

Supplementary Materials

Isolation and structure elucidation of new cytotoxic macrolides halosmysins B and C from the fungus *Halosphaeriaceae* sp. associated with a marine alga

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Table S1 NMR spectral data of **2** in CDCl₃

Position	$\delta_{\text{H}}^{\text{a}}$		J/Hz	^1H - ^1H COSY		NOESY ^b	δ_{C}	HMBC (C) ^c
2							166.2 (s)	
3	5.66	d	16.4 (4)	4	5 β , 9		124.6 (d)	2, 5
4	6.83	ddd	16.4 (3), 10.8 (5 β), 6.0 (5 α)	3, 5 α , 5 β	6		145.2 (d)	2
5 α	2.53	ddd	13.2 (5 β), 6.0 (4), 1.8 (6)	4, 5 β	15		40.0 (t)	3
5 β	2.25	ddd	13.2 (5 α), 12.6 (6), 10.8 (4)	4, 5 α , 6	3, 15			3, 4, 6
6	5.31	dqd	12.6 (5 β), 6.6 (15), 1.8 (5 α)	5 β , 15	4		69.9 (d)	
8							169.0 (s)	
9	3.59	d	3.6 (10)	10	3, 1', 7'A		52.6 (d)	8, 11, 5', 6'
10	4.81	d	3.6 (9)	9	12, S-CH ₃ , 4'		52.4 (d)	8, 11, 2', 3'
11							208.0 (s)	
12	4.56	dd	9.0 (12-OH), 8.4 (13 α)	13 α , 12-OH	10		75.1 (d)	11, 14
13 α	1.93	ddd	15.0 (13 β), 8.4 (12), 2.4 (14)	12, 13 β	16		37.3 (t)	11
13 β	2.60	ddd	15.0 (13 α), 11.4 (14), 1.2 (12)	13 α , 14	16			14
14	5.25	dqd	11.4 (13 β), 6.6 (16), 2.4 (13 α)	13 β , 16			65.8 (d)	
15	1.40	d	6.6 (6)	6	5 α , 5 β		20.5 (q)	5, 6
16	1.27	d	6.6 (14)	14	13 α , 13 β		20.3 (q)	13, 14
12-OH	3.31	br d	9.0 (12)	12				
1'(NH)	5.84	s			9, 9'			3', 5', 6', 7'
2'							165.4 (s)	
3'							68.6 (s)	
4'(NH)	6.26	s			10, S-CH ₃ ,			6'
5'							169.5 (s)	
6'							62.2 (s)	
7'A	2.99	d	14.4 (7'B)	7'B	9, 9'		34.2 (t)	9, 5', 6', 8', 9'
7'B	3.56	d	14.4 (7'A)	7'A	9'			9, 5', 6', 8', 9'
8'							125.0 (s)	
9'	6.90	d	8.4 (10')	10'	1', 7'A, 7'B		131.6 (d)	7', 11'
10'	7.30	d	8.4 (9')	9'	12'		115.6 (d)	8', 11'
11'							158.7 (s)	
12'	4.51	d	6.0 (13')	13'	10', 16'		64.8 (t)	11', 13', 14'
13'	5.50	br t	6.0 (12')	12'	15'		119.5 (d)	16'
14'							138.5 (s)	
15'	1.81	s			13'		25.8 (q)	13', 14', 16'
16'	1.76	s			12'		18.2 (q)	13', 14', 15'
S-CH ₃	2.34	s			10, 4'		12.4 (q)	3'

a ^1H chemical shift values (δ ppm from SiMe₄) followed by multiplicity and then the coupling constants (J /Hz). Figures in parentheses indicate the proton coupling with that position. b The correlations with geminal and vicinal protons are removed. c Long range ^1H - ^{13}C correlations from H to C observed in the HMBC experiment.

Table S2 NMR spectral data of **3** in MeOH-*d*₄

Position	$\delta_{\text{H}}^{\text{a}}$	<i>J</i> /Hz	¹ H- ¹ H COSY	NOESY ^b	δ_{C}	HMBC (C) ^c
2					166.9 (s)	
3	5.78 d	16.2 (4)	4	5 α	126.8 (d)	2, 5
4	6.68 ddd	16.4 (3), 12.0 (5 α), 4.2 (5 β)	3, 5 α , 5 β	6	146.0 (d)	2, 5
5 α	2.27 ddd	13.2 (5 β), 12.0 (4), 12.0 (6)	4, 5 β , 6	3, 15	42.0 (t)	3, 4, 6, 15
5 β	2.58 ddd	13.2 (5 α), 4.2 (6), 4.2 (4)	4, 5 α	15		
6	5.29 dqd	12.0 (5 α), 6.0 (15), 4.2 (5 β)	5 α , 15	4	70.1 (d)	
8					167.8 (s)	
9	6.08 dd	15.6 (10), 0.6 (11)	10	11, 5'	125.2 (d)	8, 11
10	6.77 dd	15.6 (9), 6.0 (11)	9, 11	12, 13 α	147.2 (d)	8, 9, 11
11	3.96 ddd	9.0 (12), 6.0 (11), 0.6 (9)	10, 12	9, 13 α , 1'	83.8 (d)	9, 10, 12, 1'
12	3.76 ddd	9.0 (11), 6.6 (13 β), 1.2 (13 α)	13 β	10, 16	72.6 (d)	10, 11, 13, 14
13 α	1.92 dd	16.2 (13 β), 4.2 (14)	14, 13 β	10, 11	37.1 (t)	11, 14, 16
13 β	1.48 ddd	16.2 (13 α), 6.6 (12), 2.4 (14)	12, 13 α	16		12
14	5.13 qdd	6.6 (16), 4.2 (13 α), 2.4 (13 β)	13 α , 16		69.7 (d)	12
15	1.36 d	6.0 (6)	6	5 α , 5 β	20.6 (q)	5, 6
16	1.33 d	6.6 (14)	14	12, 13 β	18.4 (q)	13, 14
1'	5.00 d	3.6 (2')	2'	11	102.9 (d)	11, 2', 5'
2'	3.43 dd	9.6 (3'), 3.6 (1')	1', 3'	4'	73.9 (d)	3'
3'	3.67 dd	9.6 (2'), 9.6 (4')	2', 4'	5'	75.1 (d)	2', 4', 5'
4'	3.34 dd	10.2 (5'), 9.6 (3')	3', 5'	2', 6'	71.4 (d)	3', 5', 6'
5'	3.55 dt	10.2 (4'), 3.6 (6')	4', 6'	9, 3'	74.3 (d)	
6'	3.64 d	3.6 (5')	5'	4'	62.1 (t)	4', 5'

a ¹H chemical shift values (δ ppm from SiMe₄) followed by multiplicity and then the coupling constants (*J*/Hz). Figures in parentheses indicate the proton coupling with that position. b The correlations with geminal and vicinal protons are removed. c Long range ¹H-¹³C correlations from H to C observed in the HMBC experiment.

Figure S1 14-membered macrodiolides associated to halosmysins

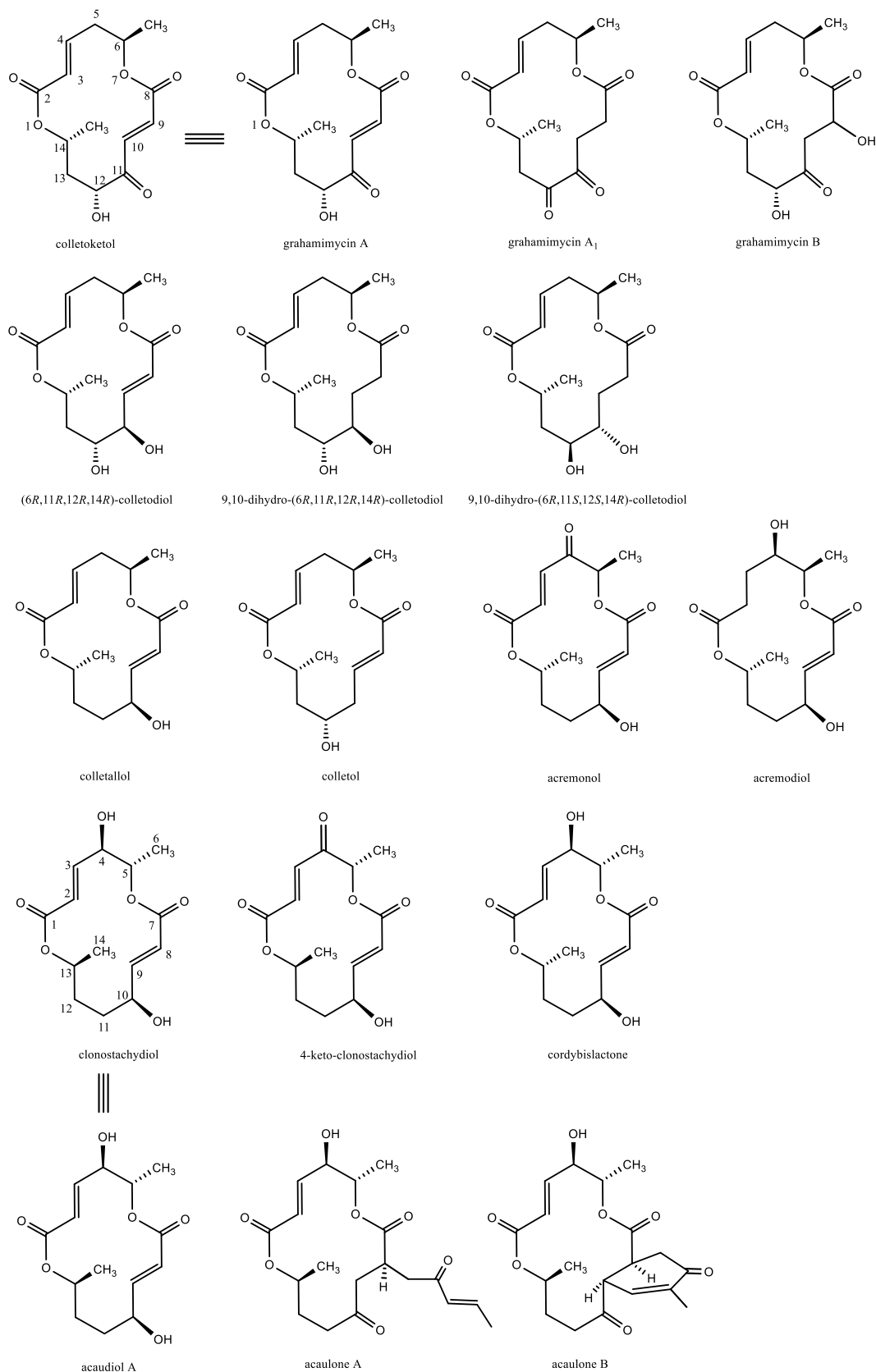
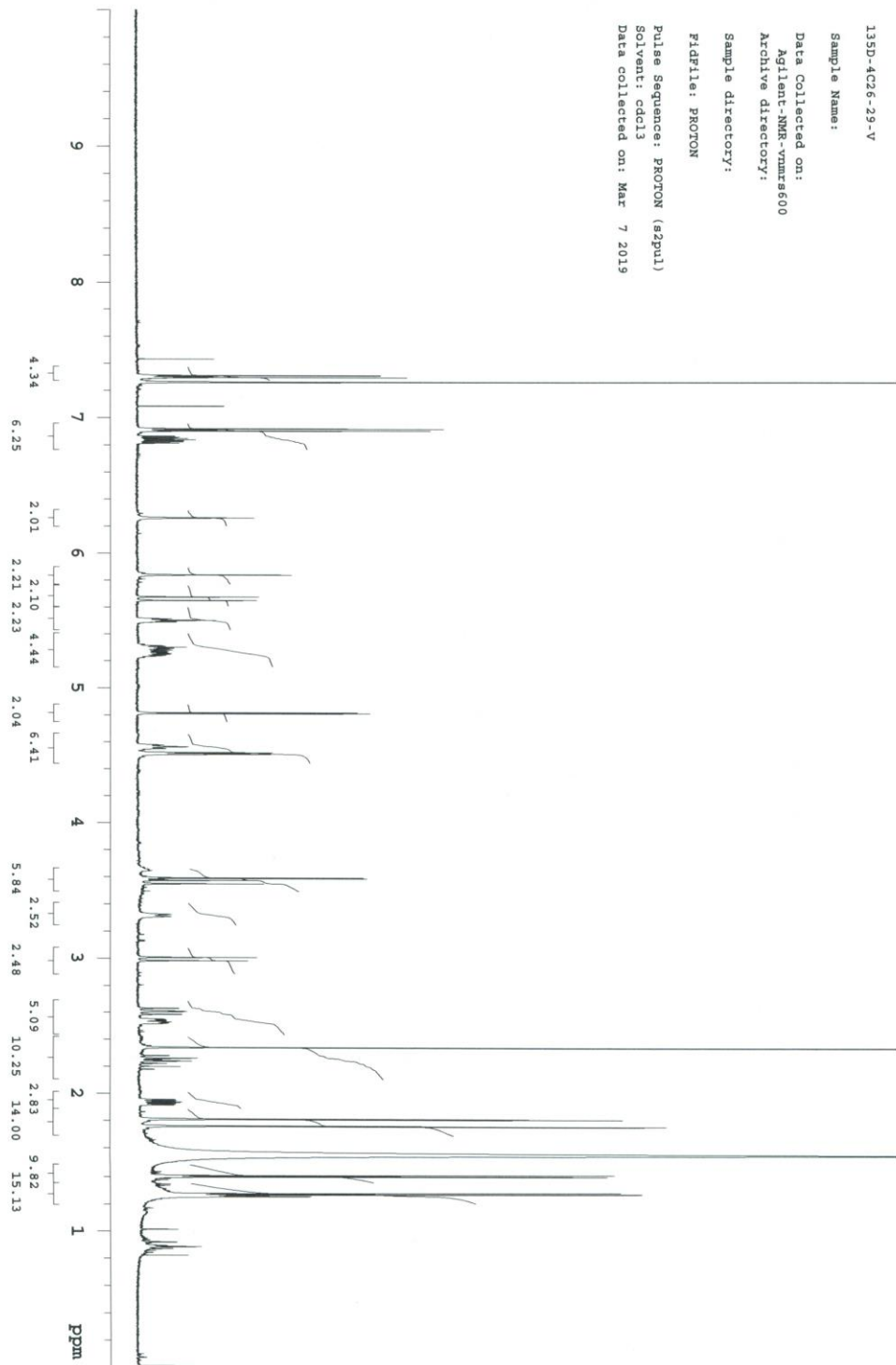
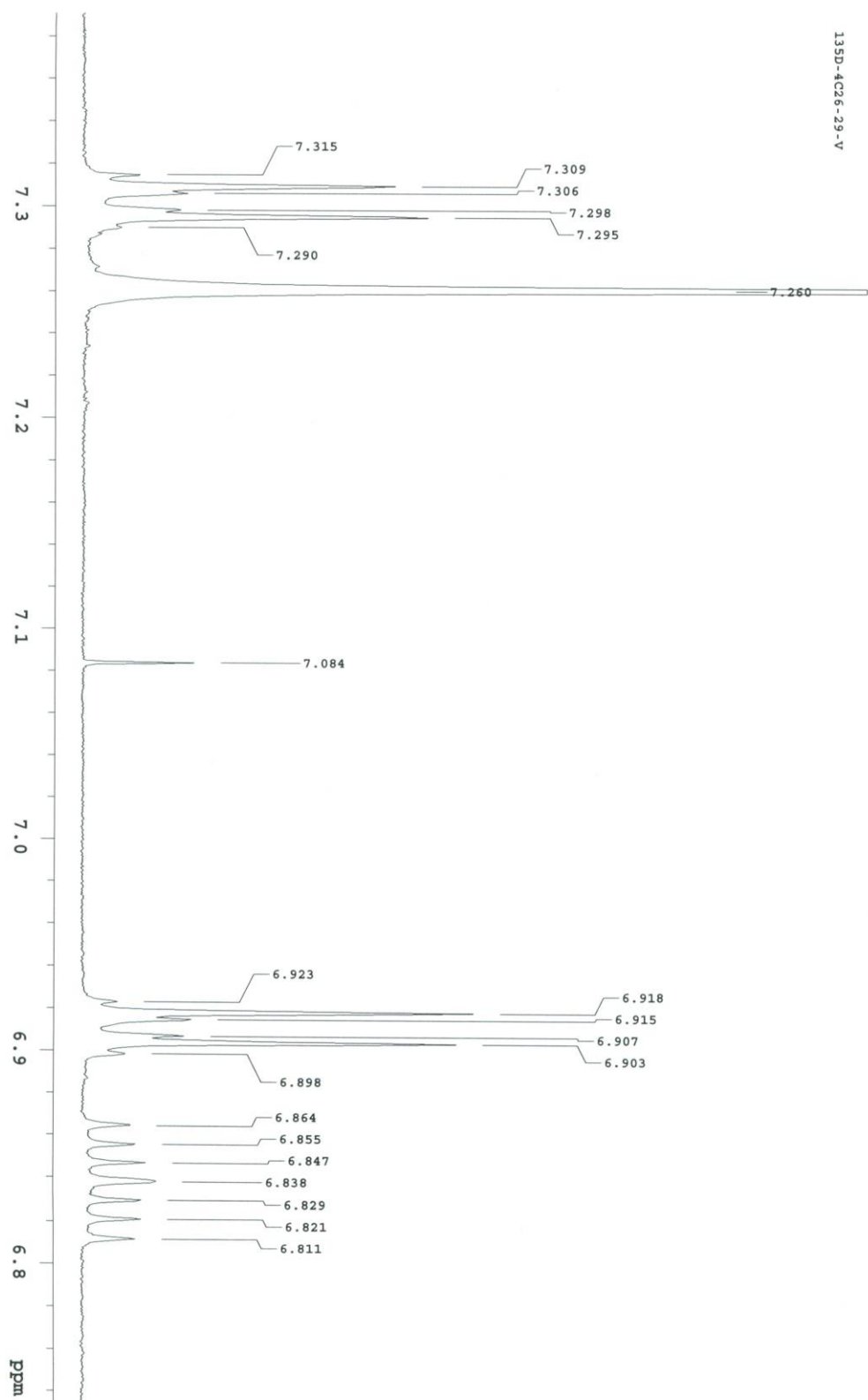
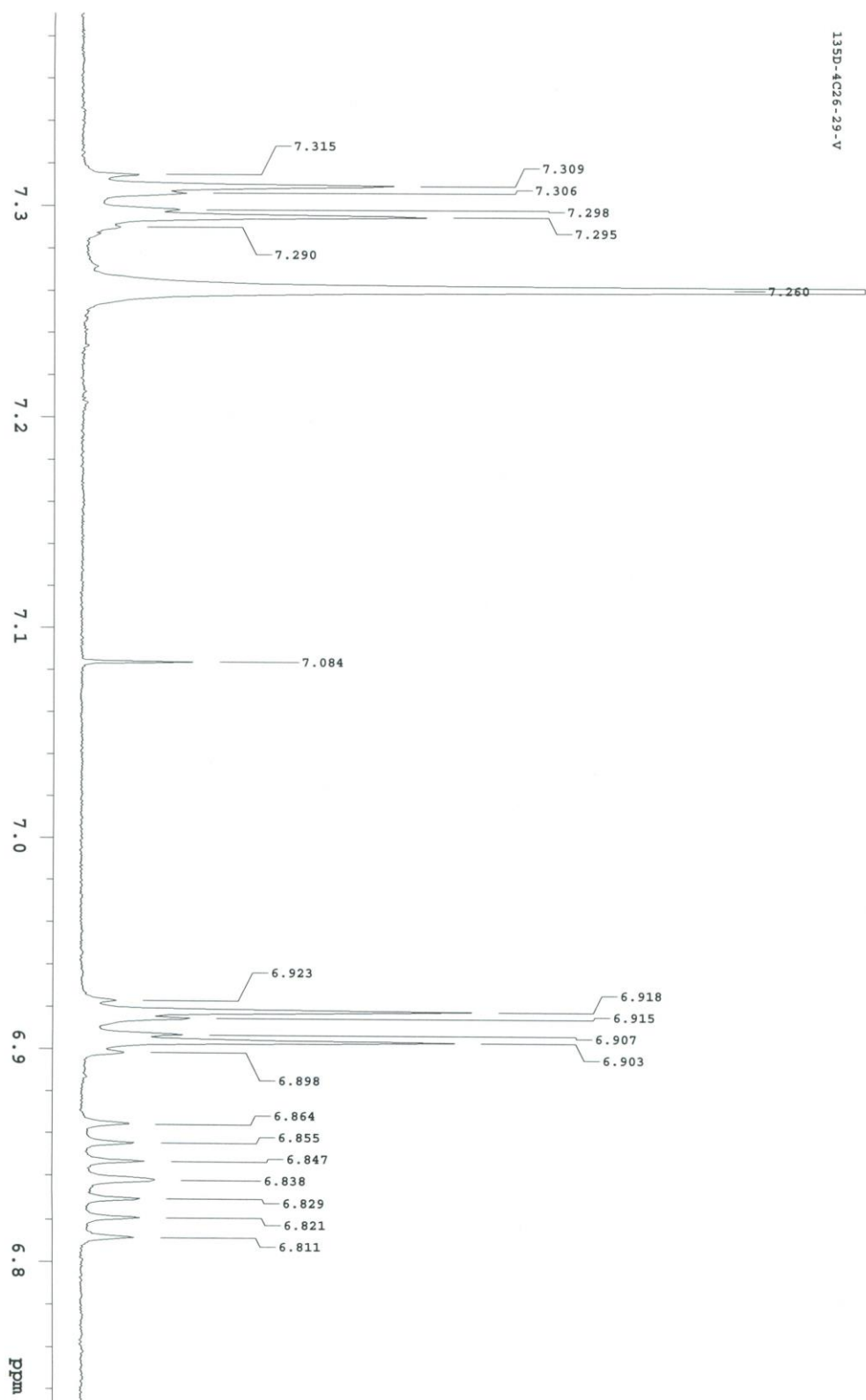
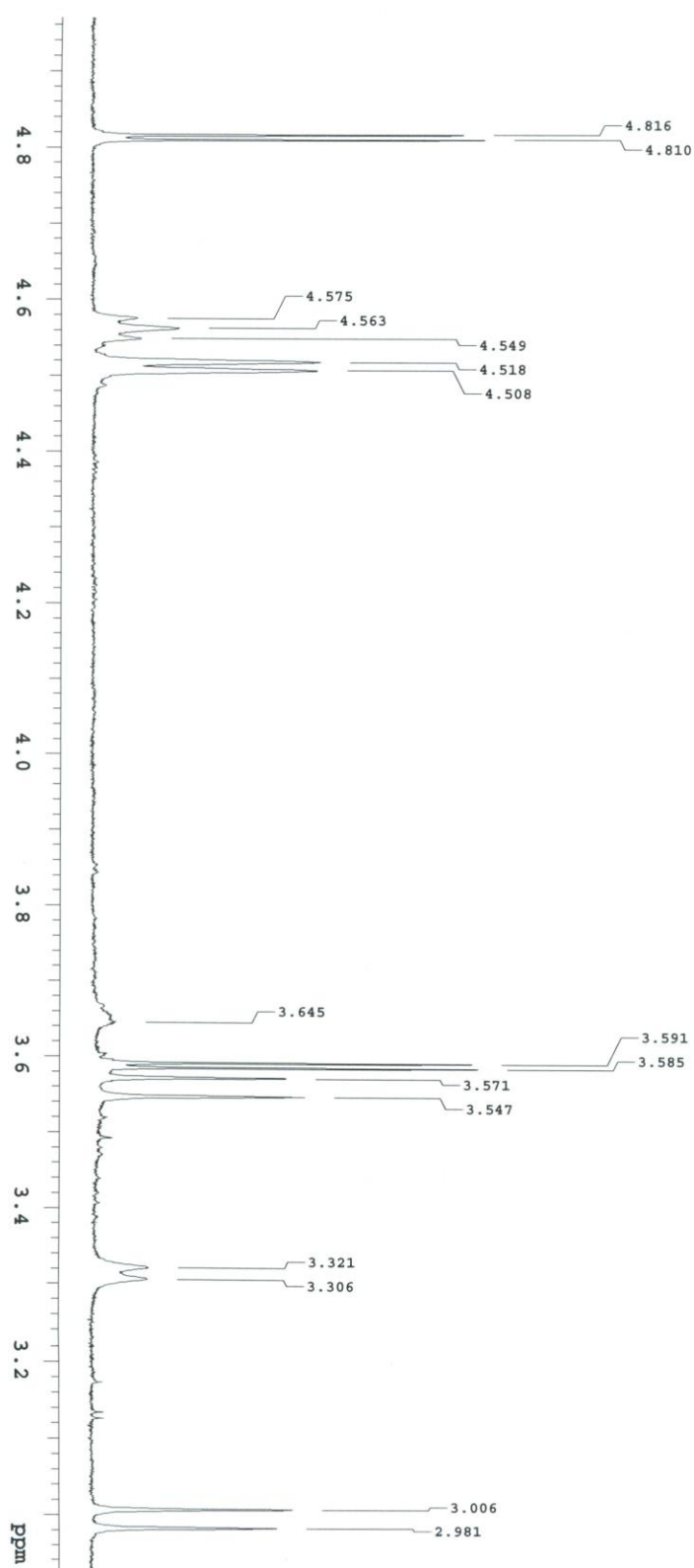


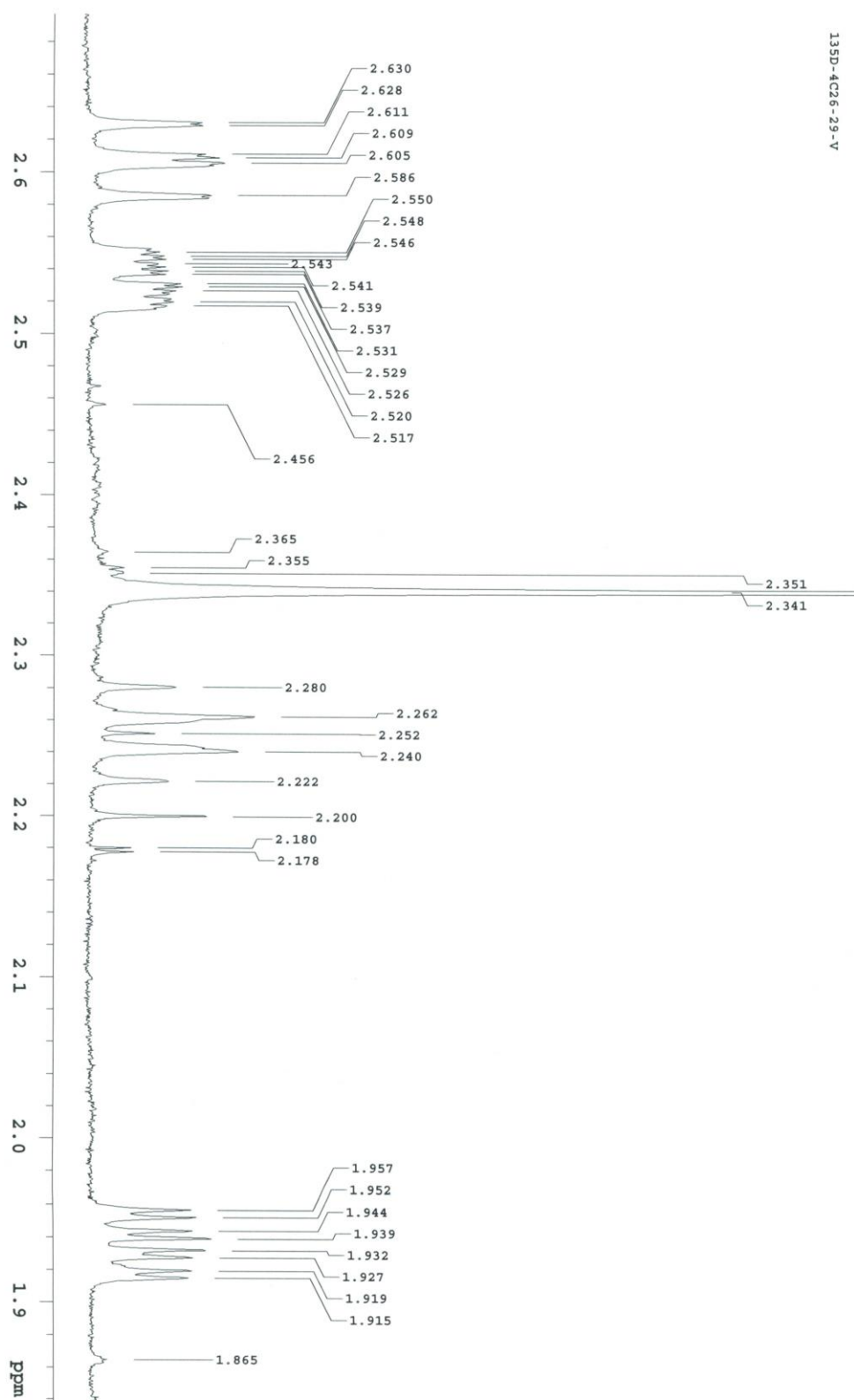
Figure S2 ^1H NMR spectrum of 2 in CDCl_3











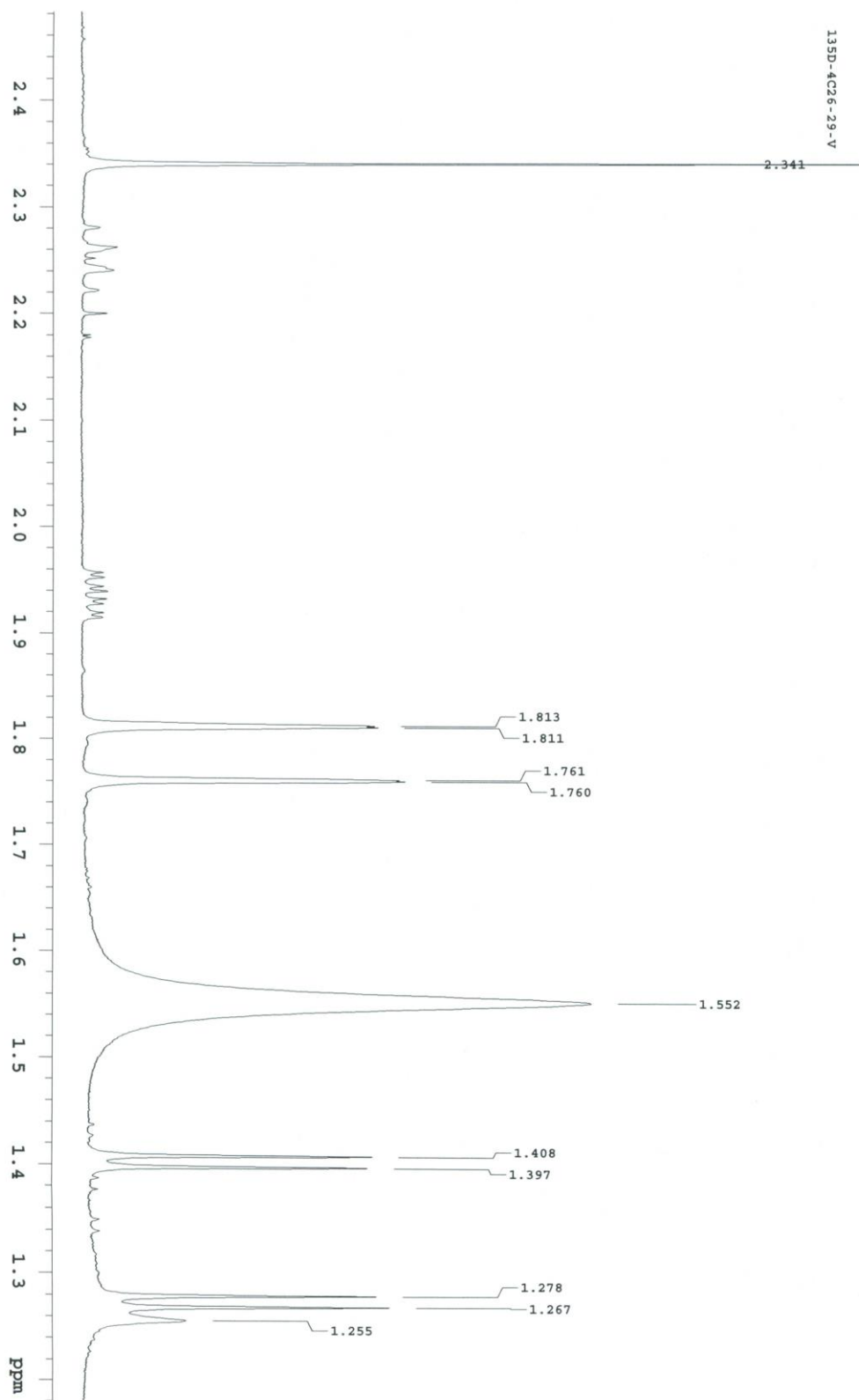
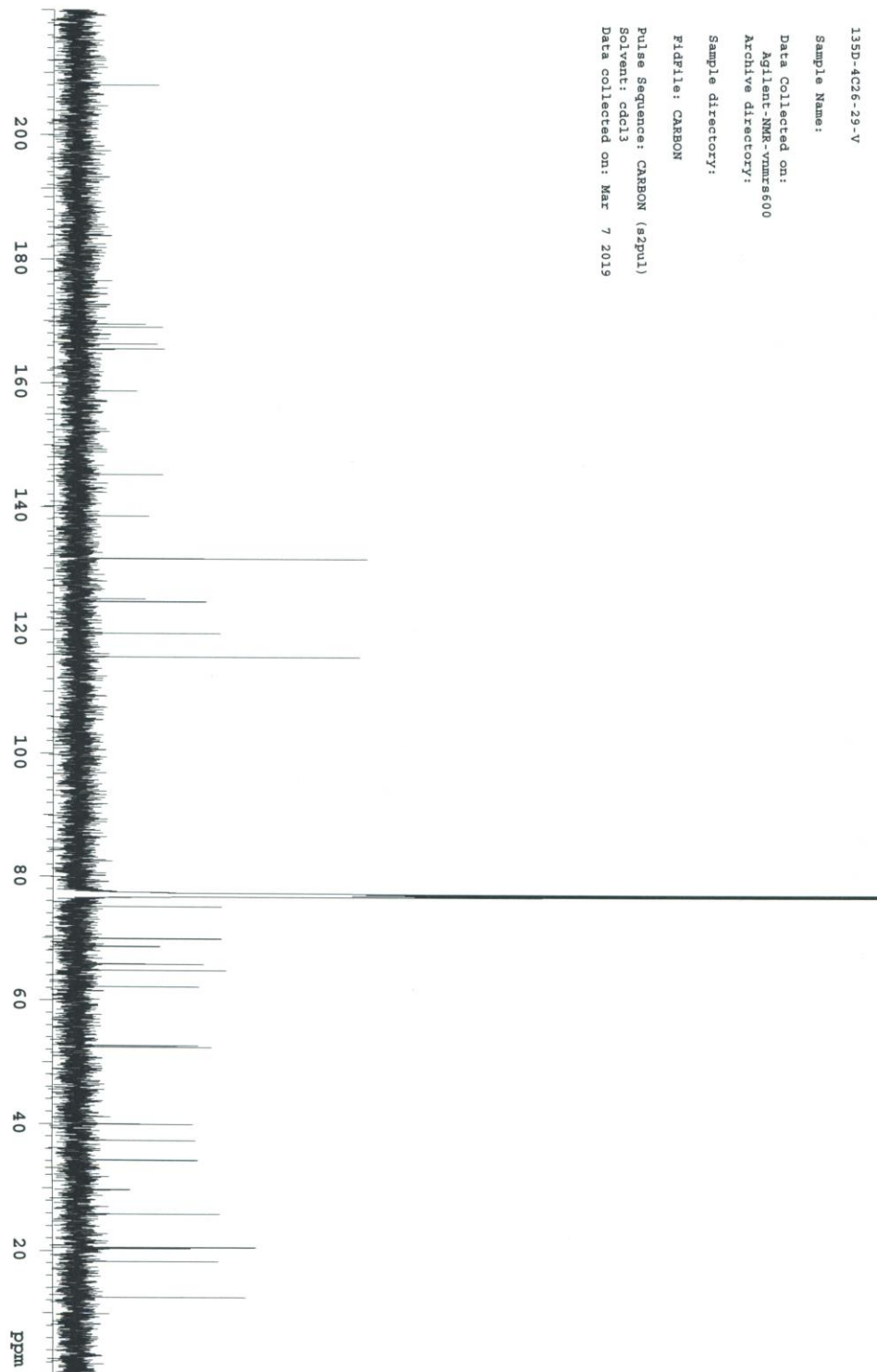


Figure S3 ^{13}C NMR spectrum of 2 in CDCl_3



13D-4C26-29-V			
INDEX	FREQUENCY	PPM	HEIGHT
1	3170.5	207.967	15.1
2	28865.5	191.360	-7.6
3	25561.7	169.458	12.5
4	25486.6	168.960	15.8
5	25069.3	166.194	14.9
6	24956.0	165.443	16.2
7	23937.6	158.691	10.9
8	23366.6	154.906	-6.9
9	21899.6	145.181	15.8
10	21126.3	140.054	-7.0
11	20891.6	138.498	13.2
12	19848.9	131.586	55.4
13	18862.9	125.049	12.5
14	18795.9	124.605	24.3
15	18021.4	119.470	27.0
16	17441.1	115.623	54.0
17	11646.2	77.207	3903.0
18	11615.0	77.000	4264.6
19	11582.6	76.785	4182.1
20	11324.8	75.077	27.4
21	10591.9	70.218	-6.6
22	10541.1	69.881	27.3
23	10351.5	68.624	15.5
24	9928.4	65.819	23.5
25	9777.0	64.815	28.3
26	9315.9	62.156	23.0
27	7939.0	52.631	22.9
28	7902.0	52.385	25.5
29	6028.2	39.963	21.9
30	5631.7	37.335	22.4
31	5154.3	34.170	22.9
32	4977.4	32.997	-6.4
33	4479.2	29.694	9.9
34	3896.6	25.832	27.1
35	3088.6	20.475	34.0
36	3059.7	20.284	21.4
37	2748.7	18.222	26.7
38	1871.3	12.406	32.0

13SD-4C26-29-V

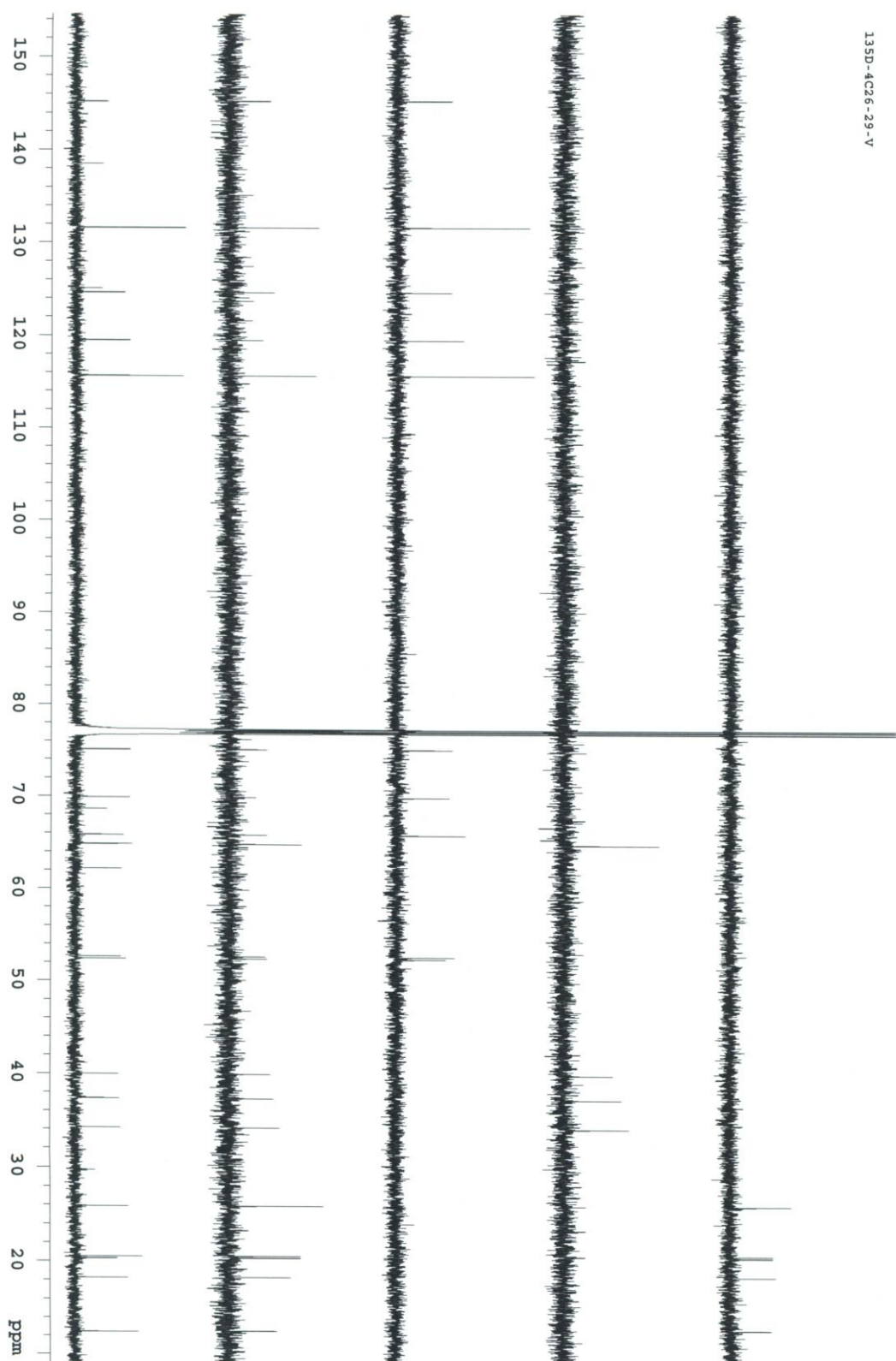
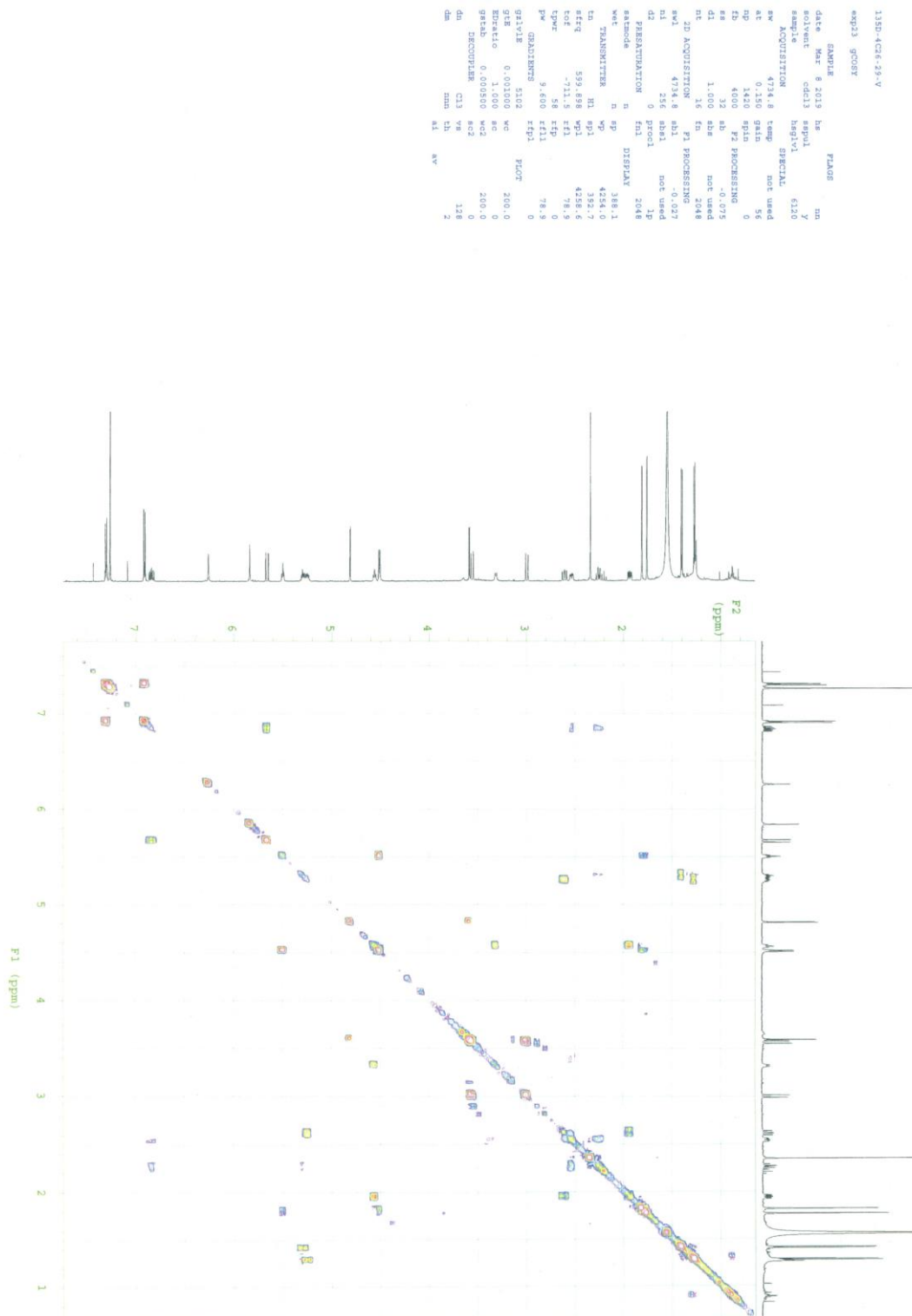


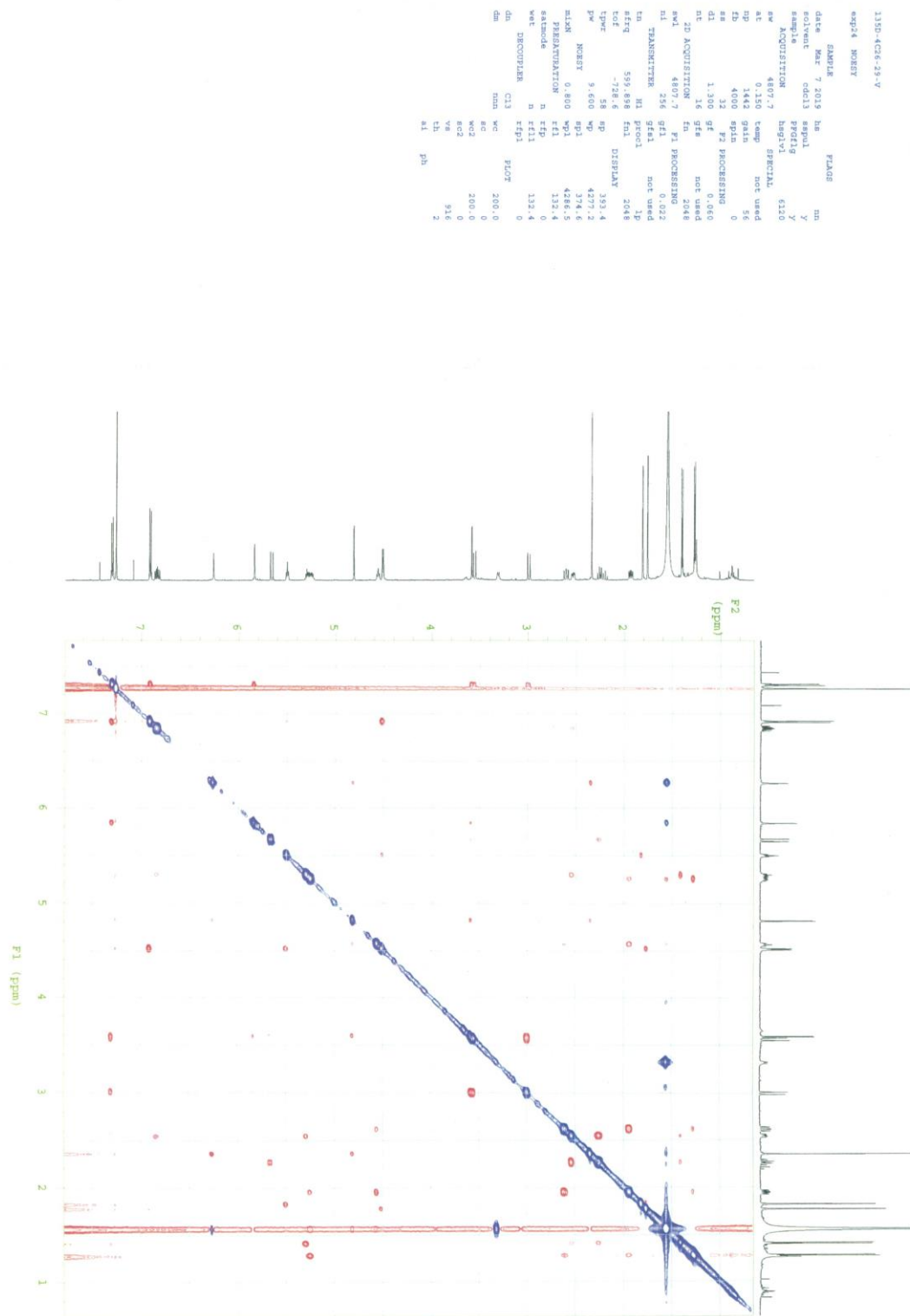
Figure S4 ^1H - ^1H COSY of 2



exp23 gcosx

[illegible]

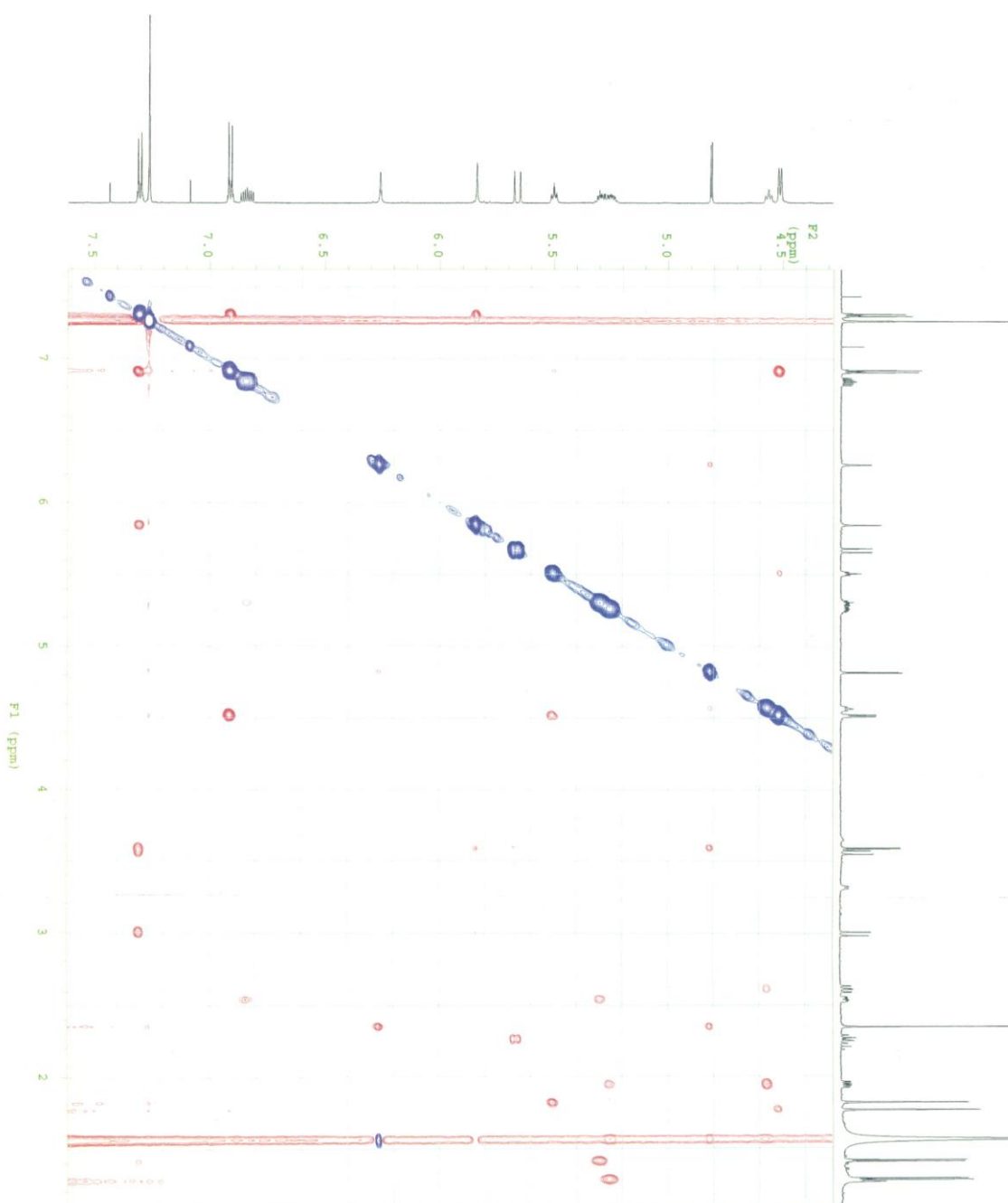
Figure S5 NOESY of 2



exp24 NOESY

exp24 NOESY

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Mar 7 2015	hs		
sample	cdcl3	spul	y
		poft	y
ACQUISITION	hsq11	6110	
4607.7		SPECTAL	
at	0.150	temp	not used
dp	1442	gain	56
td	4000	sp	
sc	12	f2	PROCESSING 0
ds	1.300	se	0.060
nt	1442	gfe	not used
2D ACQUISITION			
sw1	4607.7	p1	PROCESSING 0.032
al	256	gfe1	not used
tn	hl	pref1	1p
rfc	559.886		
tdc	-728.6		
type	56	sp	2867.2
nmey	9.600	wp1	2000.1
		wp2	664.9
mlsn	0.800	wp3	3925.0
PRESAMPLATION		rf1	133.4
salmodc	n	rf2	0
wet	n	rf11	133.4
DECOUPLER		rfp1	0
dm	cl13		
nm	wc		256.0
sc	0		
wd	200.0		
sc2	0		
vs	516		
th	2		
ph			



135D-4C76-28-V

exp24 NOSEY

SAMPLE		FLAGS	
date	Mar 7 2019	hs	nm
solvent	cdcl3	sepol	y
sample		proc1g	y
acq1	4897.7	hqv1g	6120
acq2	4897.7	spc1g	6120
at	0.110	temp	not used
nd	1443	gals	56
fb	4000	sp1g	0
ss	32	f2	PROCESSED
d1	1.300	g2	0.060
nt	16	gfs	not used
2d	4897.7	fz	2048
sw1	256	g1	0.022
ni	TRANSMITTER	gfs1	not used
tn	559.888	fm1	2048
rtq	-7.288	sp	2571.9
lwr	9.600	wp1	1981.3
pw	NOSEY	wp2	668.7
mln	0.800	wp1	1572.8
presat	0.800	wp2	132.4
satmode	n	rfp	0
wet	n	rf1	132.4
decoupler	cl3	rfp1	0
dm	nm	wc	250.0
hc	0	wc2	200.0
vc2	0	vc2	916
lb	2	at	ph

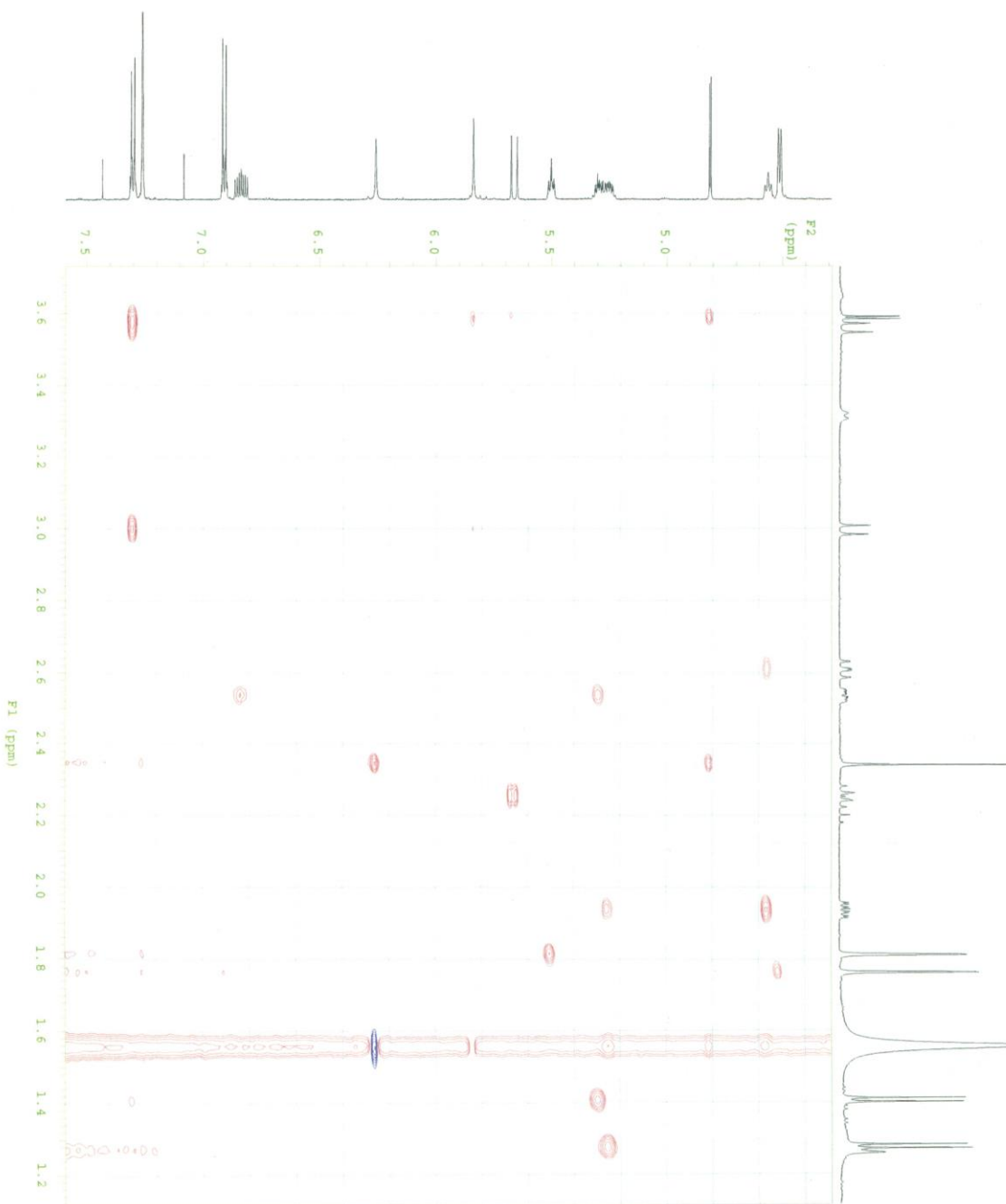
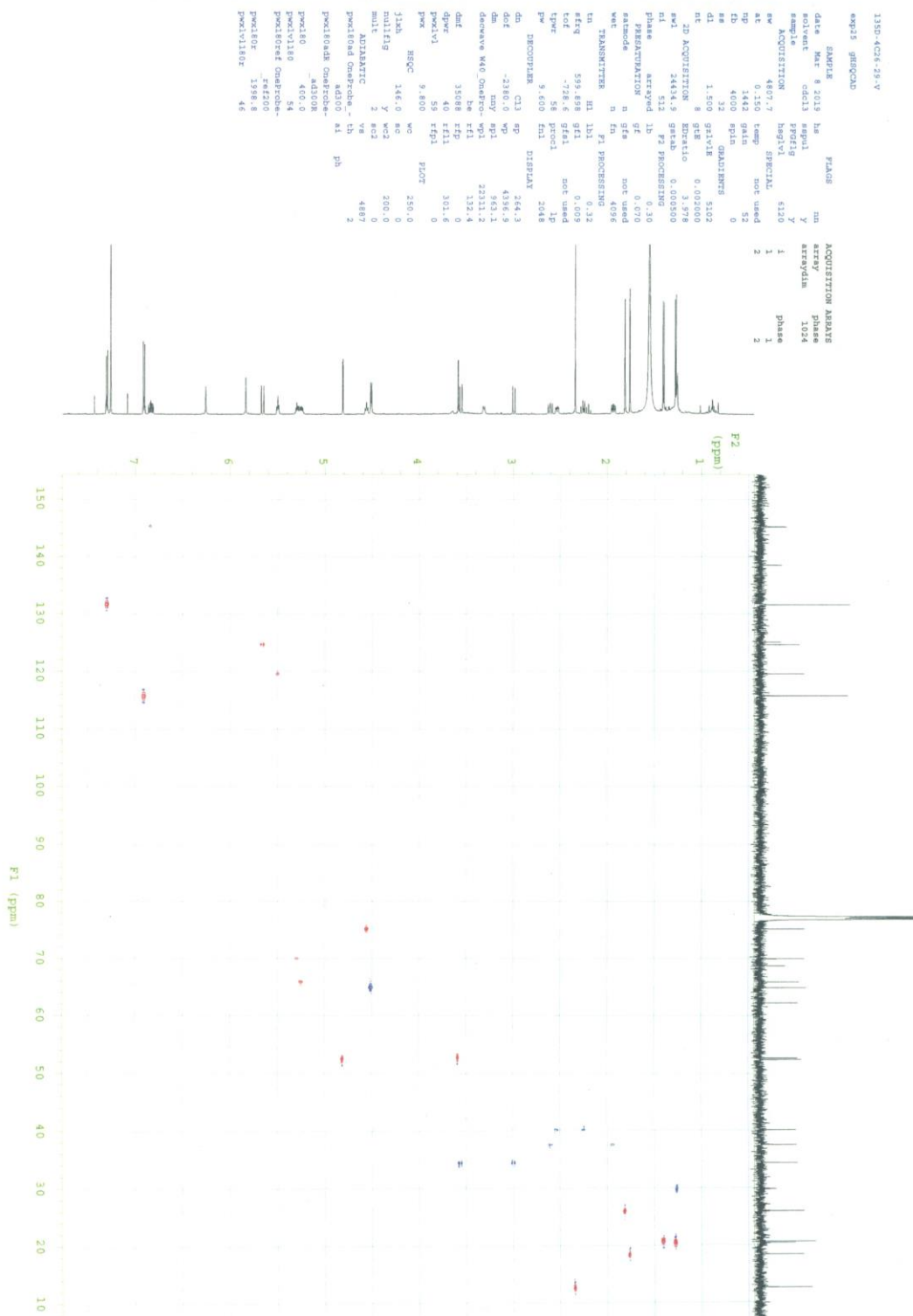


Figure S6 HMQC of 2



113D-4C26-28-V

exp25 SHOCAD

SAMPLE Mar 8 2019 ha
 solvent cdcl3 specul
 sample y
 ACQUISITION 107.7
 at 0.110 temp SPECIAL 6120
 th 1442 gals not used 1
 sp 4000 spins 52 2
 as 32 GRADIENTS 0
 d1 1.500 g1v1e 5102
 nt 8 g1e 0.002000
 2D ACQUISITION ED-ratio 3.378
 w1 24434.9 g1ab 0.000500
 n1 512 F2 PROCESSING
 phase arrayed 1b 0.10
 PREPARATION g1e 0.170
 BIRDGE n g1e not used
 w1 1000 spins 52
 THINNING n 1b1 0.12
 efq 599.898 g1i 0.005
 tot -728.6 g1e1 not used
 lpr 58 proc1 1p
 pw 9.600 fml 2048
 DECOUPLER C13 EP DISPLAY 510.8
 d0 -2380.0 wp 1673.0
 dm 1673.0
 decouple w10 OnePfc-wp1 4531.8
 dnt 35048 f1i 121.4
 dpr 40 f1i 301.6
 pw1v1 59 f1i 0
 pxc 9.800 F2OT 280.0
 j1ch 146.0 mc 0
 nullf1g y wc2 200.0
 mult 2 sc2 0
 ADJNATYC vs 4887
 pw1f0ad OnePfc-lb ph 2
 pw1f0ad OnePfc-
 pw1f0 -adion
 pw1v1180 40034
 pw1f0e OnePfc-
 pw1f0e -ref200
 pw1f0e -1988.8
 pw1v1180r 46

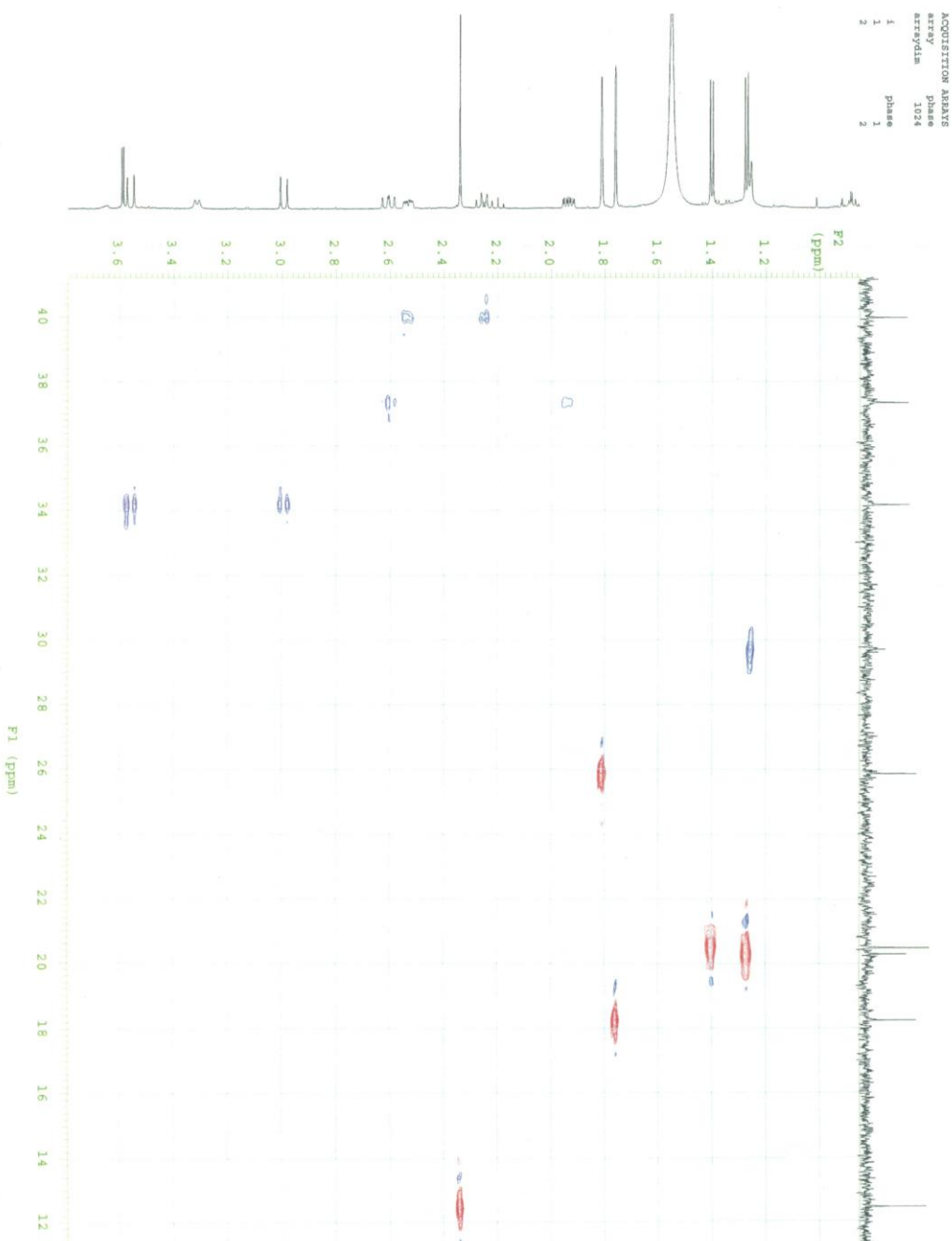
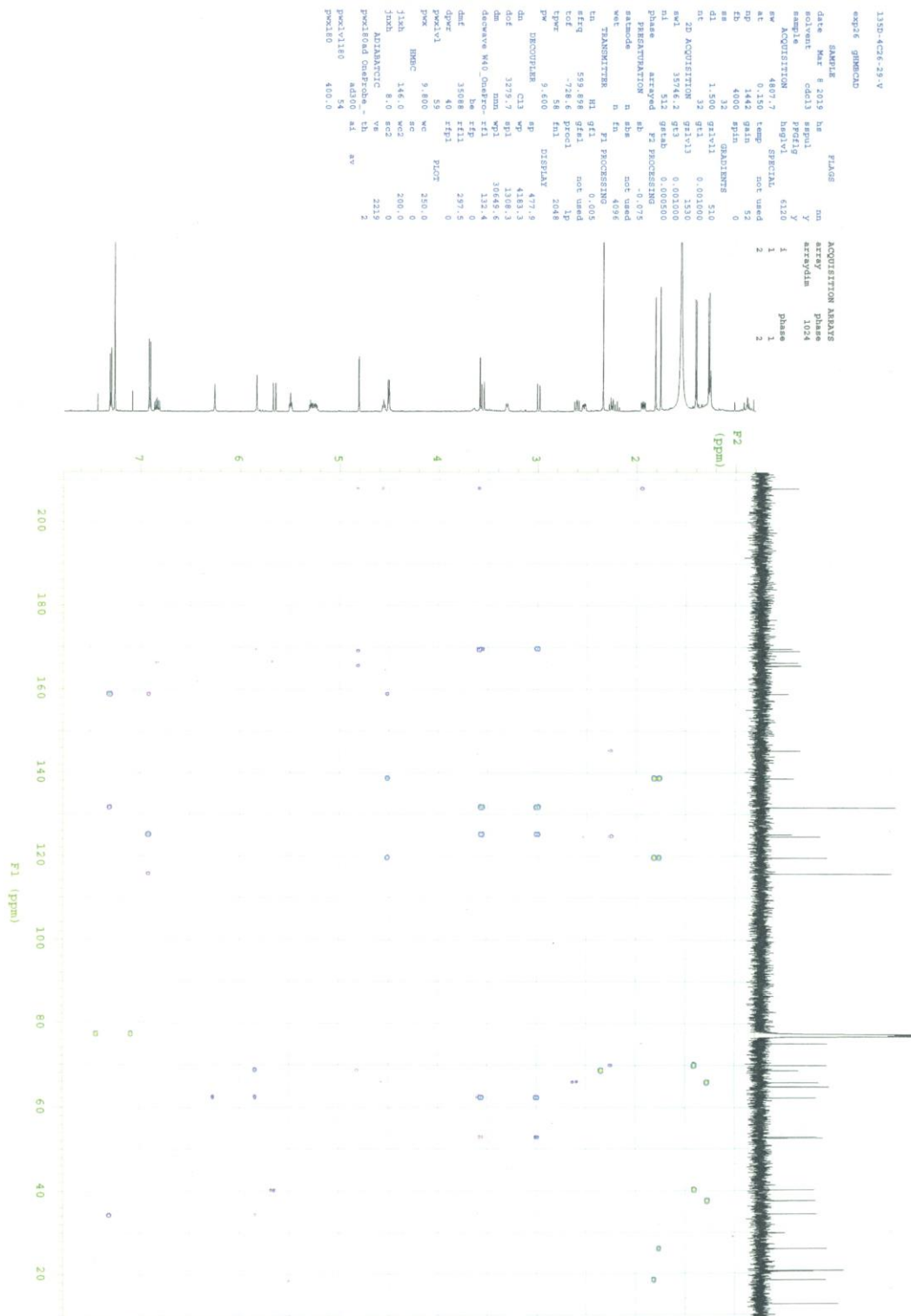
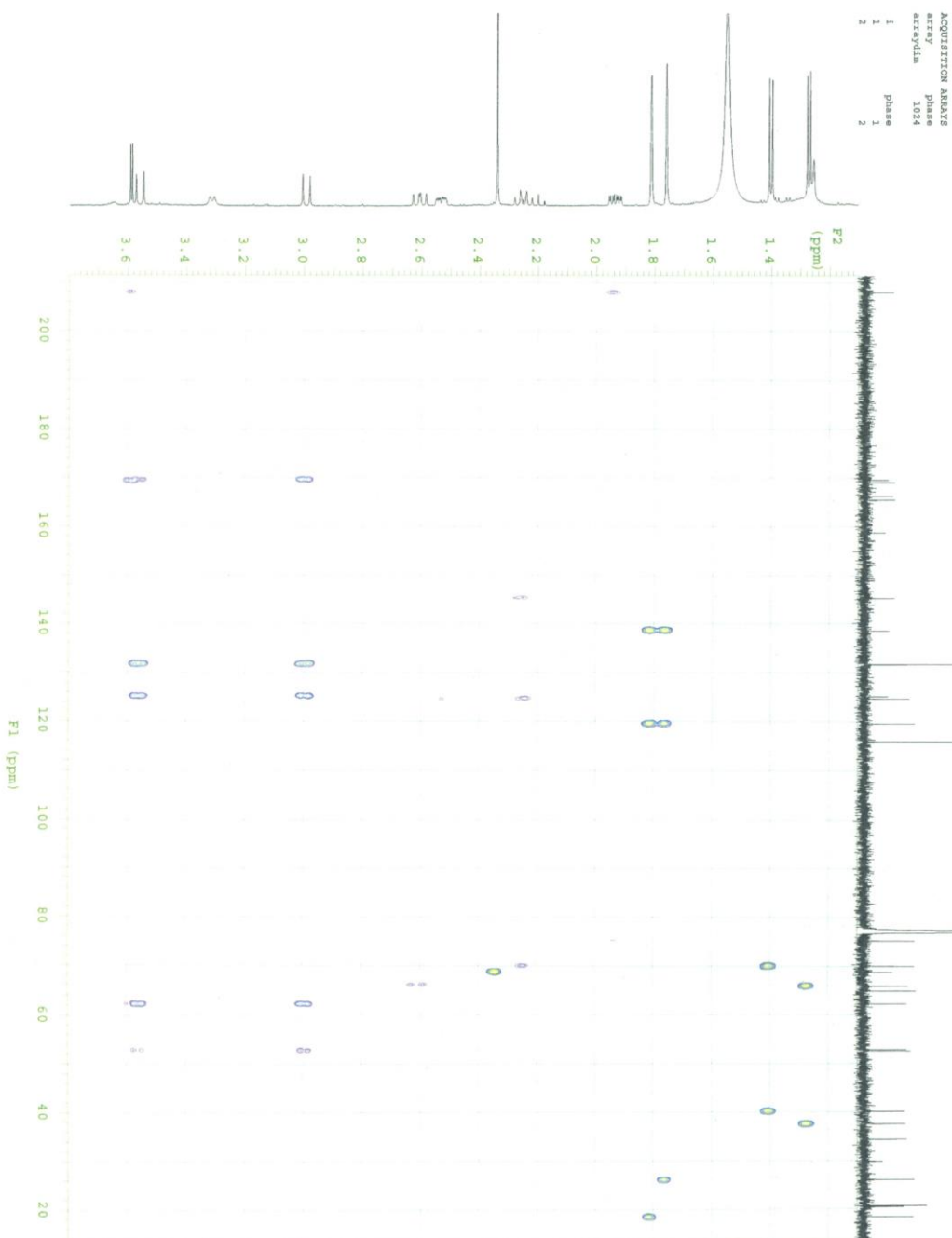


Figure S7 HMBC of 2



135D-4C26-29-V
exg26 gsmcnd

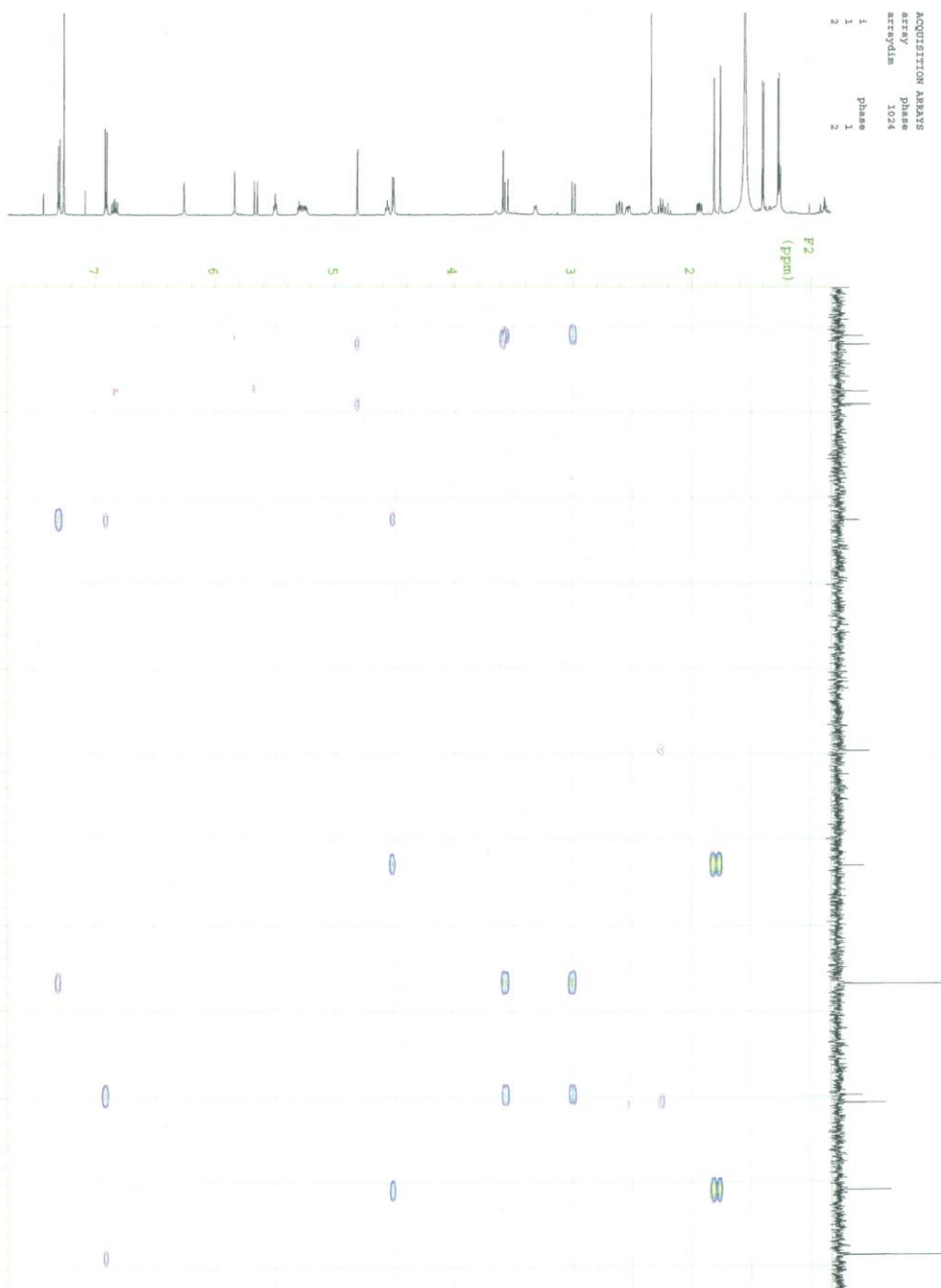
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date Mar 6 2015 hr
subject C013 spm1
sample C013 spm1
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fb 4000 spm 0
ss 32 GRADIENTS 510
dl 1.500 g1v11 0.001000
nt 32 g1v13 1530
2D ACQUISITION
w1 35746.2 g13 0.001000
w2 512 g1ab 0.000500
Phase arrayed F2 PROCESSING
PRESATURATION 0 sbs not used
satmode n sbs not used
wet n f0 4096
TRANSMITTER n f0
ln n1 g1 0.005
sfreq 559.898 gfa1 not used
tot -728.6 proc1 lp
tpr 58 fml
pw 9.600 DISPLAY 2048
DECOUPLER C13 sp 661.0
ddt 3279.7 w1 1617.4
ddt 3279.7 w2 2911.5
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be rfd 128.4
dms 35088 rfd1 297.5
dpr 40 rfd1 0
pwr1v1 59 PLOT
pwr 9.800 wc 250.0
J1kh 145.0 wc2 200.0
J1kh 8.0 wc2 0
ADJANALYTIC va 2219
pwr1v180 A1800 A1
pwr1v180 450.0



135D-4C26-28-V

exg26 gbmcdm

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acq	46077	hagv1	y	arraydim	1024
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np	1442	gain	52		
fb	4000	mpis	0		
ss	32	GRADIENTS	0		
d1	1.500	gr1v11	510		
nt	32	gr1	0.001000		
2D ACQUISITION	gr1v13	1530			
sw1	35746.2	gr1	0.001000		
nl	512	gr1ab	0.000500		
phase	arrayed	f2	PROCESSING		
PREPARATION	ab	-0.075			
balance	n	not used			
weight	n	not used			
TRANSMITTER	n	not used			
tn	599.698	gr1	not used		
efrq	599.698	gr1	not used		
tot	-728.6	gr1	not used		
lpr	58	fn1	2048		
pw	9.600	DISPLAY	494.7		
dn	494.7	sp	414.0		
ddt	3579.7	sp1	17084.9		
dm	nm	wp1	8901.6		
decoupler	W40	gr1	132.4		
dmr	3508.4	rfp	287.5		
dpr	40	rfp1	0		
lprv1	58	PCOT	250.0		
pw	9.800	mc	0		
lprh	146.0	mc2	200.0		
lprh	8.0	mc2	0		
ADDITIONAL	vs	2119			
pw1v1b6	ad100	al	2		
pw1v1b6	54	av			
pw1v1b6	400.0				



exp26 ghmbcad

exp26 ghmbcad

ACQUISITION ARRAYS	
array	phase
arraydim	1024
1	phase
1	1
2	2



135D-4C26-28-V

exp26 gsmcchd

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date Mar 8 2015
solvent CDCl3
sample 1
ACQUISITION 4807.7
nu 0.150 temp not used
ap 1442 gain 52
fp 4000 spia 0
ss 32
d1 1.500 g1v11 510
nt 32 g1v1 0.001000
2D ACQUISITION
w1 35746.2 g13 0.001000
w2 512 g1ab 0.000500
Phase arrayed F2 PROCESSING 0.075
PRESATURATION 1b not used
saturated n shs not used
wet n fo 4956
TRANSMITTER n F1 PROCESSING
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sfreq 599.898 g1a1 not used
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lpr 58 f1 2048
pw 9.600
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dn 496.7
ddt 3279.7 sp1 4141.0
dm 2250.8
dms 9704.5
decrease W40 Gain 124.4
b rfo 132.0
dnt 35088 r11 297.5
dpr 40 rfp1 0
fpe1v1 59 PLOT
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f1h 146.0 wc2 200.0
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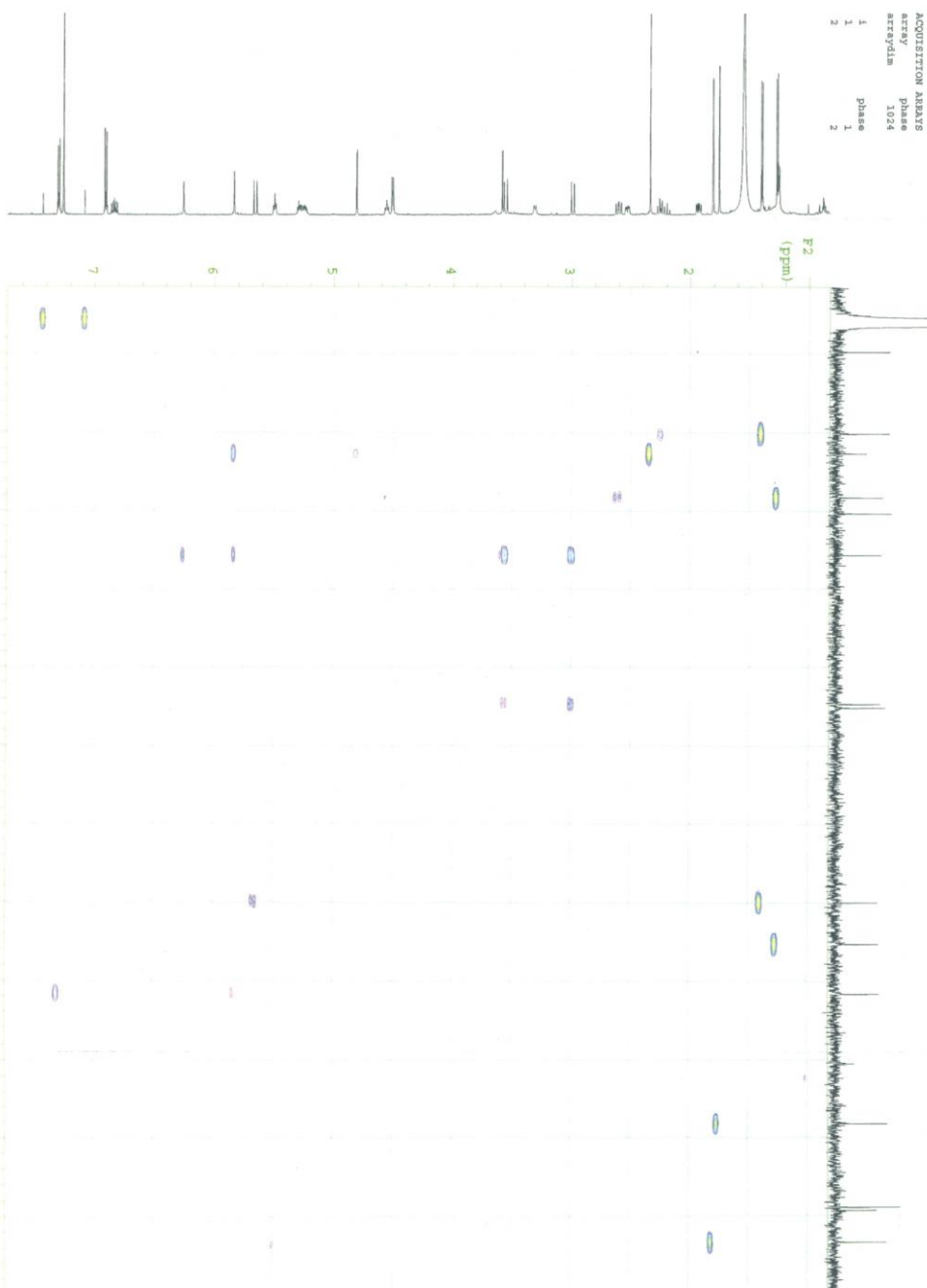
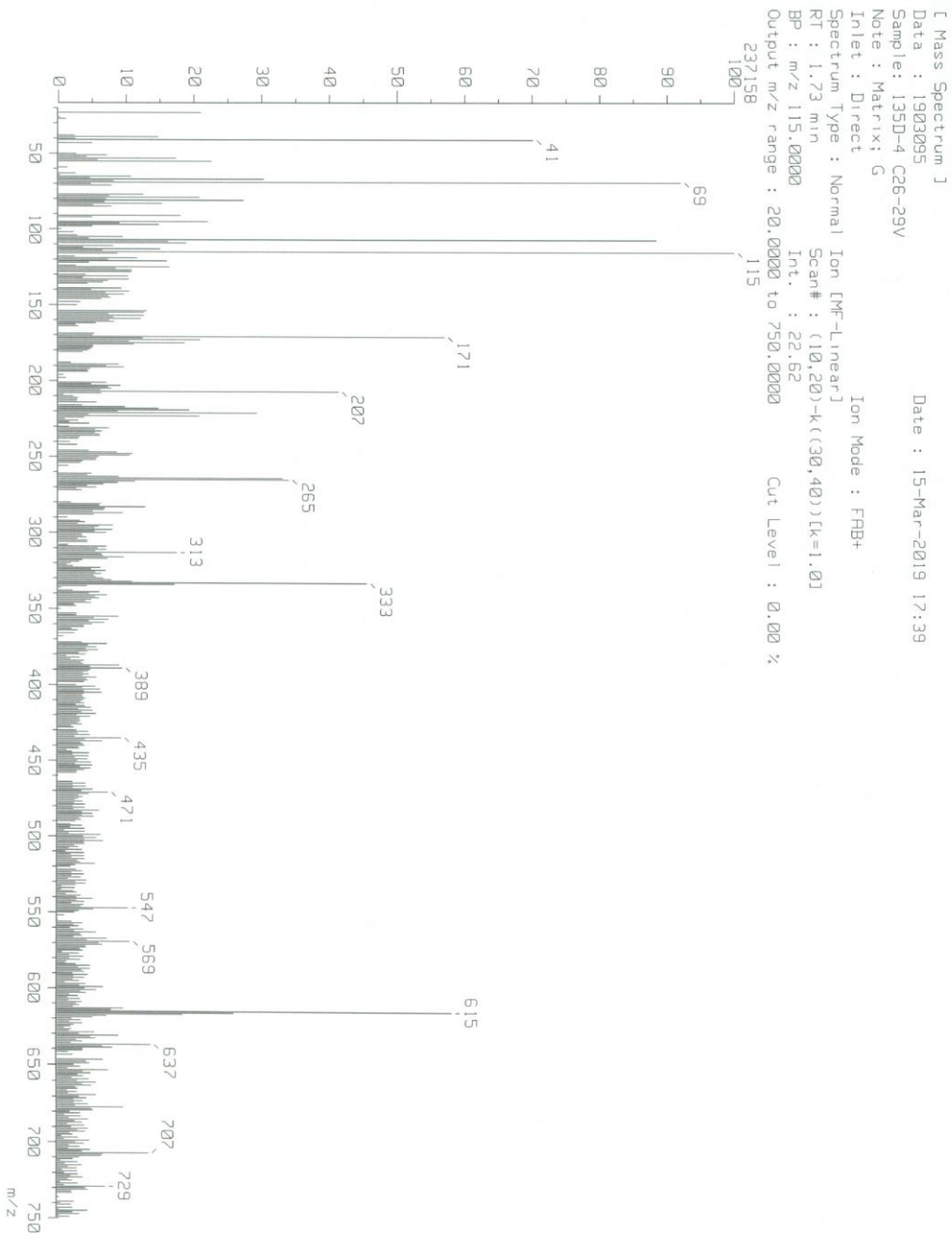


Figure S8 FABMS of 2



[Mass Spectrum]

Data : 1903095

Date : 15-Mar-2019 17:39

Page: 1

Sample: 135D-4 C26-29V

Note : Matrix; G

Inlet : Direct

Ion Mode : FAB+

Spectrum Type : Normal Ion [MF-Linear]

RT : 1.73 min Scan# : (10,20)-k((30,40)) [k=1.0]

BP : m/z 115.0000 Int. : 22.62

Output m/z range : 20.0000 to 750.0000 Cut Level : 10.00 %

m/z	Int.	Norm.	m/z	Int.	Norm.
23.0000	4.75	21.00	332.0000	2.45	10.85
39.0000	3.33	14.72	333.0000	10.30	45.55
41.0000	15.86	70.10	334.0000	3.88	17.15
53.0000	3.93	17.38	547.0000	2.36	10.44
55.0000	5.10	22.56	569.0000	2.42	10.69
65.0000	2.43	10.74	615.0000	13.19	58.31
67.0000	6.86	30.32	616.0000	5.90	26.10
69.0000	20.82	92.06	617.0000	4.19	18.52
77.0000	2.84	12.56	637.0000	3.13	13.84
79.0000	4.71	20.84	707.0000	3.04	13.45
81.0000	6.17	27.30			
83.0000	3.47	15.32			
91.0000	4.08	18.03			
95.0000	4.99	22.07			
97.0000	3.36	14.85			
107.0000	20.01	88.45			
108.0000	3.66	16.18			
109.0000	4.26	18.85			
113.0000	3.40	15.03			
115.0000	22.62	100.00			
119.0000	2.63	11.65			
121.0000	3.61	15.97			
125.0000	3.71	16.42			
127.0000	2.45	10.85			
128.0000	2.44	10.78			
131.0000	2.32	10.28			
133.0000	2.36	10.43			
141.0000	2.37	10.47			
154.0000	2.94	13.02			
155.0000	2.86	12.65			
157.0000	2.86	12.64			
159.0000	2.76	12.20			
171.0000	12.90	57.02			
172.0000	2.82	12.45			
173.0000	4.74	20.96			
174.0000	2.35	10.38			
175.0000	4.22	18.66			
176.0000	2.52	11.15			
207.0000	9.35	41.34			
218.0000	3.34	14.77			
219.0000	4.37	19.31			
221.0000	6.63	29.32			
223.0000	4.71	20.82			
248.0000	2.48	10.96			
249.0000	2.38	10.52			
264.0000	7.49	33.11			
265.0000	7.69	34.02			
266.0000	2.57	11.34			
283.0000	2.91	12.85			
313.0000	3.96	17.51			

[Elemental Composition]

Data : 1904044

Date : 11-Apr-2019 18:01

Page: 1

Sample: 135D-4 C26-29 V

Note : Matrix; G

Inlet : Direct

Ion Mode : FAB+

RT : 6.47 min

Scan#: (50,55)

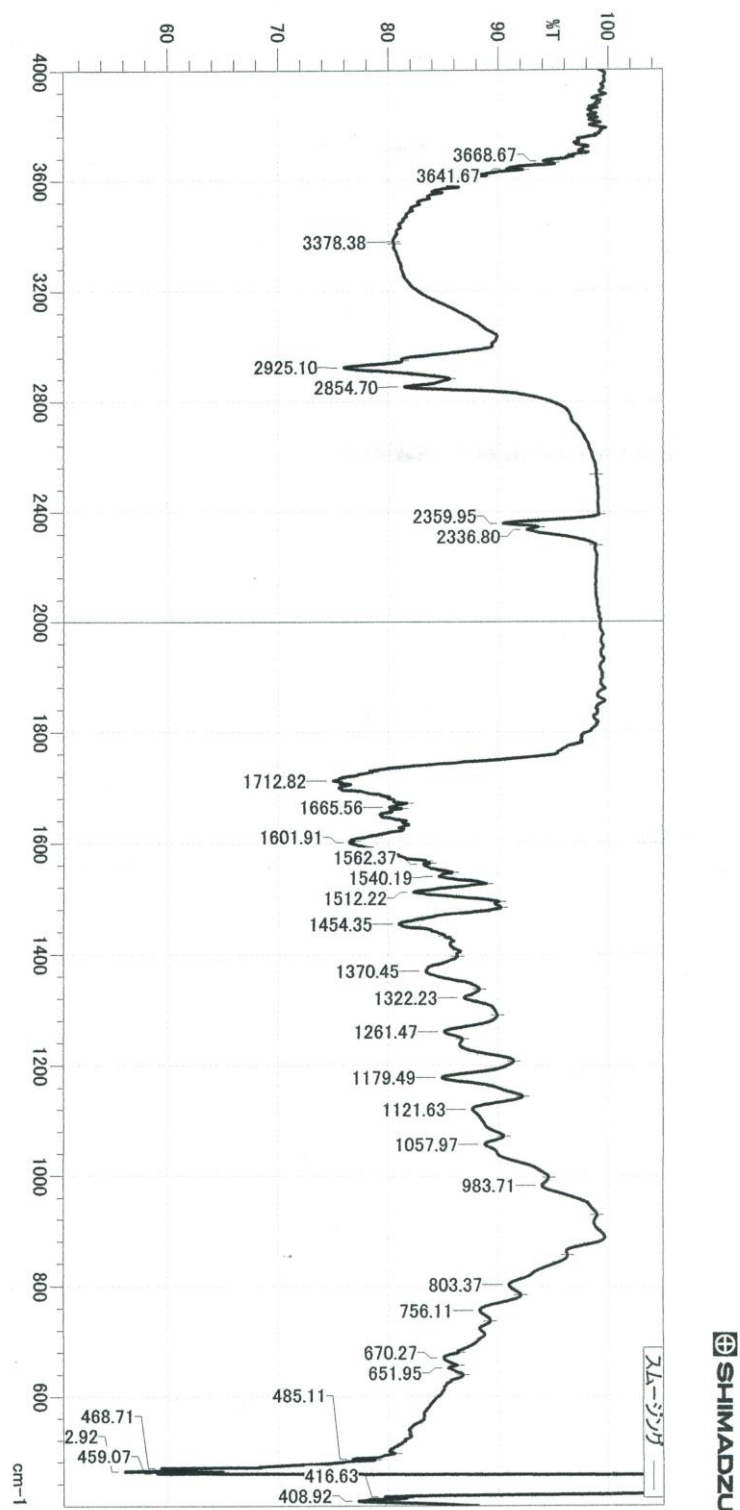
Elements : C 35/25, H 45/35, N 4/0, O 12/0, S 1/0

Mass Tolerance : 20ppm, 2mmu if m/z > 100

Unsaturation (U.S.) : -1.0 - 40.0

Observed m/z	Int%	Err[ppm / mmu]	U.S.	Composition
615.2378	100.0	+0.3 / +0.2	14.5	C 31 H 39 N 2 O 9 S

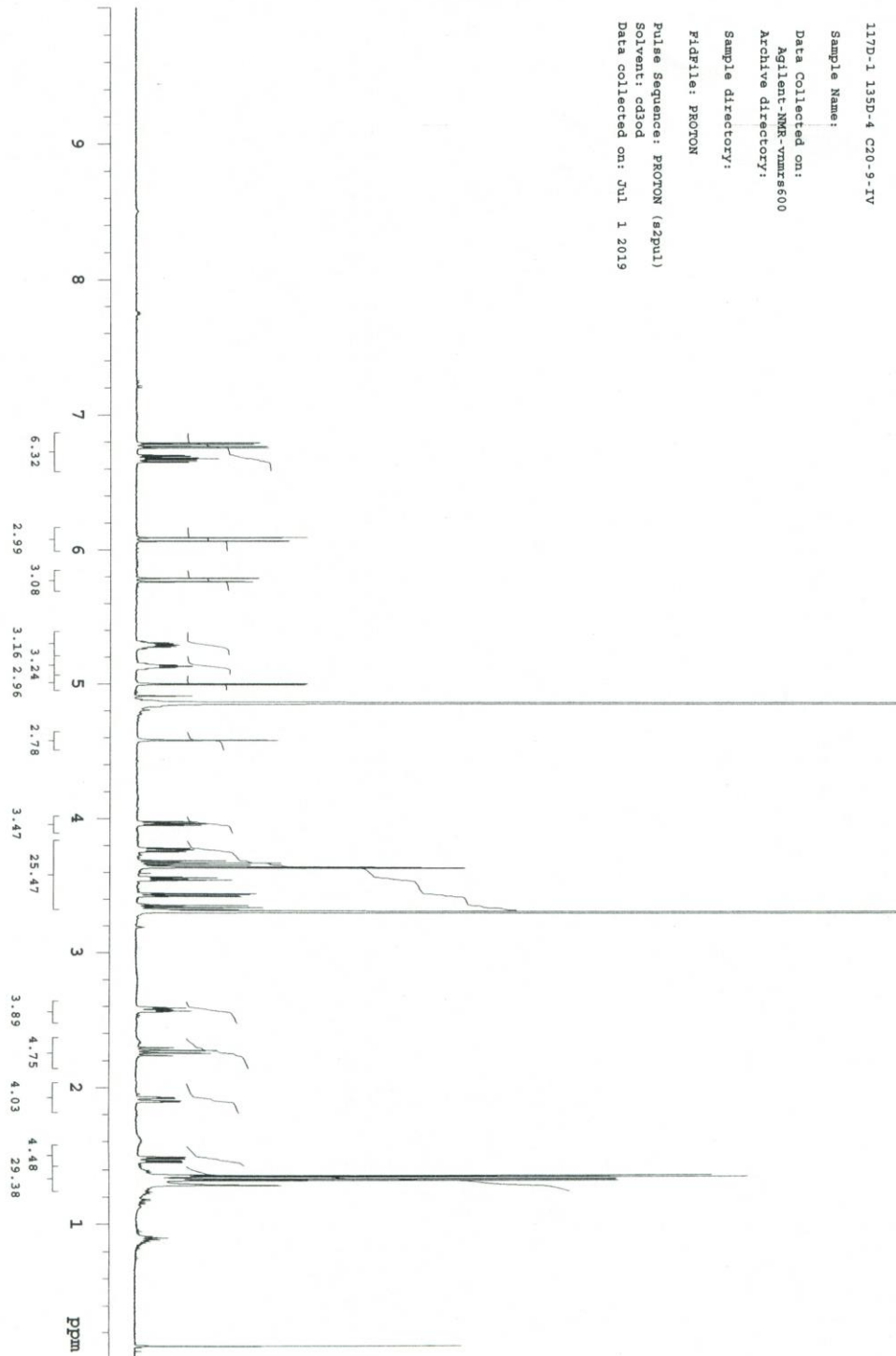
Figure S9 IR spectrum of 2



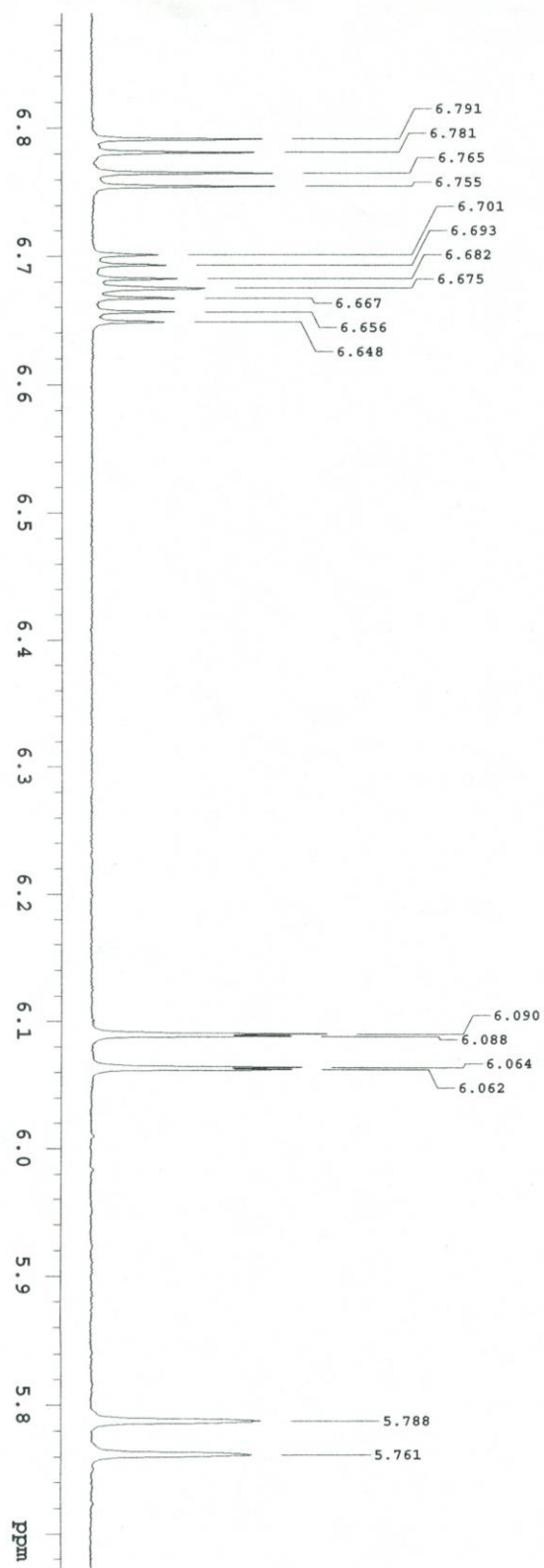
D:*医薬分子化学\山田\135D-4 c26 29 V-2 3.ispd

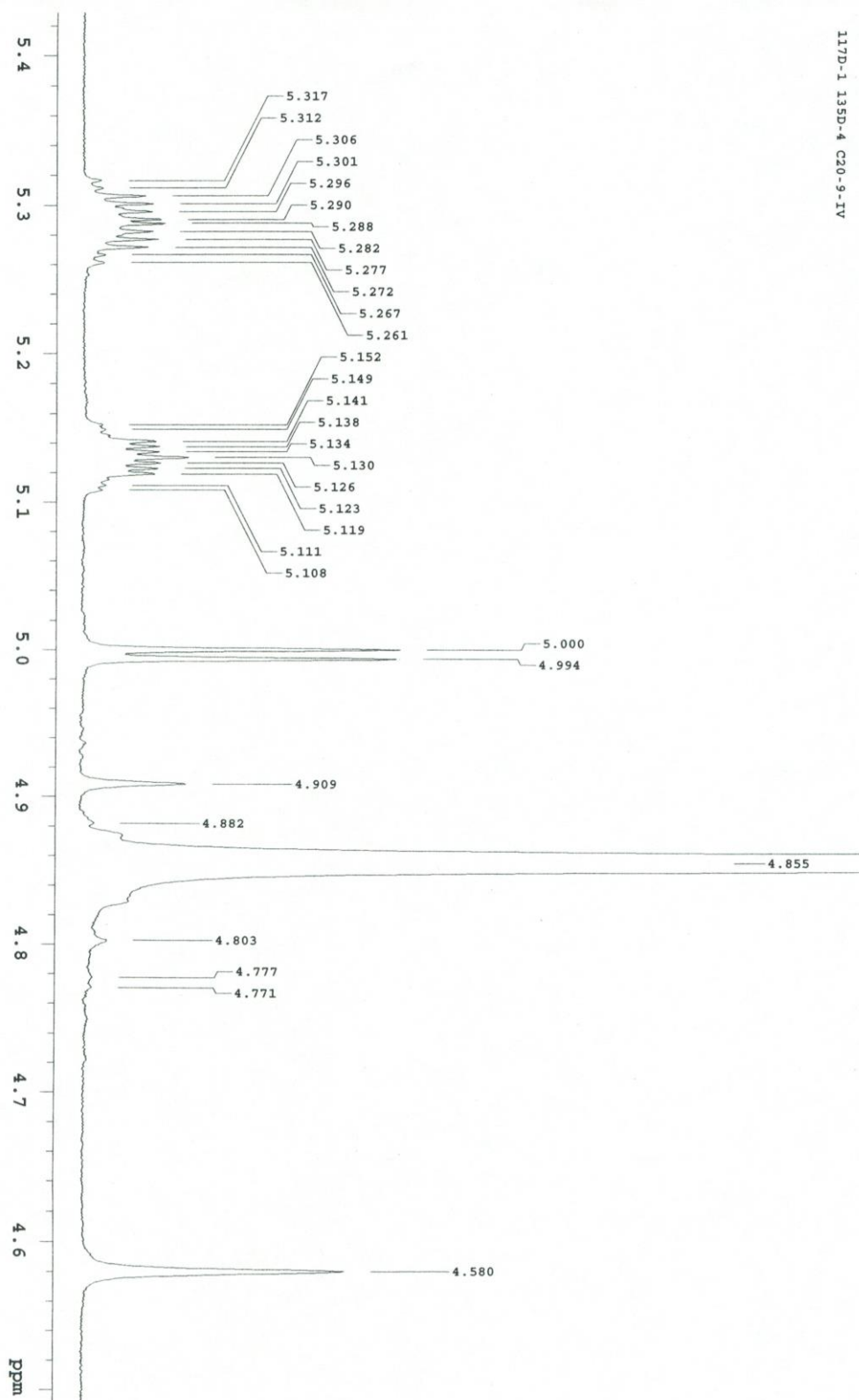
2	サンプル名	135D-4 c26 29 V
3	サンプルID	
4	オプティ	
5	測定モード	透過率
6	アトメックス関数	Happ-Genzel
9	掃査回数	32
10	分解	2 cm⁻¹

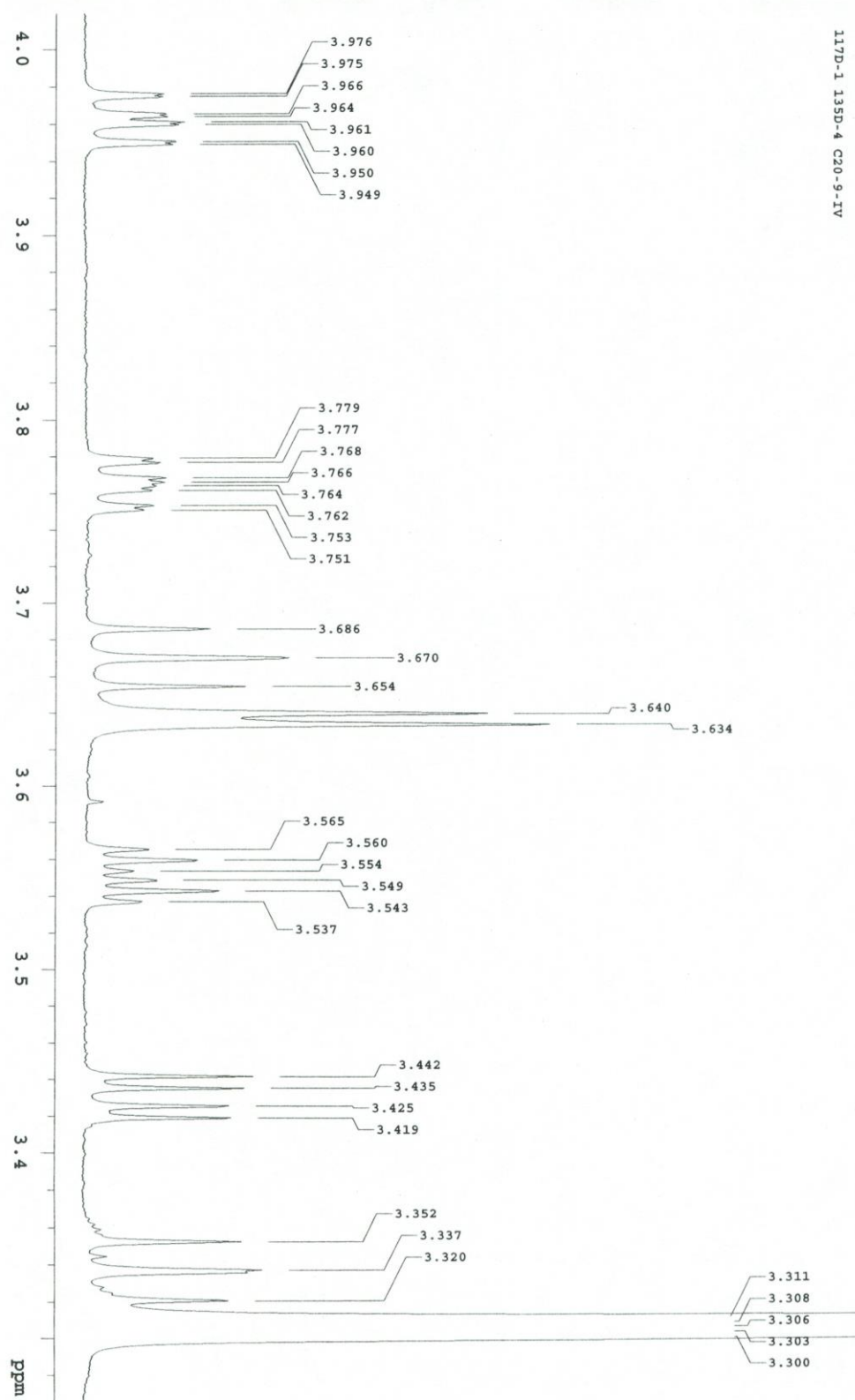
Figure S10 ^1H NMR spectrum of 3 in $\text{MeOH-}d_4$



117D-1 135D-4 C20-9-IV







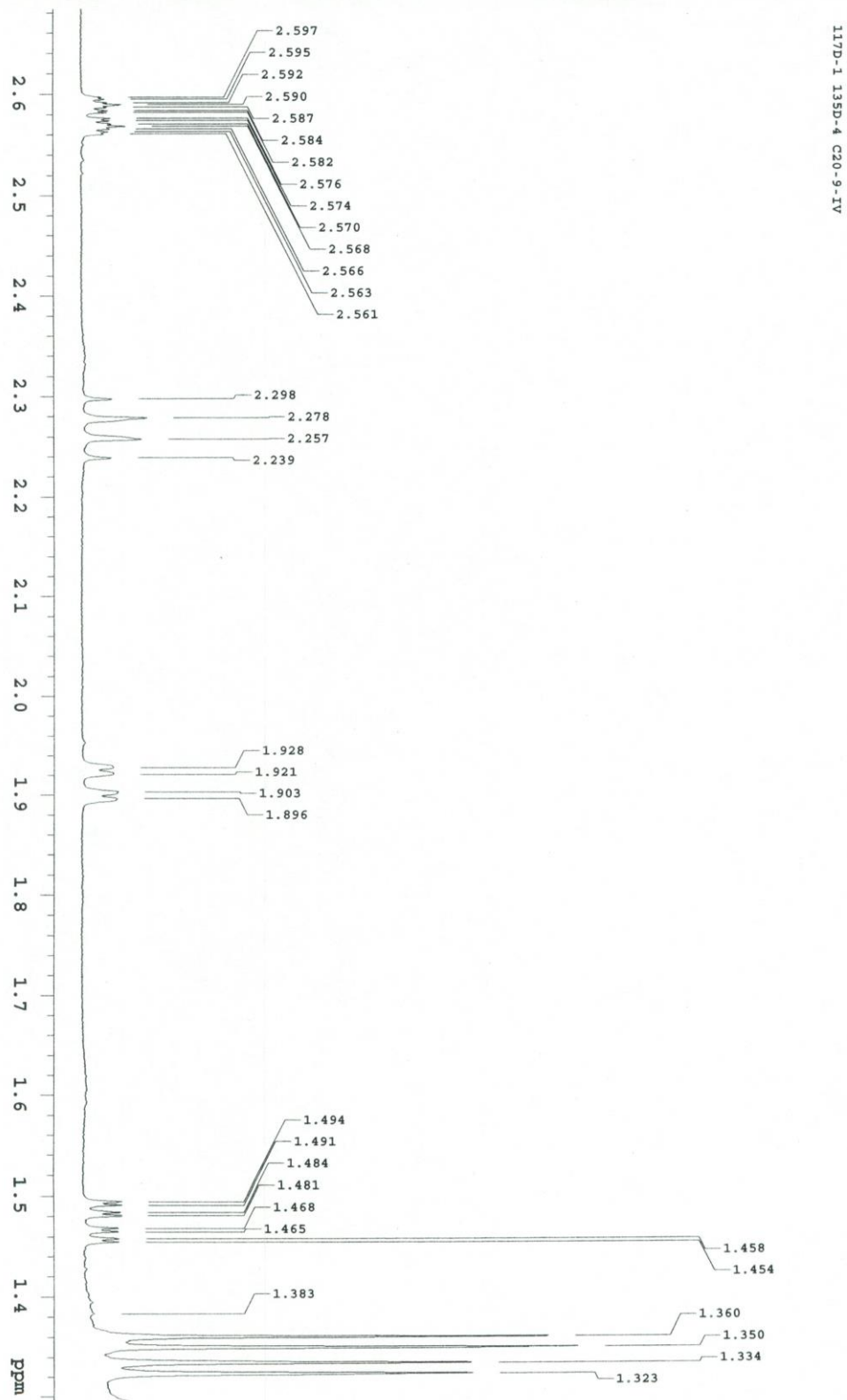
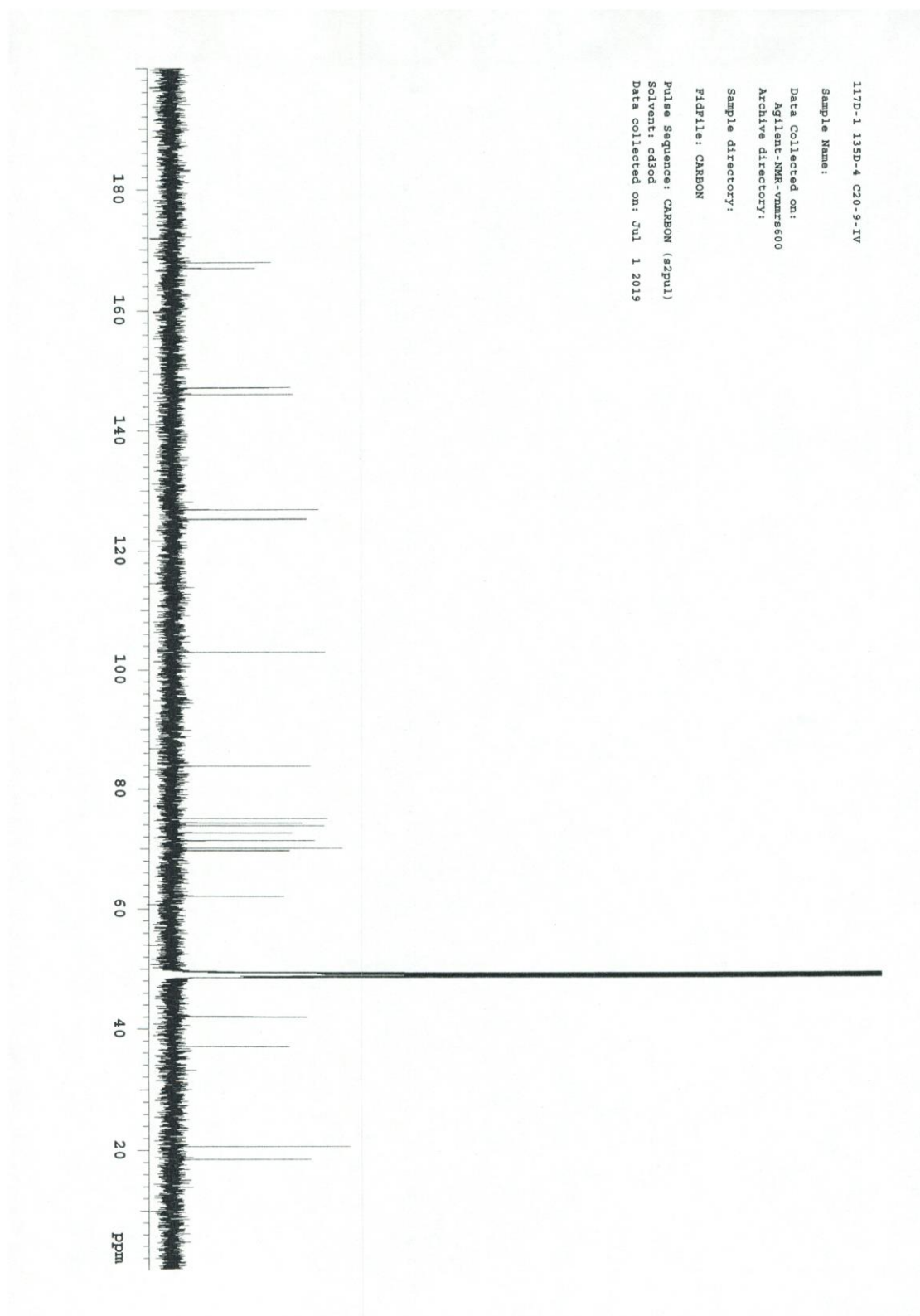


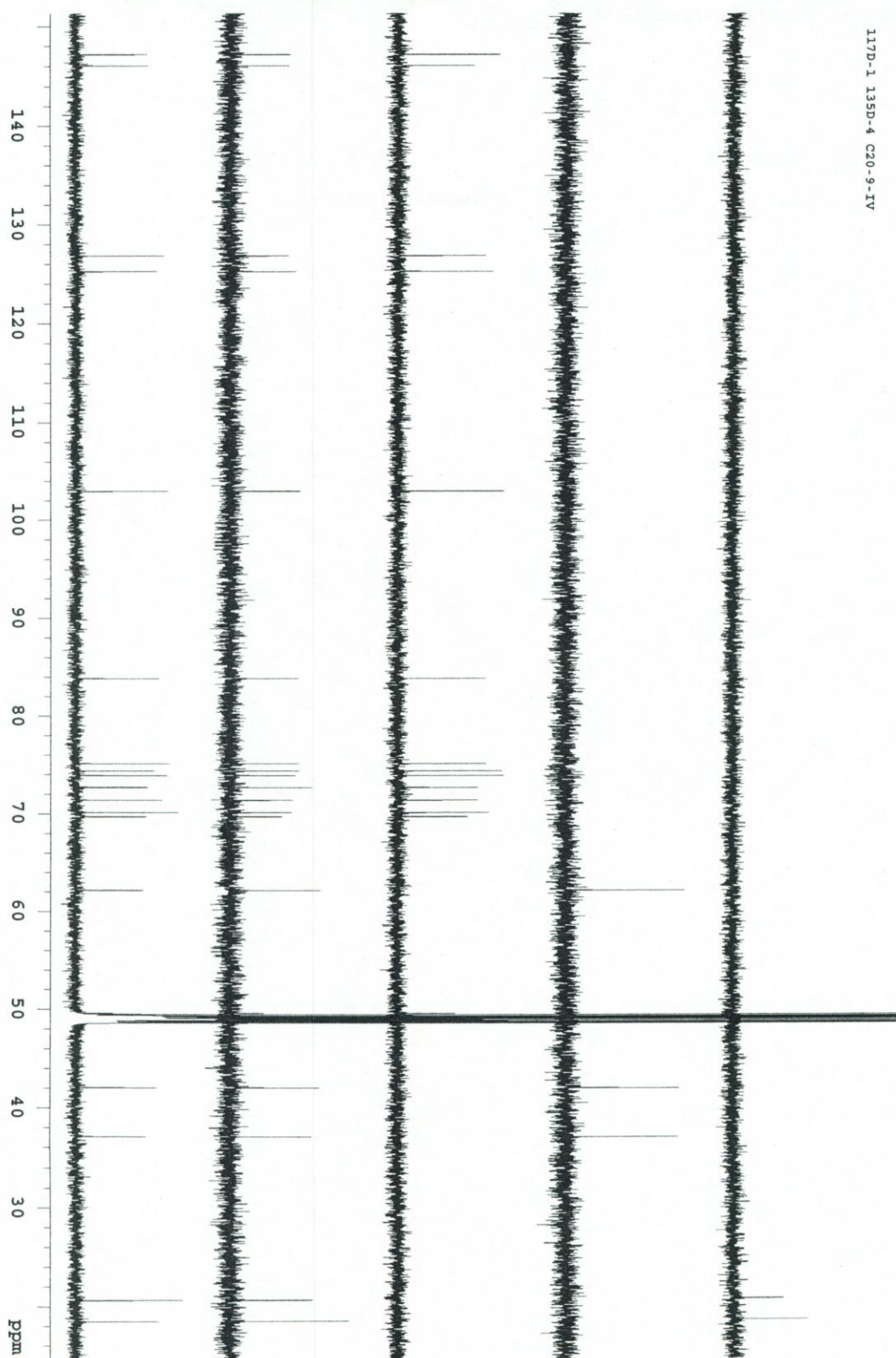
Figure S11 ^{13}C NMR spectrum of 3 in $\text{MeOH-}d_4$



117D-1 135D-4 C20-9-IV

INDEX	FREQUENCY	PPM	HEIGHT
1	25316.9	167.835	21.7
2	25174.7	166.892	18.1
3	22196.9	147.151	25.7
4	22029.3	146.040	26.3
5	19127.8	126.805	31.9
6	18888.5	125.219	29.4
7	15528.1	102.941	33.4
8	12644.0	83.821	30.2
9	11323.9	75.070	33.9
10	11211.7	74.327	28.4
11	11141.2	73.859	33.2
12	10958.6	72.648	26.1
13	10764.4	71.361	31.1
14	10574.8	70.104	37.2
15	10512.4	69.690	25.5
16	9370.3	62.119	24.4
17	7483.7	49.612	13.5
18	7462.9	49.474	65.9
19	7441.0	49.339	1572.9
20	7420.2	49.191	3779.8
21	7398.2	49.045	3878.7
22	7377.4	48.907	3692.4
23	7355.4	48.762	1810.0
24	7334.6	48.624	589.1
25	6342.8	42.049	29.5
26	5592.6	37.075	25.6
27	3108.4	20.607	38.9
28	2782.4	18.446	30.2

117D-1 13SD-4 C20-9-IV



117D-1 13SD-4 C20-9-IV

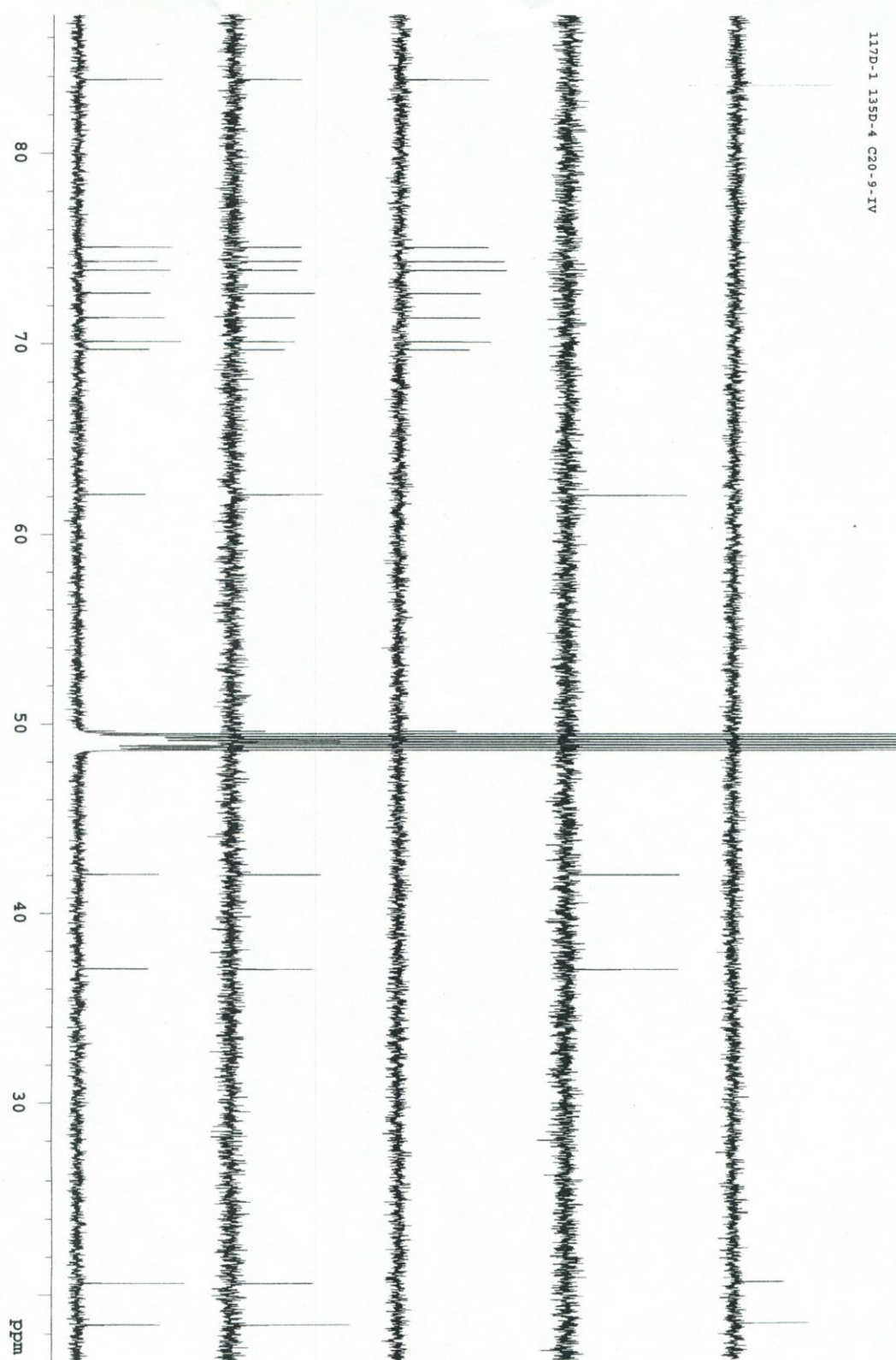
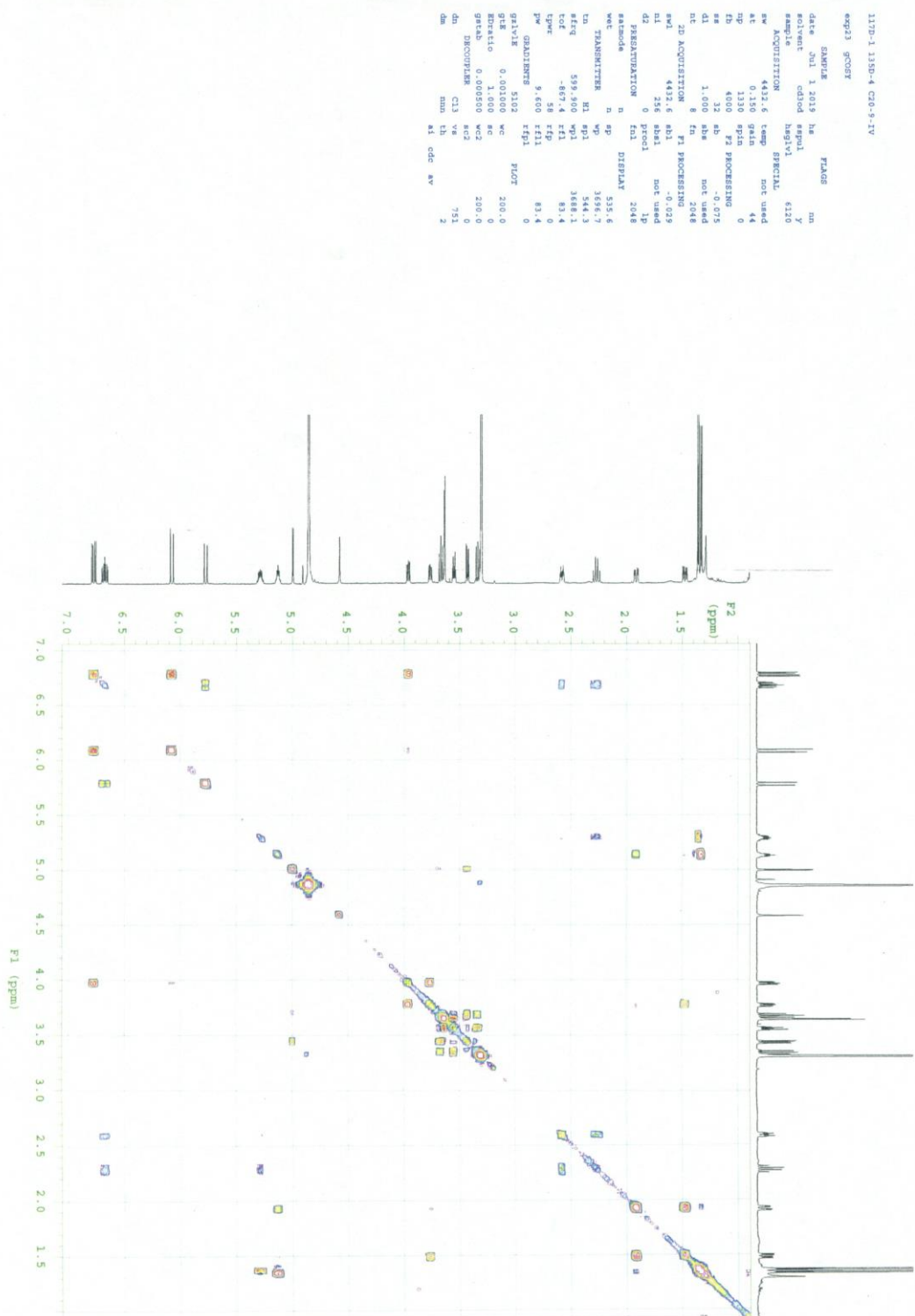


Figure S12 ^1H - ^1H COSY of 3

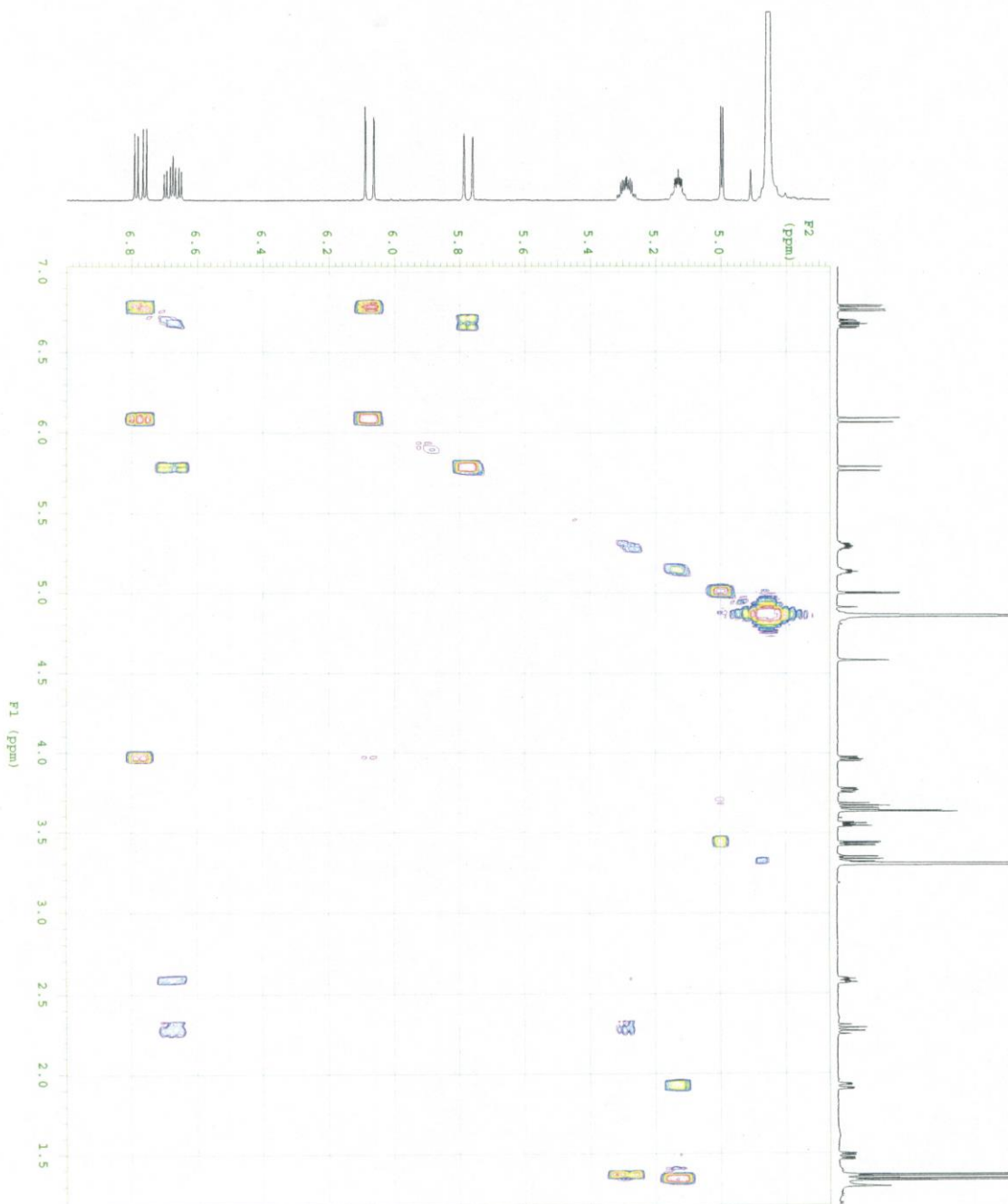



```

117D-1 115D-4 C2D-9-1V
exp23 gcosy

SAMPLE
date Jul 1 2019 hs
solvent ctdod sepi1 y
sample hagi1 6120
ACQUISITION SPECIAL
wv 4432.6 temp not used
at 0.150 gain 44
ap 1150 spin
fp 4000 F2 PROCESSING 0
wv 4432.6 ab not used
dt 1.000 sbw not used
nt 8 F2 2048
2D ACQUISITION 8 F1 PROCESSING -0.029
wv1 4432.6 sb1 not used
nl 256 sbal1 not used
d2 PRESENTATION 0 proc1 1p
PARAMODE n F1 DISPLAY 2048
wet n ap 2799.5
TANDEMATTER n1 wp 1398.2
n2wq 559.800 wpi 3708.1
n3wq 467.441 wpi 3212.4
tprc 58 rfp 83.6
pw GRADIENTS 9.600 rfp1 83.6
g1v1e 5102 F1 F2 0
g1v1e 0.001000 wc 250.0
g1v1e 1.000 wc 0
g1v1e 0.000500 wc2 200.0
DECOUPLER C13 v8 751
dm mm ch al cdc ny 2

```



```

117D-1 135D-4 C20-9-IV
exp23 gCOSY
=====
SAMPLE          FLAG
date  Jul 1 2019  hr
solvent  cdcl3  repul  Y
sample  6130
ACQUISITION
  av  4432.6  temp  not used
  at  0.150  gain  44
  np  1330  spin  0
  fb  4000  p2 PROCESSING 0
  ss  32  sb  -0.075
  d1  1.000  sbc  not used
  n1  8  fm  2048
  n1d ACQUISITION 8  fm  2048
  n1d 4432.6  sb  0.025
  n1  256  sbal  not used
  d2  0  proc1  lp
  PRESENTATION  fml  DISPLAY 2048
=====
PARAMETERS
  wet  n  sp  674.1
  tn  n1  wp  1824.7
  efrq  599.900  wpl  678.4
  tot  -867.4  r1  1831.1
  fpcr  38  rfp  83.4
  pw  GRADIENTS 5132  rfp1  0
  g1v1x  0.001000  PLOT 200.0
  g1v1y  0.001000  wc  0
  g1v1z  1.000  mc  0
  grab  0.000500  wcd  200.0
  DECOUPLER  c13  vs  0
  dm  mm  th  751
  al  cdc  av  2

```

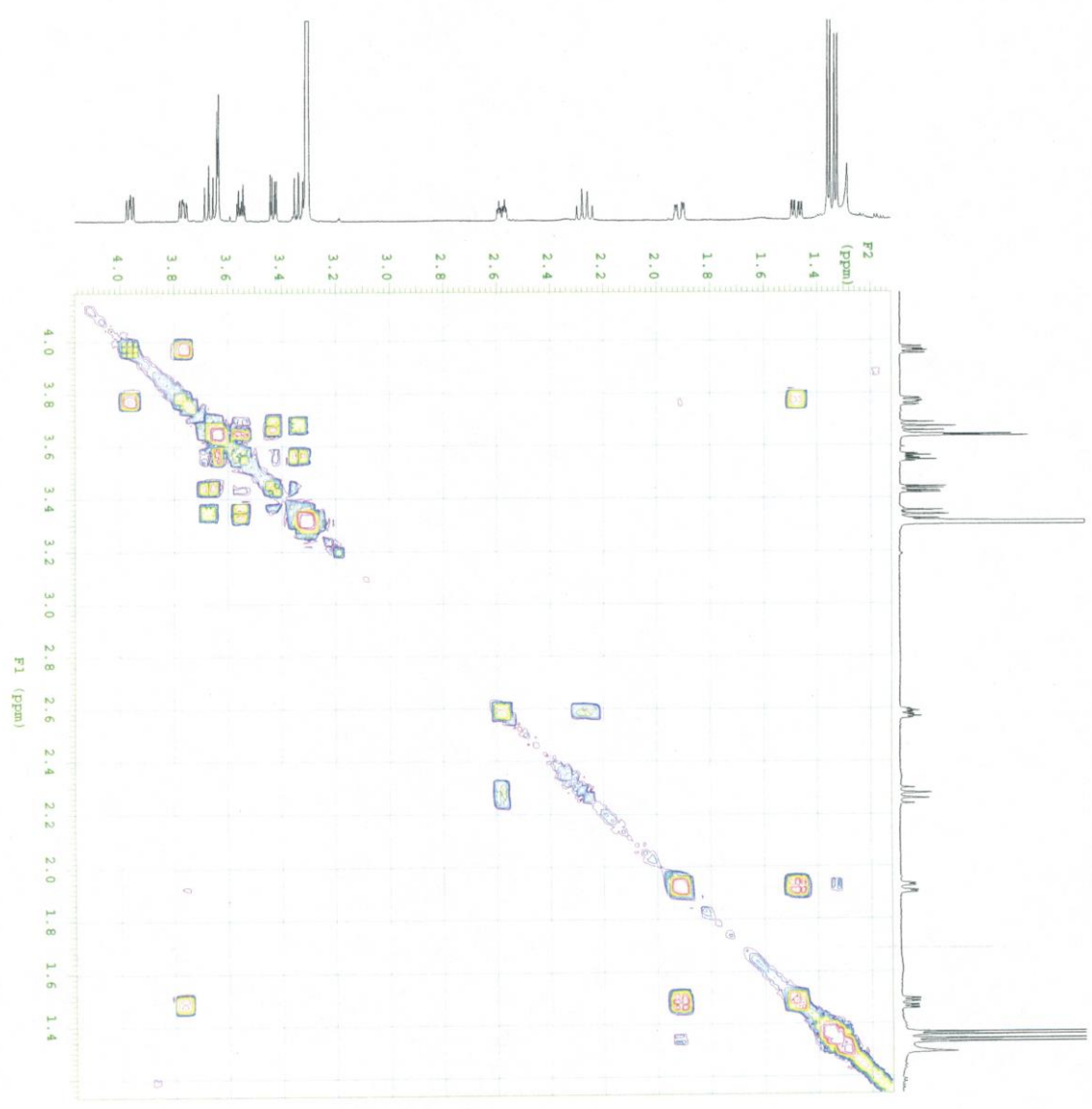
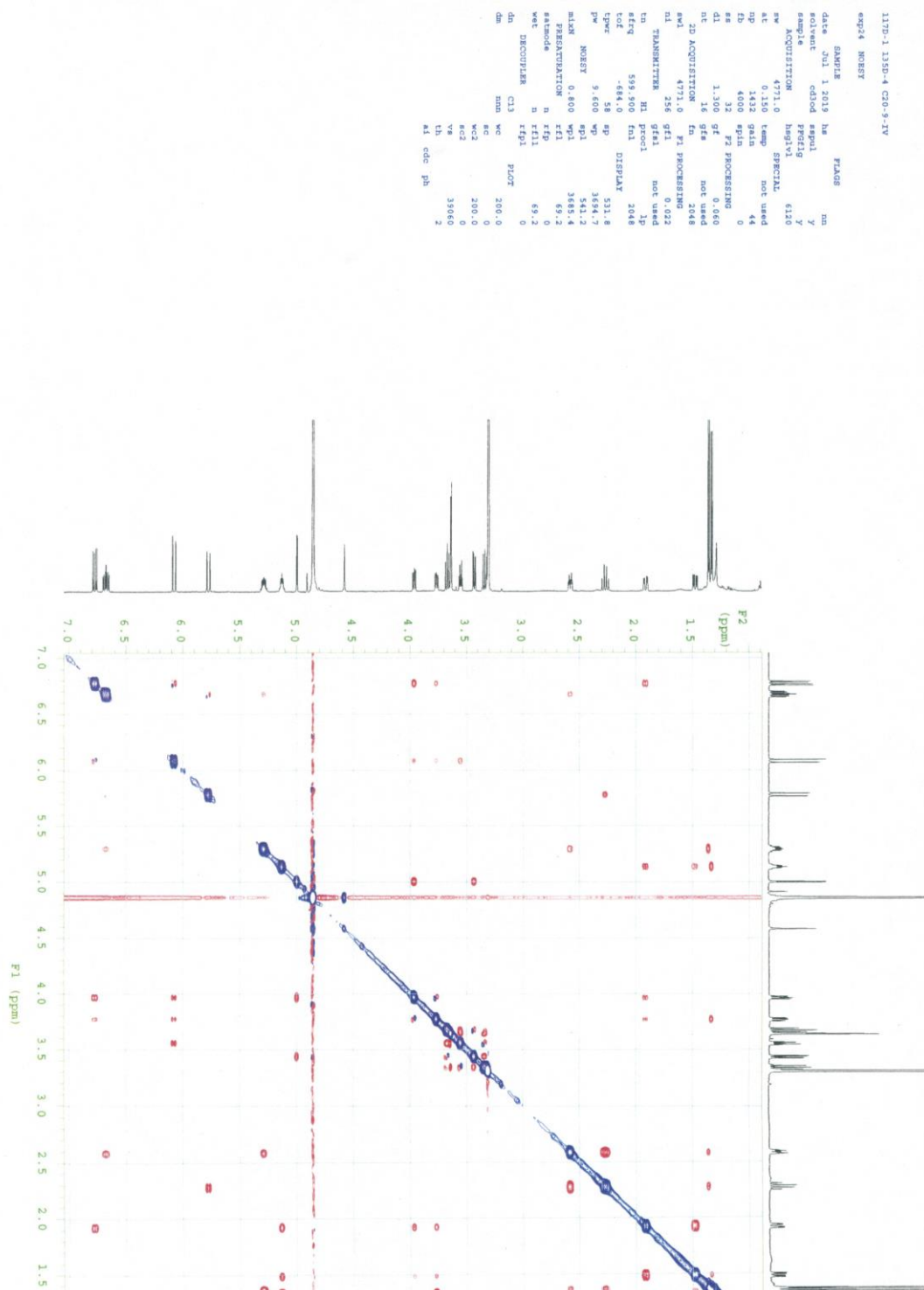
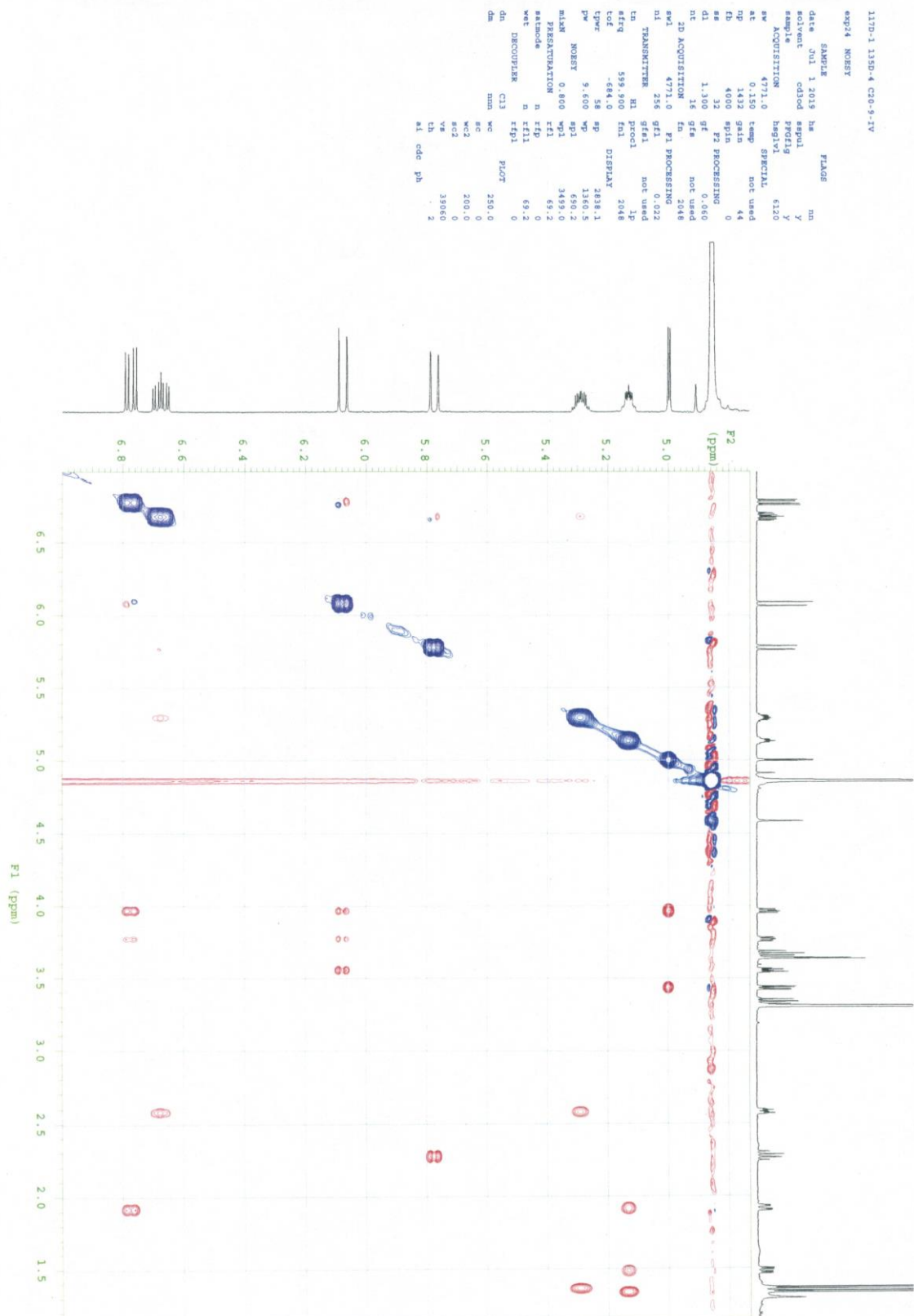


Figure S13 NOESY of 3





```

117D-1 135D-4 C20-9-IV
exp24 ROBERT

SAMPLE
date Jul 1 2015 hm
solvent cdcl3 aspl
sample y
PROG y
ACQUISITION hsq1v1 6120
sw 4771.0 SPECIAL
at 0.150 temp not used
np 1432 gain 44
fb 4000 spin 0
sa 32 F2 PROCESSING 0
ol 1.300 gf 0.060
nl 1.300 gf 0.060
F1 ACQUISITION 4771.0 not used
F2 ACQUISITION 4771.0 not used
F1 PROCESSING 256 gf1 not used
F2 PROCESSING 256 gf1 not used
TRANSMITTER H1 proc1 1p
ln 599.900 fml 2048
sfreq 599.900 fml 2048
tof -684.0 DISPLAY
tpwr 58 sp 699.6
pw 9.600 wp 1789.1
mkn ROBERT 0.400 ap1 694.8
PRESATURATION D 1779.8
satpwr 4000 fml 69.2
wet n rft1 69.2
DECOUPLE C13 rfp1 0
dm C13 FLOT 200.0
nm wc 200.0
nc 0
wc2 200.0
nc2 0
vs 35060
lh 2
al cdc ph

```

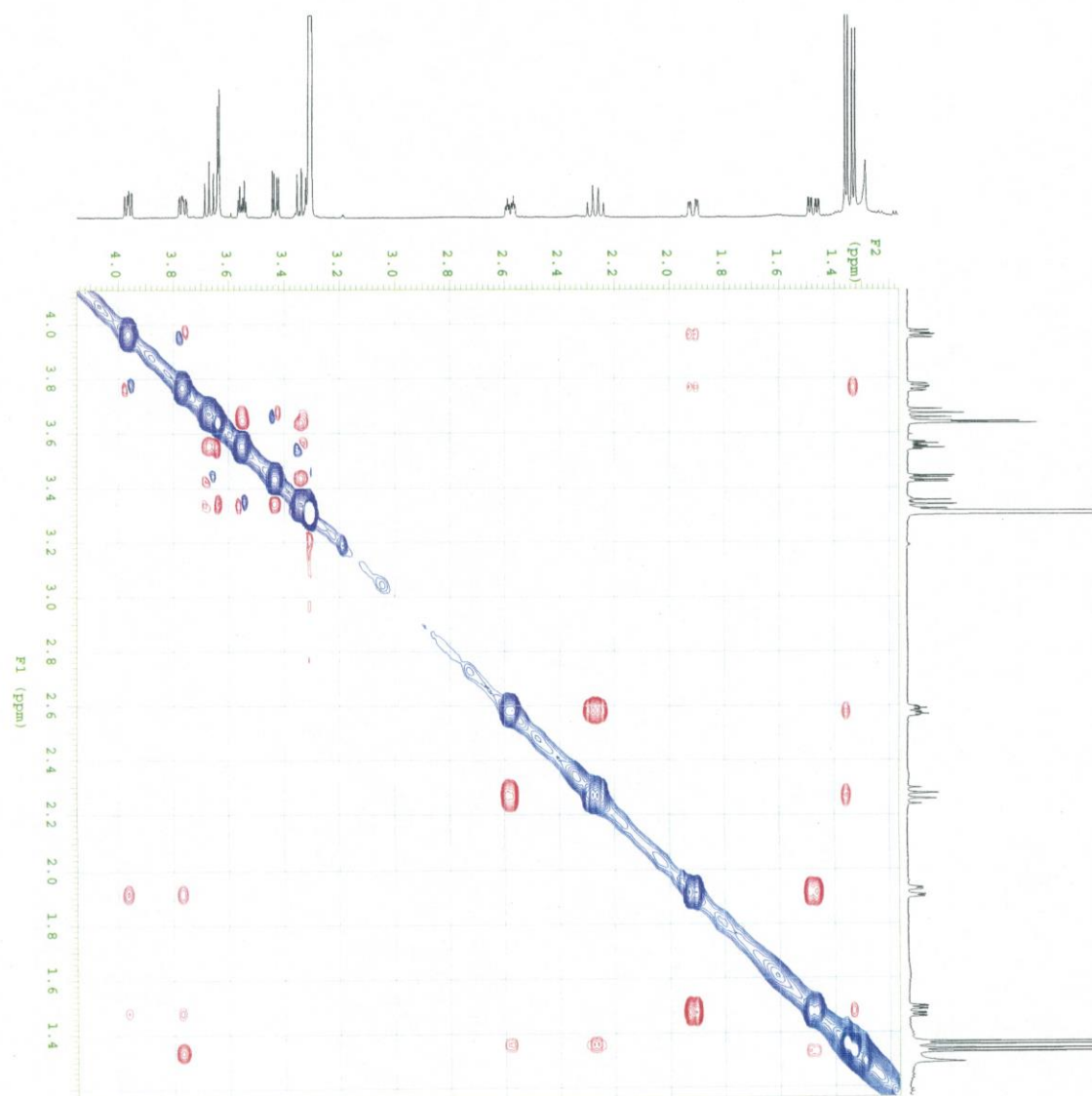
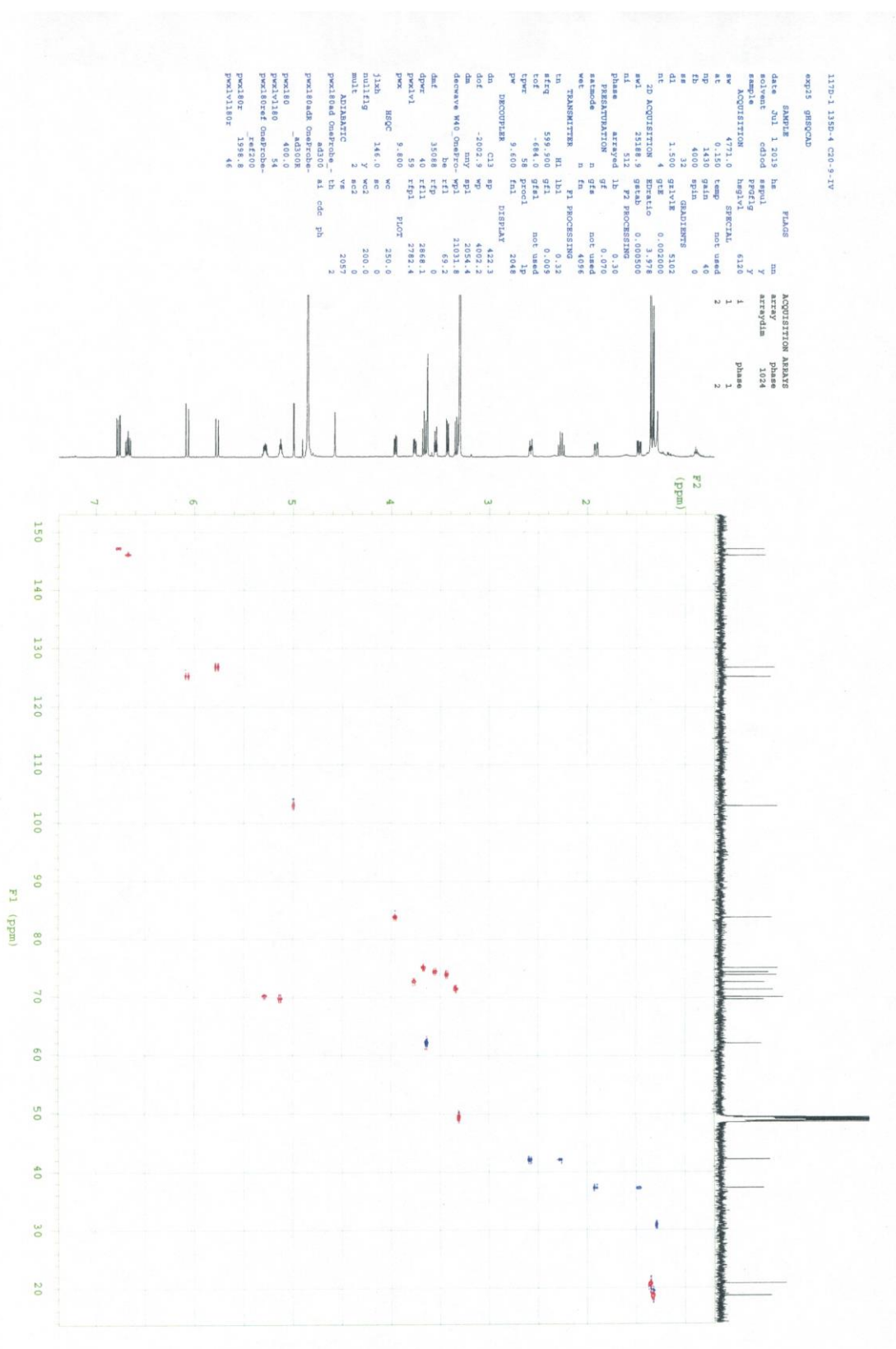


Figure S14 HMQC of 3



exp25 ghsqcal

ACQUISITION ARRAYS	
array	phase
arraydim	1024
1	phase
1	1
2	2

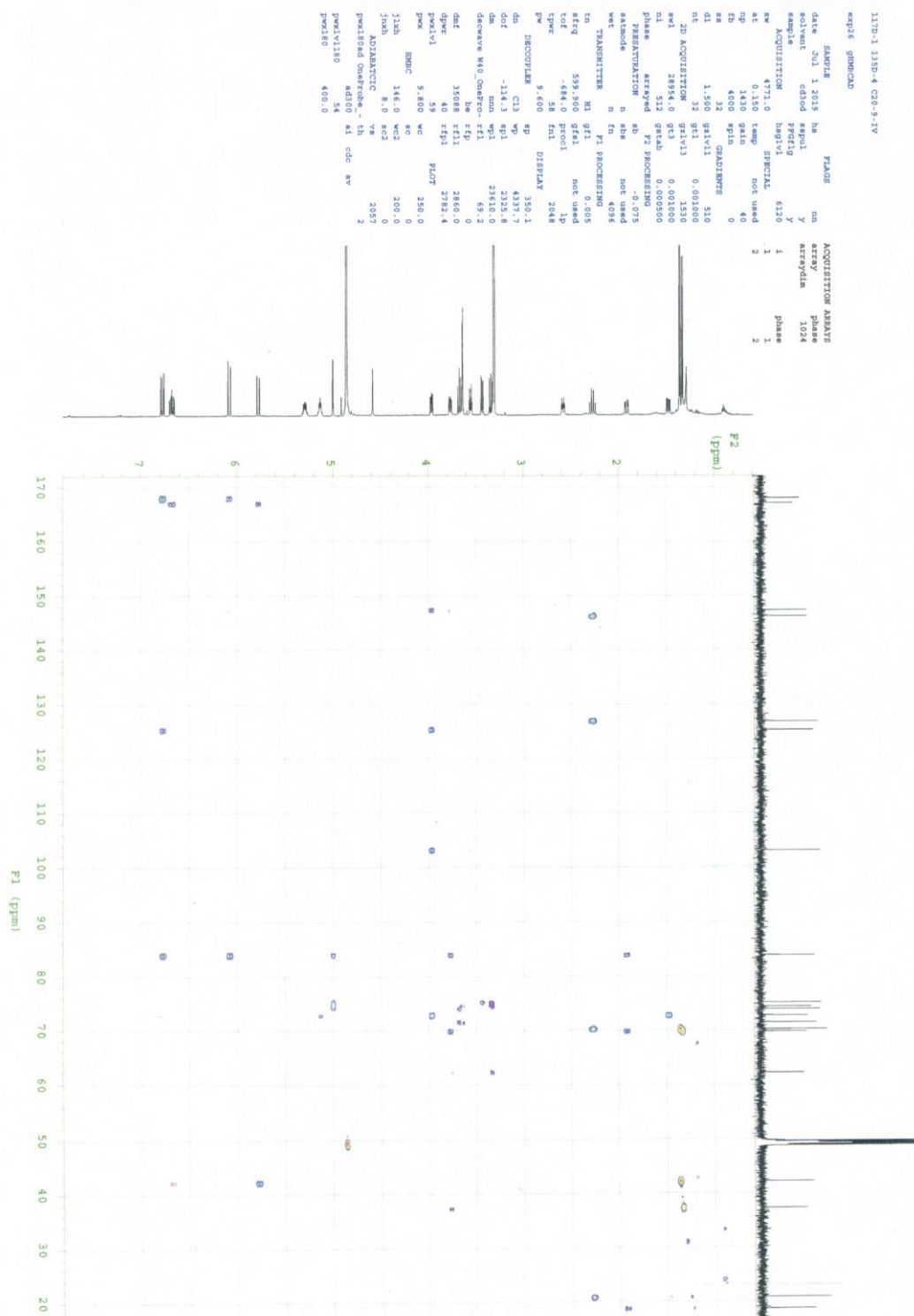


exp25 gnsqcad

pwk1v1180r 46



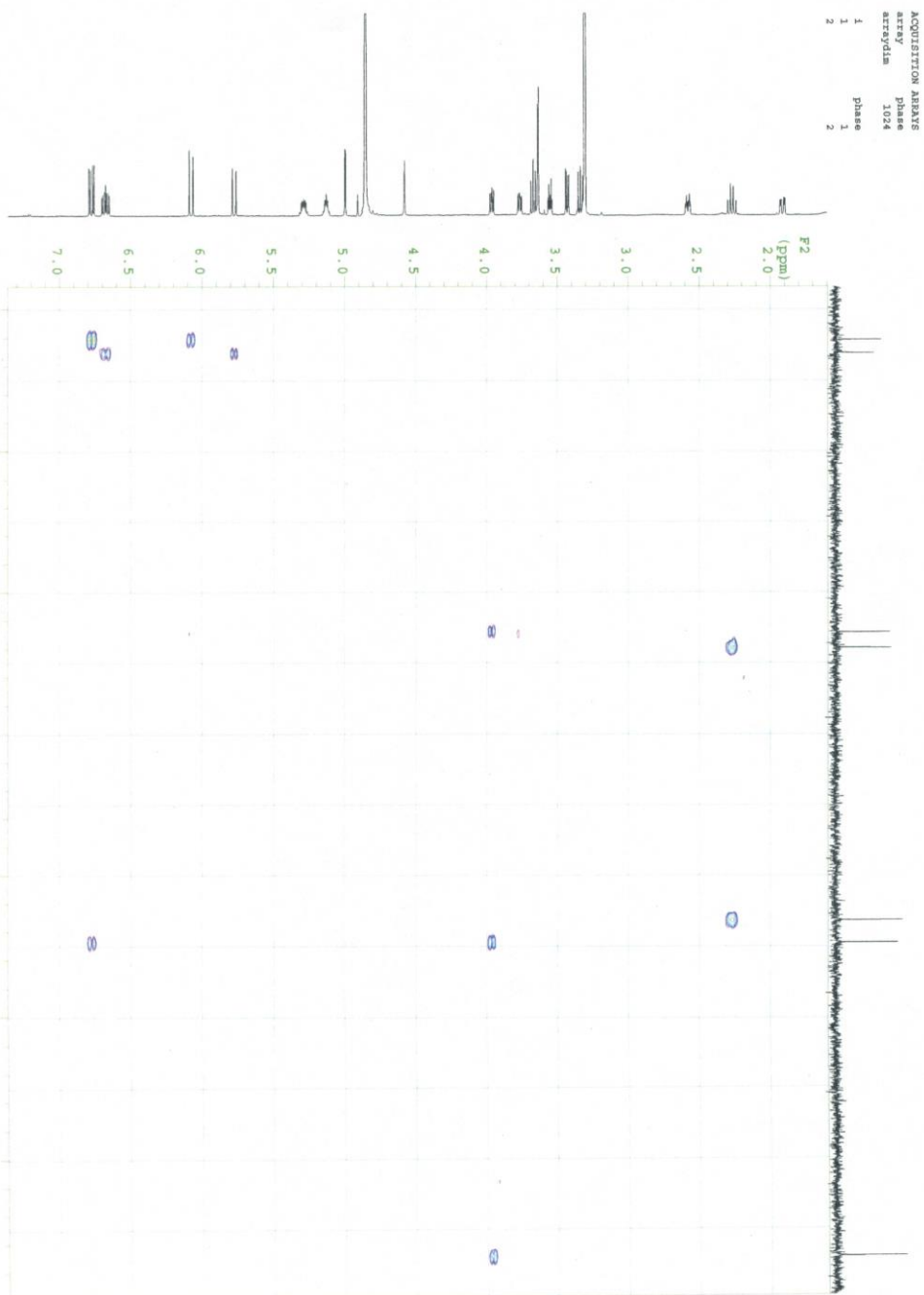
Figure S15 HMBC of 3



117D-1 135D-4 C20-9-IV

exp26 gbrmclad

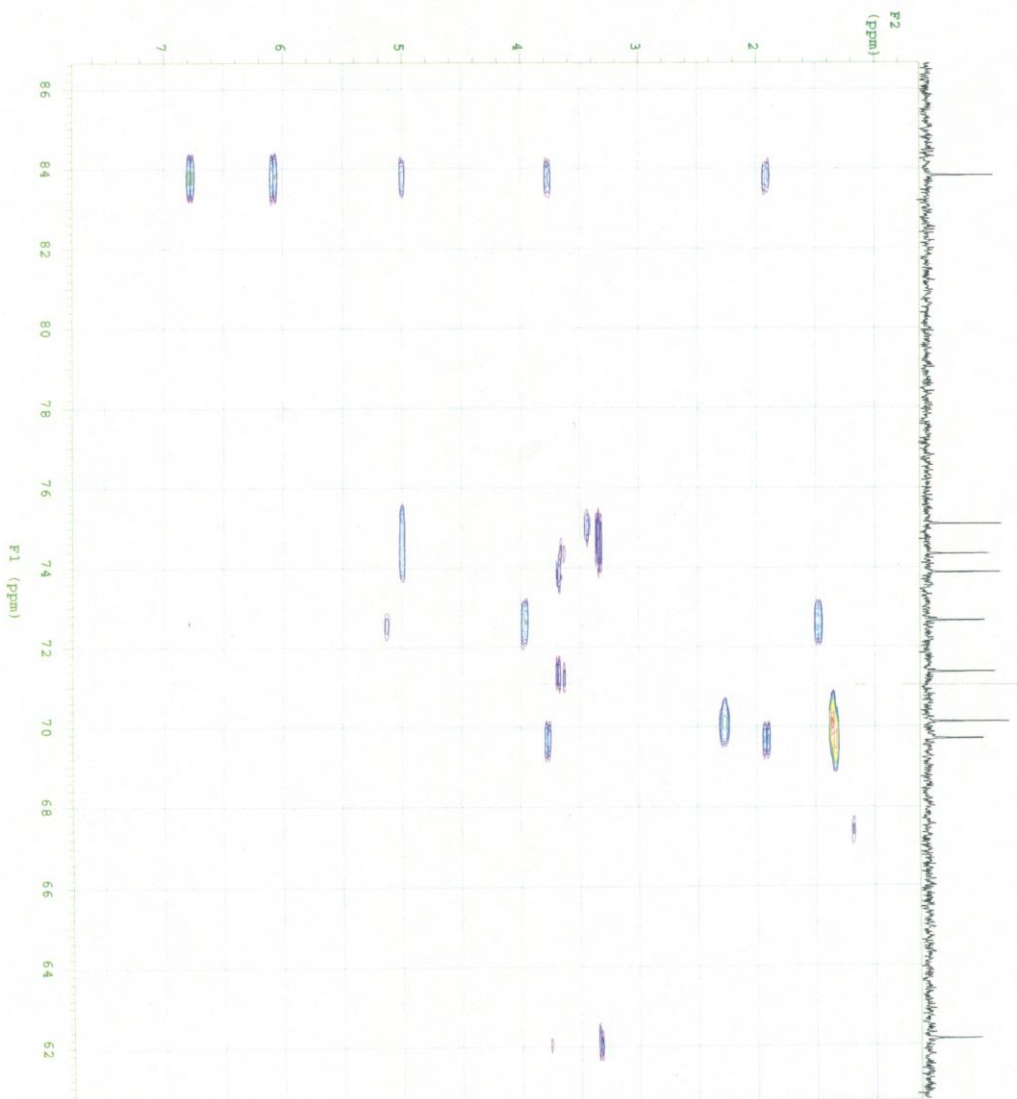
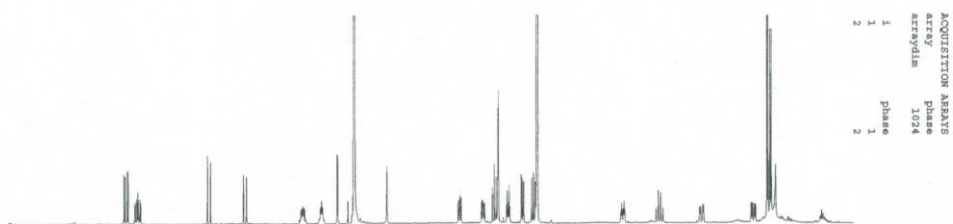
NAME: FLAG: nm
date Jul 1 2019
solvent cdcl3
sample y
ACQUISITION hsq1v1 6120
sw 4771.0 SPECIAL
at 0.150 temp not used
np 1410 gain 40
f2 32 4000 spin 0
d1 1.500 g1v1 510
d2 0.125 g1v1 0.001000
F2 ACQUISITION g1v1 1350
sw1 2854.0 g1v1 0.001000
rl 512 g1v1 0.000500
phase arrayed F2 PROCESSING
PRESATURATION ab -0.075
satmode n abe not used
wet n fn not used
TRANSMITTER F1 PROCESSING
tx 81 g1v1 0.005
strq 559.500 g1v1 not used
tcf -644.0 f1v1 1p
tpr 9.600 f1v1 2048
DECOUPLER SP DISPLAY 558.1
dn C13 wp 3457.1
dot -114.3 wp 15106.3
dm nm wp1 10772.9
decave W40 OnaPro- r1 59.2
dnt Be rfp 0
dpr 35088 r1v1 2860.0
dpr 40 rfp1 2782.4
pex1v1 59 p1v1 250.0
pex 9.600 wc 250.0
j1v1 146.0 wc2 200.0
j1v1 8.0 wc2 2057
ADJUNCTIC va
pex1v1 OnaPro- th 2
pex1v1 54 ad100 al cdc av
pex1v1 400.0



117D-1 115D-4 C20-5-IV

exp26 gsmcnd

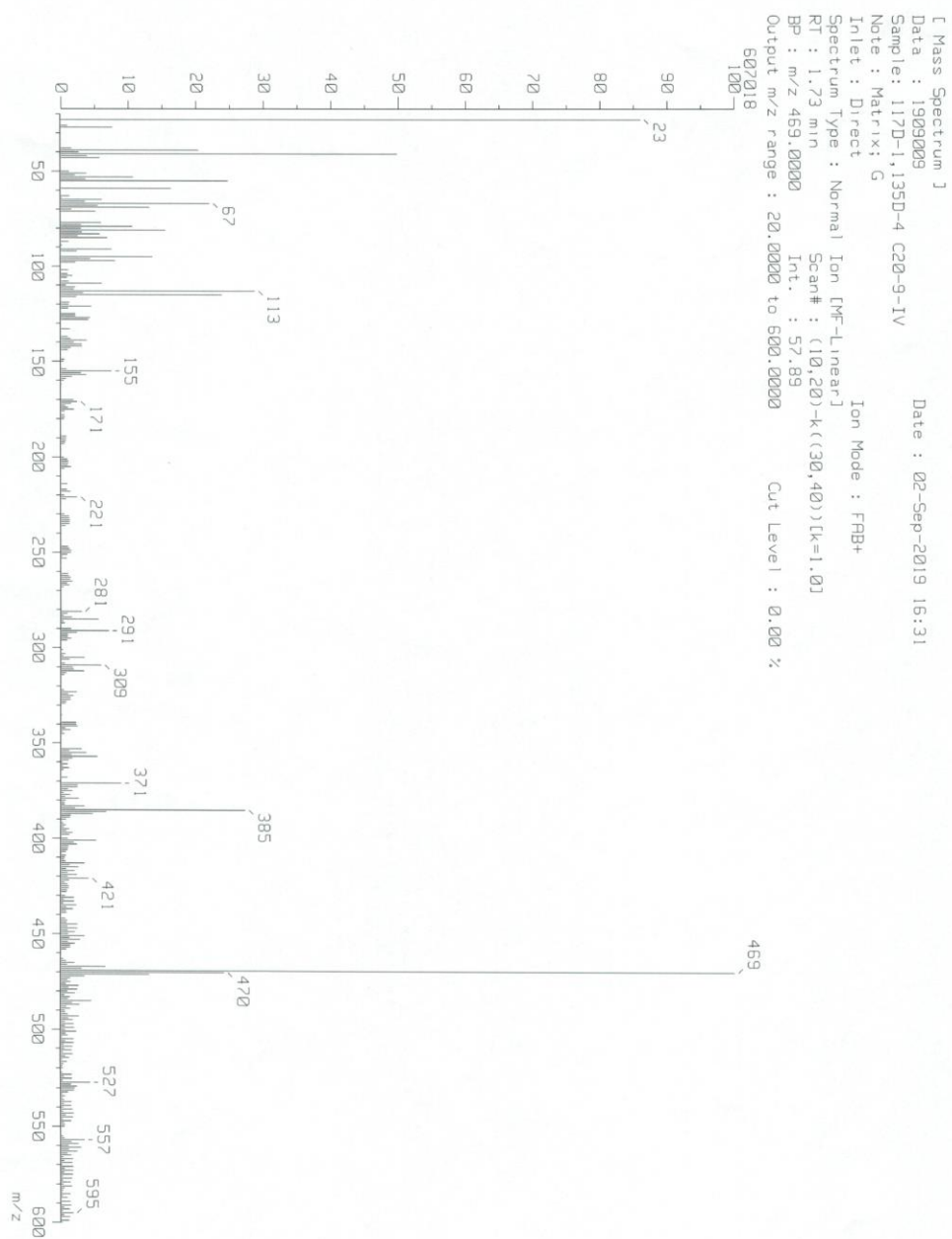
SAMPLE FLAG
date Jul 1 2015
solvent CDCl3
sample C20-5-IV
ACQUISITION hmgv1 6120
nu 4771.0 SPECIAL 1
at 0.150 temp not used 2
np 1410 gain 40
fb 4000 spin 0
ss 32 GRABBER 510
dl 1.500 g1v11 0.001000
nt 32 g1v13 1530
2D ACQUISITION g1v13 1530
w1 26934.0 g1b 0.001000
w2 512 g1v1b 0.001000
Phase Arrived 2 PROCESSING
PRESENTATION ab not used
astmode n ab not used
wet n fn 4096
TRANSMITTER F1 PROCESSING
tn n1 g1 0.005
afq 599.900 g1a1 not used
tof -564.0 p1a1 1p
lpr 58 fn1 2048
pw 9.600 DISPLAY 373.4
DECOUPLER C13 ap 4286.4
dn -114.3 ap1 3150.2
de 3150.2
decoupe W40 Gmptco-rf1 69.2
dnt 3508 rf1 2860.0
dpr 40 rf1 2782.4
pwa1 59 PLOT 250.0
pwx 9.800 wc 0
jsh 146.0 wc2 200.0
jsh 8.0 wc2 0
ADDITIONAL CH 2057
pwa10 Gmptco-rf1 2
pwa10 400.0



117D-1 135D-4 C20-9-IV
exp26 gbmecad

SAMPLE		FLAGS		ACQUISITION ANALYSIS	
date	Jul 1 2015	ba	nm	acq	phase
solvent	cdcl3	aspl	y	array	1024
sample	precig	ba	y	array	1024
ACQUISITION	beg1v1	SPECIAL	6120	1	1
ac	4771.0	temp	not used	2	2
ap	0.150	gain	40		
bp	1430	spin	0		
cb	4000	GRABINITS	0		
cc	32				
cd	1.500	g1v1	510		
de	0.001000				
df	0.001000				
dg	0.001000				
dh	0.001000				
di	0.001000				
dj	0.001000				
dk	0.001000				
dl	0.001000				
dm	0.001000				
dn	0.001000				
do	0.001000				
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dr	0.001000				
ds	0.001000				
dt	0.001000				
du	0.001000				
dv	0.001000				
dw	0.001000				
dx	0.001000				
dy	0.001000				
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ed	0.001000				
ee	0.001000				
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fc	0.001000				
fd	0.001000				
fe	0.001000				
ff	0.001000				
fg	0.001000				
fh	0.001000				
fi	0.001000				
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fn	0.001000				
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fp	0.001000				
fq	0.001000				
fr	0.001000				
fs	0.001000				
ft	0.001000				
fu	0.001000				
fv	0.001000				
fw	0.001000				
fx	0.001000				
fy	0.001000				
fz	0.001000				
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gb	0.001000				
gc	0.001000				
gd	0.001000				
ge	0.001000				
gf	0.001000				
gh	0.001000				
gi	0.001000				
gj	0.001000				
gk	0.001000				
gl	0.001000				
gm	0.001000				
gn	0.001000				
go	0.001000				
gp	0.001000				
gq	0.001000				
gr	0.001000				
gs	0.001000				
gt	0.001000				
gu	0.001000				
gv	0.001000				
gw	0.001000				
gx	0.001000				
gy	0.001000				
gz	0.001000				
ha	0.001000				
hb	0.001000				
hc	0.001000				
hd	0.001000				
he	0.001000				
hf	0.001000				
hg	0.001000				
hh	0.001000				
hi	0.001000				
hj	0.001000				
hk	0.001000				
hl	0.001000				
hm	0.001000				
hn	0.001000				
ho	0.001000				
hp	0.001000				
hq	0.001000				
hr	0.001000				
hs	0.001000				
ht	0.001000				
hu	0.001000				
hv	0.001000				
hw	0.001000				
hx	0.001000				
hy	0.001000				
hz	0.001000				
ia	0.001000				
ib	0.001000				
ic	0.001000				
id	0.001000				
ie	0.001000				
if	0.001000				
ig	0.001000				
ih	0.001000				
ii	0.001000				
ij	0.001000				
ik	0.001000				
il	0.001000				
im	0.001000				
in	0.001000				
io	0.001000				
ip	0.001000				
iq	0.001000				
ir	0.001000				
is	0.001000				
it	0.001000				
iu	0.001000				
iv	0.001000				
iw	0.001000				
ix	0.001000				
iy	0.001000				
iz	0.001000				
ja	0.001000				
jb	0.001000				
jc	0.001000				
jd	0.001000				
je	0.001000				
jf	0.001000				
jj	0.001000				
jk	0.001000				
jl	0.001000				
jm	0.001000				
jn	0.001000				
jo	0.001000				
jp	0.001000				
jq	0.001000				
jr	0.001000				
js	0.001000				
jt	0.001000				
ju	0.001000				
jv	0.001000				
jw	0.001000				
jx	0.001000				
jy	0.001000				
jz	0.001000				
ka	0.001000				
kb	0.001000				
kc	0.001000				
kd	0.001000				
ke	0.001000				
kf	0.001000				
kg	0.001000				
kh	0.001000				
ki	0.001000				
kj	0.001000				
kk	0.001000				
kl	0.001000				
km	0.001000				
kn	0.001000				
ko	0.001000				
kp	0.001000				
kq	0.001000				
kr	0.001000				
ks	0.001000				
kt	0.001000				
ku	0.001000				
kv	0.001000				
kw	0.001000				
kx	0.001000				
ky	0.001000				
kz	0.001000				
la	0.001000				
lb	0.001000				
lc	0.001000				
ld	0.001000				
le	0.001000				
lf	0.001000				
lg	0.001000				
lh	0.001000				
li	0.001000				
lj	0.001000				
lk	0.001000				
ll	0.001000				
lm	0.001000				
ln	0.001000				
lo	0.001000				
lp	0.001000				
lq	0.001000				
lr	0.001000				
ls	0.001000				
lt	0.001000				
lu	0.001000				
lv	0.001000				
lw	0.001000				
lx	0.001000				
ly	0.001000				
lz	0.001000				
ma	0.001000				
mb	0.001000				
mc	0.001000				
md	0.001000				
me	0.001000				
mf	0.001000				
mg	0.001000				
mh	0.001000				
mi	0.001000				
mj	0.001000				
mk	0.001000				
ml	0.001000				
mm	0.001000				
mn	0.001000				
mo	0.001000				
mp	0.001000				
mq	0.001000				
mr	0.001000				
ms	0.001000				

Figure S16 FABMS of 3



[Mass Spectrum]

Data : 1909009

Date : 02-Sep-2019 16:31

Page: 1

Sample: 117D-1,135D-4 C20-9-IV

Note : Matrix; G

Inlet : Direct

Ion Mode : FAB+

Spectrum Type : Normal Ion [MF-Linear]

RT : 1.73 min Scan# : (10,20)-k((30,40)) [k=1.0]

BP : m/z 469.0000 Int. : 57.89

Output m/z range : 20.0000 to 600.0000

Cut Level : 3.00 %

m/z	Int.	Norm.	m/z	Int.	Norm.
23.0000	49.79	86.02	387.0000	2.68	4.63
27.0000	4.40	7.60	401.0000	2.97	5.14
39.0000	11.78	20.35	413.0000	1.99	3.43
41.0000	28.83	49.80	421.0000	2.33	4.03
42.0000	2.21	3.82	451.0000	1.97	3.41
43.0000	3.32	5.74	467.0000	3.79	6.55
51.0000	2.16	3.73	468.0000	1.74	3.00
53.0000	6.19	10.69	469.0000	57.89	100.00
54.0000	2.09	3.61	470.0000	13.96	24.12
55.0000	14.31	24.71	471.0000	7.52	12.99
59.0000	9.43	16.28	472.0000	1.97	3.41
65.0000	3.51	6.06	485.0000	2.56	4.43
66.0000	2.06	3.56	527.0000	2.45	4.24
67.0000	12.72	21.98	557.0000	1.96	3.39
68.0000	3.14	5.42			
69.0000	7.57	13.08			
71.0000	2.95	5.10			
77.0000	3.44	5.94			
79.0000	6.12	10.57			
81.0000	8.94	15.45			
82.0000	1.76	3.05			
83.0000	3.32	5.73			
85.0000	3.97	6.85			
91.0000	4.32	7.46			
95.0000	7.84	13.55			
96.0000	2.50	4.31			
97.0000	4.64	8.02			
109.0000	3.48	6.02			
113.0000	16.60	28.67			
114.0000	1.88	3.24			
115.0000	13.78	23.80			
121.0000	2.59	4.47			
127.0000	2.47	4.27			
128.0000	2.33	4.02			
139.0000	2.19	3.79			
141.0000	1.74	3.01			
142.0000	1.76	3.04			
155.0000	4.31	7.44			
157.0000	2.14	3.69			
285.0000	3.17	5.47			
291.0000	4.05	6.99			
305.0000	1.96	3.39			
309.0000	3.39	5.85			
312.0000	1.90	3.29			
355.0000	2.12	3.66			
357.0000	3.07	5.30			
371.0000	5.10	8.82			
383.0000	1.96	3.39			
385.0000	15.74	27.18			
386.0000	3.83	6.61			

[Elemental Composition]

Page: 1

Data : 1909155 Date : 18-Sep-2019 16:52
Sample: 117D-1,135D-4 C20-9-IV
Note : Matrix; G
Inlet : Direct Ion Mode : FAB+
RT : 8.37 min Scan#: (75,80)
Elements : C 25/15, H 35/25, O 15/0, Na 1/0
Mass Tolerance : 20ppm, 1mmu if m/z > 50
Unsaturation (U.S.) : -1.0 - 40.0

Observed m/z	Int%	Err[ppm / mmu]	U.S.	Composition
469.1692	100.0	+1.2 / +0.6	5.5	C 20 H 30 O 11 Na

469.1697

Figure S17 IR spectrum of 3

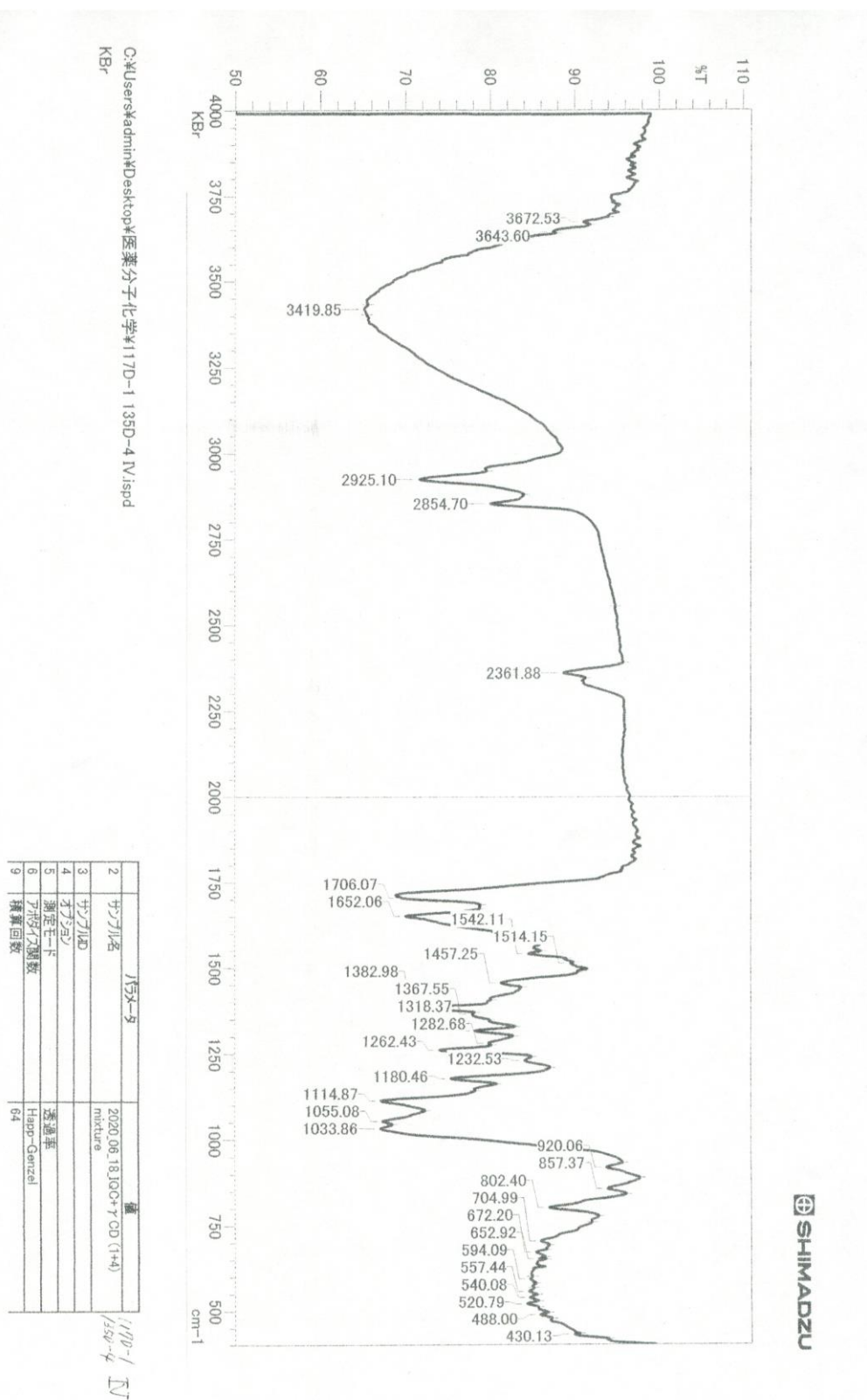


Figure S18 HPLC chromatograms of halosmycins A, B, and C

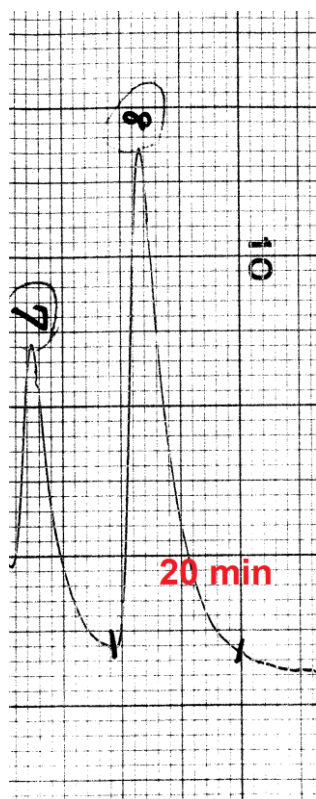


Peak 19: halosmycins B (r.t. 28.8 min)

Peak 21: halosmycins A (r.t. 31.7 min)

Column: Cosmosil Packed Column 5C18-MSII (25 cm X 20 mm i.d.)

Mobile phase: 80% MeOH in H₂O



Peak 8: halosmycins C (r.t. 18.3 min)

Column: Cosmosil Packed Column 5C18-MSII (25 cm X 20 mm i.d.)

Mobile phase: 80% MeOH in H₂O

Figure S19 ^1H NMR spectrum of 5-hydroxy-(2*E*)-hexenoic acid in $\text{MeOH-}d_4$

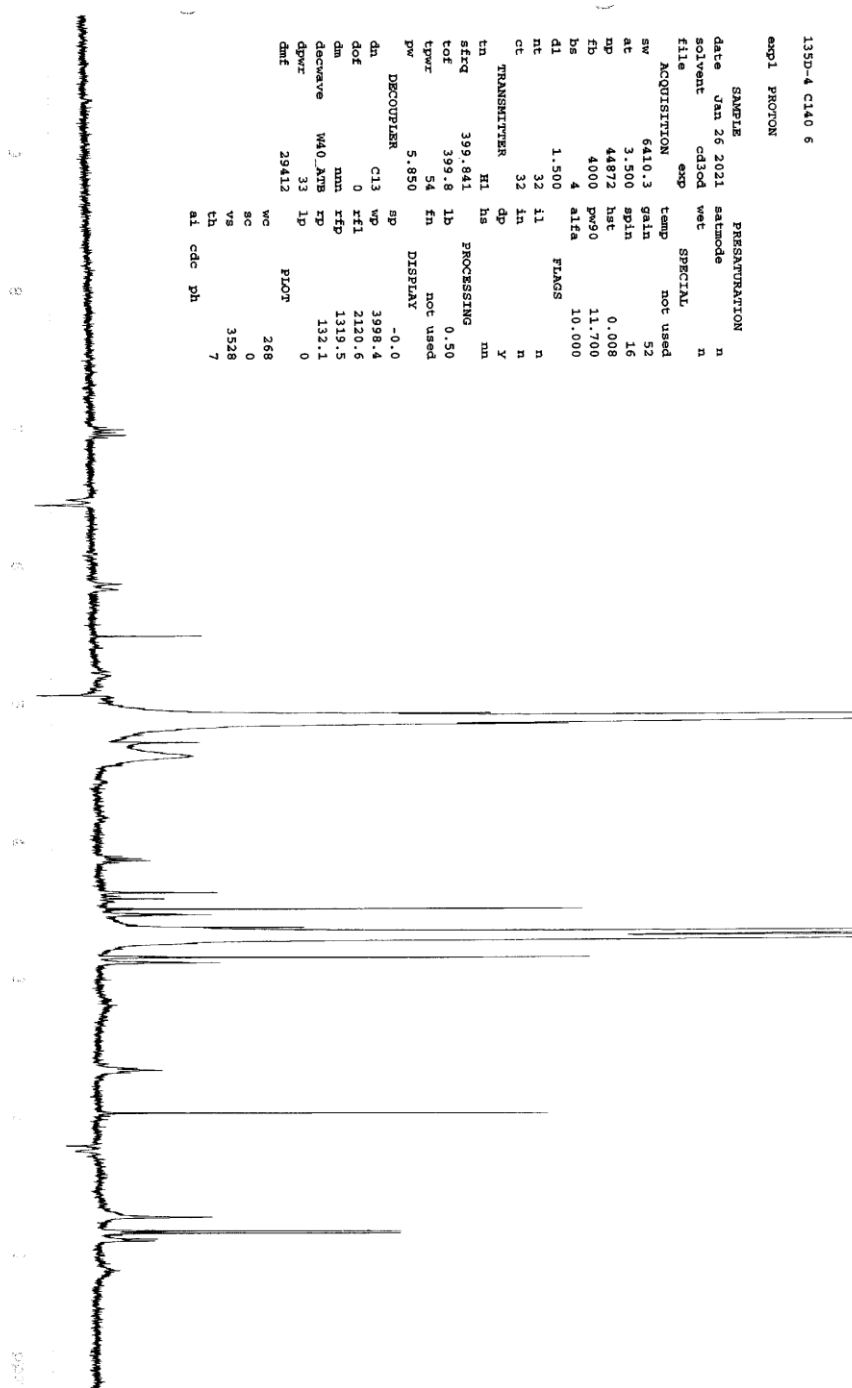


Figure S20 ^1H NMR spectrum of 4,5,7-trihydroxy-(2*E*)-octenoic acid in $\text{MeOH-}d_4$

