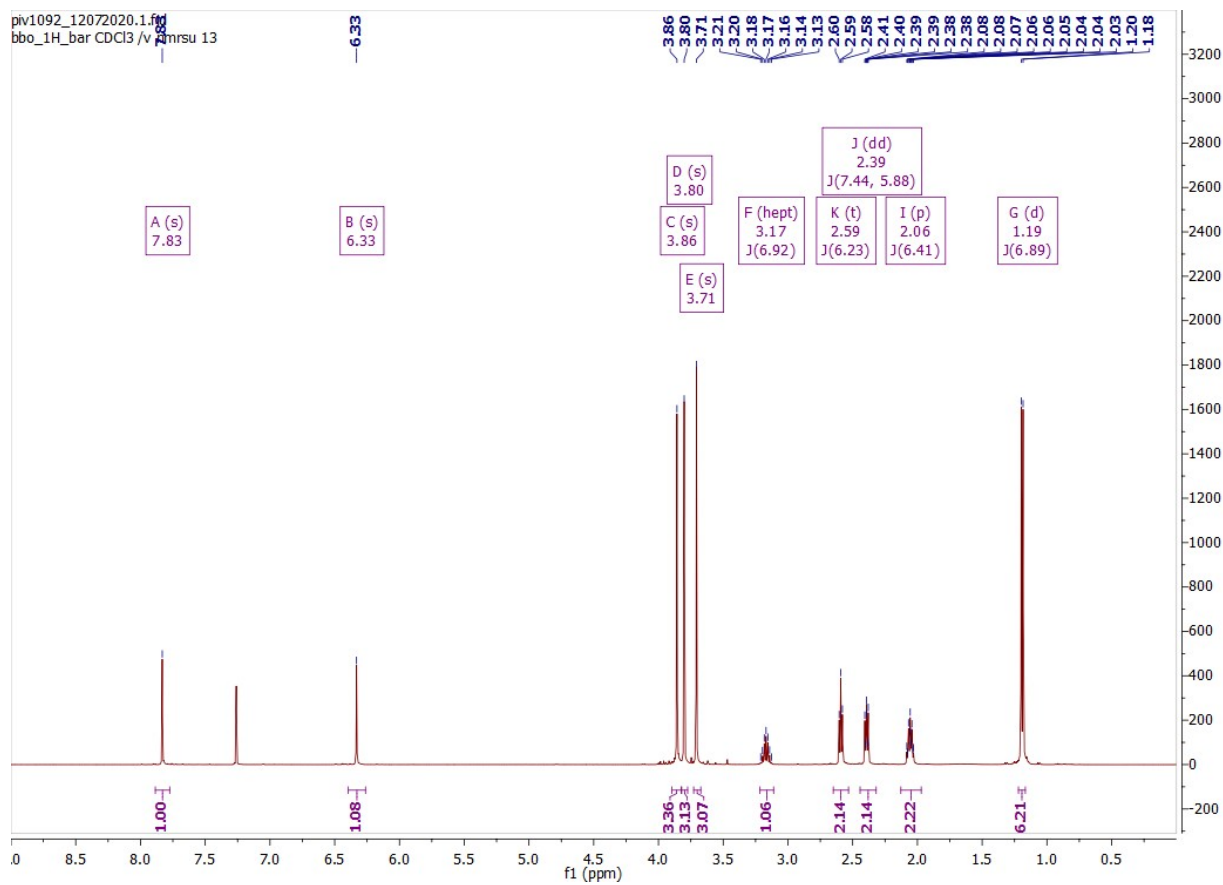
Chemical Formula: C₁₉H₂₄O₅

Exact Mass: 332,1624

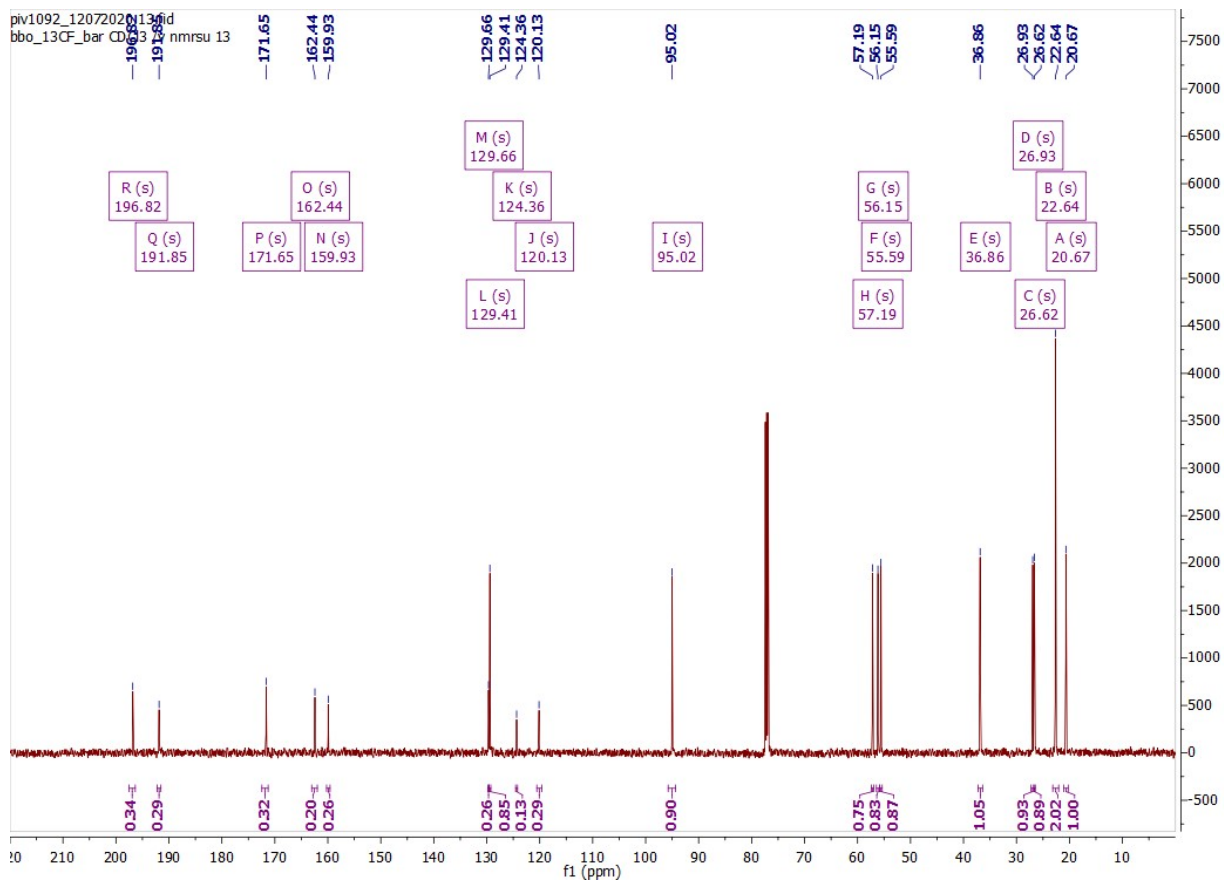
Molecular Weight: 332,3960

16

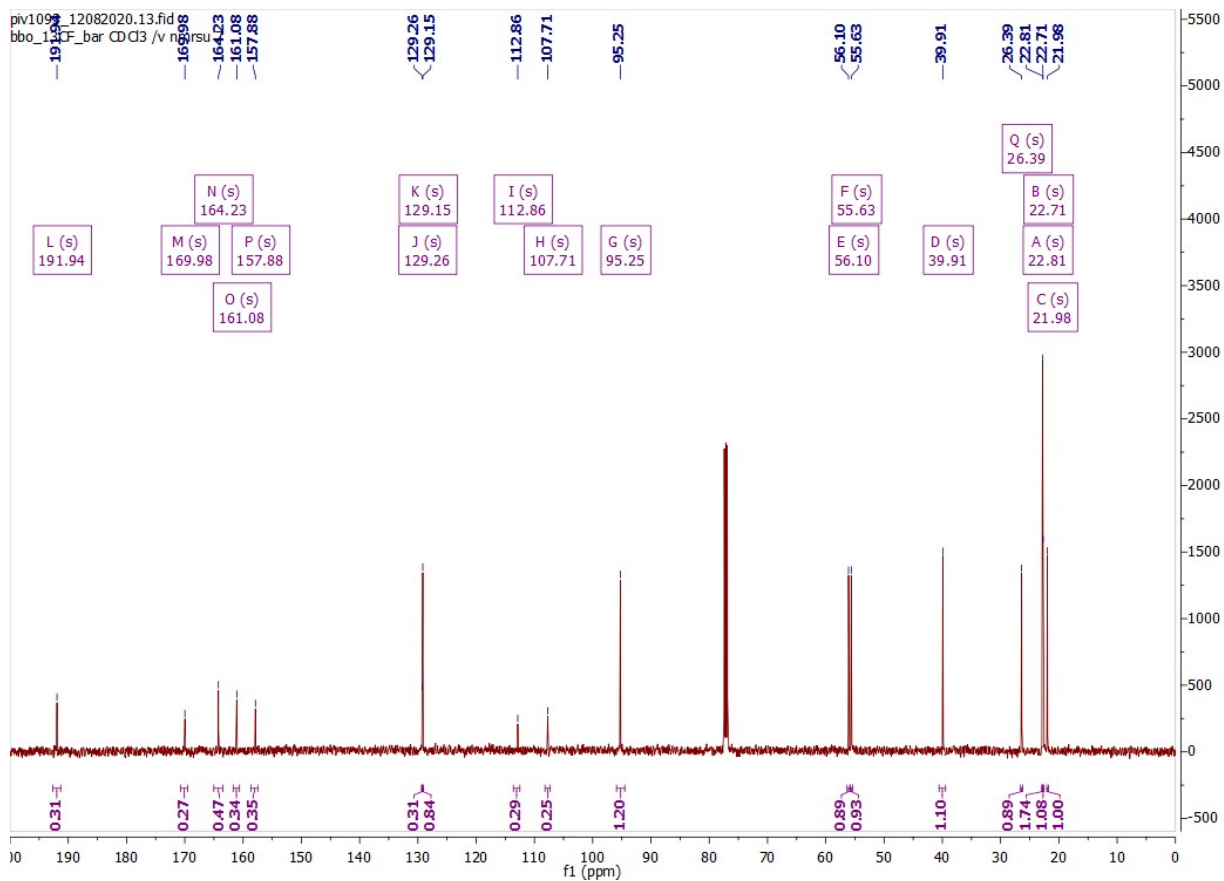
¹H spectra of 16:



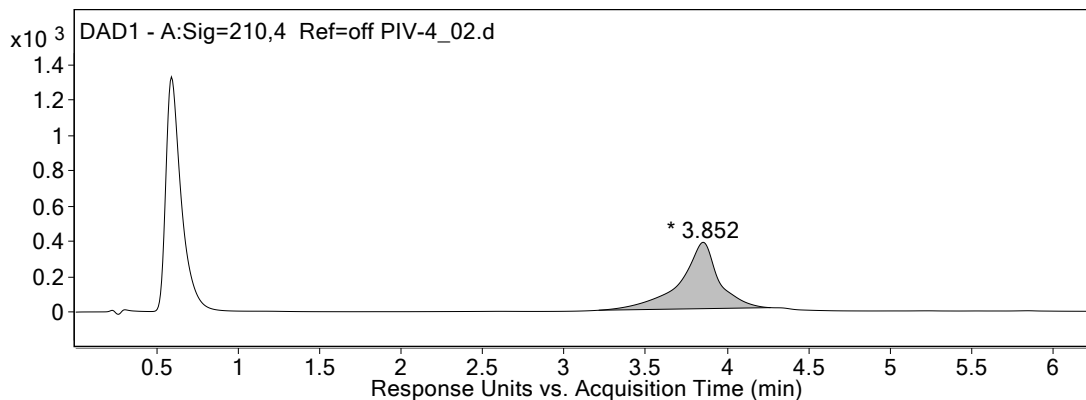
¹³C spectra of 16:



¹H spectra of 5d:



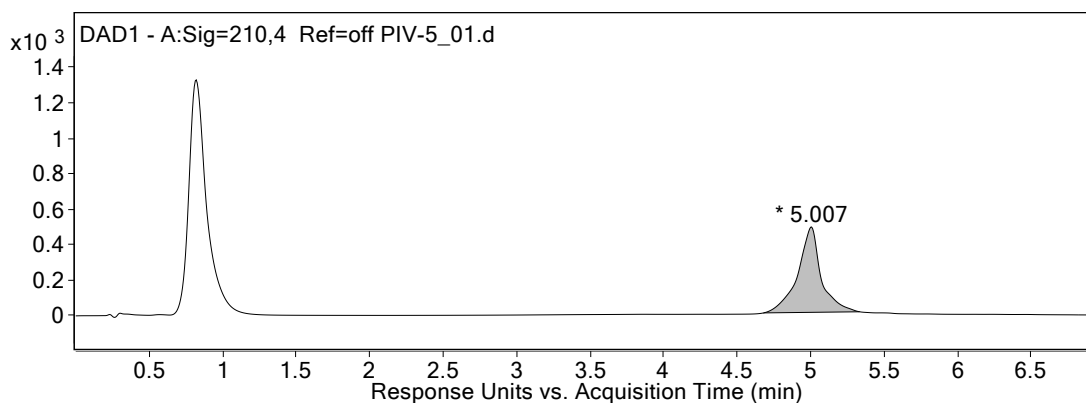
HPLC of 8b



Integration Peak List

Peak	Start	RT	End	Height	Area	Area %
1	3.212	3.852	4.266	376.73	6144.71	100

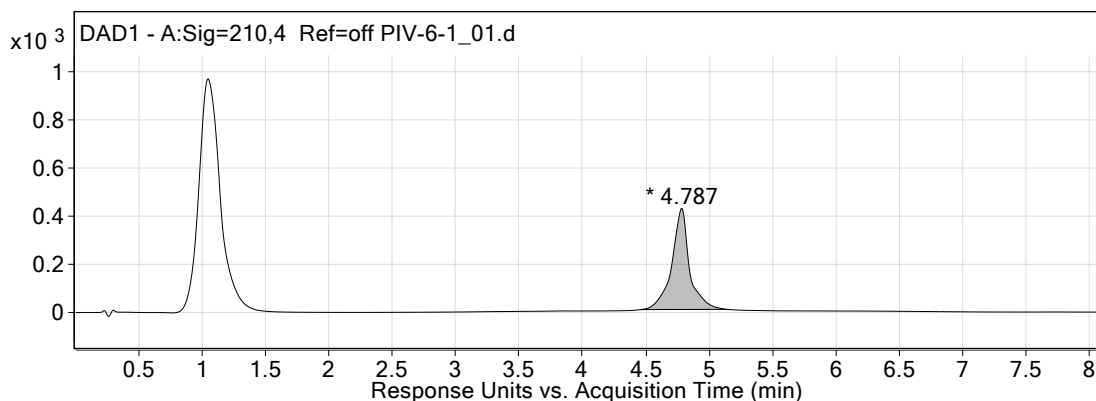
HPLC of 8c



Integration Peak List

Peak	Start	RT	End	Height	Area	Area %
1	4.681	5.007	5.334	481.3	5310.47	100

HPLC of 8q

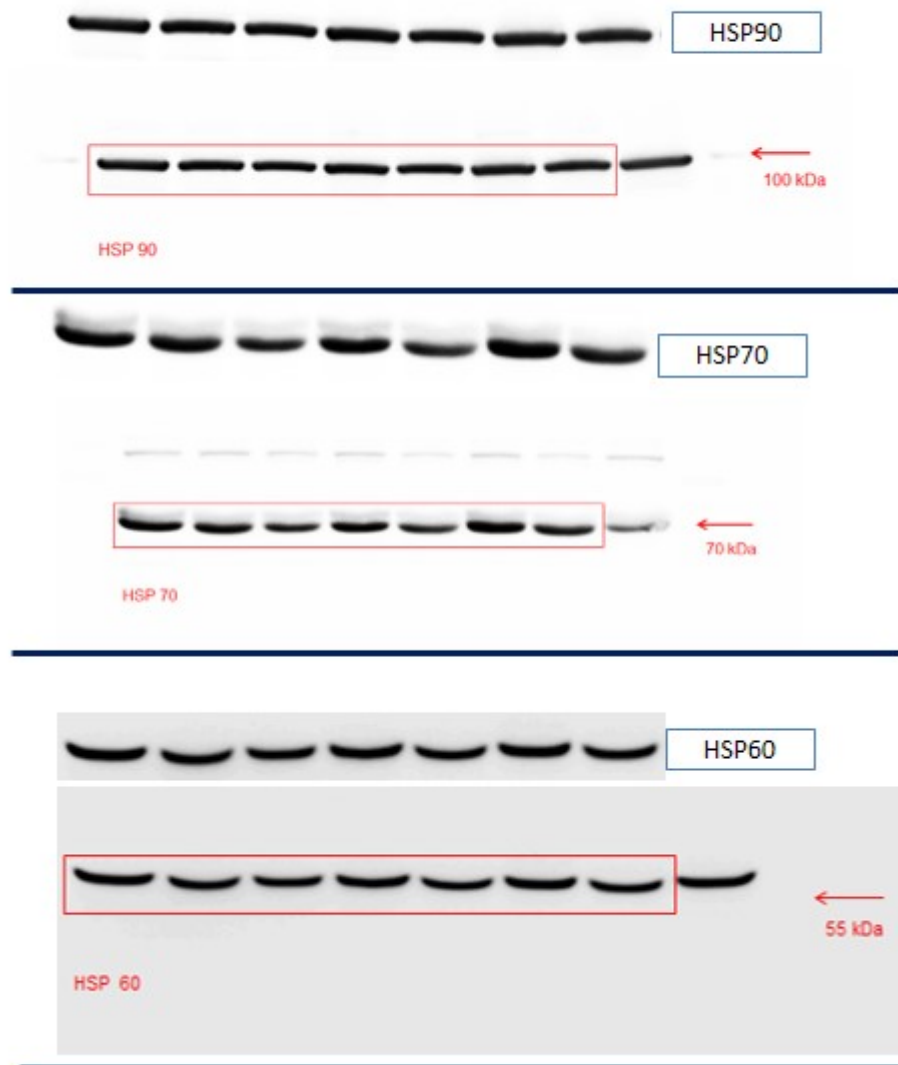


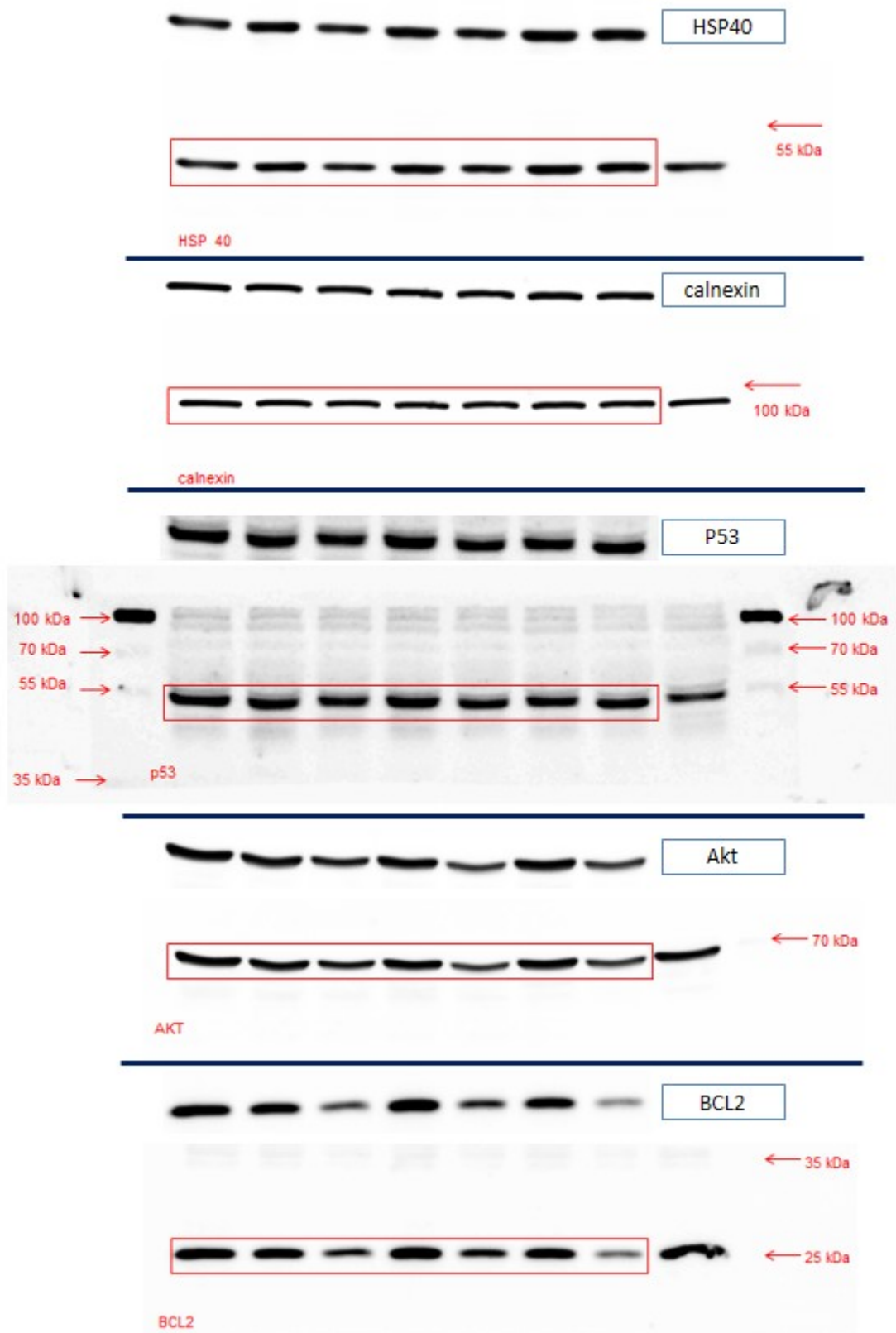
Integration Peak List

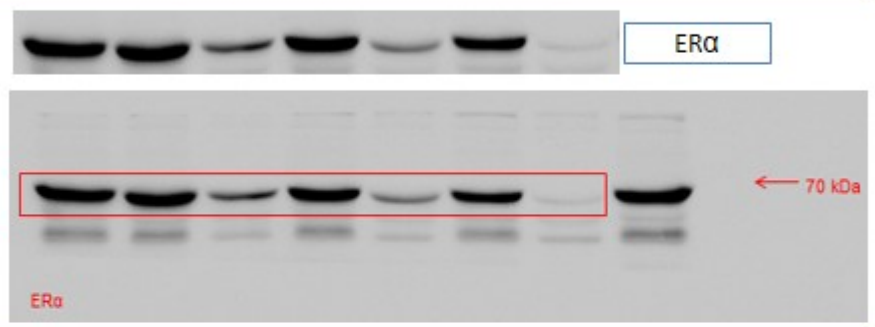
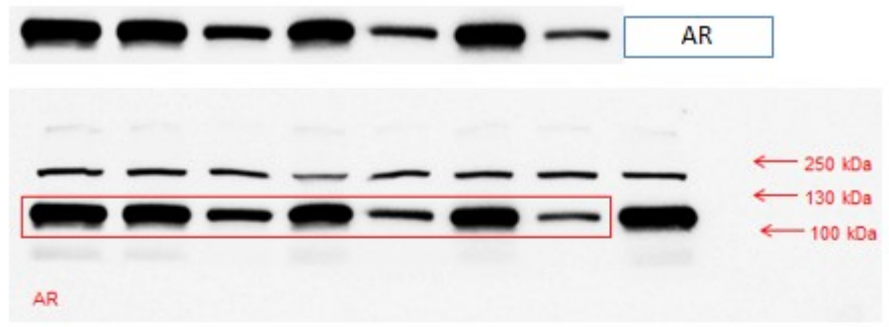
Peak	Start	RT	End	Height	Area	Area %
1	4.46	4.787	5.14	419.6	4066.23	100

Western blots

Uncropped images







References

1. A. A. Akhrem, F. A. Lakhvich, S. I. Budai, T. S. Khlebnicova and I. I. Petrusevich, *Synthesis*, 1978, **1978**, 925-927.
2. T. S. Khlebnikova, V. G. Isakova, A. V. Baranovskii, F. A. Lakhvich and A. S. Lyakhov, *Russian Journal of General Chemistry*, 2007, **77**, 1724-1731.
3. Y. Tamura, M. Inoue, A. Wada, M. Fujita and Y. Kita, *Chemical and Pharmaceutical Bulletin*, 1981, **29**, 3226-3231.