NAME _____

ID # _____

INTERPRETATION OF ORGANIC SPECTRA (4361/8361)

1:30 – 3:30 pm, December 15, 2012

Final Exam

This exam is open book and open note. You are permitted to use any written materials you have brought as aids on this exam. You may also use a simple calculator. Other than this, please do not use any other electronic devices (cell phones, computers, recording devices, etc.) during the exam.

You may use pen or pencil. However, re-grades will be considered only for exams completed in pen.

Please write your answers in the boxes/spaces provided. If your answer is not in the appropriate space (say, for example, it's on the back of the page), draw us an arrow and/or note telling us where to look.

Feel free to remove the corner staple if this helps you analyze the spectra; you will have the opportunity to re-staple your exam at the end. You will be given 2 hours total to finish the test. This exam contains <u>two</u> main problems, which are split into parts. Many of these parts can be answered independently. *Do not get stuck* on one part and then assume that you will be unable to answer the rest of the question—move on. In addition, partial credit will be given for incorrect but still plausible answers, so *guess* on problems you cannot answer perfectly.

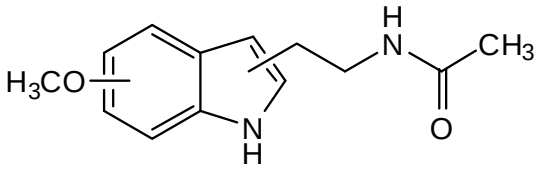
At the end of the 2 hour exam period you will be asked to return your exam to the proctor. Please do not take any part of the exam packet with you when you are done; everything will be returned to you after the exams are graded. This packet should contain 22 pages, including this one. Please check to make sure that your packet contains 22 pages before beginning your exam.

NAME _____

Scoring:			
	1. _____ / 5	5. _____ / 39	9. _____ / 4
	2. _____ / 5	6. _____ / 5	10. _____ / 10
	3. _____ / 48	7. _____ / 7	11. _____ / 18
	4. _____ / 16	8. _____ / 15	12. _____ / 18

Total Score: _____ / 200

Melatonin is a human hormone that varies regularly in concentration over the course of a day, and has been implicated in the regulation (via "circadian rhythms") of a variety of biological processes. Melatonin is a substituted indole, with the incompletely determined structure shown at right. In this structure, the -OCH_3 group could be attached to any available carbon on the indole benzene ring; and the alkylamide group could be attached to either free carbon of the indole pyrrole ring. In the first part of this exam, you will use ^{1}H, ^{13}C, ^{1}H-^{1}H COSY, ^{1}H-^{13}C HSQC, and ^{1}H-^{13}C HMBC spectra (attached to the back of this exam) to determine the correct structure of melatonin.

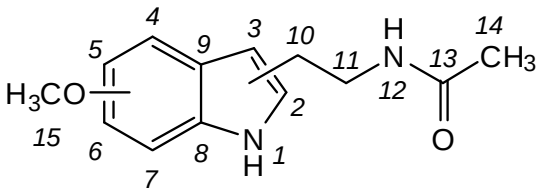


melatonin

1. All of the NMR spectra were collected on samples of melatonin dissolved in deuterated DMSO-d_6, which has molecular formula C_2D_6SO. Deuterium atoms are invisible to ^{1}H NMR, and yet we still see a multiplet at δ = 2.5 ppm corresponding to the NMR solvent. Why?

2. The ^{1}H resonance at δ = 8.0 ppm corresponds to a proton attached to a nitrogen atom. This resonance looks sharper, better resolved, than we would expect for an N-H proton. What must be true of the NMR sample to achieve this unusually sharp N-H resonance? *(Answer on the next page.)*

3. In the chart below, assign chemical shift values (to within 0.05 ppm) to every proton in melatonin, using the numbering scheme on the right. I have provided a box for every possible location of a proton on the melatonin skeleton, but remember that two of these locations will be occupied by other groups (the -OCH_3 and alkylamide groups). As a result, you should **leave two of the boxes empty**.



proton	δ (ppm)
H1	
H2	
H3	
H4	

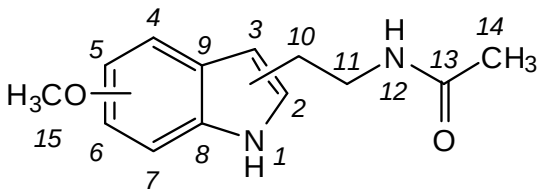
proton	δ (ppm)
H5	
H6	
H7	
H10	

proton	δ (ppm)
H11	
H12	
H14	
H15	

4. In the boxes on the right, name four unique coupling constants that you observe in the ^{1}H NMR spectrum of melatonin, and give the *J* value for each (to within 1 Hz).

coupling constant name "*J*(H#,H#)"	***J* (Hz)**

5. In the chart below, assign chemical shift values (to within 0.1 ppm) to every carbon atom in melatonin, using the same numbering scheme as on the previous page.



proton	δ (ppm)
C2	
C3	
C4	
C5	
C6	

proton	δ (ppm)
C7	
C8	
C9	
C10	

proton	δ (ppm)
C11	
C13	
C14	
C15	

6. In principle, ^{13}C-^{13}C INADEQUATE NMR could be used to determine the carbon-carbon connectivity of melatonin. But INADEQUATE is an NMR technique that is almost never used. Why not?

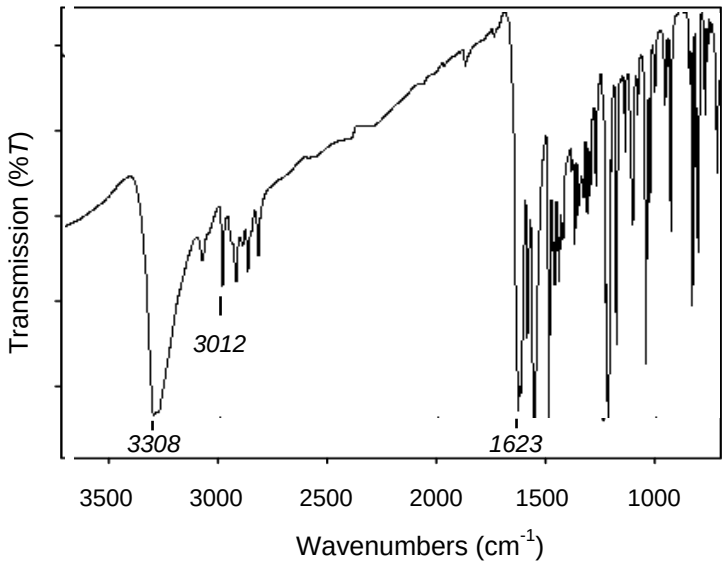
7. Melatonin absorbs UV light, with a fairly large extinction coefficient at λ = 275 nm. Both the aromatic indole and the amide group contribute to absorption at this wavelength. Which would you expect contributes more?

indole **amide** *or* **both contribute equally** *? (Circle one.)*

Assuming that the contributions of these two parts of the molecule to the extinction coefficient at 275 nm (ε_{275}) are additive, what would you estimate the extinction coefficient to be closest to?

10 **100** **1000** **10000** *or* **100000** ? *(Circle one.)*

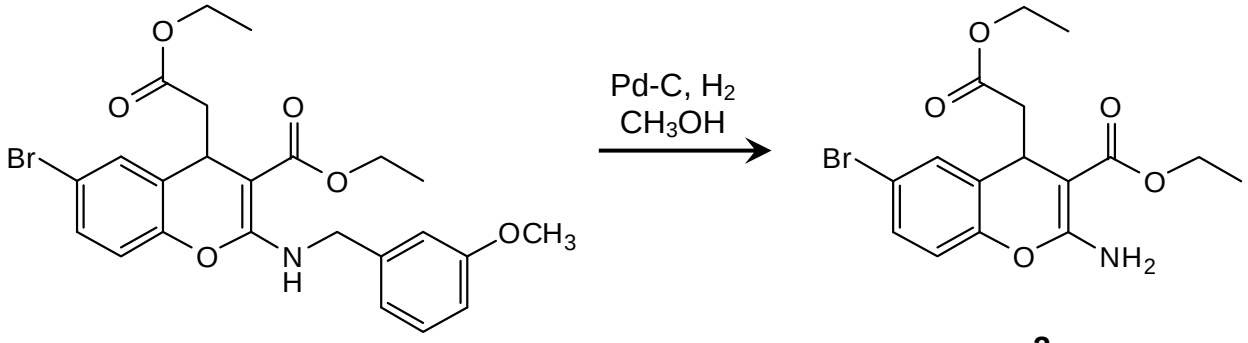
8. The IR spectrum of melatonin is shown below. What functional groups are responsible for each of the peaks labeled on the spectrum?



functional group
ν = 3308 cm^{-1}
ν = 3012 cm^{-1}
ν = 1623 cm^{-1}

9. There are a lot of unanswered questions about how melatonin is distributed in tissues, and how that distribution may be affected by the daily cycle of sunlight and darkness. It would be great to be able to image melatonin in cells and tissues. Unfortunately, melatonin is not fluorescent, so this distribution cannot be imaged by fluorescence microscopy. Propose an alternate technique to image the distribution of melatonin in a tissue sample. You do not need to describe the technique—just a name is enough.

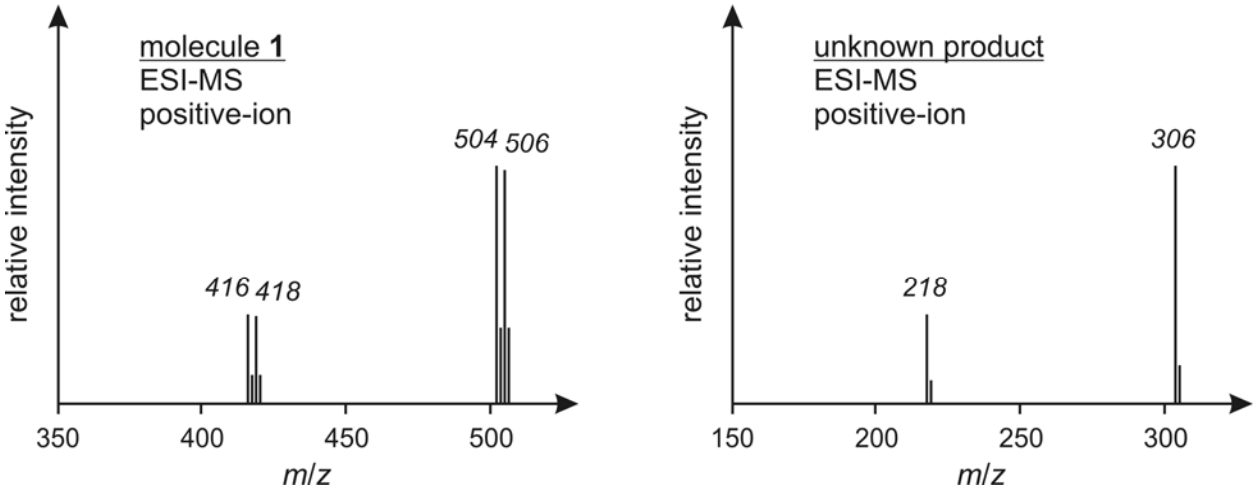
Bo Zhou (Xing group, Med. Chem.) conducted a small-scale, Pd-catalyzed hydrogenolysis of starting material **1**, with the hope of removing its methoxybenzoyl protecting group and obtaining product **2**. Bo analyzed the crude reaction mixture by electrospray-ionization mass spectrometry (ESI-MS), in positive-ion mode. He found that the starting material had indeed been consumed in the reaction, but that the parent mass of the primary product was not consistent with structure **2**. He suspected, however, that the product was probably structurally related to **2**. In the second part of this exam, you will analyze Bo's MS data to determine the structure of the actual product obtained in this reaction.



1
($C_{24}H_{26}BrNO_6$)

2
($C_{16}H_{18}BrNO_5$,
not observed)

The ESI-MS spectra of pure **1** and the major reaction product are shown below.



10. What is the most likely chemical formula for the ions represented by each of the peak masses listed on the next page? In each formula, give not only the atomic symbol for each element, but also the isotope number for any element that is not >90% abundant in nature. Make sure to indicate the charge state of the ion.

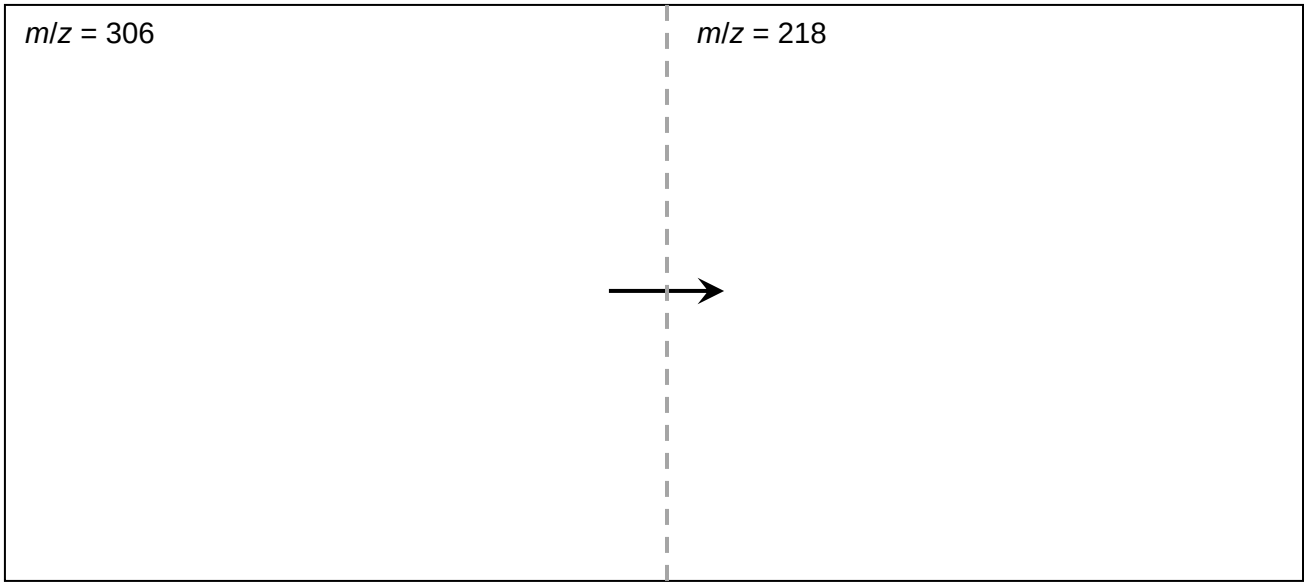
Example of answer format:

m/*z* = 42: $C_2(^{13}C)_1H_5^+$

m/*z* = 504:

m/*z* = 505:

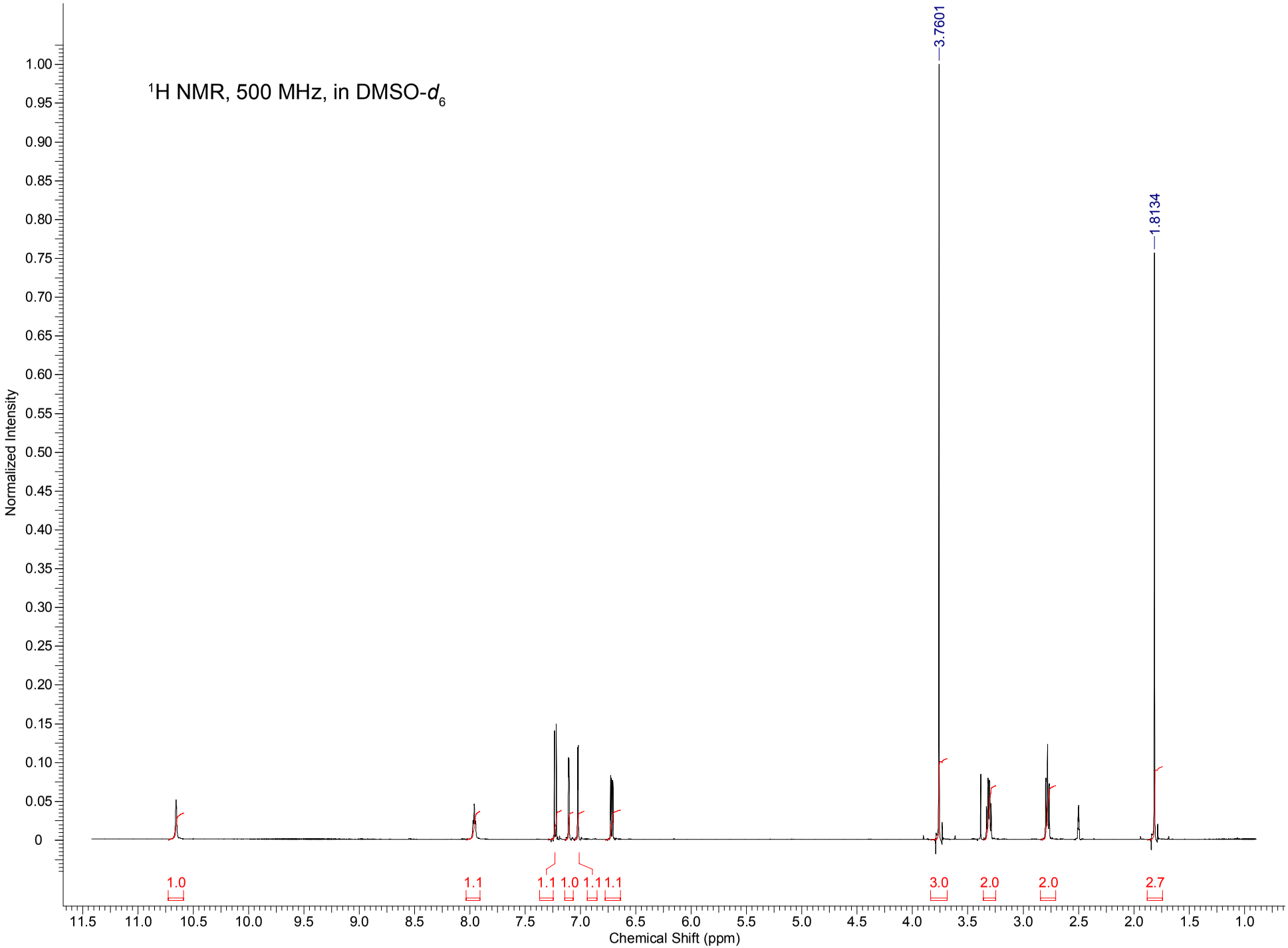
11. Assuming that the *m*/*z* = 306 peak in the ESI-MS spectrum of the reaction product corresponds to an unfragmented parent ion, what is the structure of this *m*/*z* = 306 ion? And how does it fragment to yield a *m*/*z* = 218 daughter ion? In the box below, draw both ion structures, and a mechanism (using “electron pushing”) that connects them.

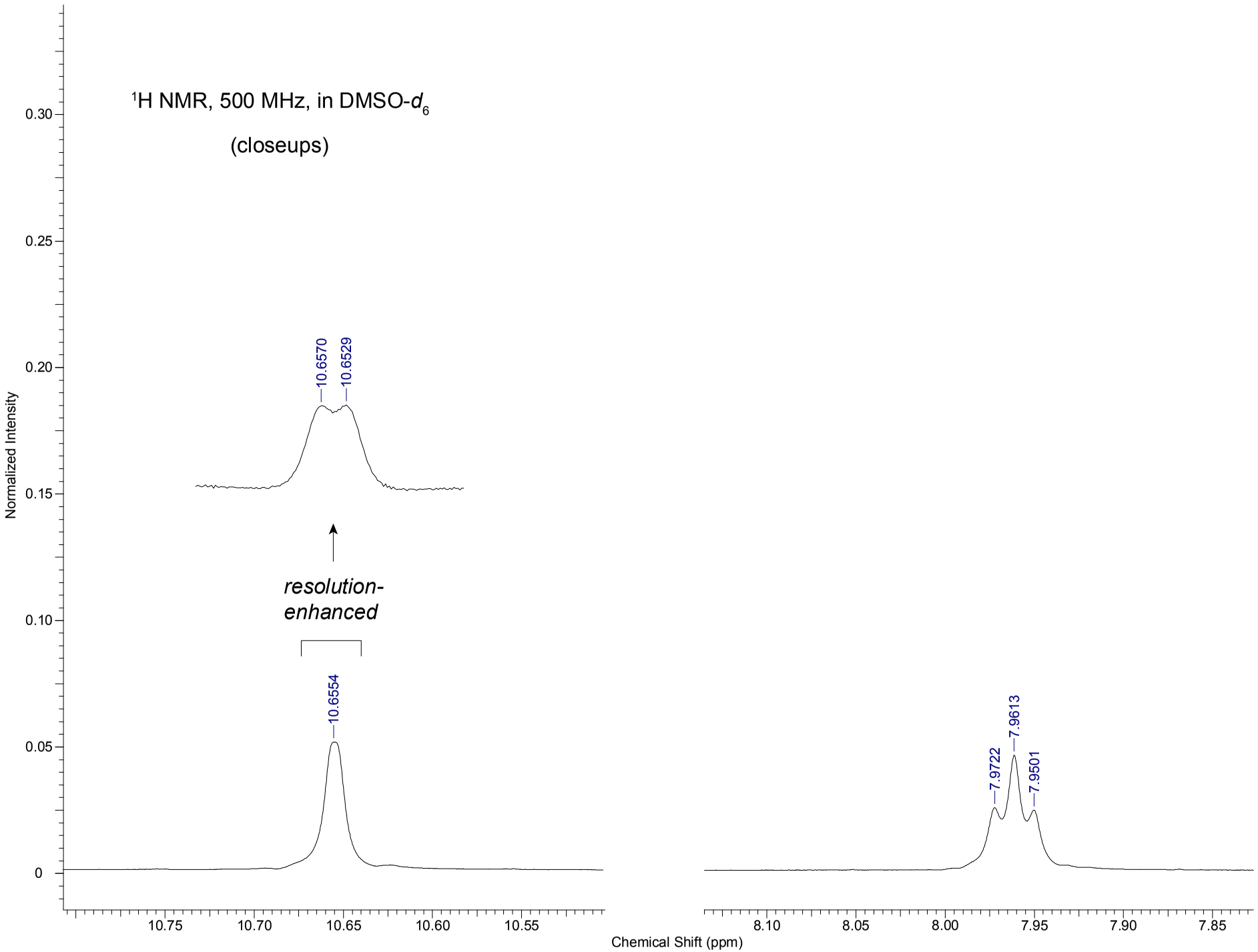


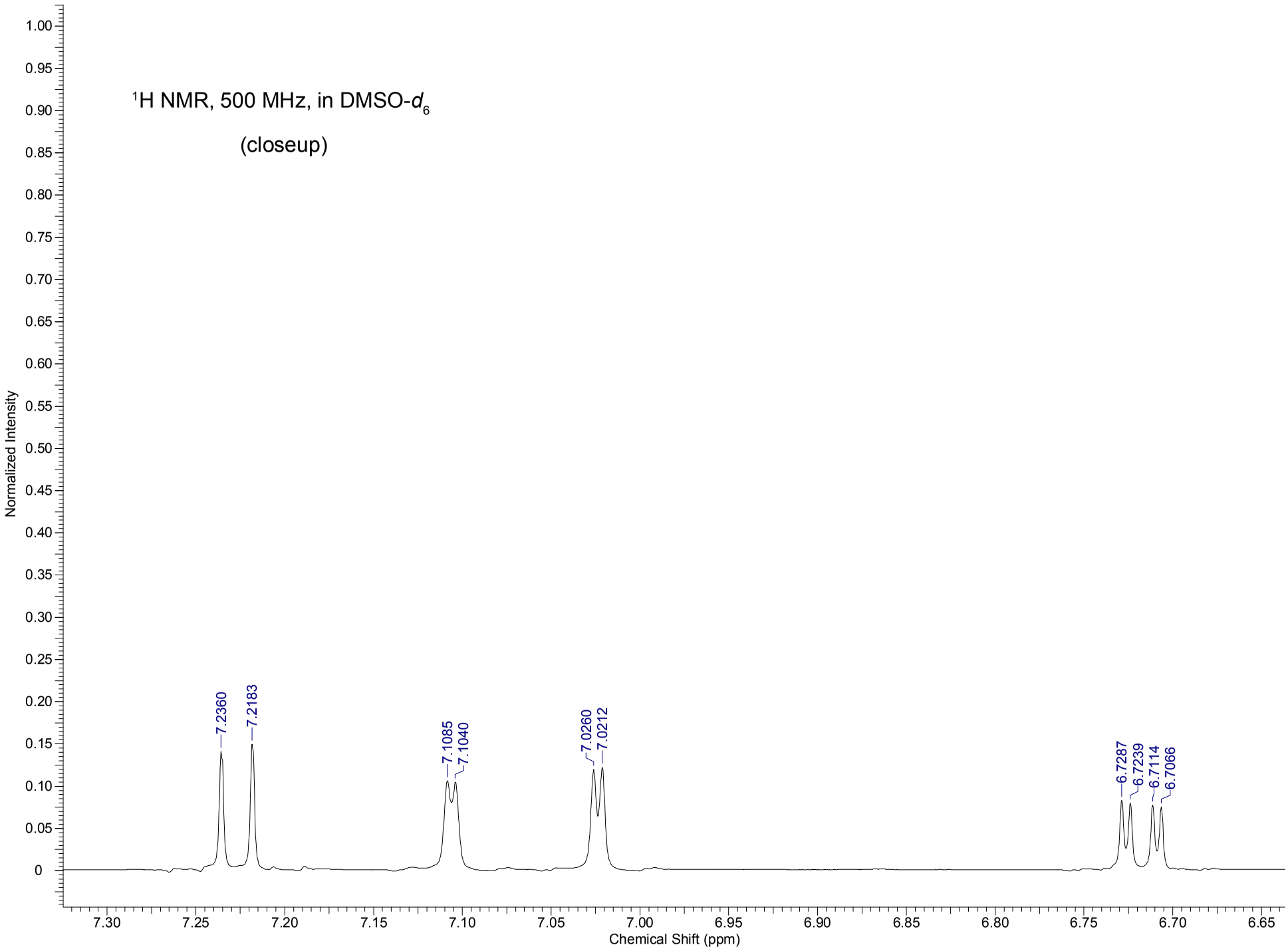
12. Bo didn’t even try analyzing his reaction via electron-ionization (EI) mass spectrometry, because he expected that both his starting material and any products would fragment too easily. But what if he had performed an EI-MS on his starting material—what fragments would we expect? In the boxes on the next page, draw mechanisms that show how electron-ionized molecule **1** might undergo two common fragmentation pathways in EI-MS: α-cleavage, and the McLafferty rearrangement. In each case, make sure you draw unpaired electrons and formal charges, and illustrate each mechanism using “electron pushing”.

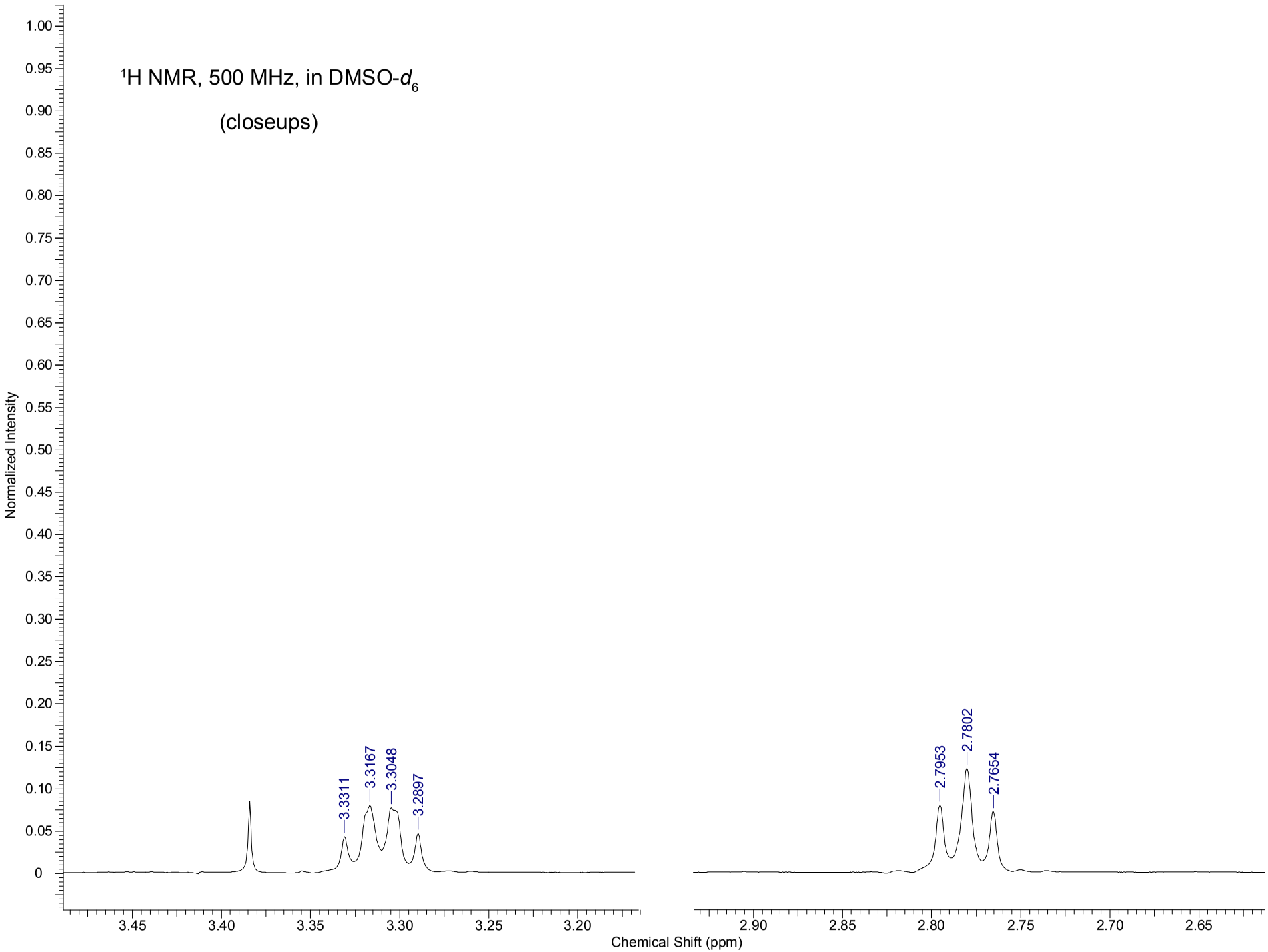
α-cleavage

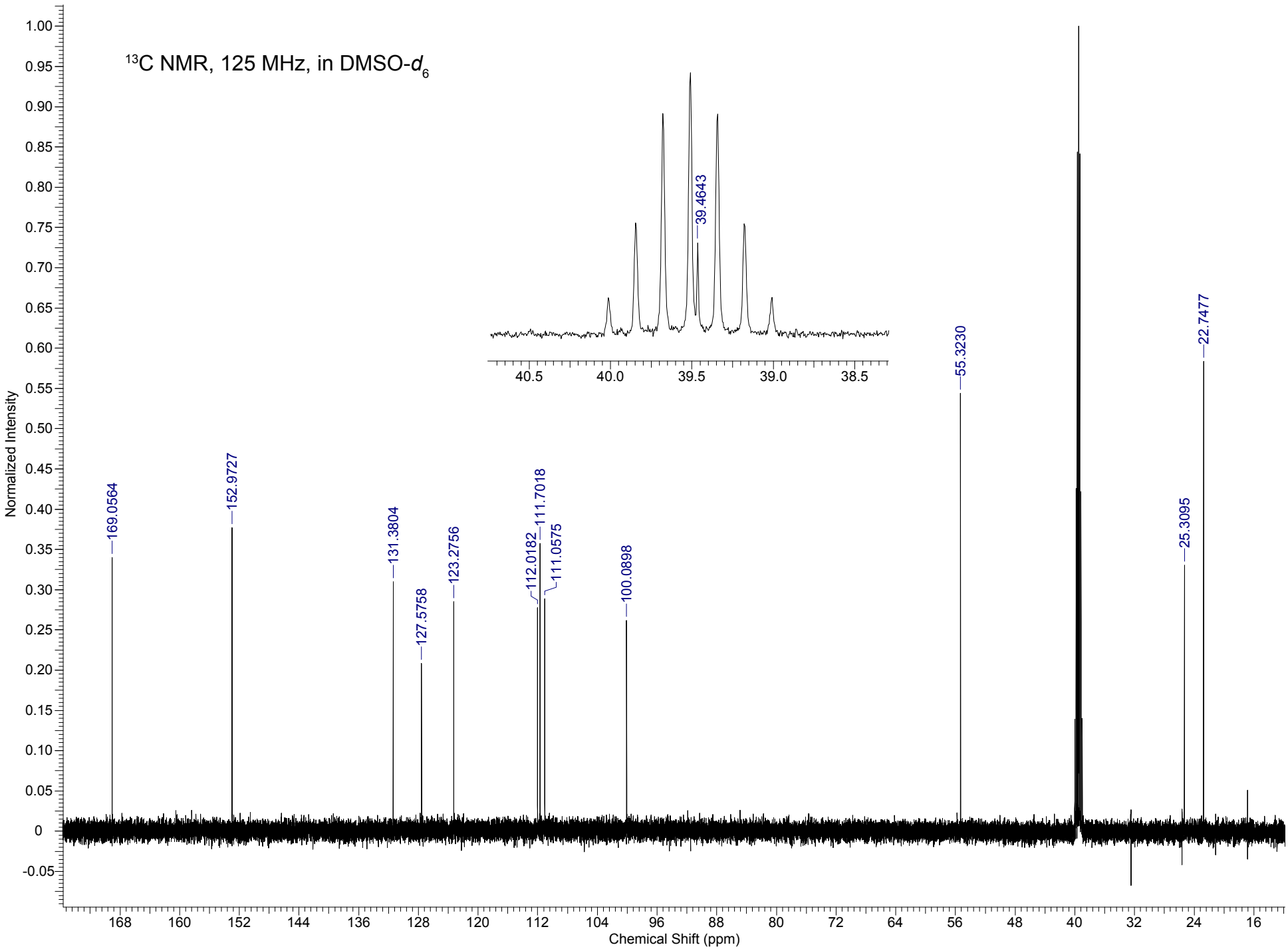
McLafferty rearrangement


^{1}H NMR, 500 MHz, in DMSO-d_6
3.7601
1.8134
Normalized Intensity
Chemical Shift (ppm)
1.0
1.1
1.1
1.0
1.1
1.1
3.0
2.0
2.0
2.7

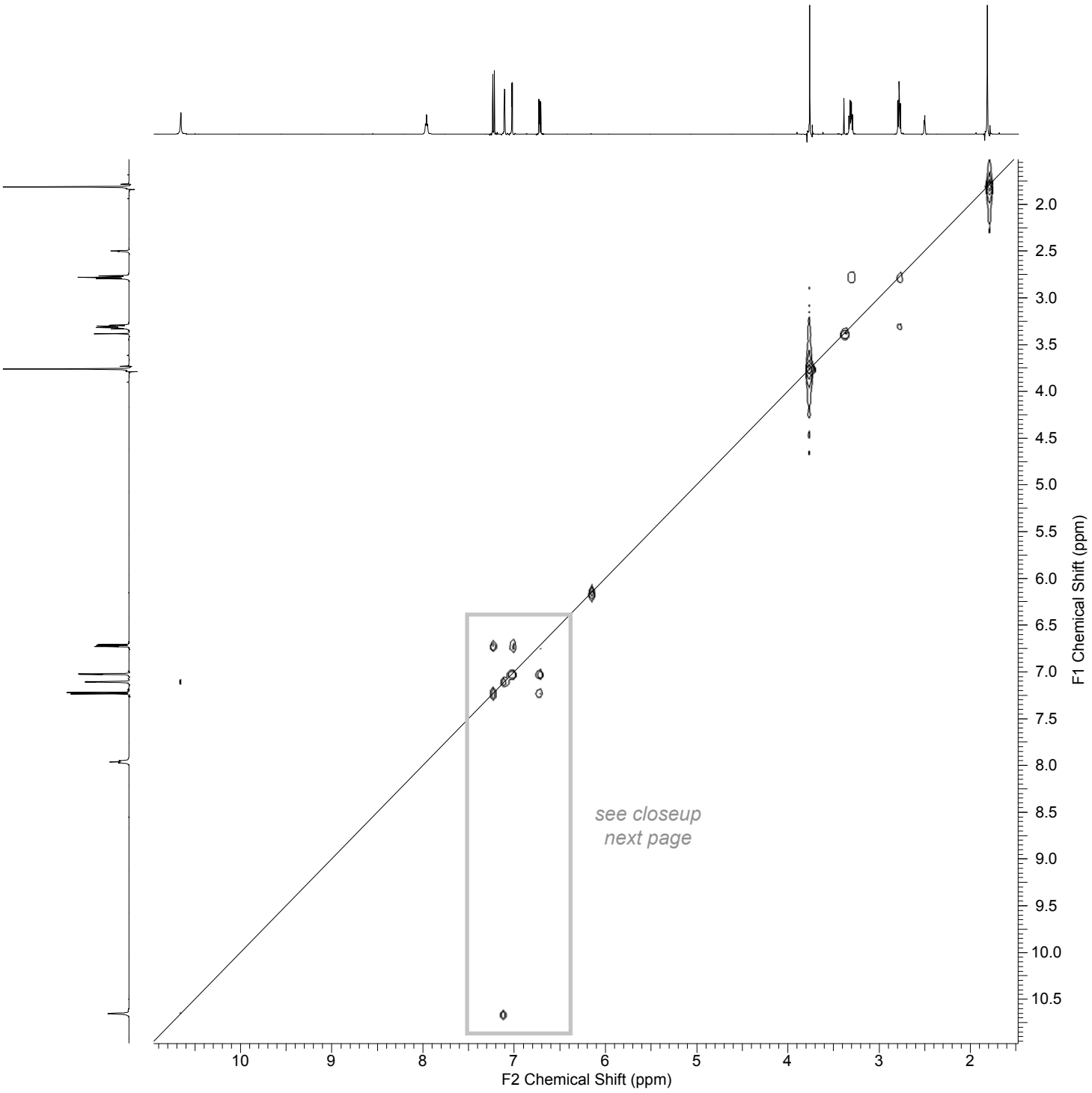

^{1}H NMR, 500 MHz, in DMSO-d_6
(closeups)
Normalized Intensity
0.30
0.25
0.20
0.15
0.10
0.05
0
10.6570
10.6529
resolution-
enhanced
10.6554
7.9722
7.9613
7.9501
10.75
10.70
10.65
10.60
10.55
8.10
8.05
8.00
7.95
7.90
7.85
Chemical Shift (ppm)


^{1}H NMR, 500 MHz, in DMSO-d_6
(closeup)
Normalized Intensity
1.00
0.95
0.90
0.85
0.80
0.75
0.70
0.65
0.60
0.55
0.50
0.45
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0
7.2360
7.2183
7.1085
7.1040
7.0260
7.0212
6.7287
6.7239
6.7114
6.7066
7.30
7.25
7.20
7.15
7.10
7.05
7.00
6.95
6.90
6.85
6.80
6.75
6.70
6.65
Chemical Shift (ppm)


^{1}H NMR, 500 MHz, in DMSO-d_6
(closeups)
Normalized Intensity
1.00
0.95
0.90
0.85
0.80
0.75
0.70
0.65
0.60
0.55
0.50
0.45
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0
3.3311
3.3167
3.3048
3.2897
2.7953
2.7802
2.7654
3.45
3.40
3.35
3.30
3.25
3.20
2.90
2.85
2.80
2.75
2.70
2.65
Chemical Shift (ppm)

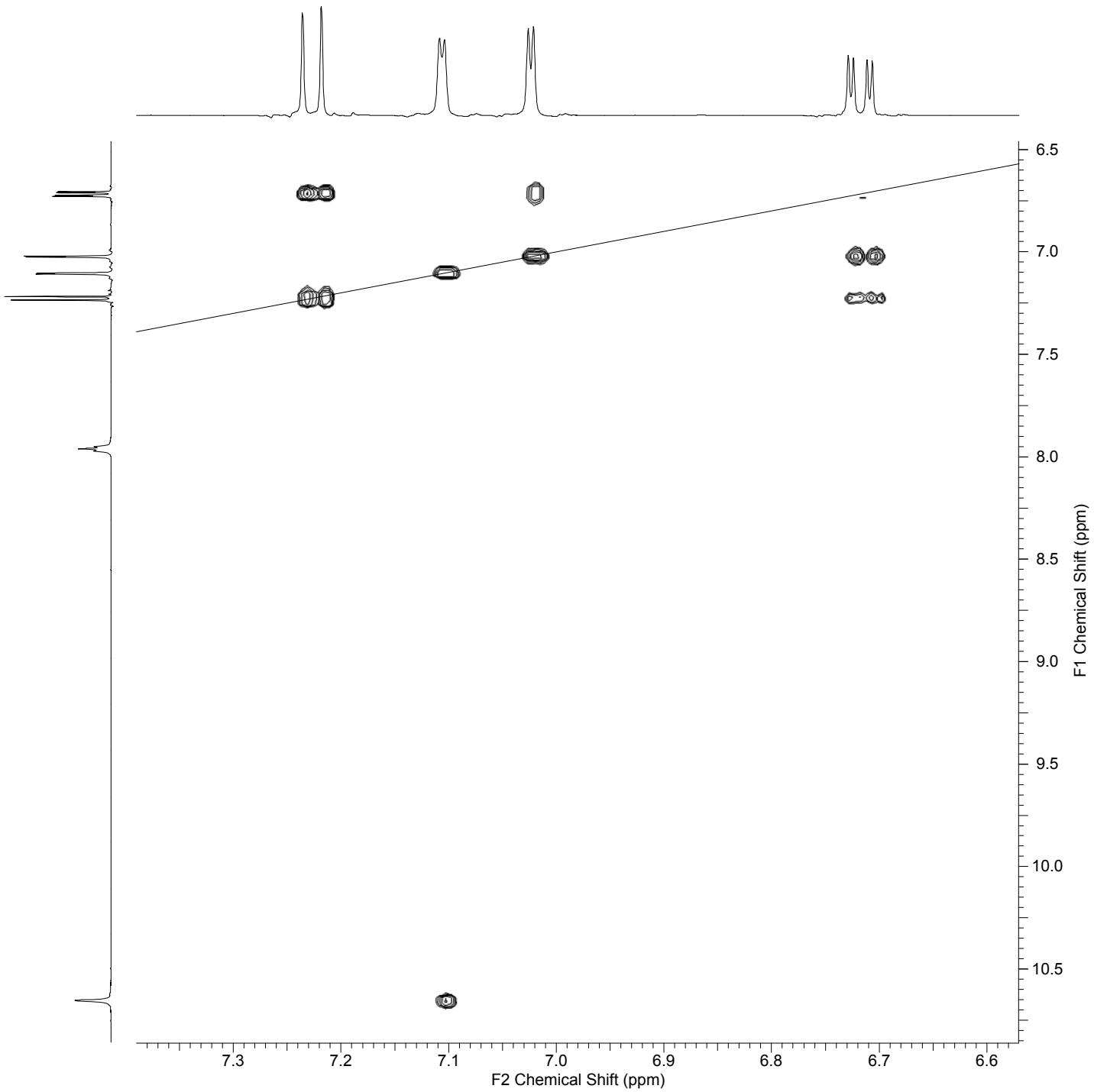

^{13}C NMR, 125 MHz, in DMSO-d_6
Normalized Intensity
Chemical Shift (ppm)
169.0564
152.9727
131.3804
127.5758
123.2756
112.0182
111.7018
111.0575
100.0898
55.3230
39.4643
25.3095
22.7477

^{1}H-^{1}H gCOSY, 500 MHz, in DMSO-d_6

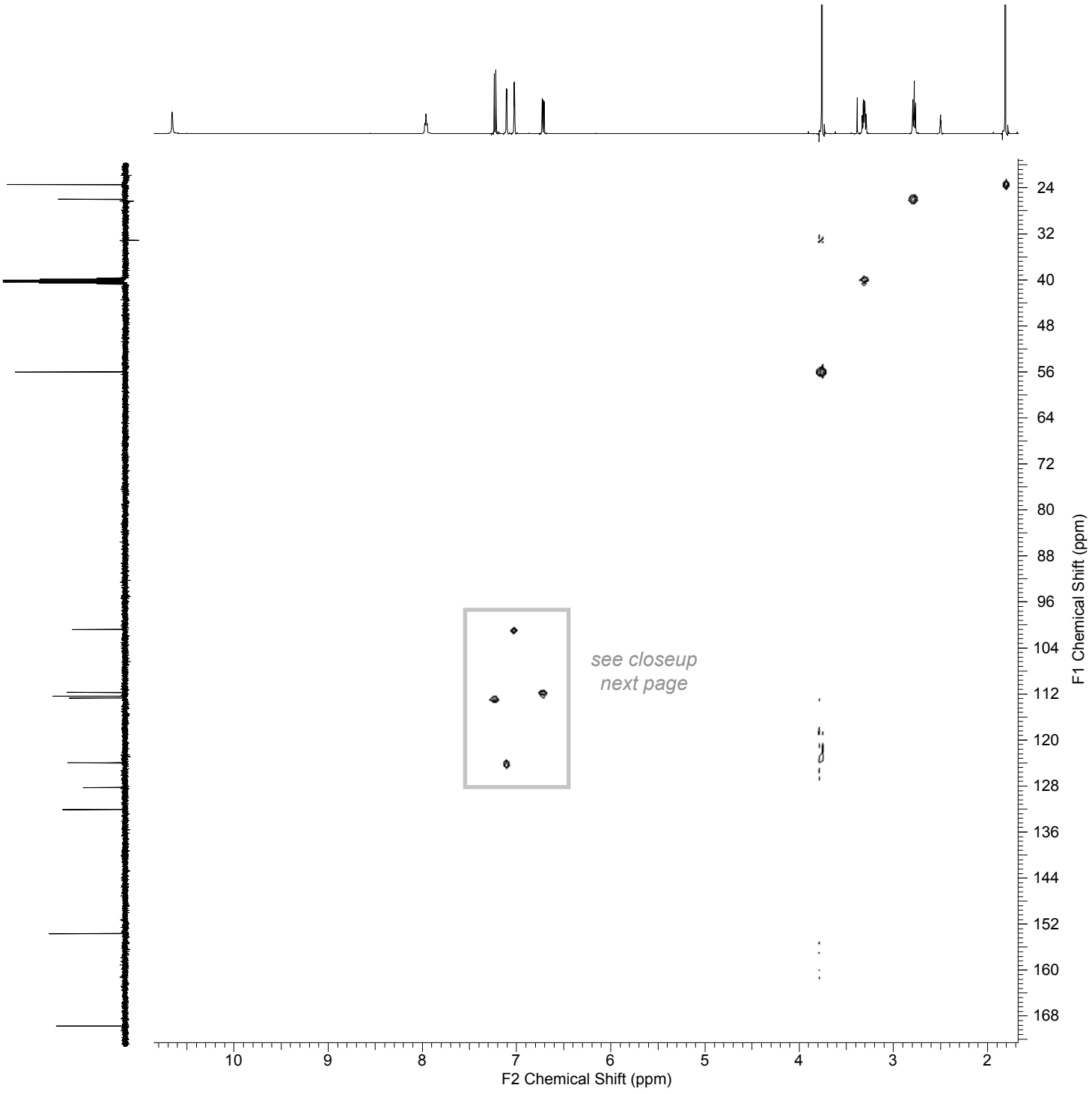


^{1}H-^{1}H gCOSY, 500 MHz, in DMSO-d_6

(closeup)

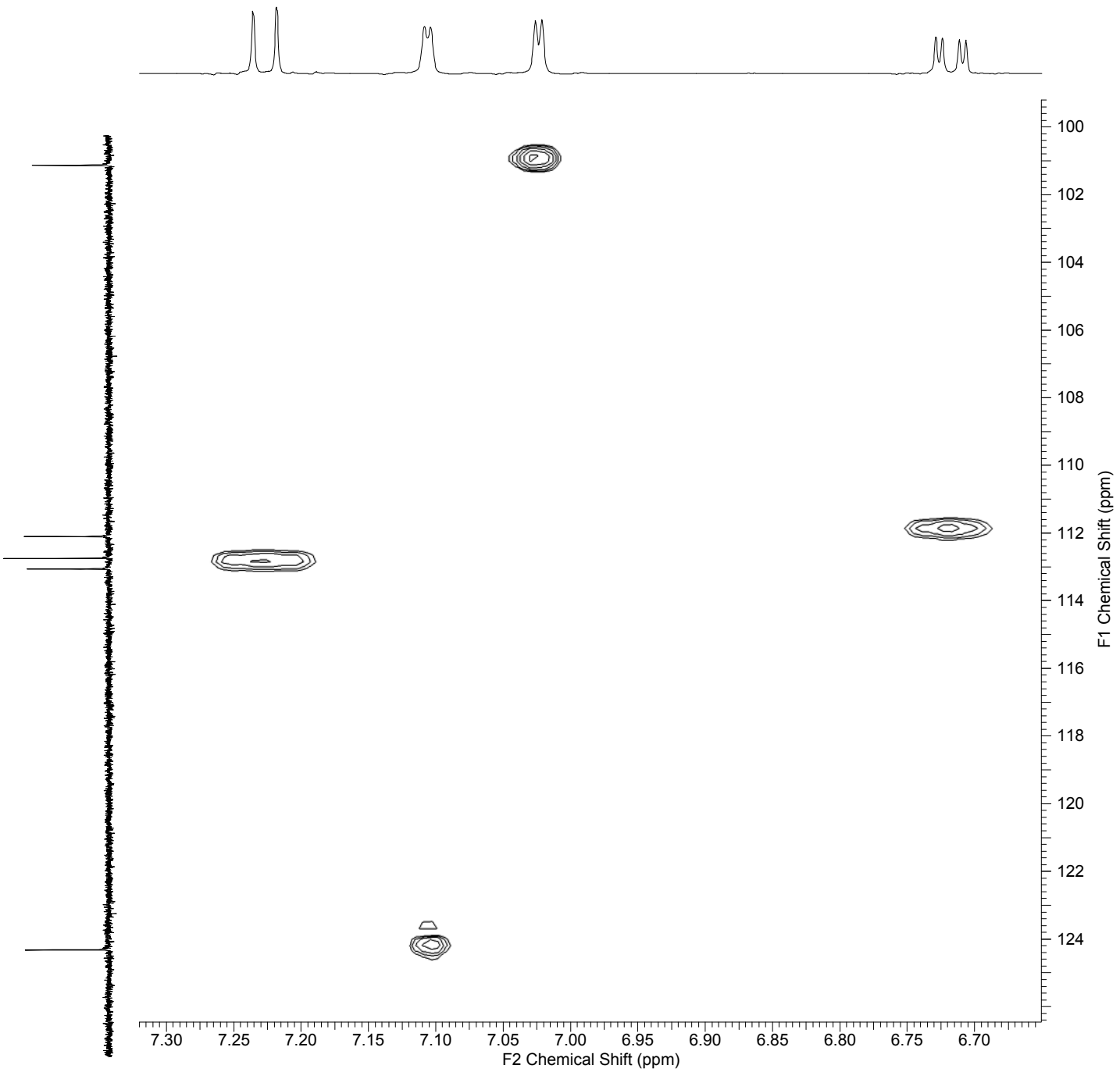


^{1}H-^{13}C HSQC, 500/125 MHz, in DMSO-d_6

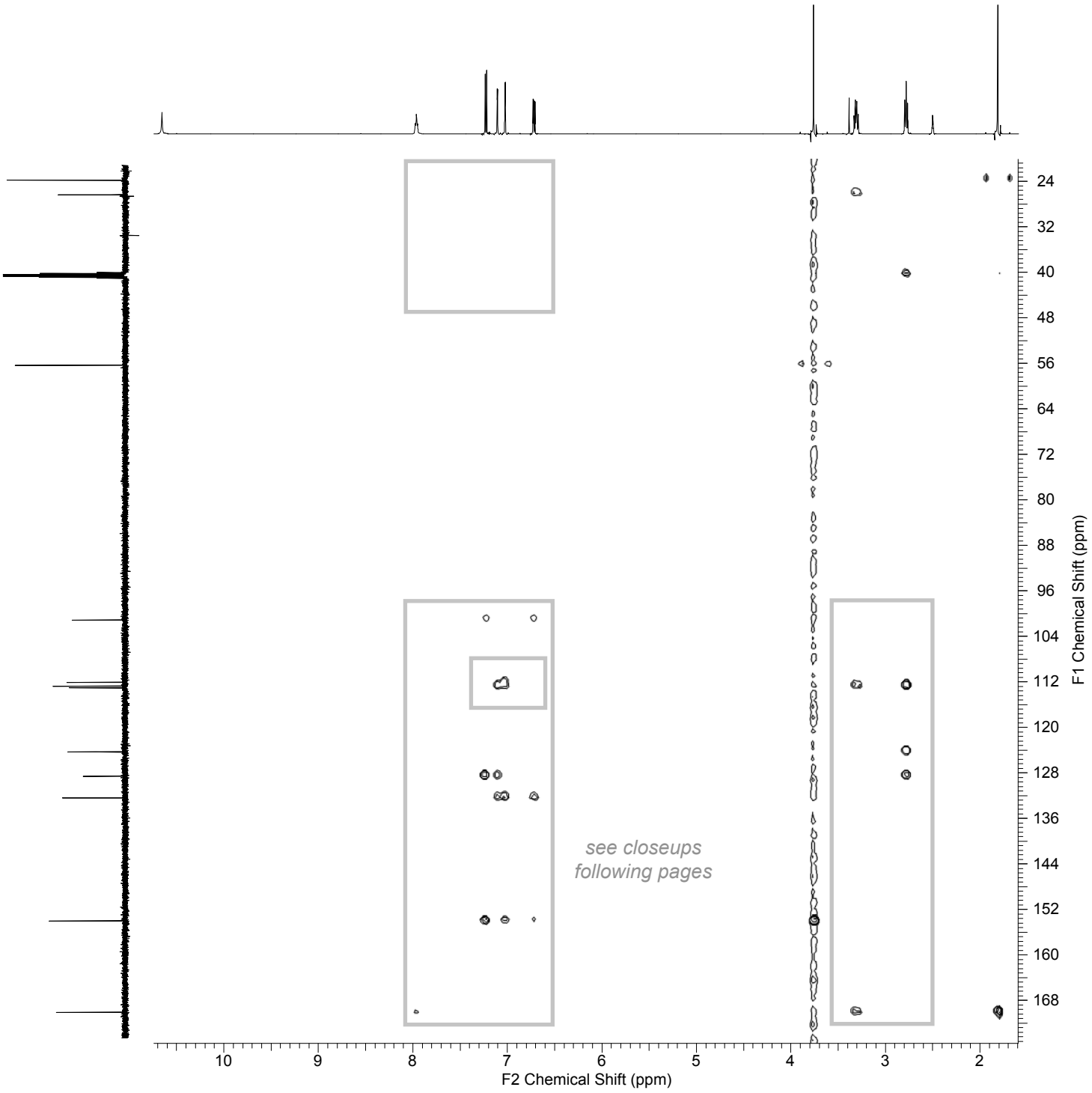


^{1}H-^{13}C HSQC, 500/125 MHz, in DMSO-d_6

(closeup)

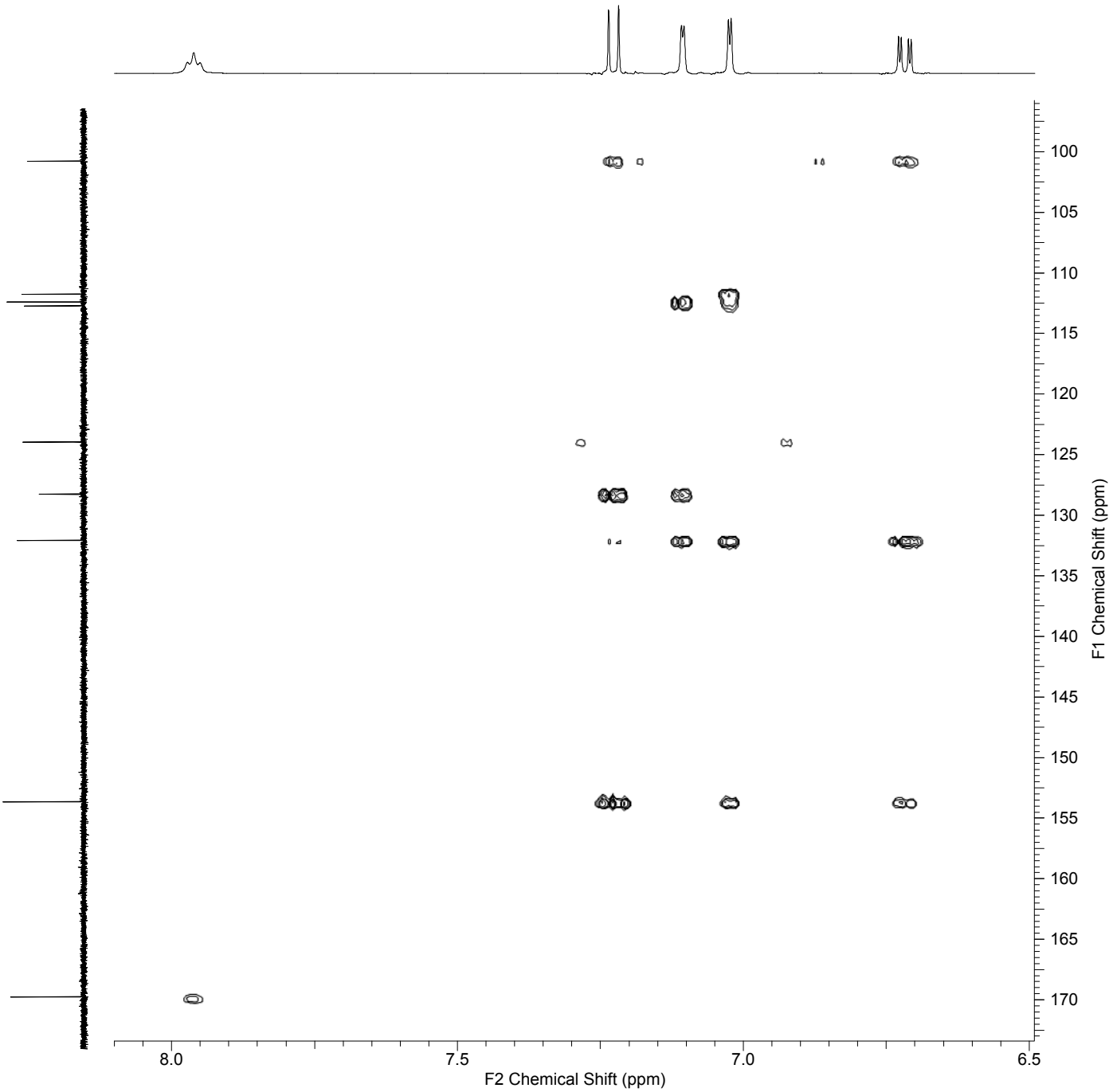


^{1}H-^{13}C gHMBC, 500/125 MHz, in DMSO-d_6



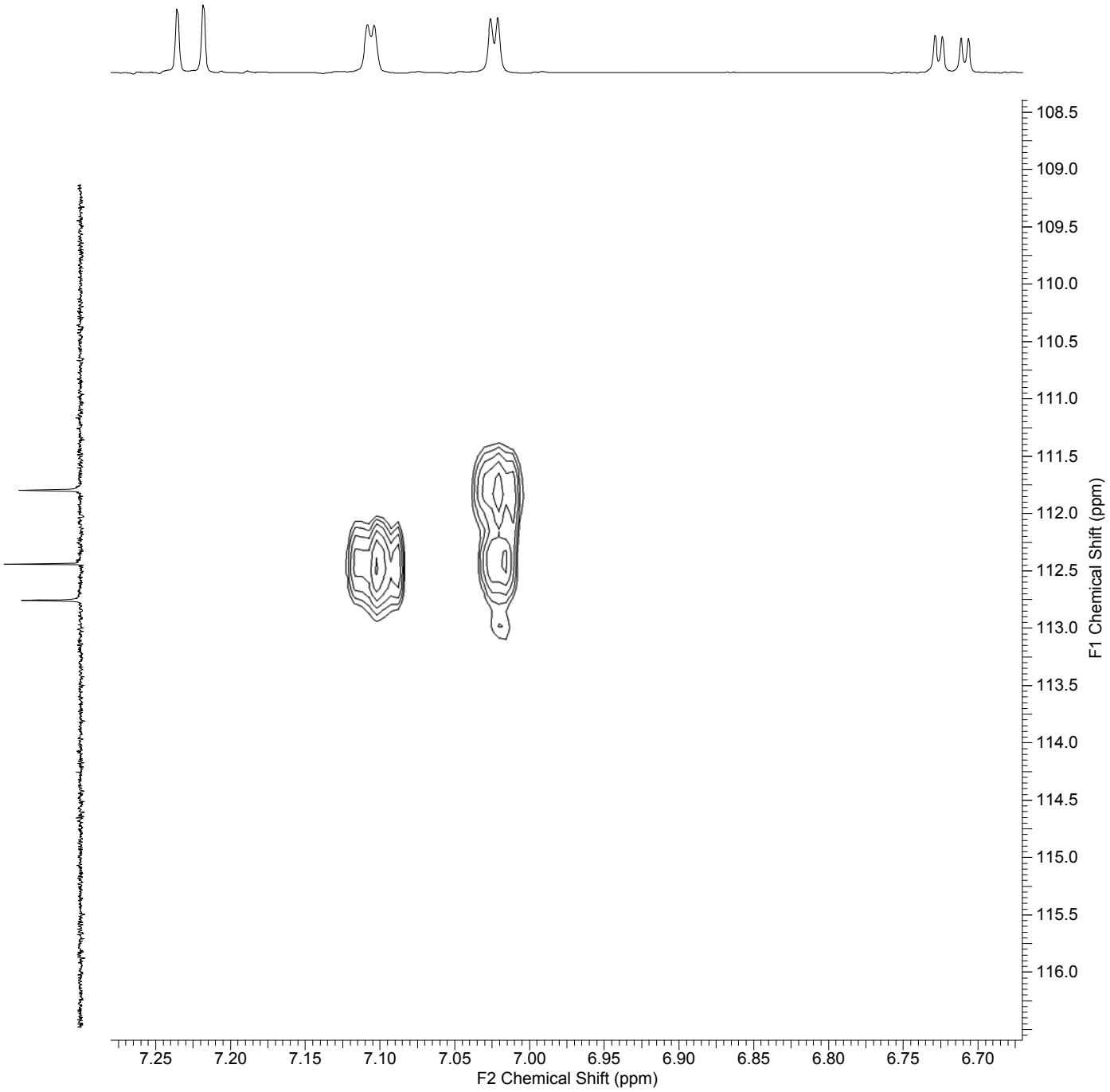
^{1}H-^{13}C gHMBC, 500/125 MHz, in DMSO-d_6

(closeup)



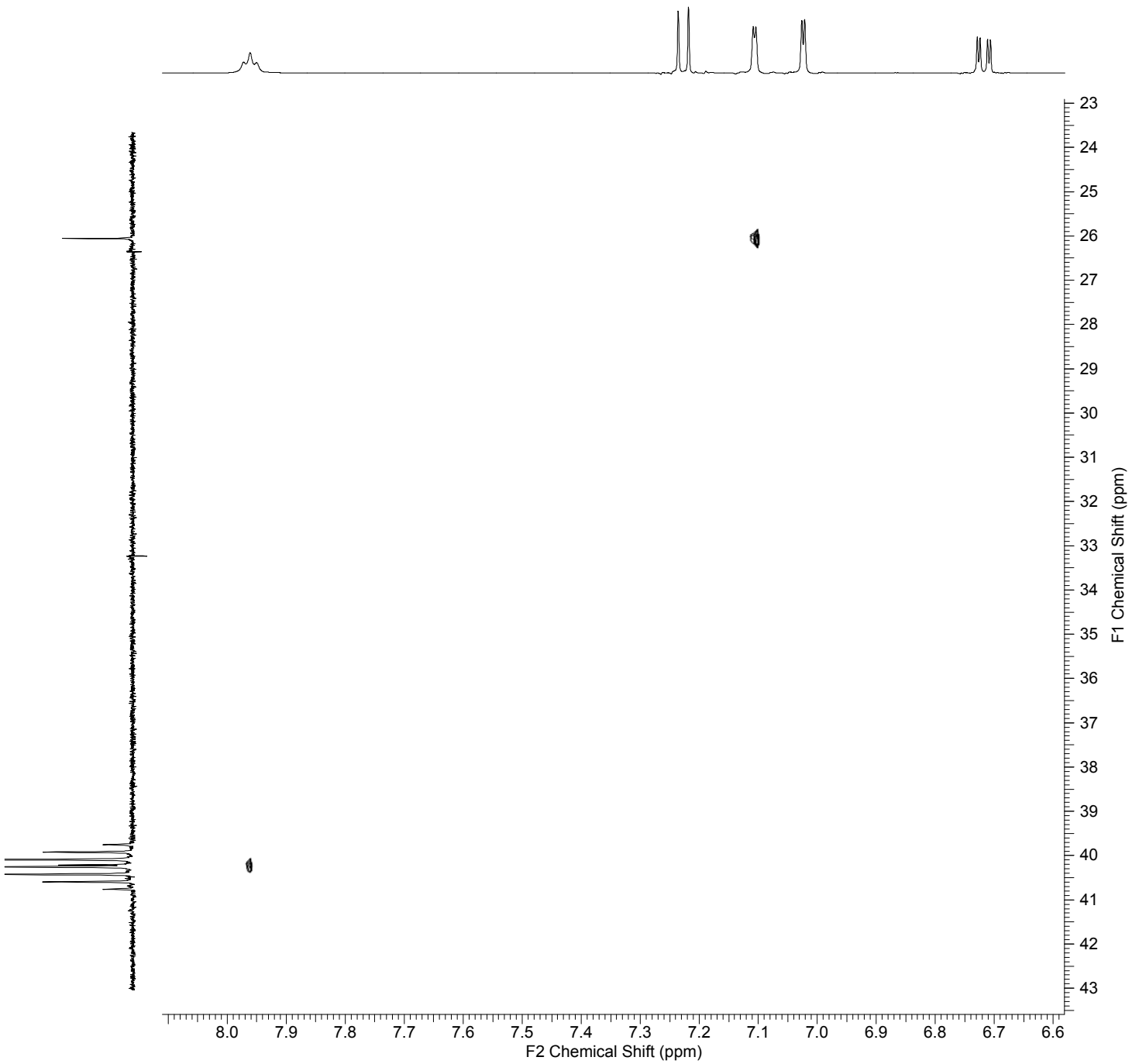
^{1}H-^{13}C gHMBC, 500/125 MHz, in DMSO-d_6

(closeup)



^{1}H-^{13}C gHMBC, 500/125 MHz, in DMSO-d_6

(closeup)



^{1}H-^{13}C gHMBC, 500/125 MHz, in DMSO-d_6

(closeup)