Electronic Supplementary Material (ESI) for Organic and Biomolecular Chemistry

Design, synthesis and photoactivation studies of fluorous photolabels

Arun Babu Kumar, Jordan Micheal Anderson and Roman Manetsch*

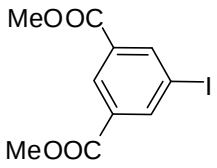
Department of Chemistry, University of South Florida, CHE205, 4202 East Fowler Ave, Tampa, Florida 33620

manetsch@usf.edu

Electronic Supplementary Information

Table of Content

Experimental Procedures



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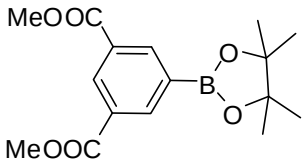
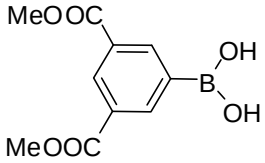
Dimethyl 5-iodoisophthalate (9): To a solution of dimethyl-5-aminoisophthalate (8.0 g, 38.2 mmol) in 20% hydrochloric acid (23 ml) a solution of $NaNO_2$ (2.7 g, 39.1 mmol) in water (46 ml) was added at -5°C. To this reaction mixture toluene (62 ml) was added followed by a solution of potassium iodide (13 g, 78.3 mmol) in water (31 ml) was added slowly and brought to room temperature and stirred for 12 hours and then refluxed for 1 hour. The reaction mixture was cooled to room temperature and extracted with toluene and washed with water. The organic layer was dried with anhydrous sodium sulfate and concentrated followed by recrystalization (with methanol) to give **9** (7.32 g, 60%) as pale yellow solid. R_f = 0.31 (9:1, hexanes and ethyl acetate). Mp: 89-92°C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.59 (s, 1H), 8.51 (s, 2H), 3.92 (s, 6H). ^{13}C NMR (100 MHz, $CDCl_3$): δC (ppm) 165.0, 142.6, 132.4, 130.0, 93.6, 52.8. HRMS (ESI^+) for $[M+H]^+$; calculated: 320.96183, found: 320.96172 (error m/z = -0.35 ppm).



Dimethyl 5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)isophthalate: 9 (15.7 g, 49.2 mmol) and dimethyl sulfoxide (150 ml) were added to a flask charged with [1,1'-bis(diphenylphosphino)ferrocene] dichloropalladium(II) (1.08 g, 1.5 mmol) and bis(pinacolato)diboron (14.99 g, 59.0 mmol), and potassium acetate (14.48 g, 147 mmol) and stirred at 80°C for 2 hours. The reaction mixture was extracted with toluene and washed with water. The resulting organic layer was dried over anhydrous sodium sulfate and concentrated under vacuum, without further purification it was used for the next step. HRMS (ESI^+) for $[M+NH4]^+$; calculated: 338.17694, found: 338.17741 (error m/z = 1.38 ppm). Mp: 96-101 °C.



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3,5-bis(methoxycarbonyl)phenylboronic acid (10): To the crude residue of boronic ester from the previous reaction THF and water (4:1, 320 ml : 80 ml), sodium periodate (31.53 g, 147 mmol) was added at room temperature and stirred for 30 mins followed by the addition of 1N hydrochloric acid (34 ml) and stirred for 17

hours. The reaction mixture was diluted with water, extracted with ethyl acetate and washed with 1:1 brine:water. After drying with anhydrous sodium sulfate the organic layer was concentrated. The resulting crude was washed with hexane and vacuum filtered to give a beige powder of **10** (9.5 g, 81% crude yield). ^{13}C NMR (100 MHz, DMSO-D6): δC (ppm) 165.7, 139.1, 135.7, 131.1, 129.4, 52.4. HRMS (ESI$^+$) for $[M+NH4]^+$; calculated: 256.09869, found: 256.09857 (error m/z = -0.49 ppm).

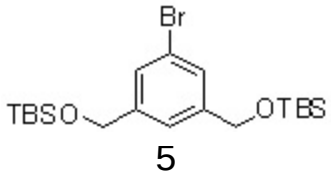
Enrichment of photoaffinity probe 3 form peptide mixture by FSPE

A mixture (0.5 ml) of photoaffinity probe **3** and peptide (KFAKFAKKFAKFAKKFAKFAK) in PBS buffer (50mM, pH = 7.4) with 0.5% Tween-20 was loaded on to a bed of fluorous silica gel in a pipet column (fluorous bed: 40 x 6 mm). Initially the loaded fluorous short column was eluted with water (1 ml) and the fractions collected were subjected to reverse phase HPLC analysis. The column was washed with furthermore water (2 ml) followed by elution with methanol and water in different ratios (1:3 and 1:1) and the resultant fractions were subjected to HPLC analysis (no peaks were found in either fractions). Finally 100% methanol (1 ml) was passed through the column and the fraction was injected in to the HPLC.

NMR parameters for ^{19}F 2D experiments

^{19}F-gCOSY : sw = 36003.6, at = 0.26, np = 18722, d1 = 3, nt = 48, ni = 256, t_n = ^{19}F

^{19}F NOESY : sw = 35698.3, at = 0.029, np = 2048, d1 = 1.8, nt = 24, ni = 256, t_n = ^{19}F, d_n = ^{1}H



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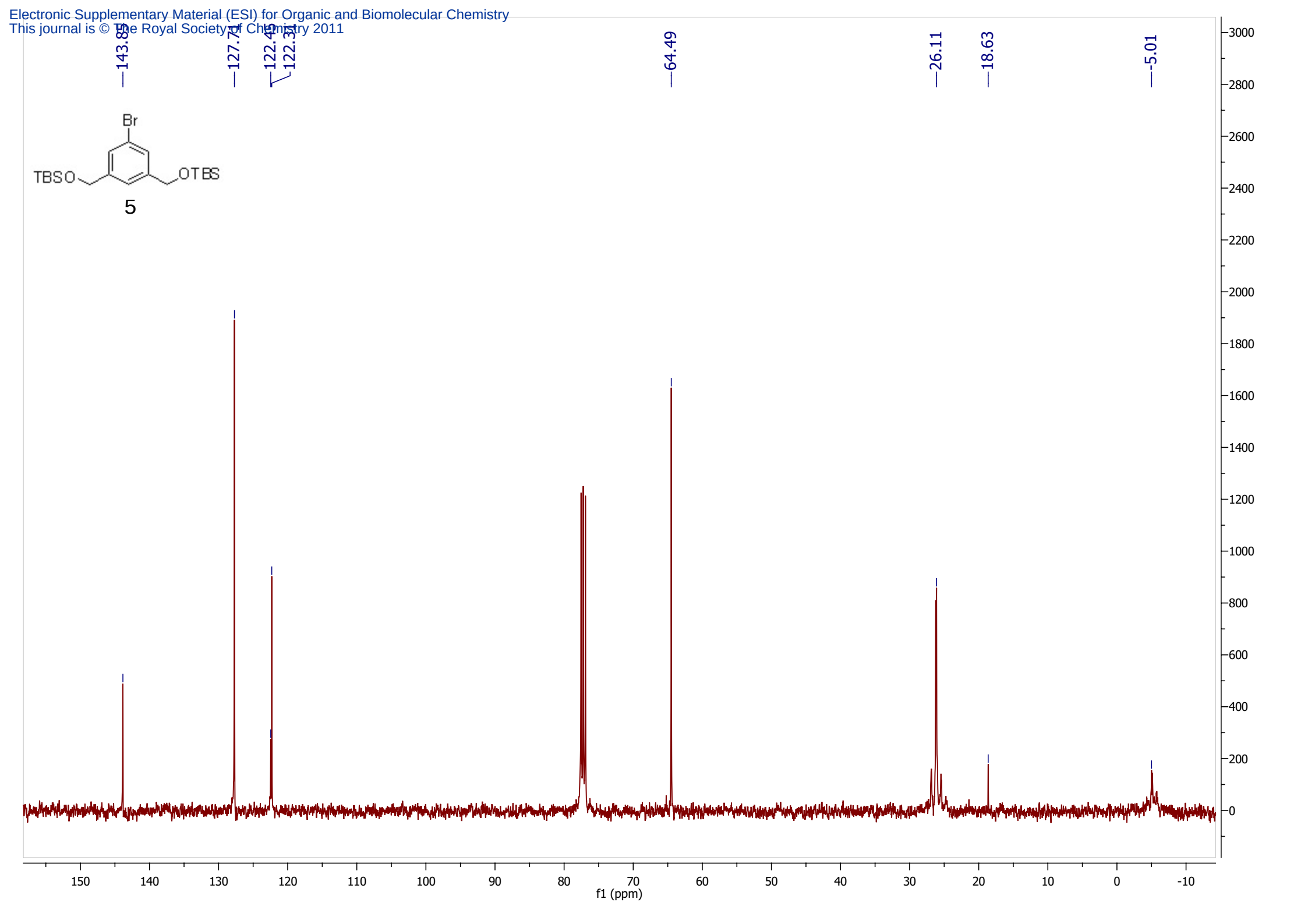

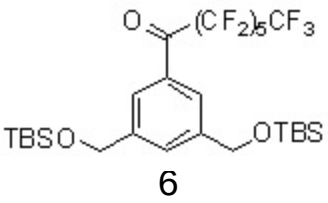

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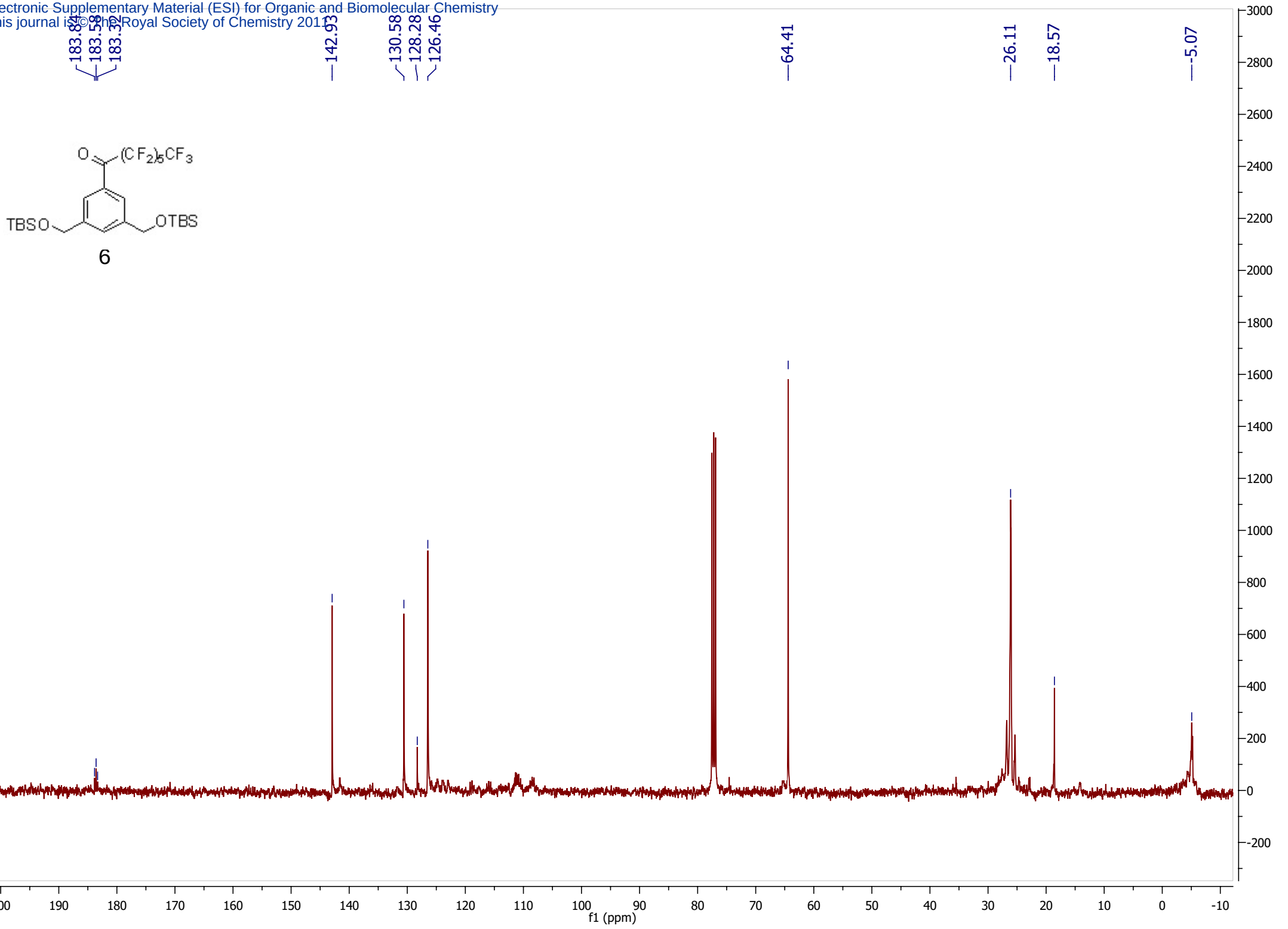

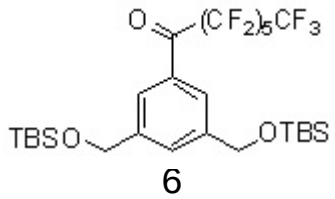

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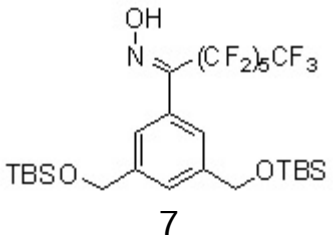

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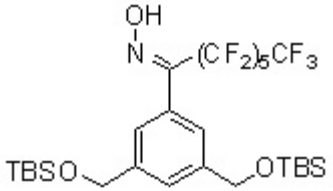

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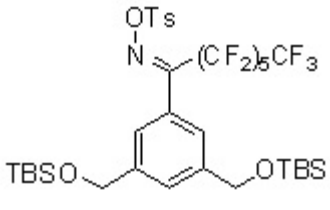

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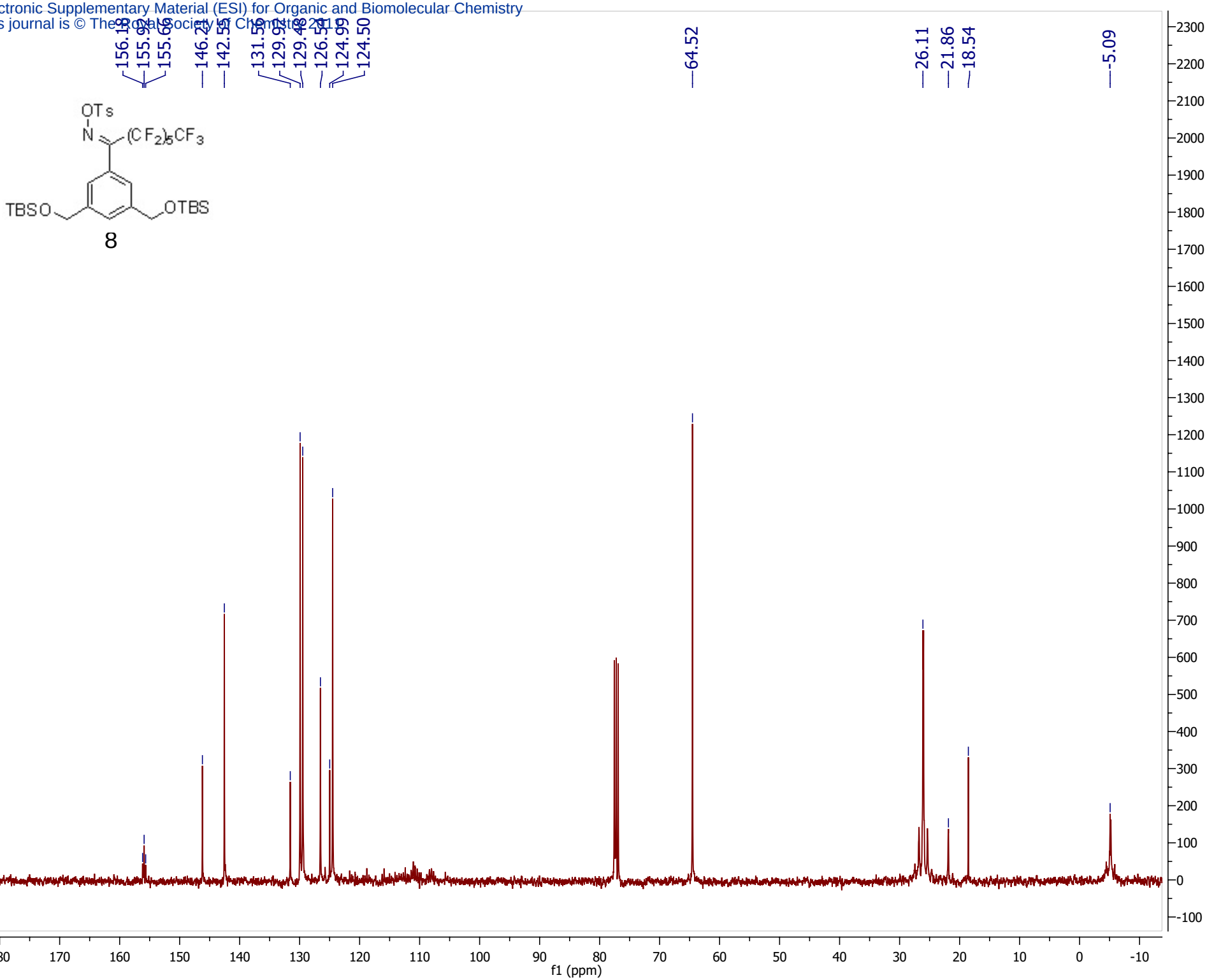

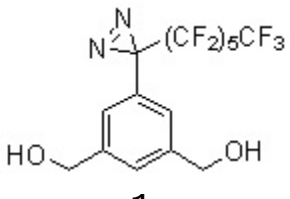

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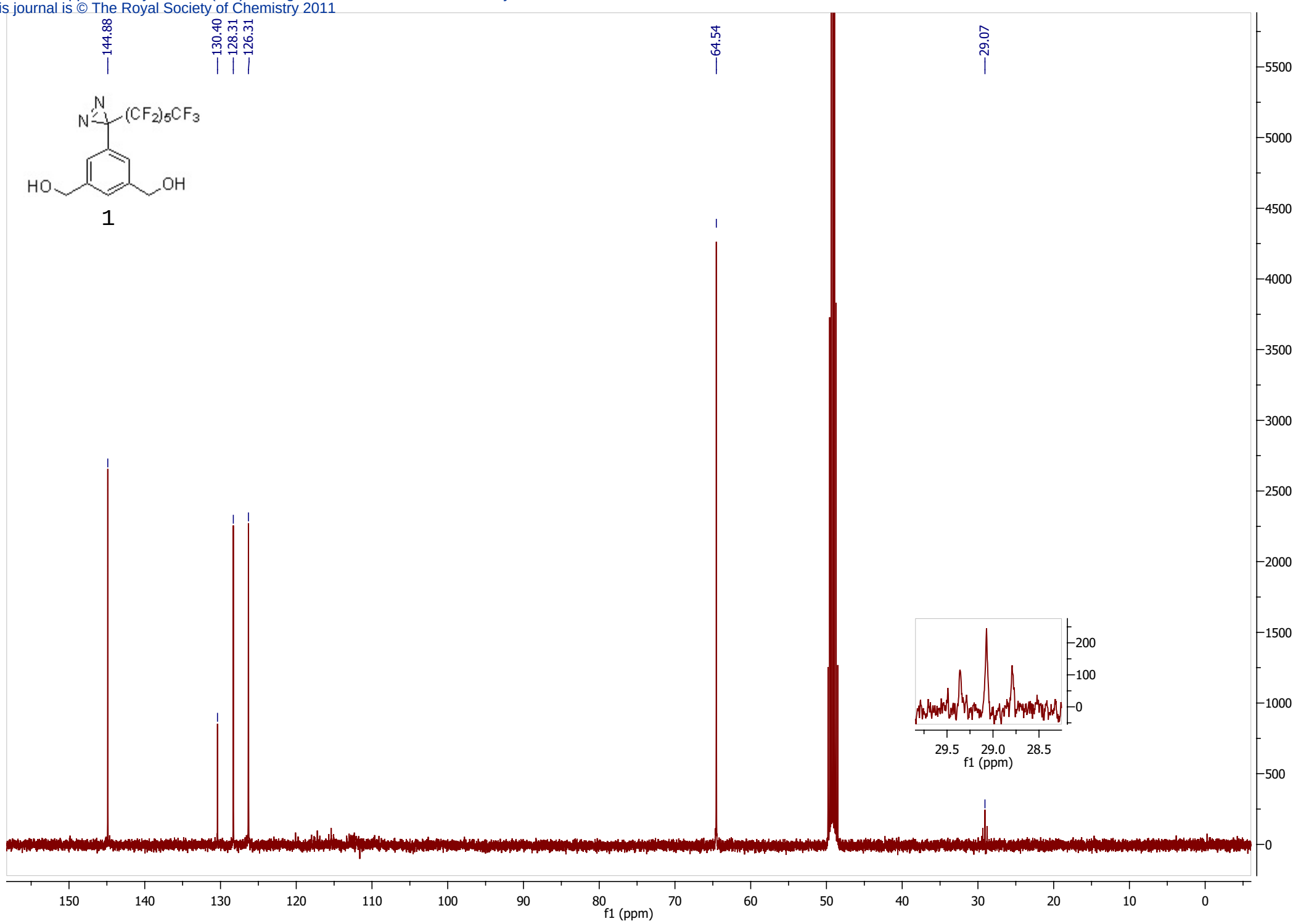

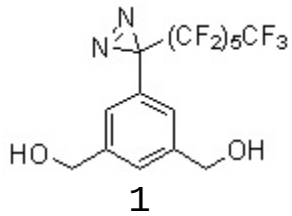

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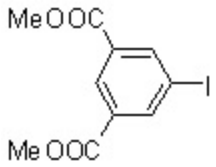

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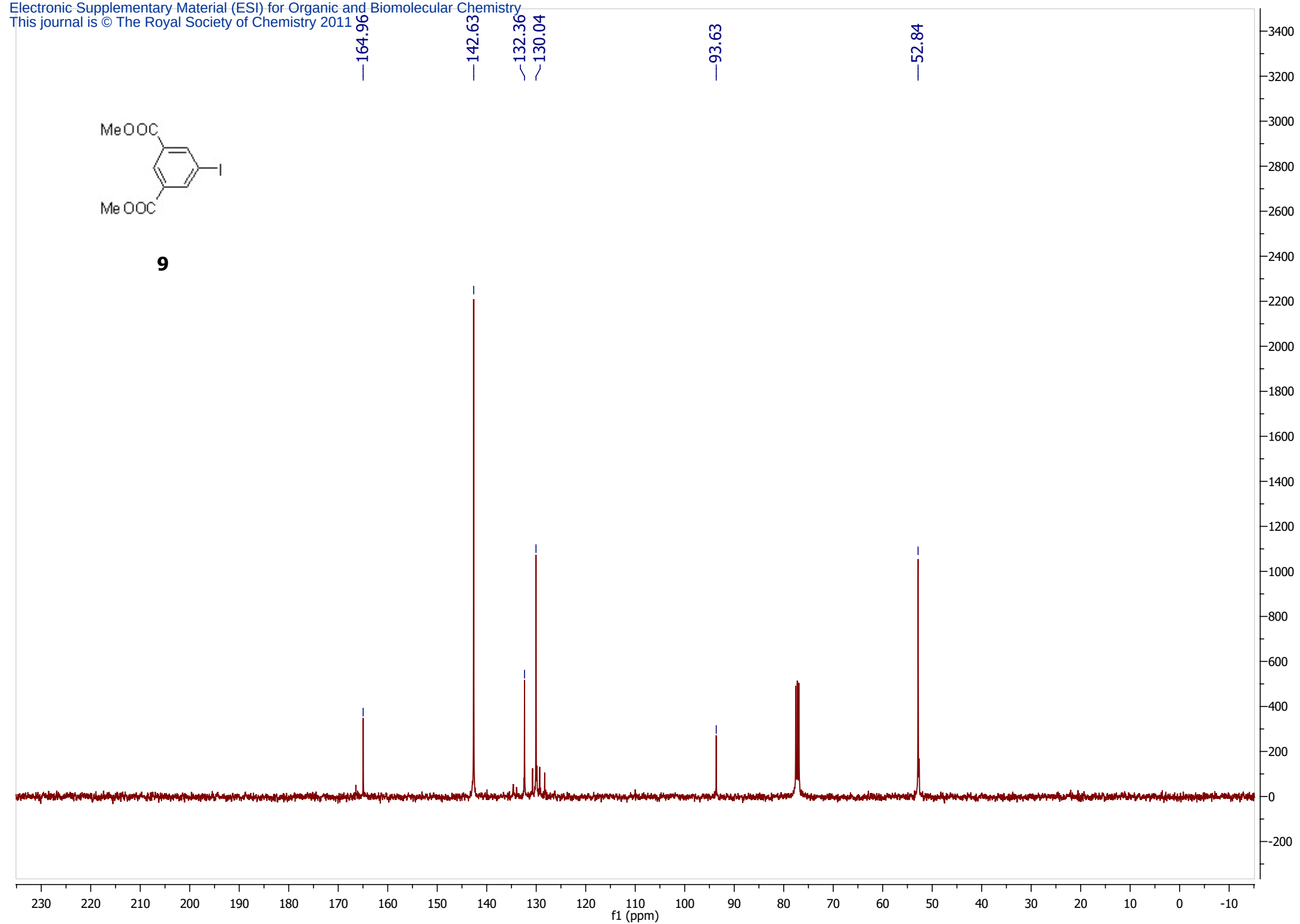

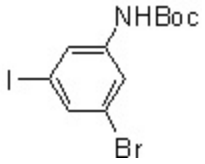

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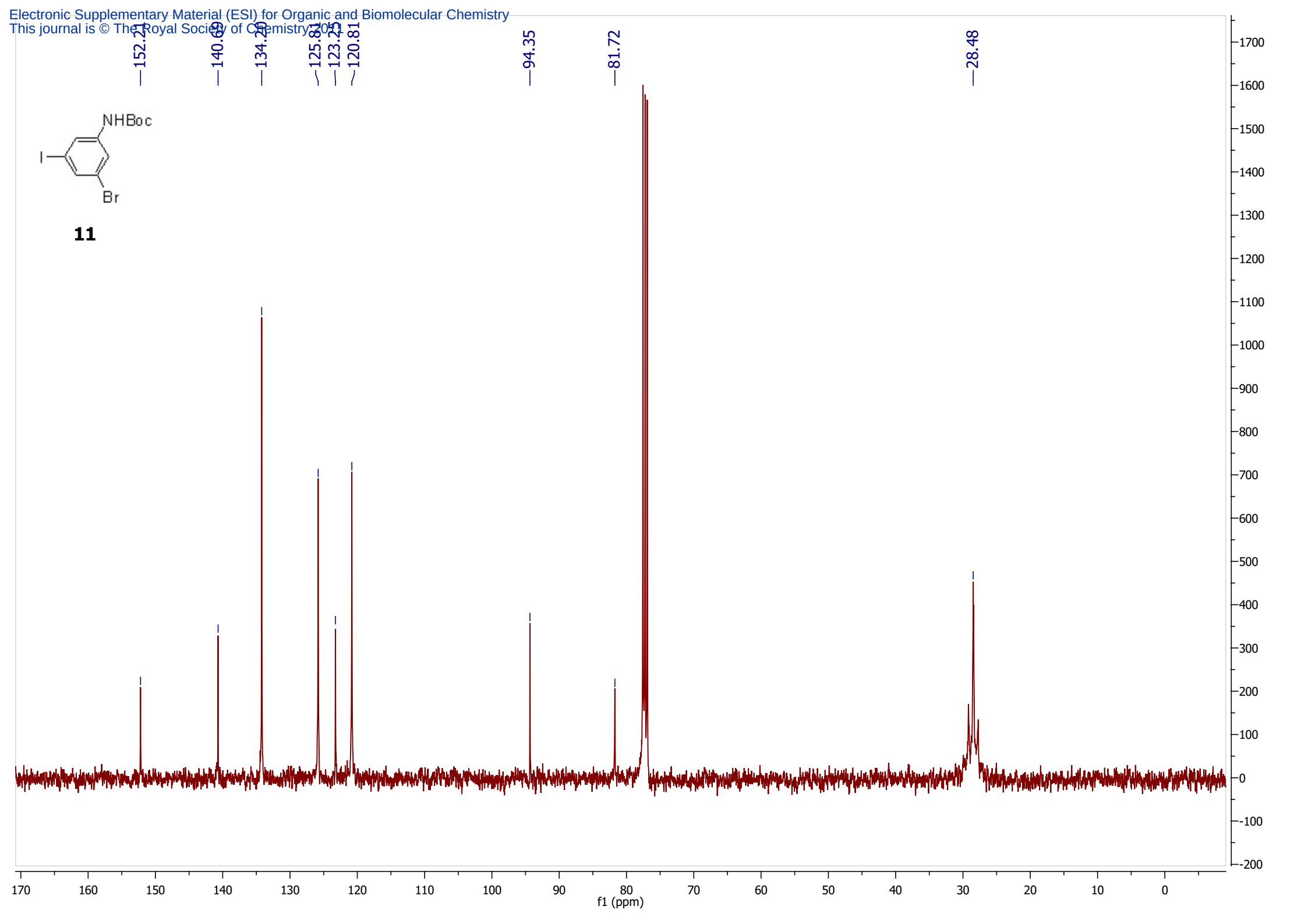

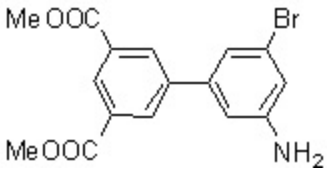

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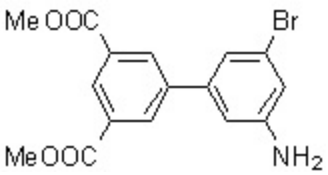

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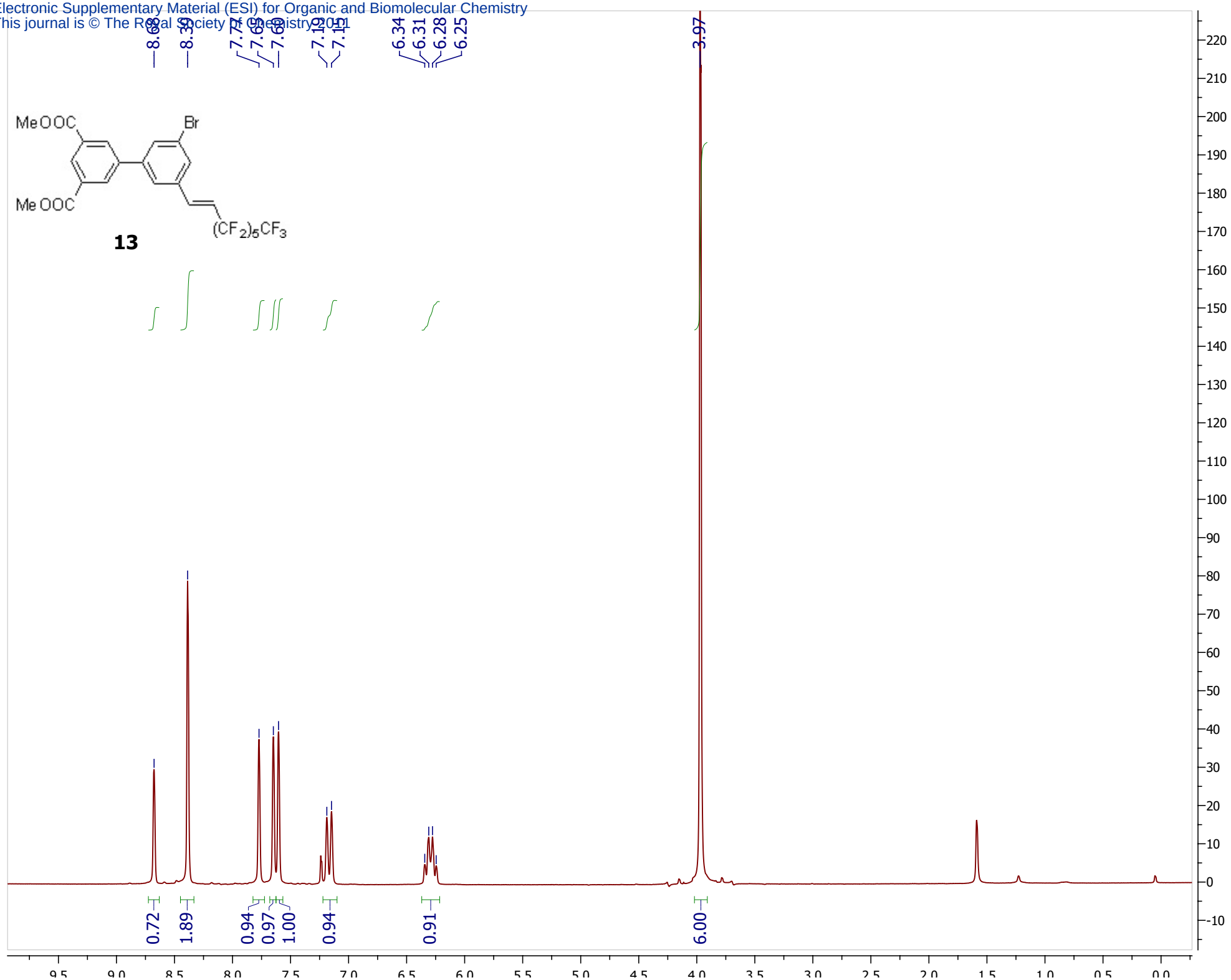

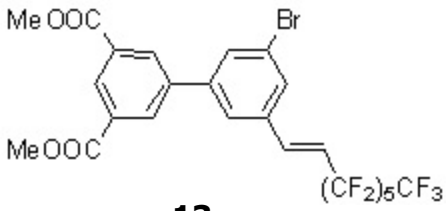

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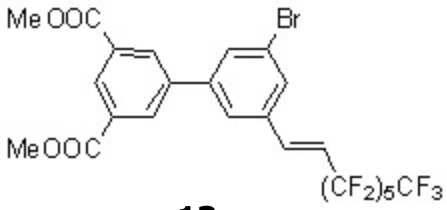

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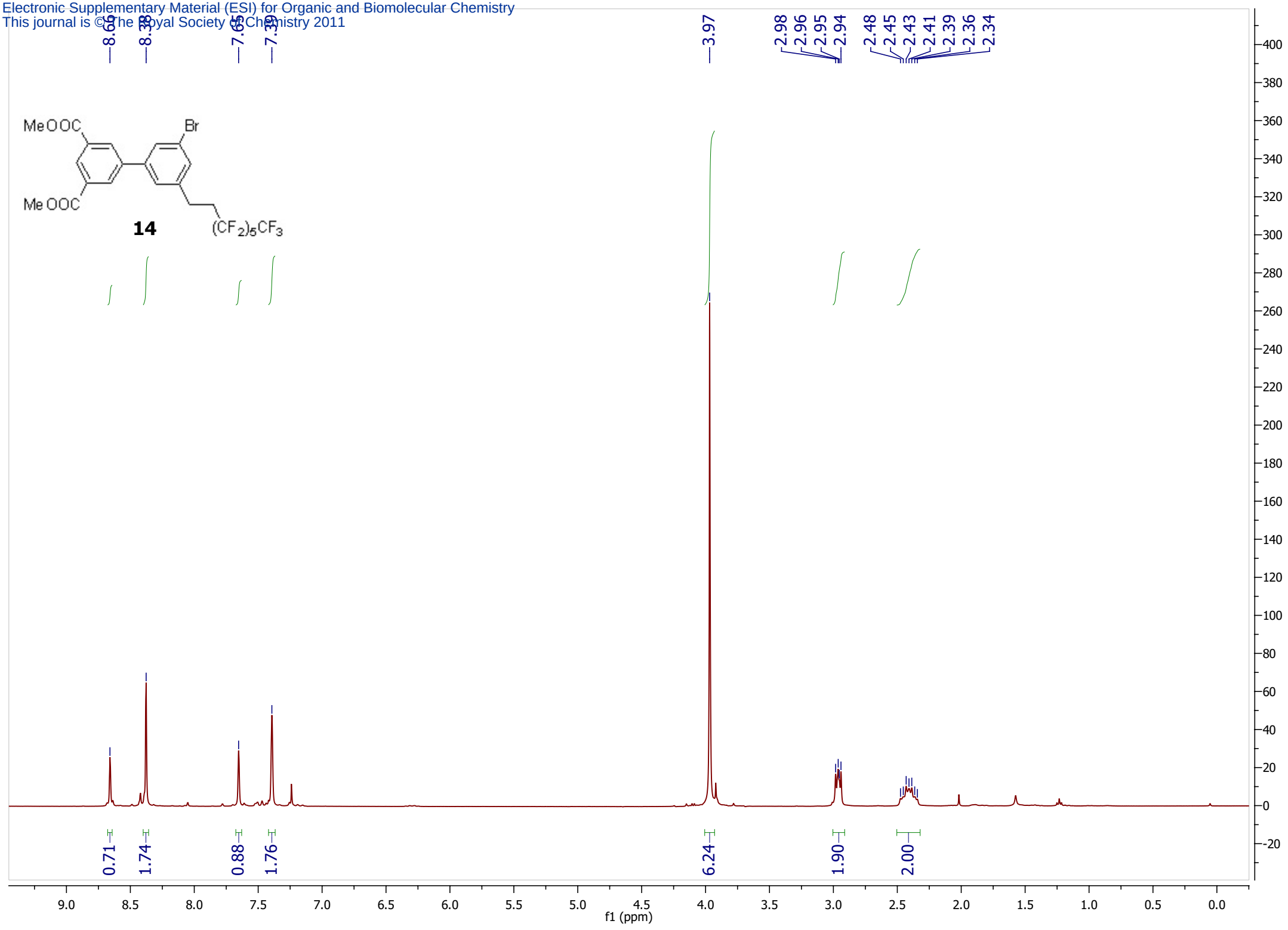

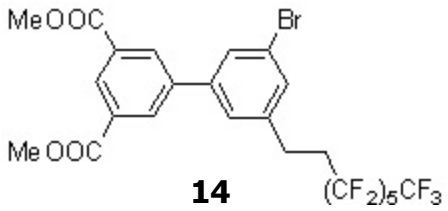

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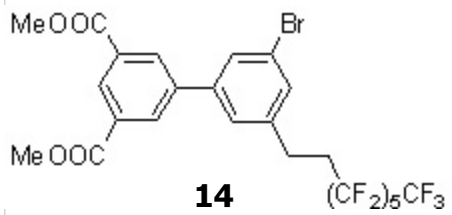

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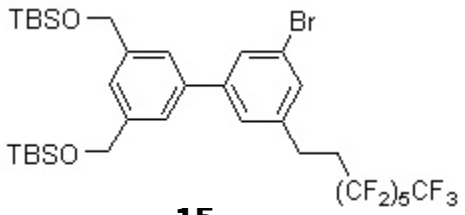

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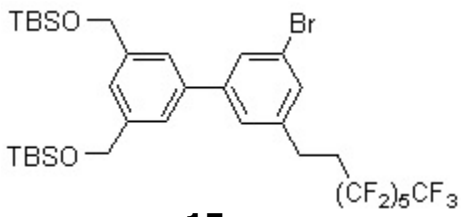

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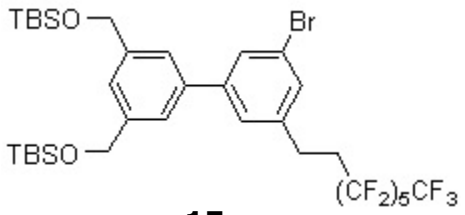

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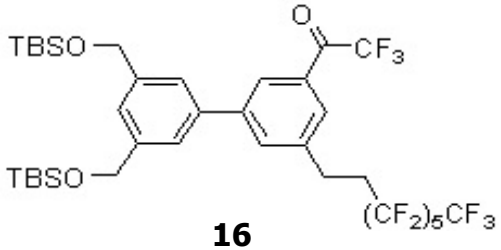

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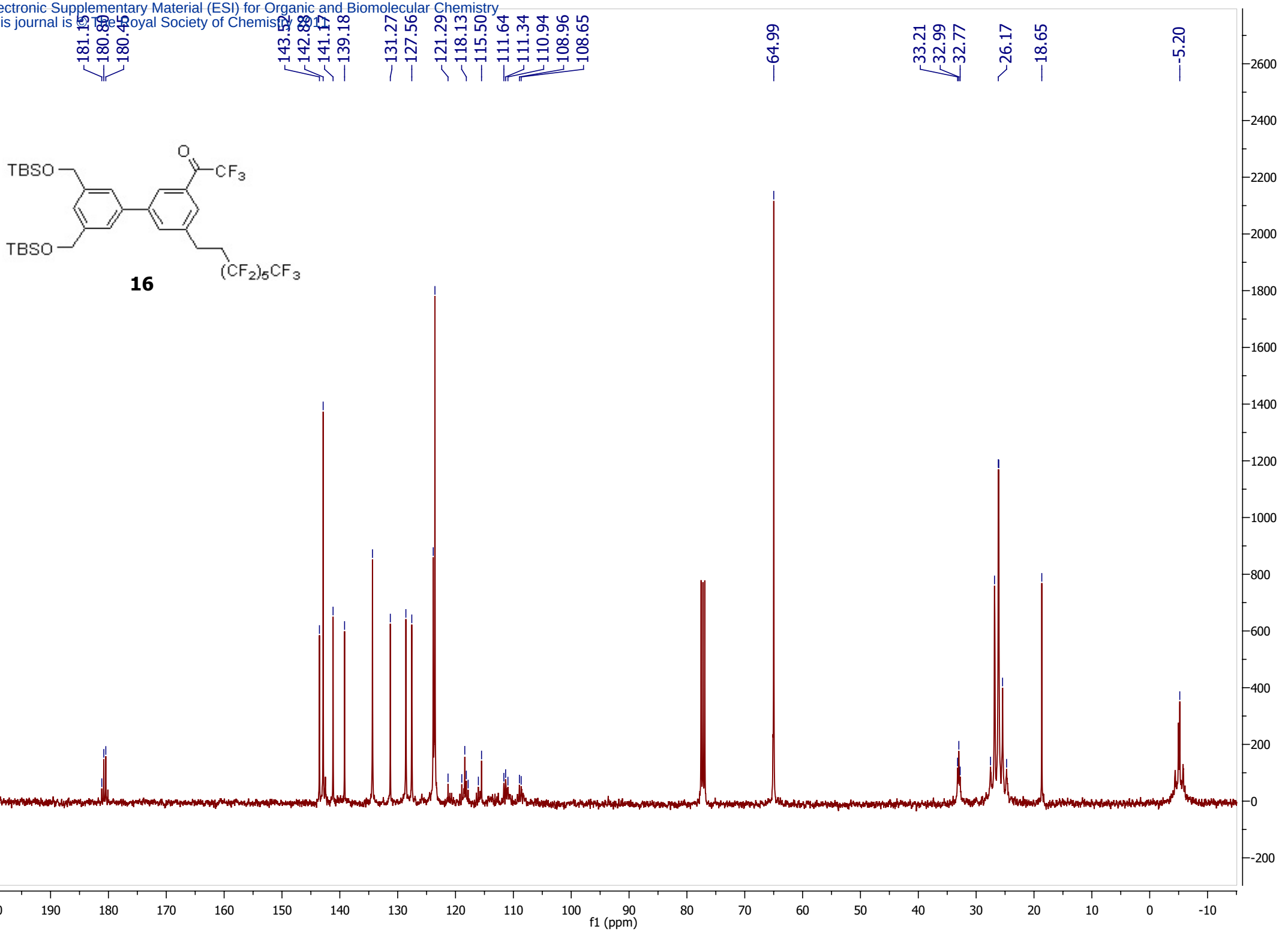

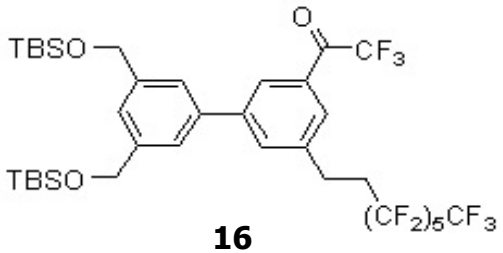

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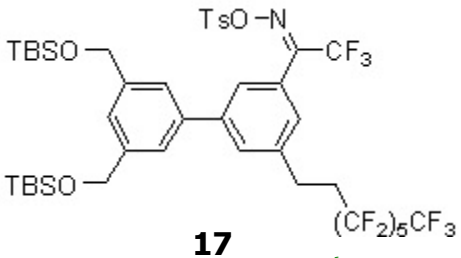

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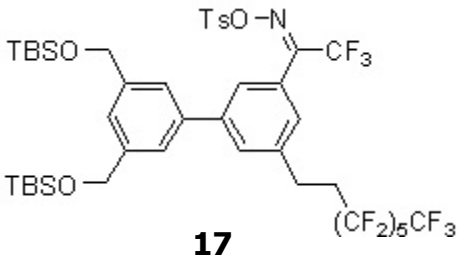

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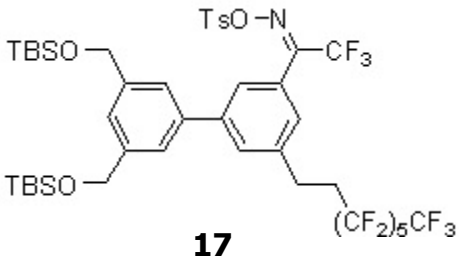

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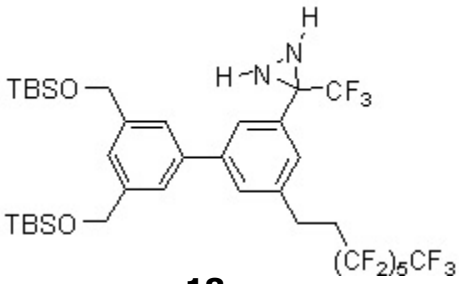

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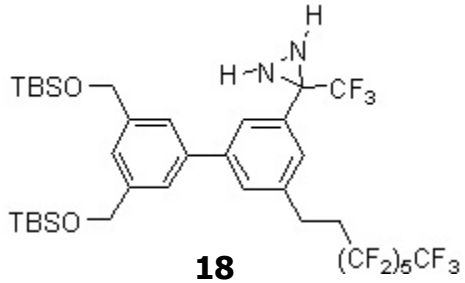

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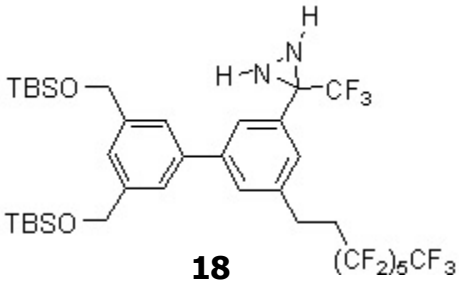

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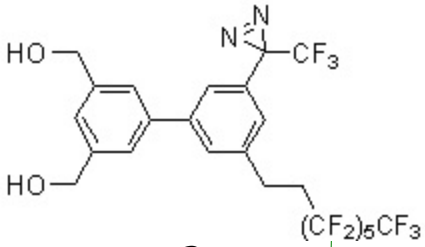

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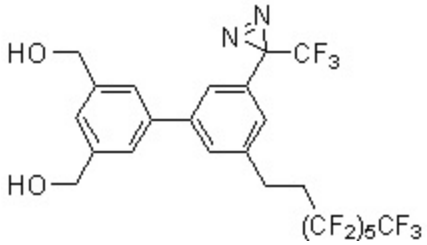
Electronic Supplementary Material (ESI) for Organic and Biomolecular Chemistry



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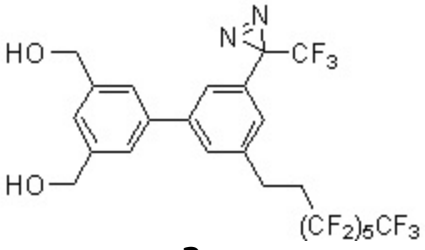

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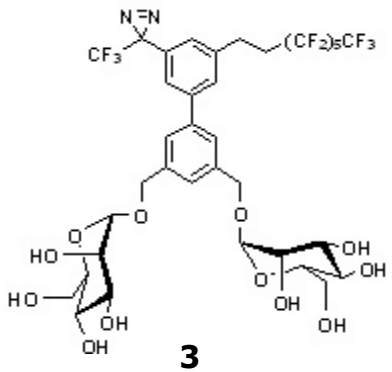

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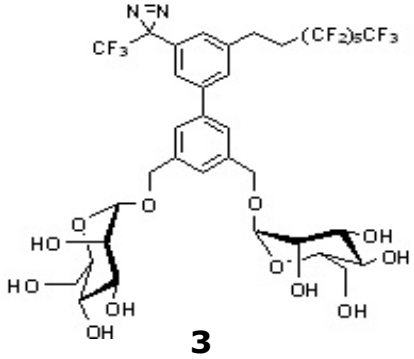

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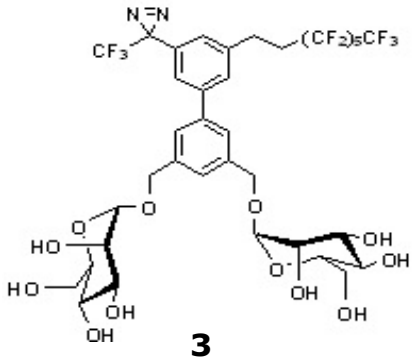

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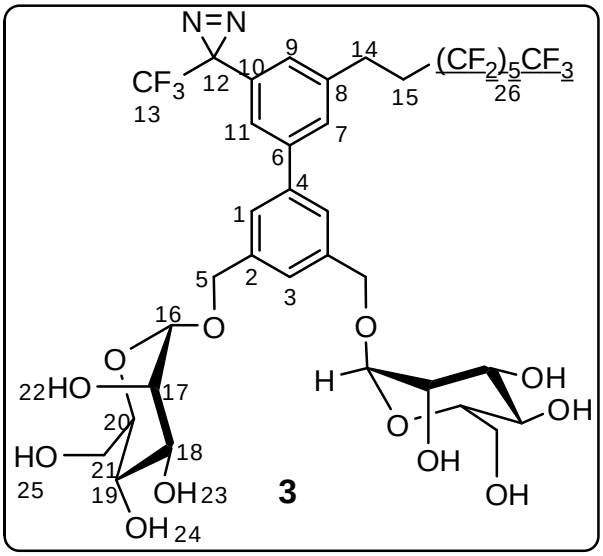

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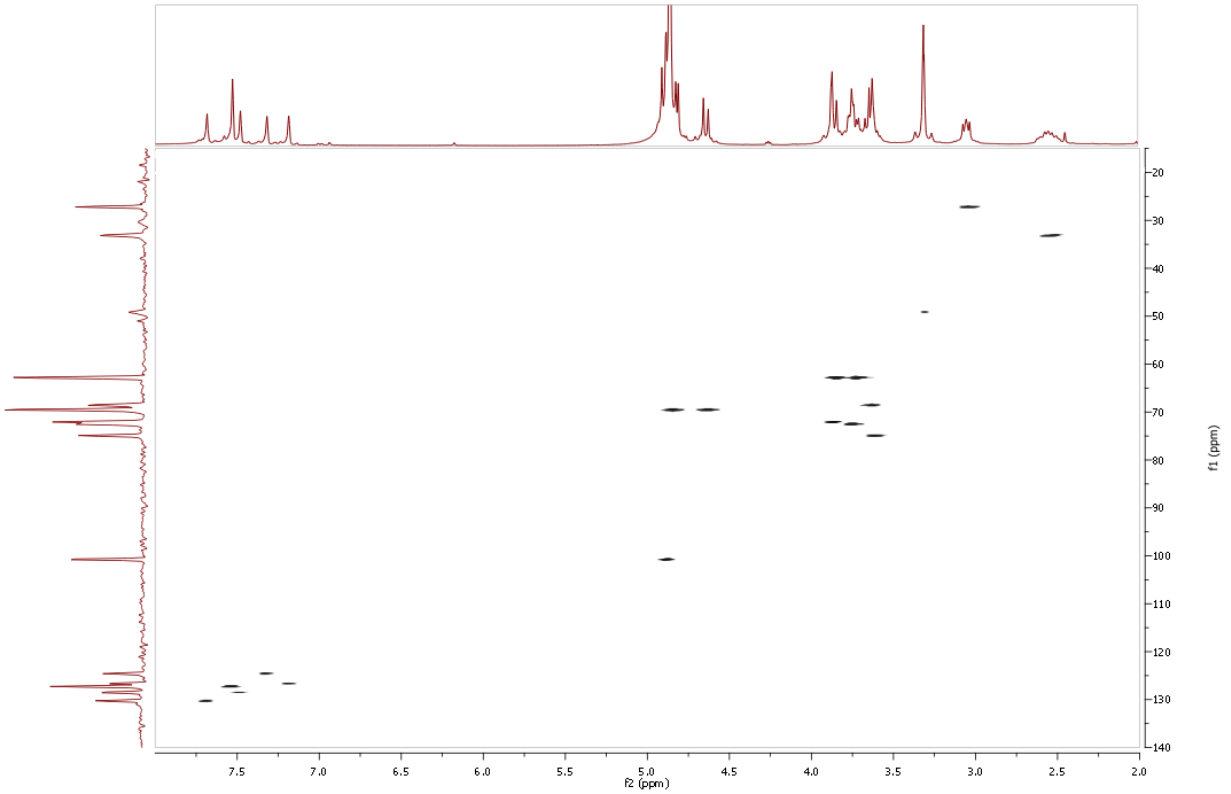
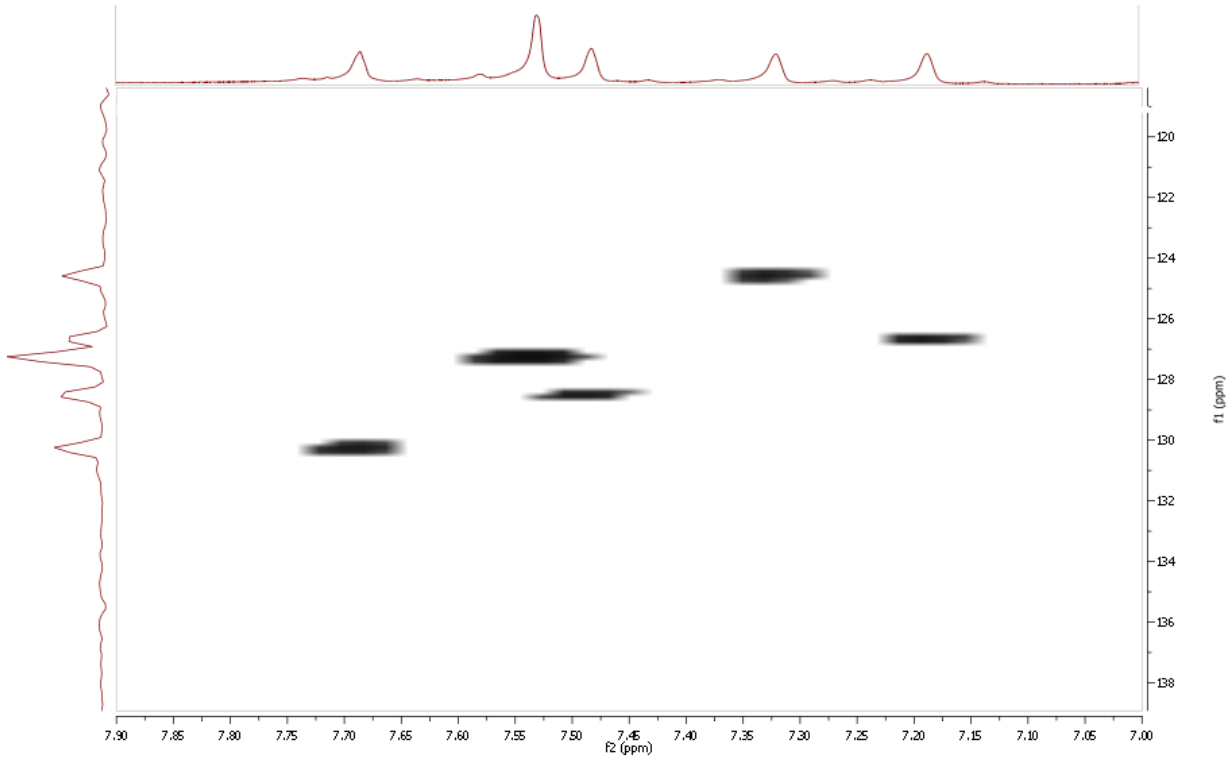
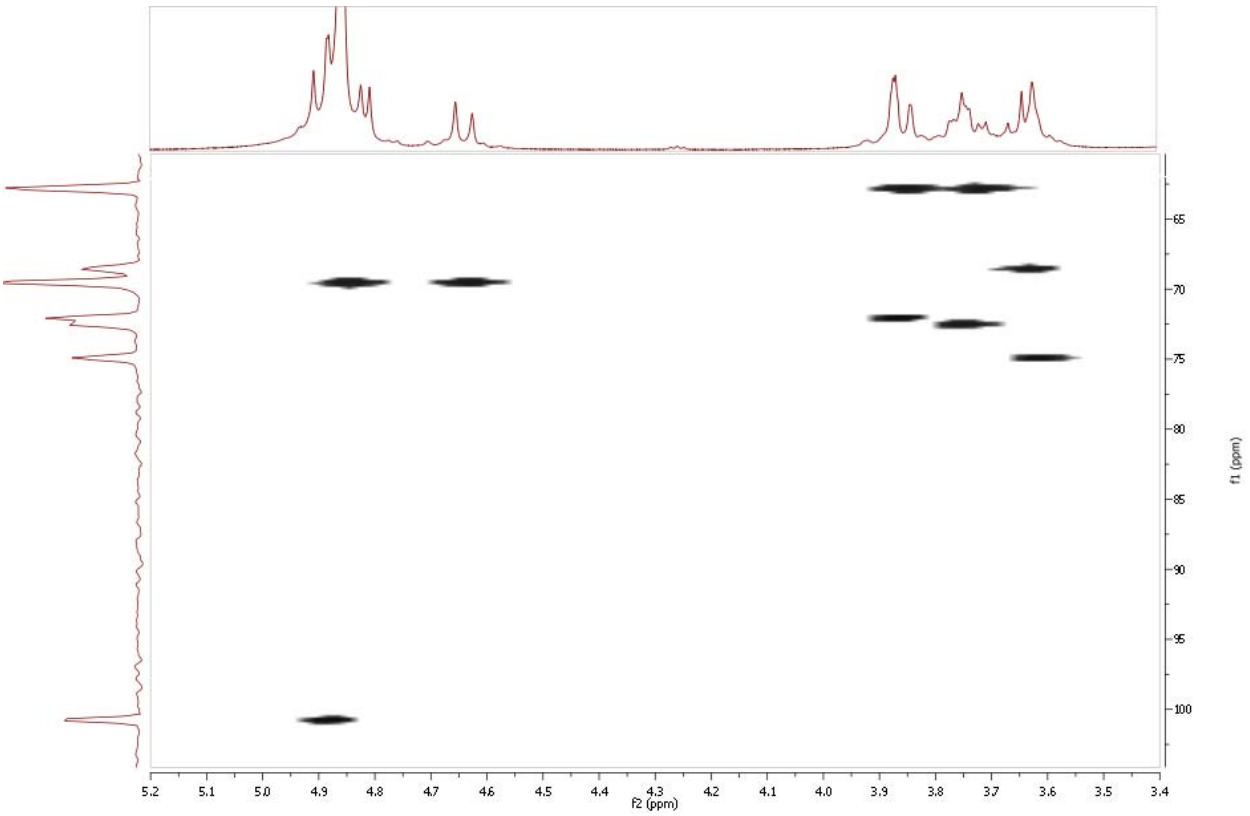
Atom	^{1}H shift (ppm)	^{1}H mult & J values in HZ	^{13}C shift (ppm)	^{13}C mult & J values in HZ	HMQC	COSY
1	7.53	s	127.5	s	CH	3, 5a, 5b
2			140.5	s	C	
3	7.48	s	128.7	s	CH	1, 5a, 5b
4			143.9	s	C	
5a	4.84	d, J=12.0Hz	69.8	s	CH_2	1, 3, 5b
5b	4.64	d, J=12.0Hz				1, 3, 5a
6			143.1	s	C	
7	7.19	s	126.9	s	CH	9, 11, 15
8			141.5	s	C	
9	7.69	s	131.2	s	CH	7, 11, 15
10			130.5	s	C	
11	7.32	s	124.8	s	CH	7, 9
12			29.7	q, J=40.4Hz	C	
13			126 - 121		C	
14	3.10-3.00	m	27.5	s	CH_2	7, 9, 15
15	2.65-2.46	m	33.4	t, J=19.2Hz	CH_2	14
16	4.88	d, J=1.7Hz	100.9	s	CH	17
17	3.91 – 3.81	m	72.3	s	CH	16, 18
18	3.79 -3.69	m	72.8	s	CH	17,19
19	3.67 -3.58	m	68.8	s	CH	18, 20
20	3.67 – 3.58	m	75.2	s	CH	19, 21a, 21b
21a	3.91 – 3.81	m	63.1	s	CH_2	21b, 20
21b	3.79 - 3.69	m				21a, 20
22, 23, 24, 25	4.84	s				
26			124.2 – 108.2	m	C	

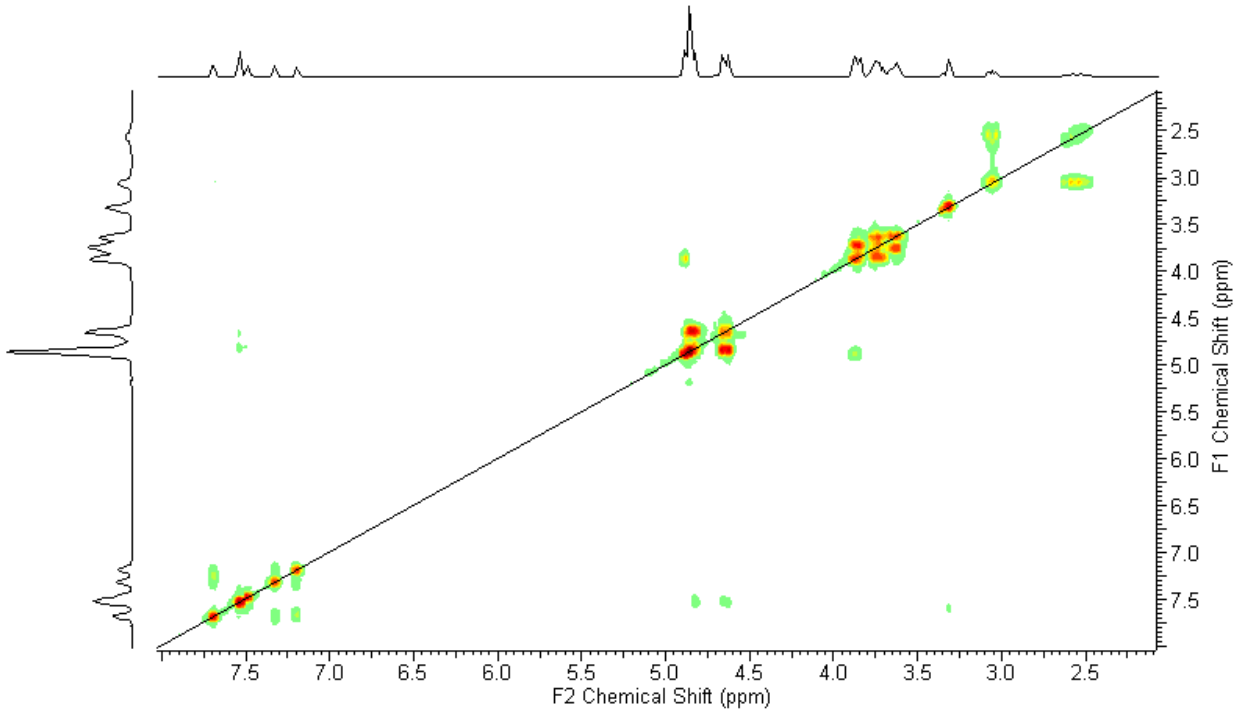
HMQC

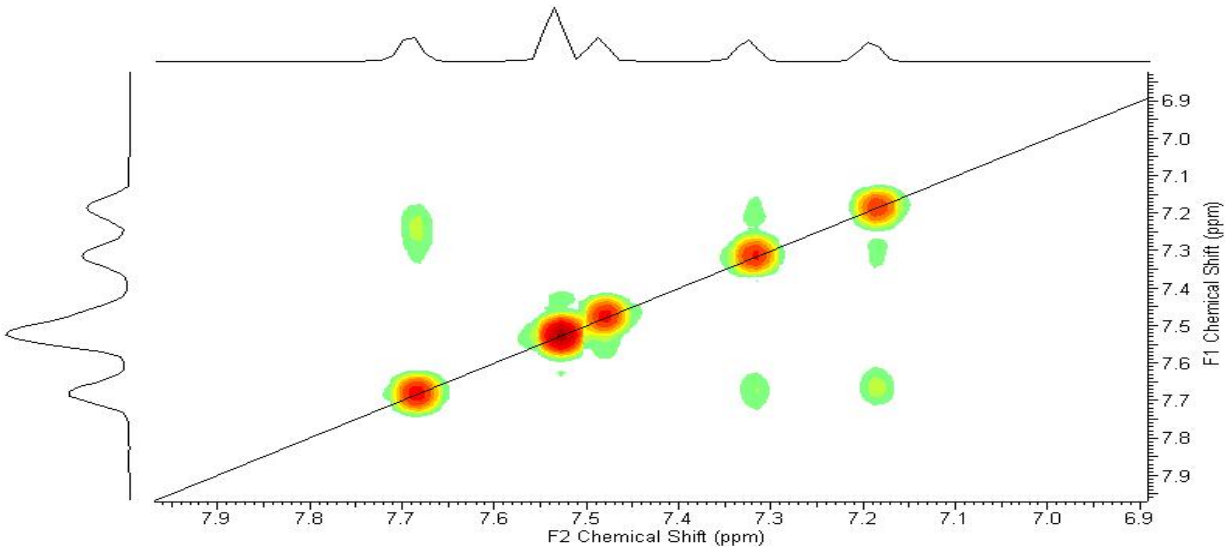


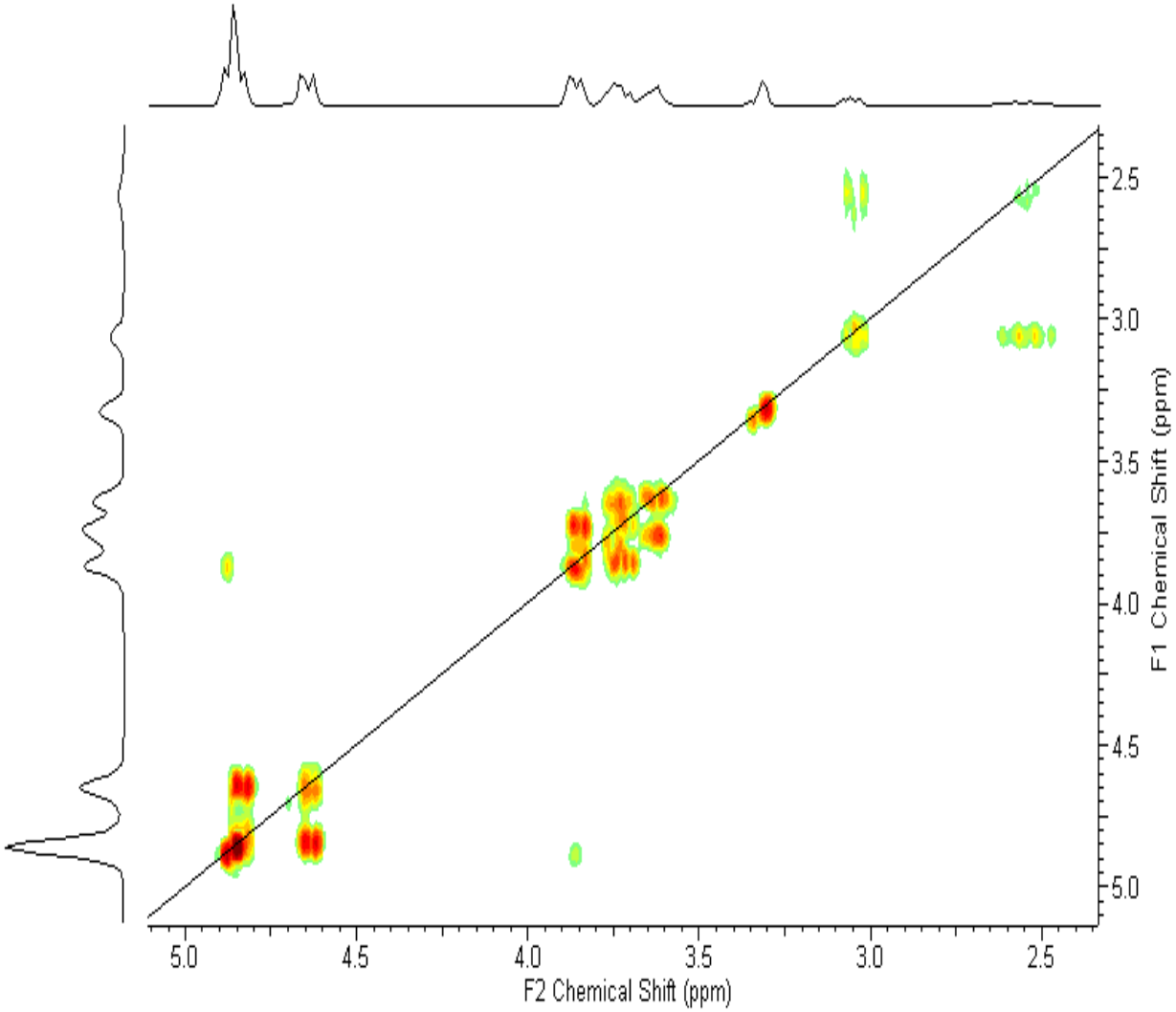


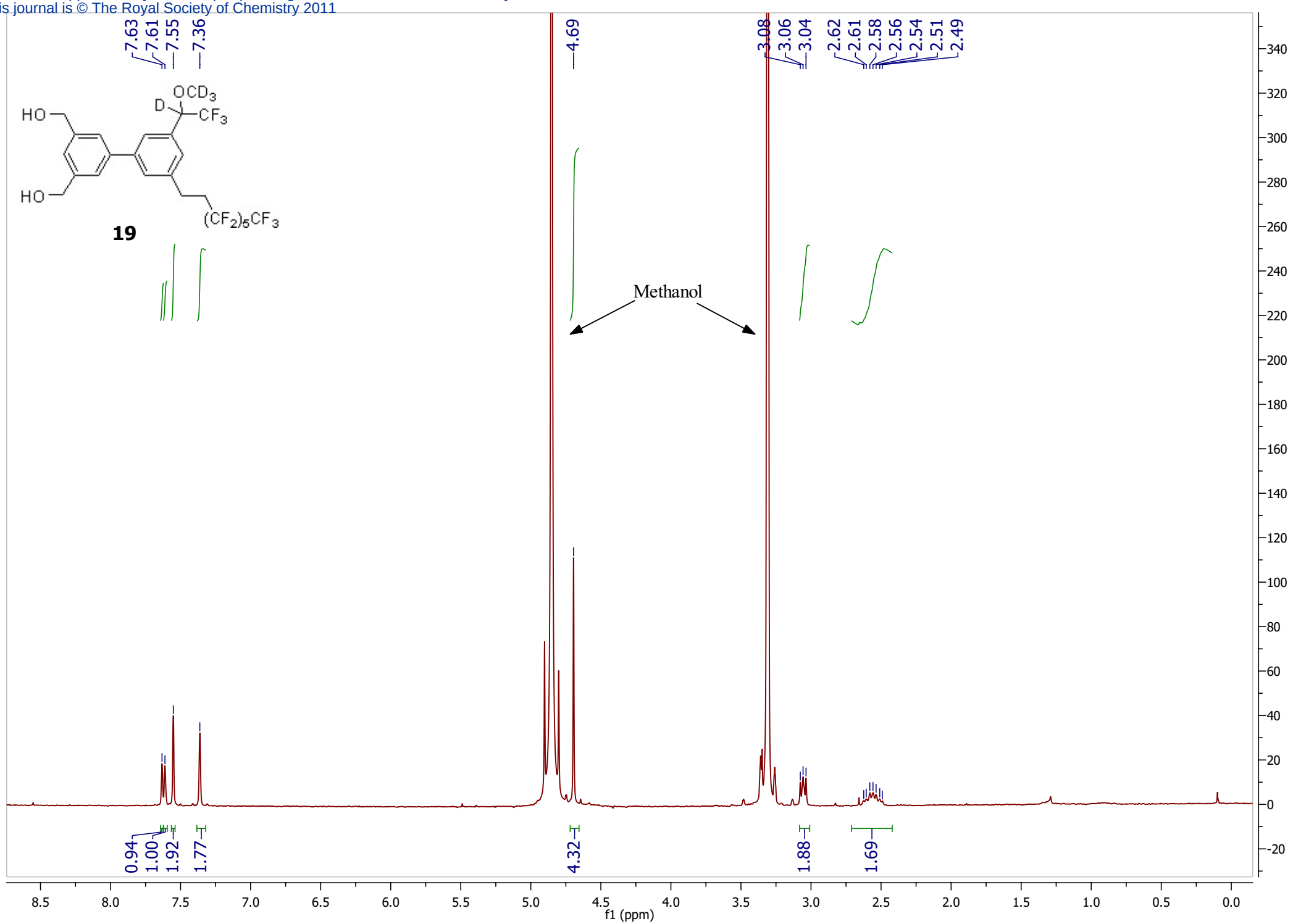


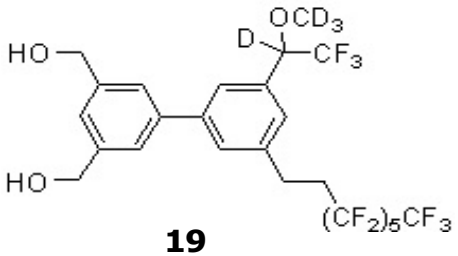
gCOSY



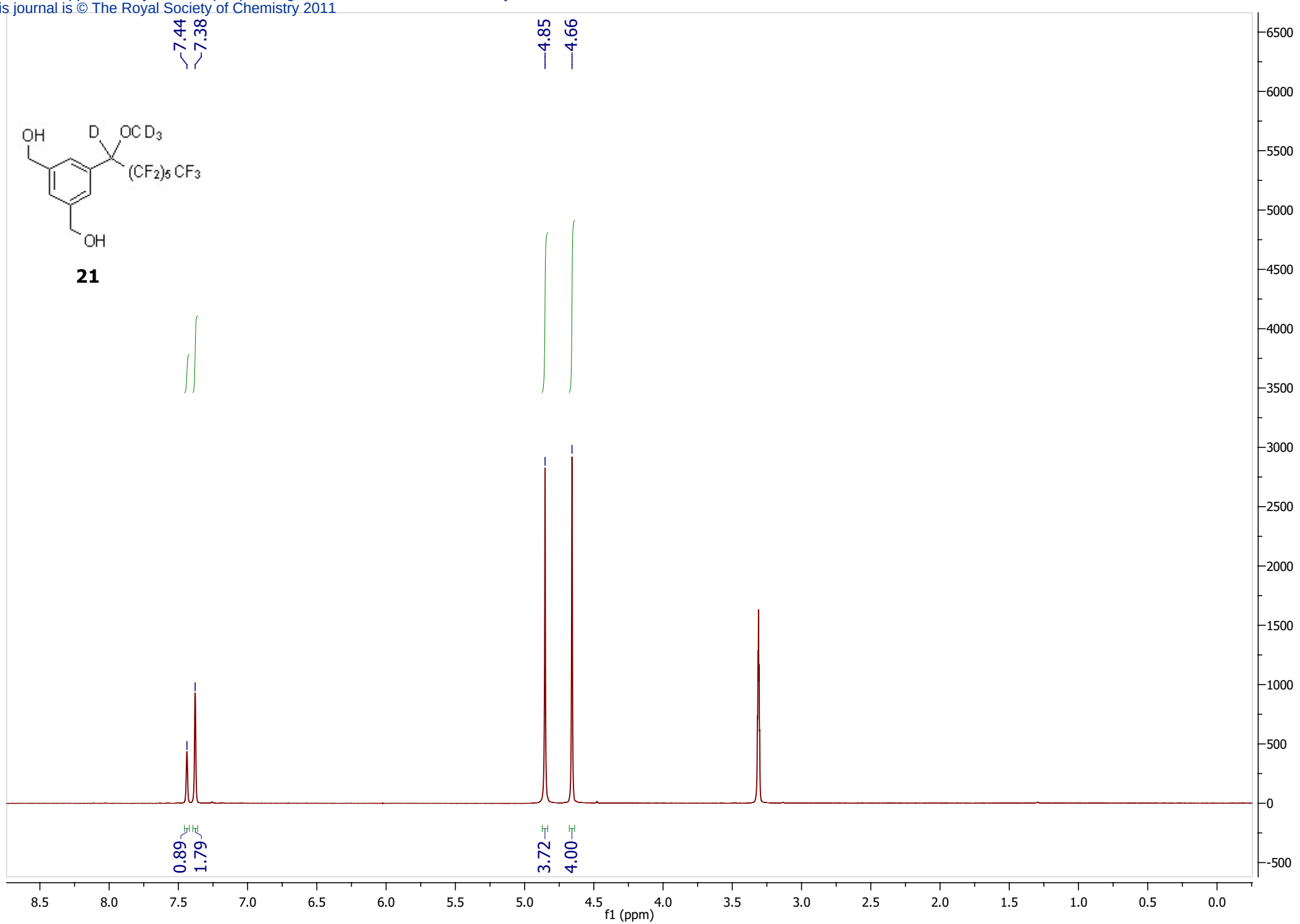


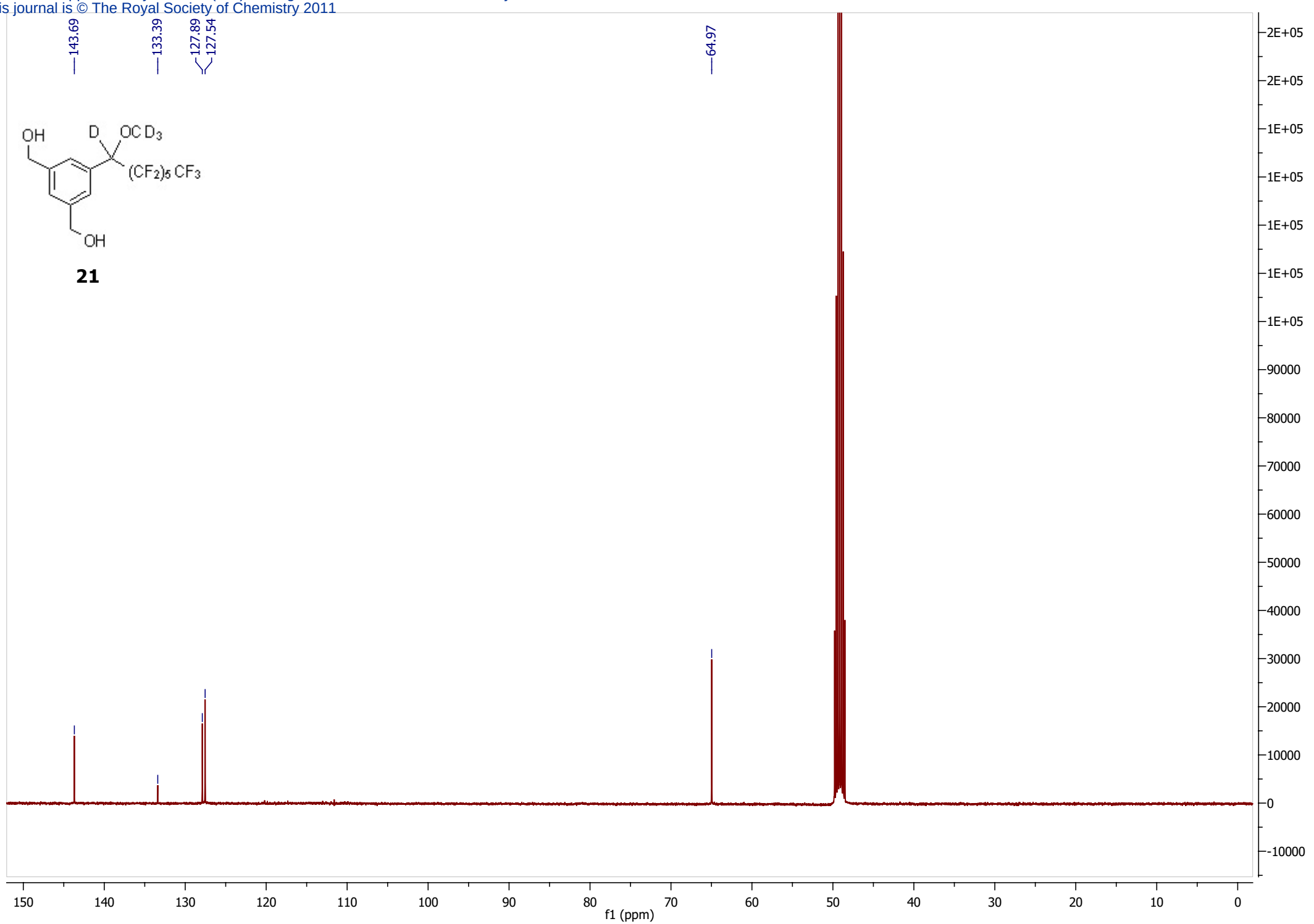


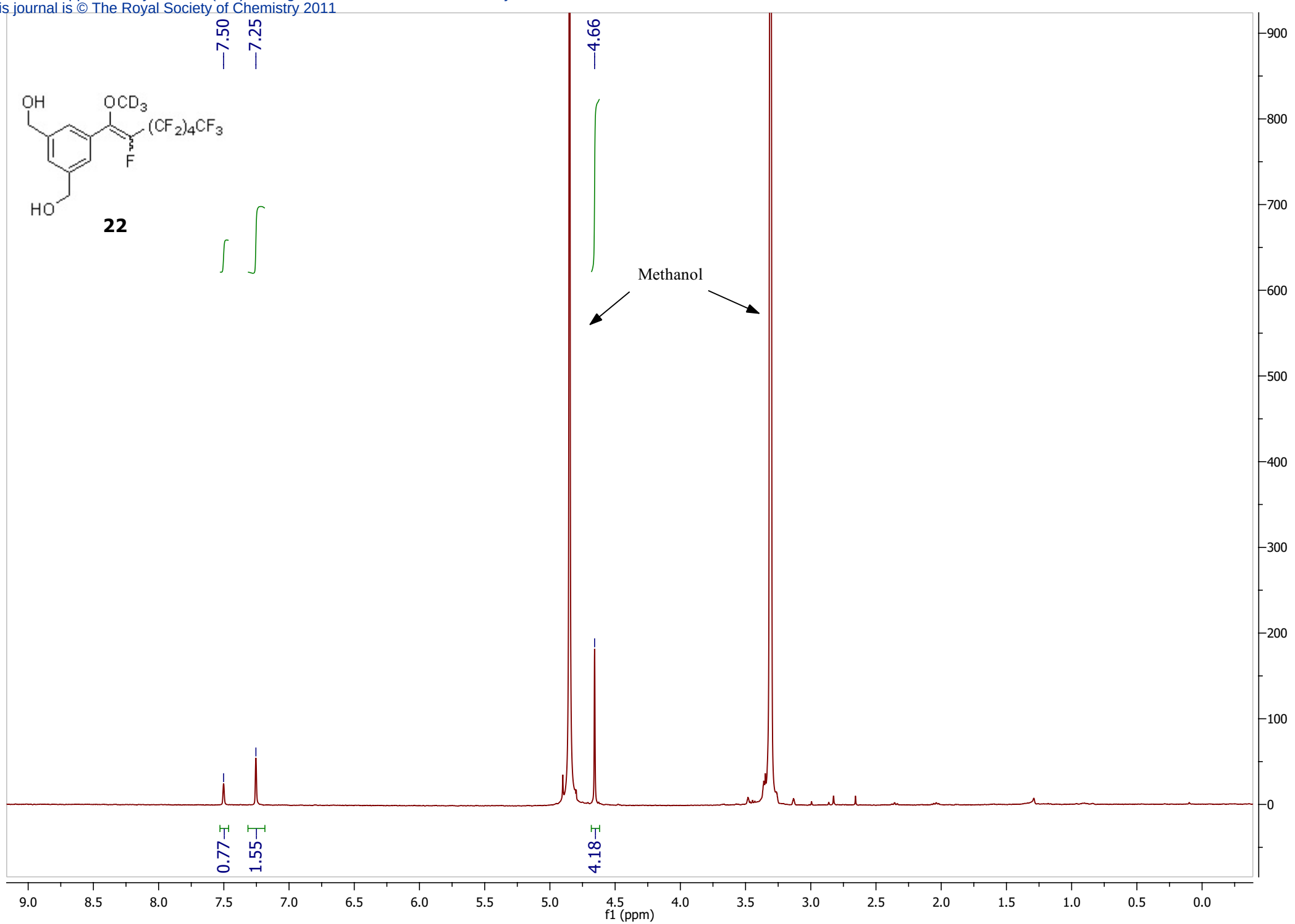


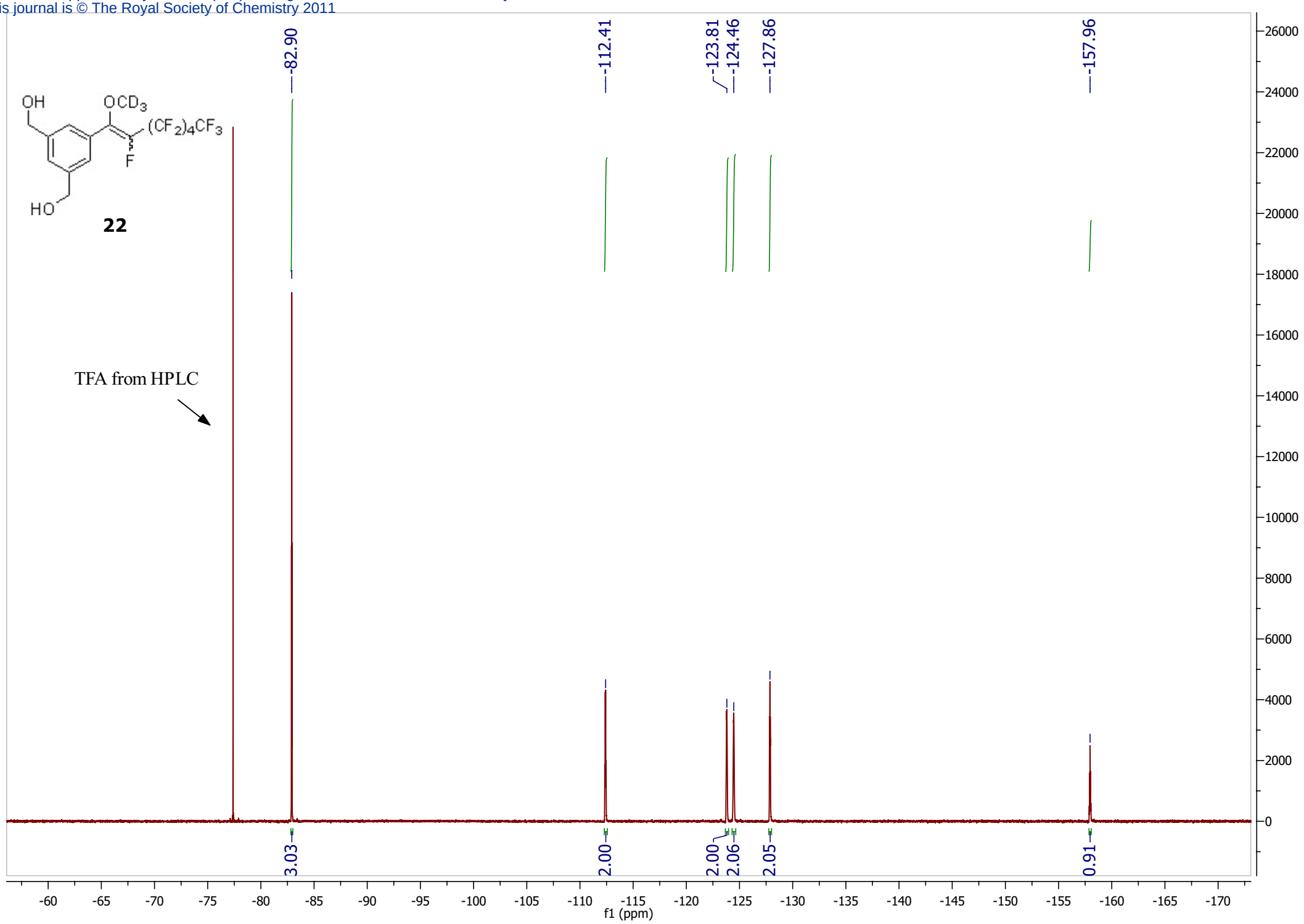


19

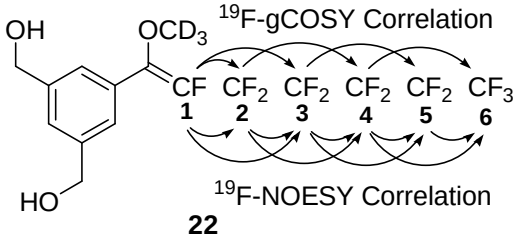








^{19}F 2D-NMR characterization of **22**

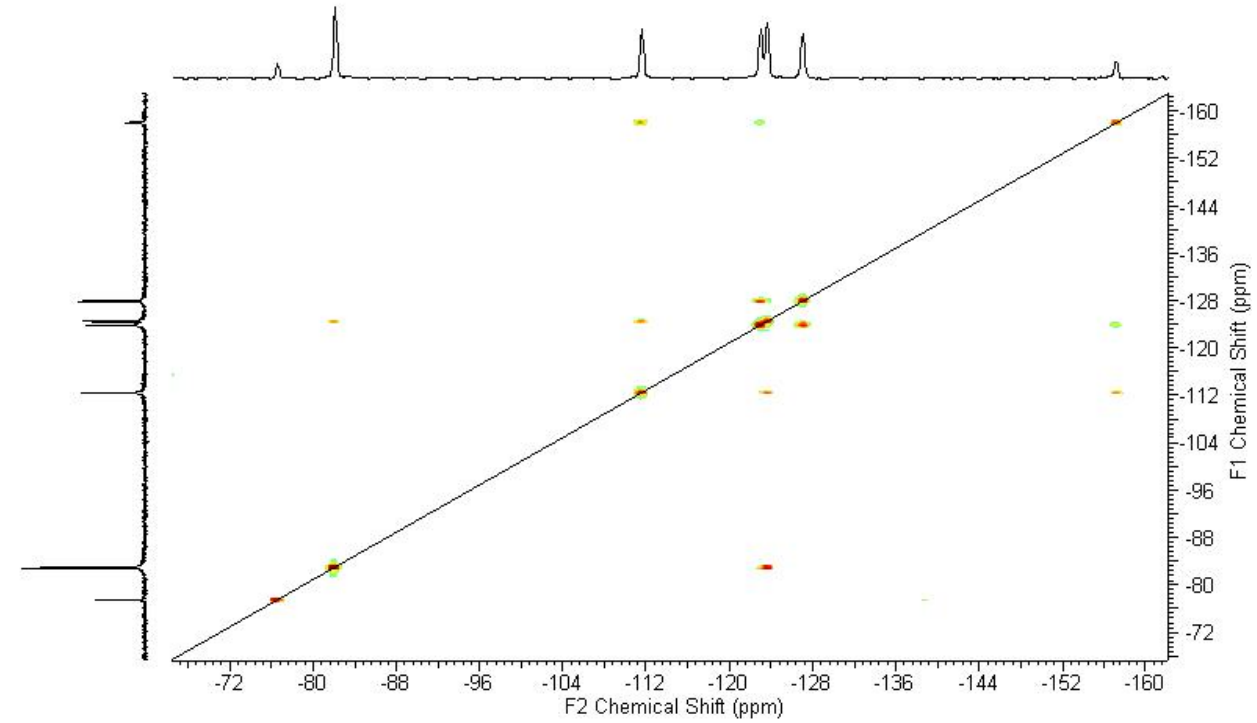


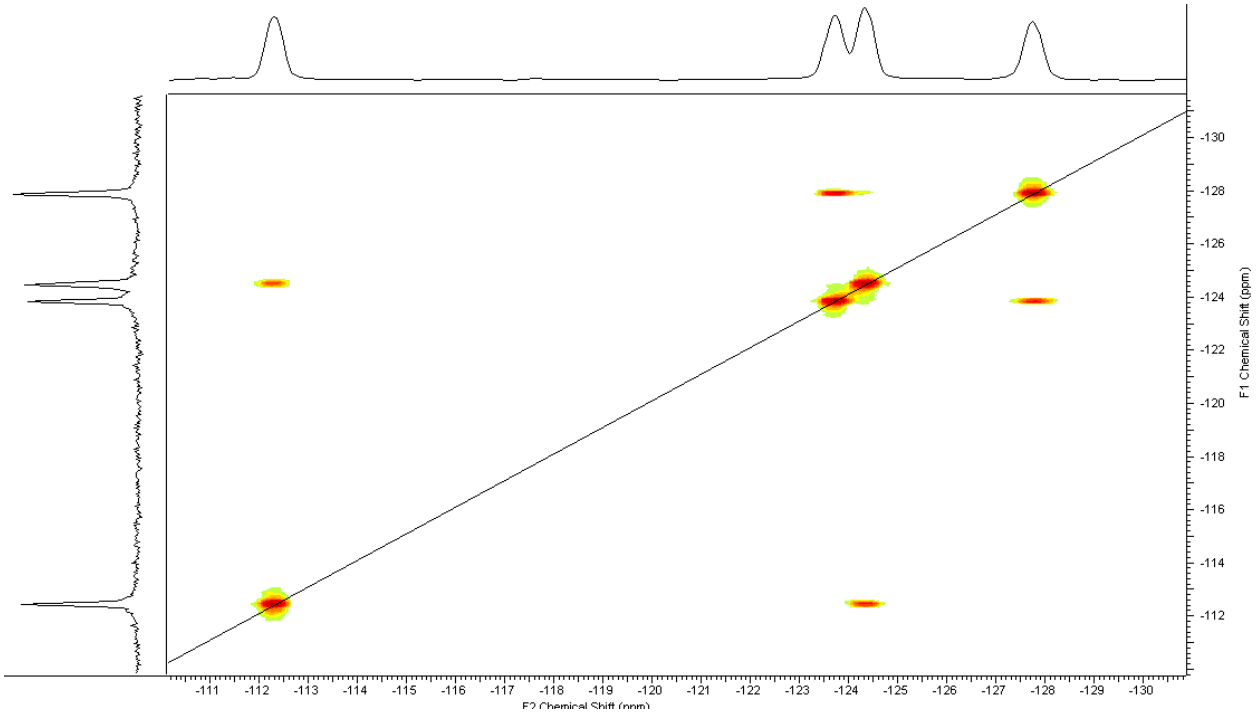
22

Atom	^{19}F NMR Shift (ppm)	^{19}F NMR integral value	^{19}F gCOSY	^{19}F NOESY
1	-157.96	1	3, 2	2, 3
2	-112.41	2	4, 1	3, 4, 1
3	-123.81	2	5, 1	2, 4, 5, 1
4	-124.46	2	2, 6	2, 3, 5, 6
5	-127.86	2	3	3, 4, 6
6	-82.90	3	4	4, 5

Note: In the case of 1 and 2, a $^{3}J_{FF}$ correlation is observed in ^{19}F gCOSY due to the sp^2 hybridization of the 1-carbon.

^{19}F-gCOSY





^{19}F NOESY