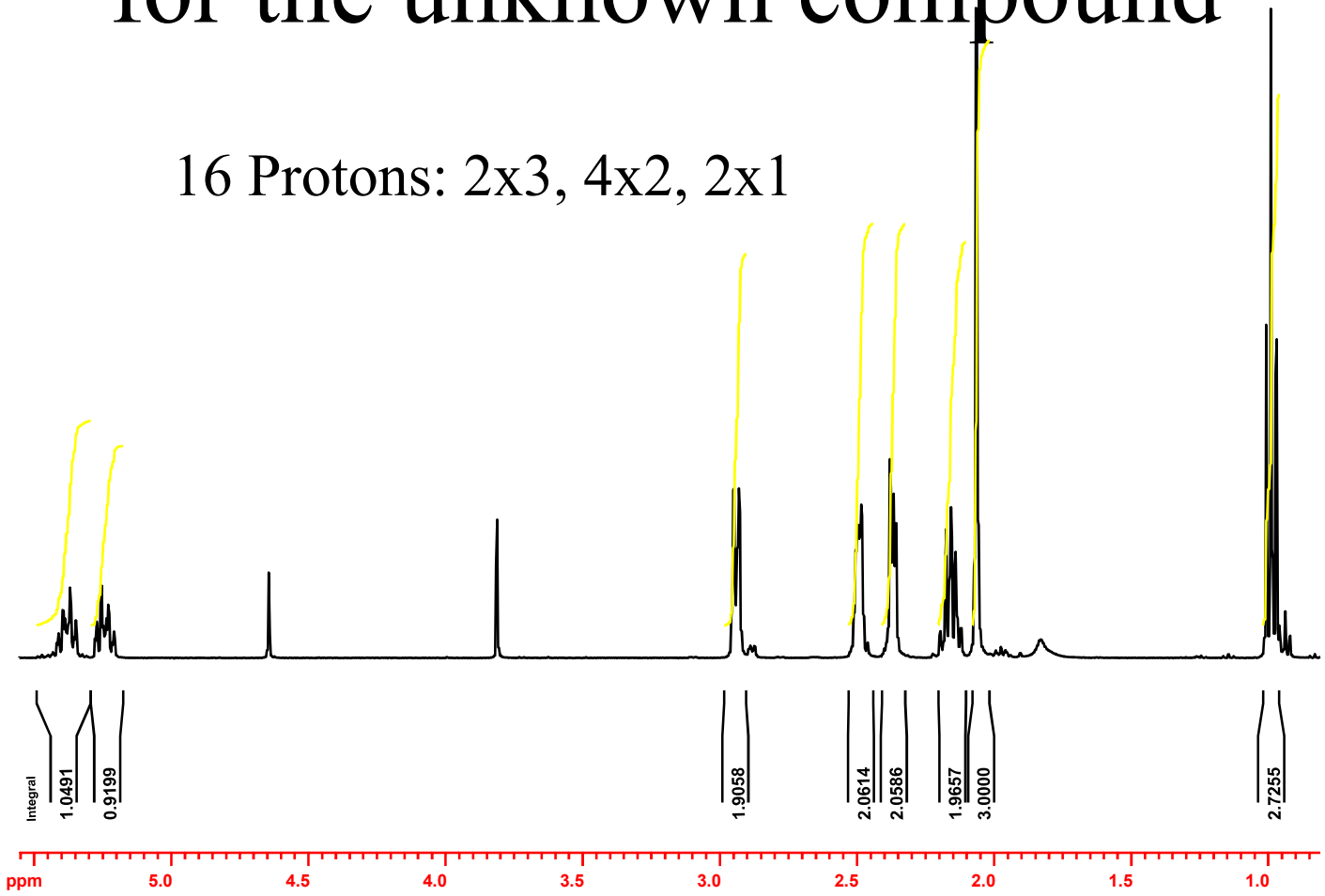
NMR assignment

By Roy Hoffman

Number of protons from integrals for the unknown compound

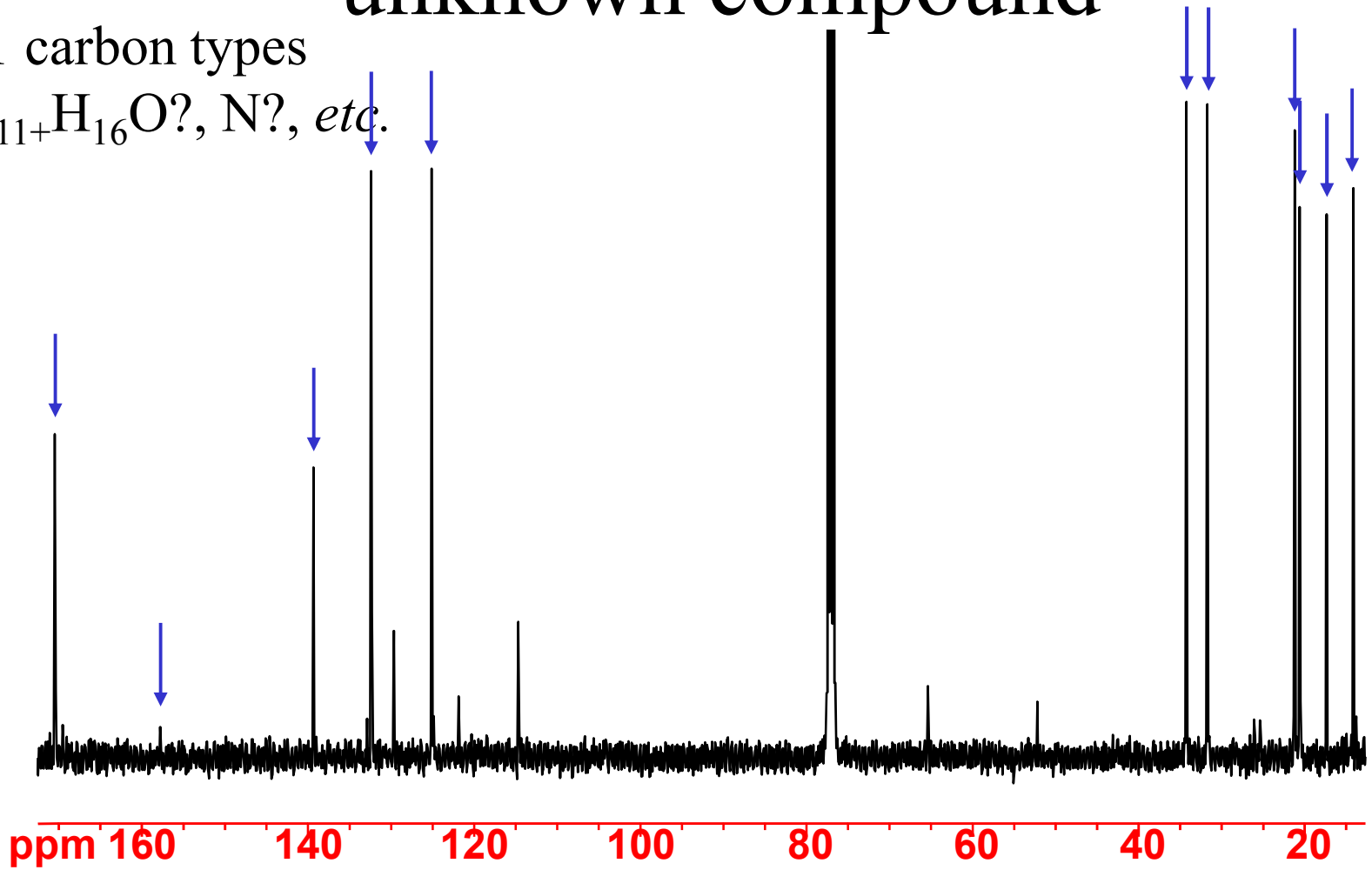
16 Protons: 2x3, 4x2, 2x1



Number of carbon types for the unknown compound

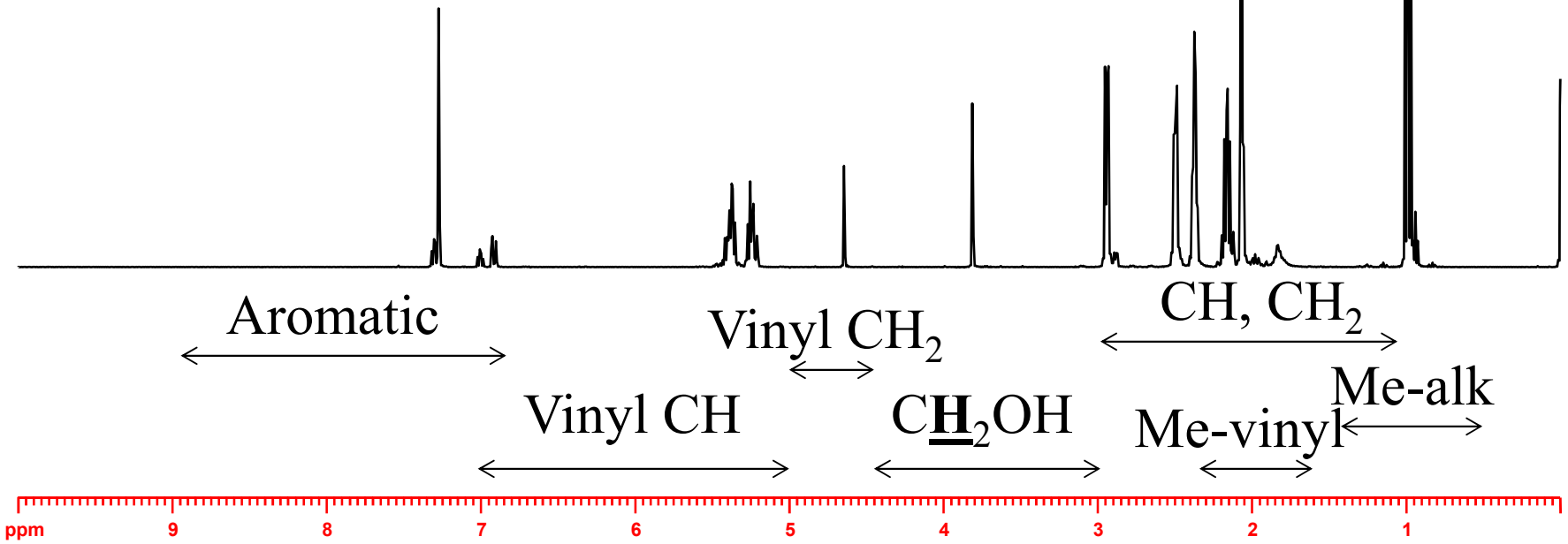
11 carbon types

$C_{11+}H_{16}O$?, N?, *etc.*



Functionality from ^{1}H chemical shifts for the unknown compound

2x1vinyl CH
1x3 methyl alkyl
4x2 CH_2



Functionality from ^{13}C chemical shifts for the unknown compound

2x1vinyl CH

1x3 methyl alkyl

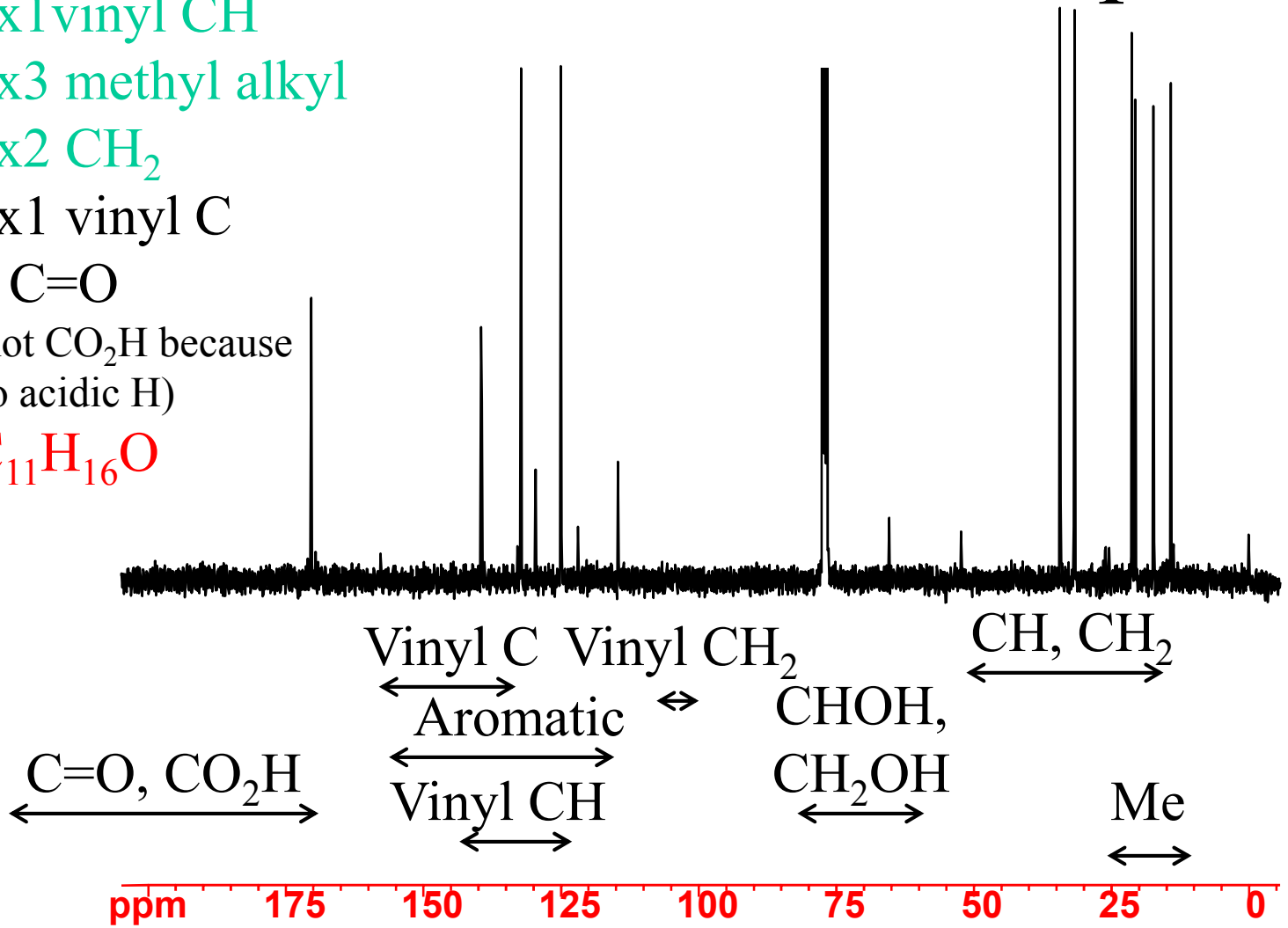
4x2 CH_2

2x1 vinyl C

1 C=O

(not CO_2H because no acidic H)

$C_{11}H_{16}O$

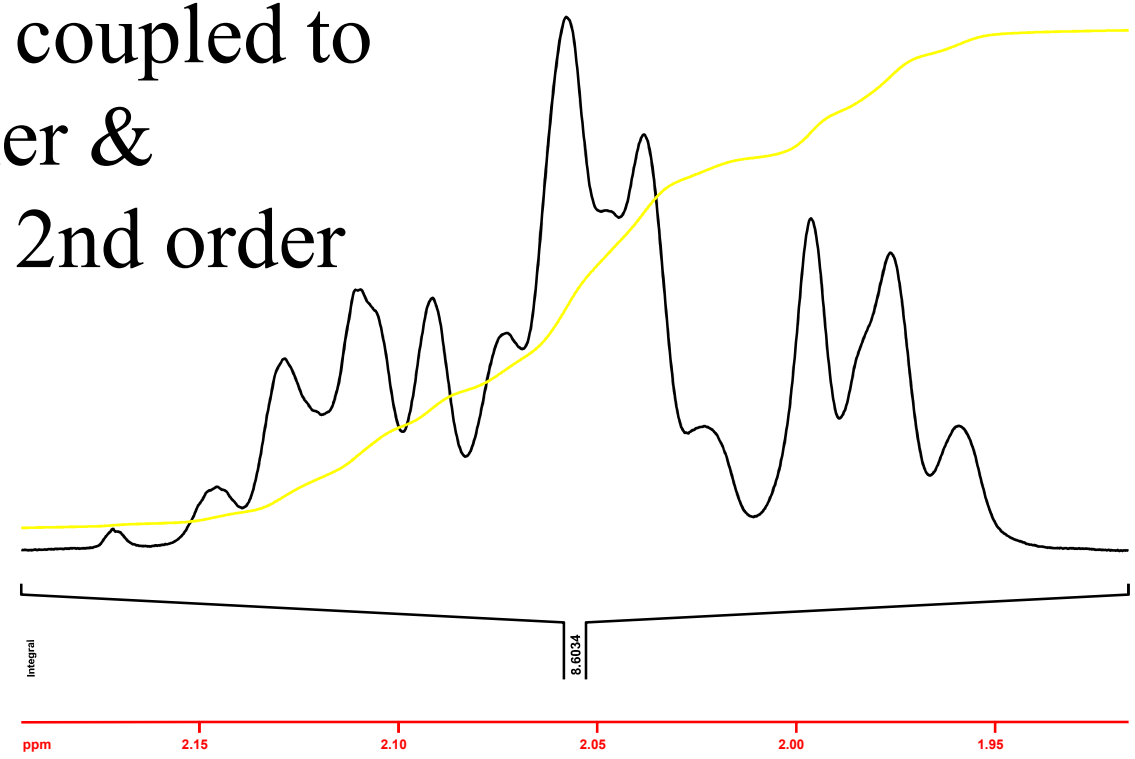


The situation so far for the unknown compound

- $C_{11}H_{16}O$ with C=O and 2 double bonds.
- This requires one ring (Without it would be $C_{11}H_{18}O$).
- There are 2 terminal methyls.
- One of the vinyls has two *trans* H's
- Look for the following ring structures

More complex structures require 2D interpretation.

•4 pairs of protons strongly coupled to each other & displays 2nd order splitting

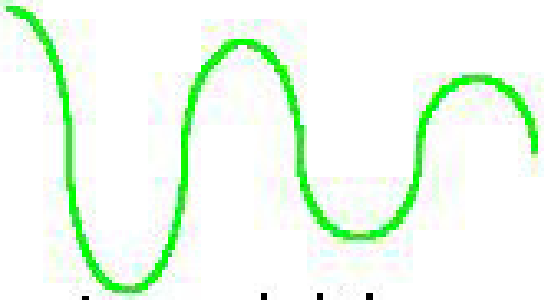


Symbols used in pulse sequences

Radio frequency (hard) pulse

Gradient pulse

Radio frequency soft pulse



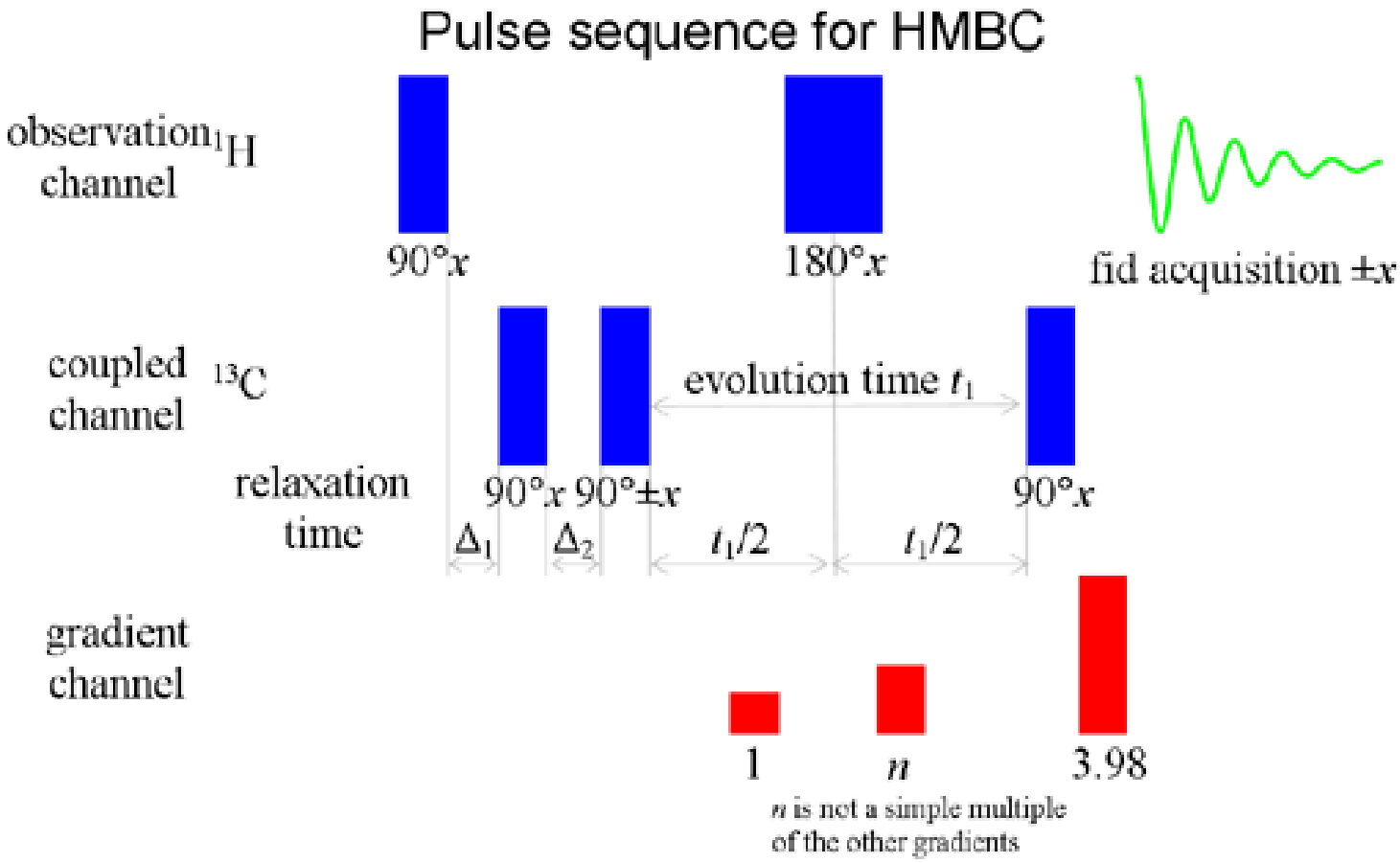
Acquisition of FID

Decoupling

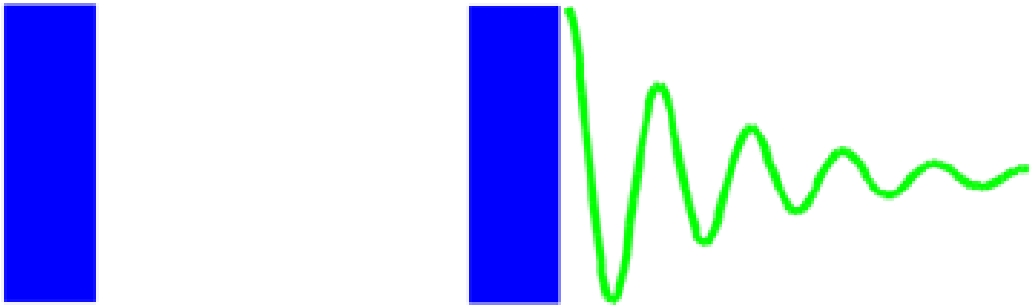
Gradient pulses

- It is possible to apply a magnetic gradient to the sample.
- A gradient affects the signal in the following manner. At the start of the experiment it disperses the signal, making it disappear. Then the application of a gradient in the opposite direction allows the signal to be seen again.
- In combination with rf pulses that act as quantum filters it is possible to observe correlations between nuclei.

Example pulse sequence

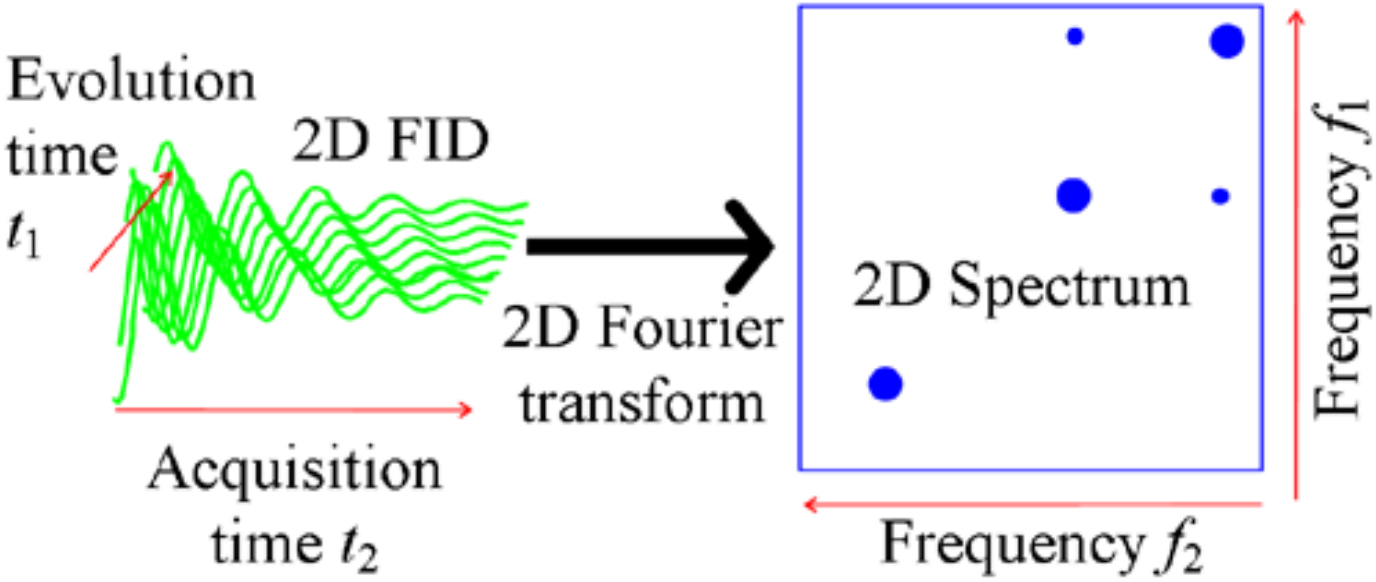


Basic pulse sequence for 2D acquisition



Relaxation time | Evolution time, t_1 | Acquisition of fid, t_2

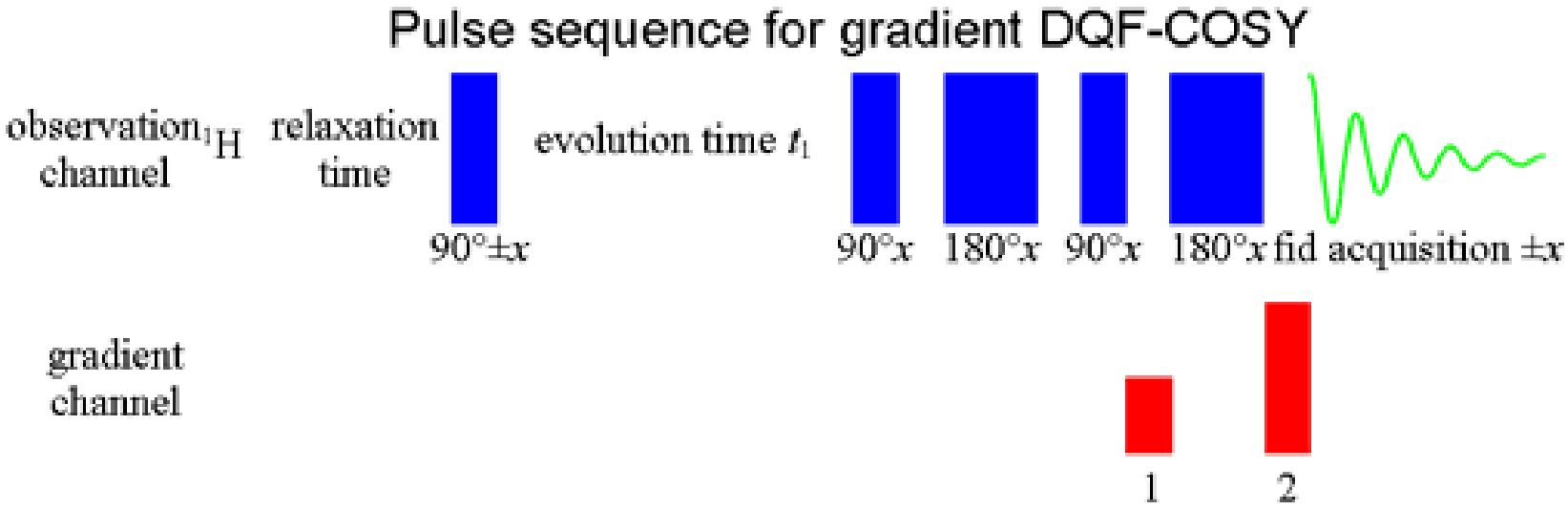
2D Fourier transform



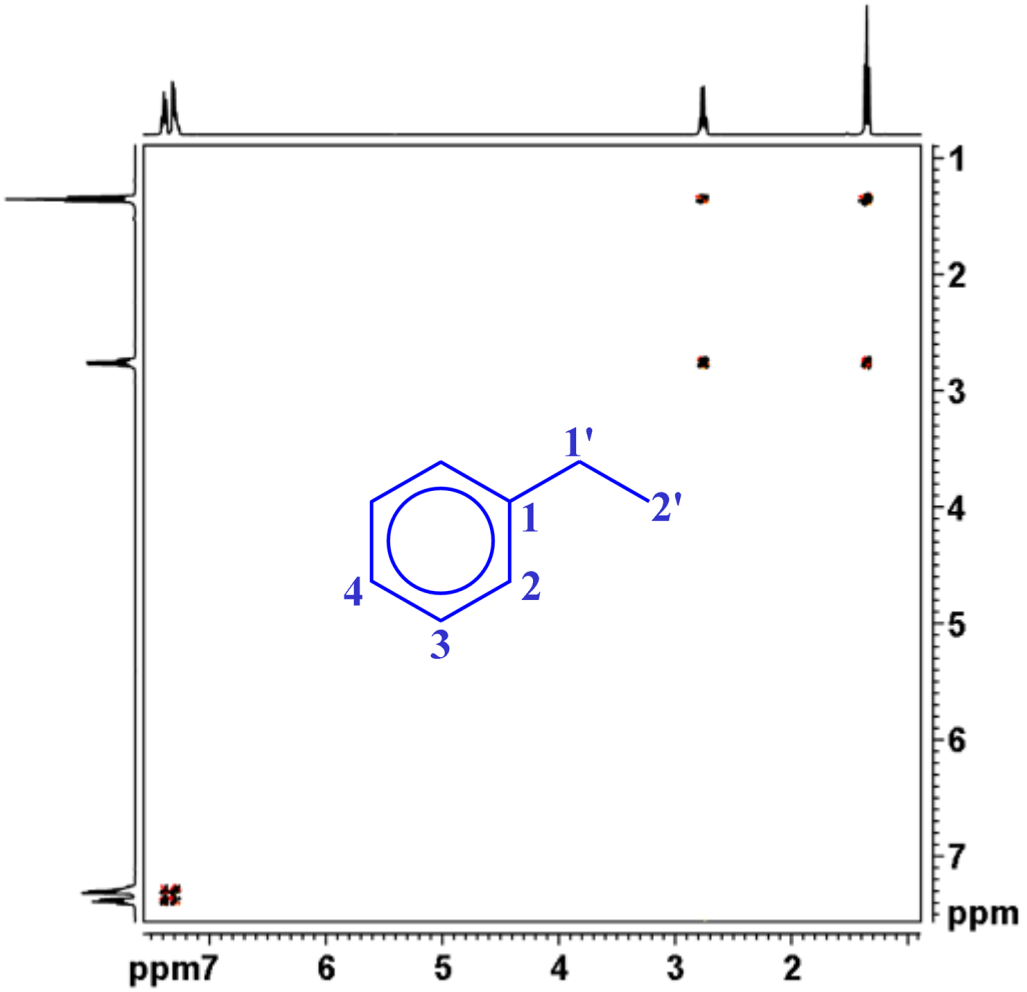
Types of 2D

- Through bond
  - COSY, TOCSY, heteronuclear correlation (HSQC, HMBC, HMQC), 2D-INADEQUATE
- Through space
  - NOESY, ROESY, HOESY
- Resolving the Hamiltonian
  - J-resolved, MQ-MAS

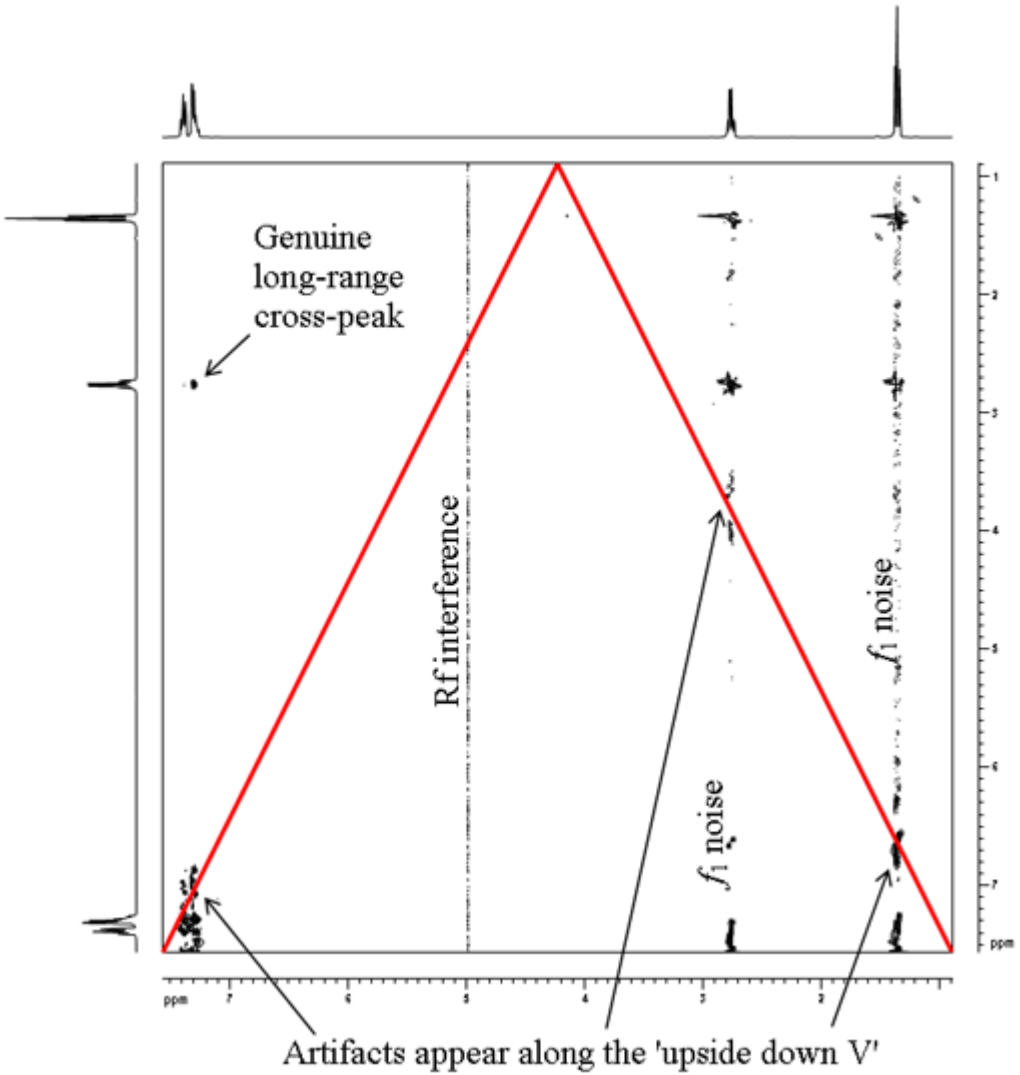
COSY


Pulse sequence for gradient DQF-COSY
observation ^{1}H channel
relaxation time
evolution time t_1
90°±x
90°x
180°x
90°x
180°x
fid acquisition ±x
gradient channel
1
2

COSY of ethylbenzene

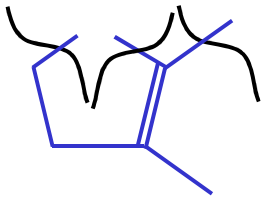


Artifacts in COSY



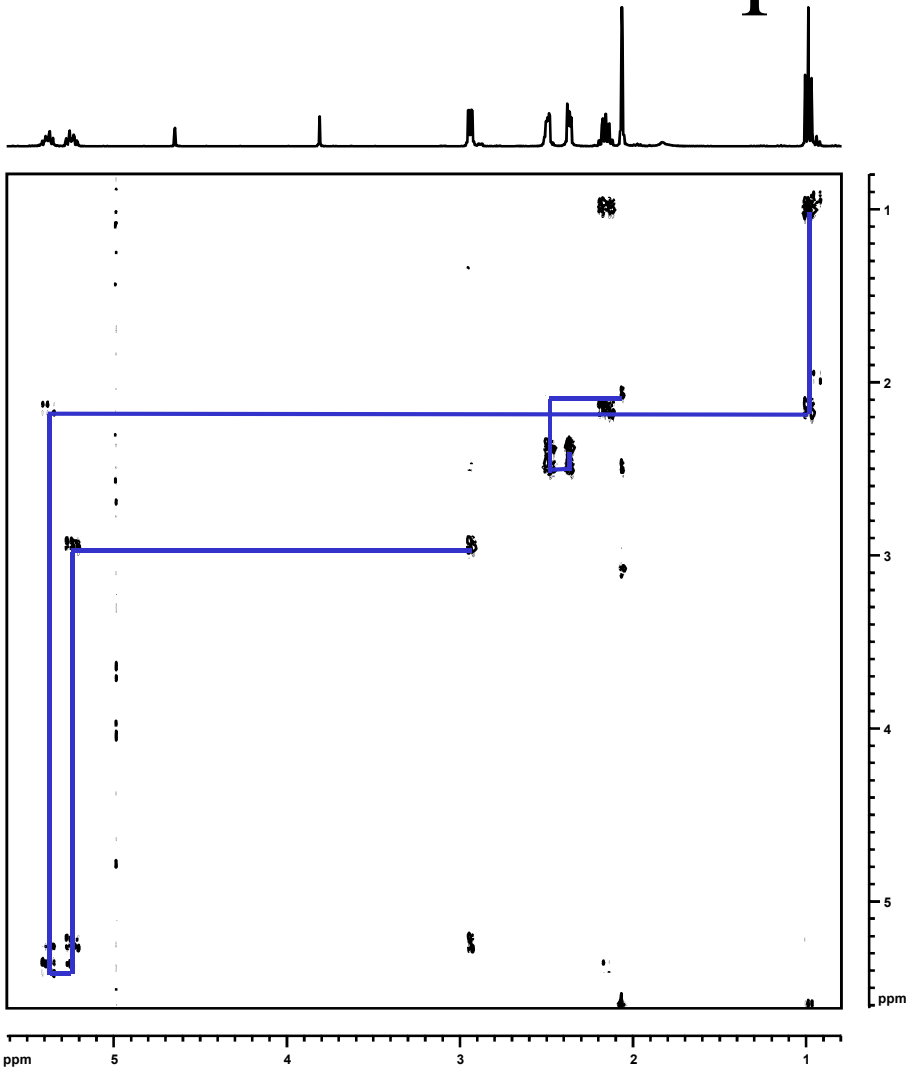
COSY of the unknown compound

Two networks:
CH3CH2CH=CHCH2

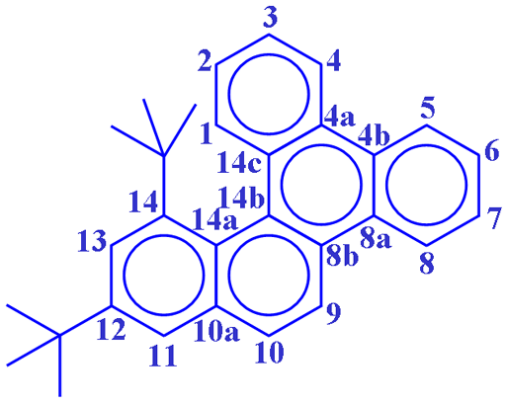


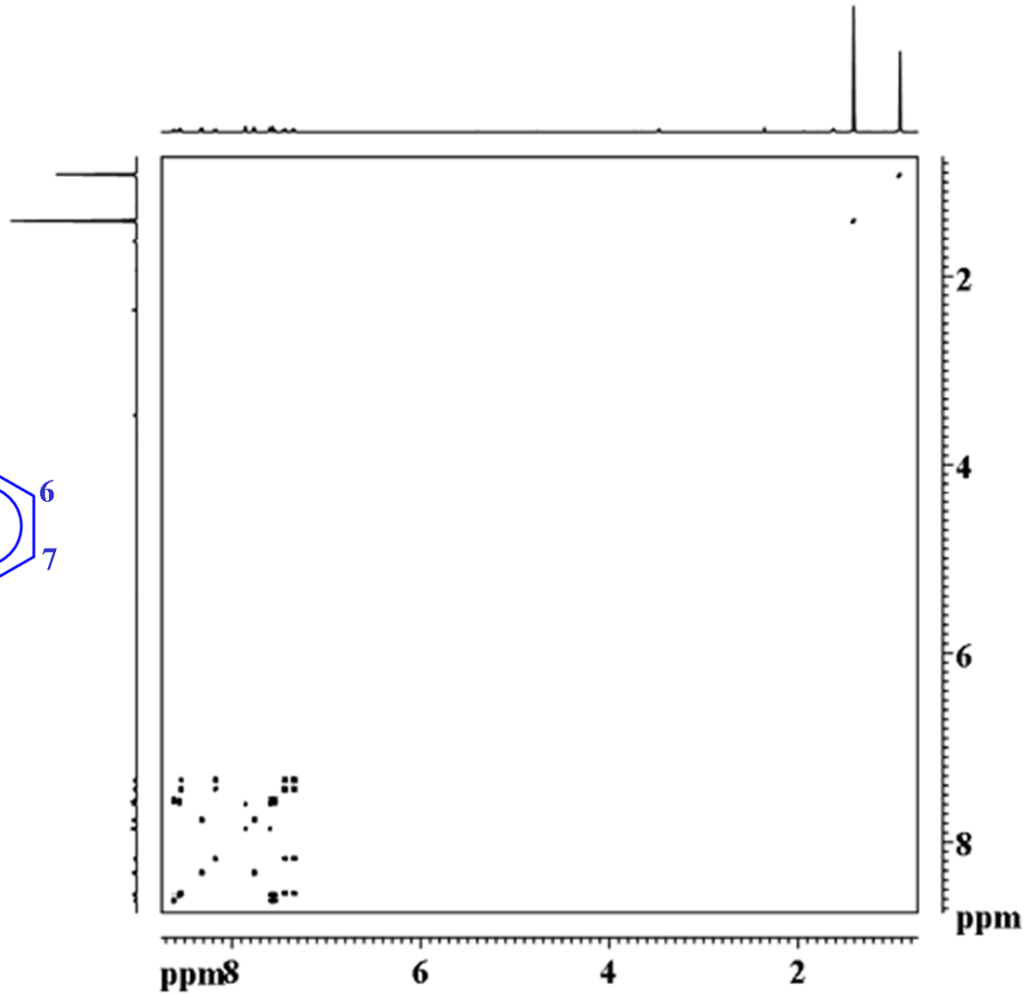
and C=O

Strong signals are 2 & 3 bond, weak signals are 4 bond

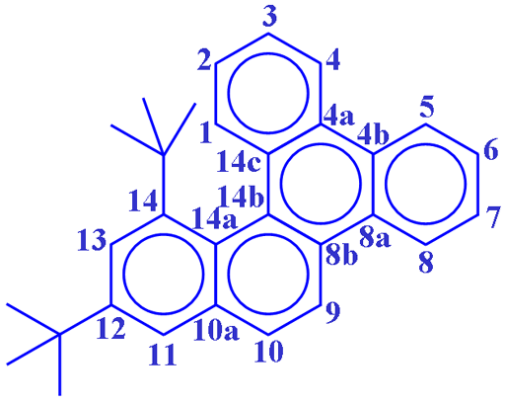


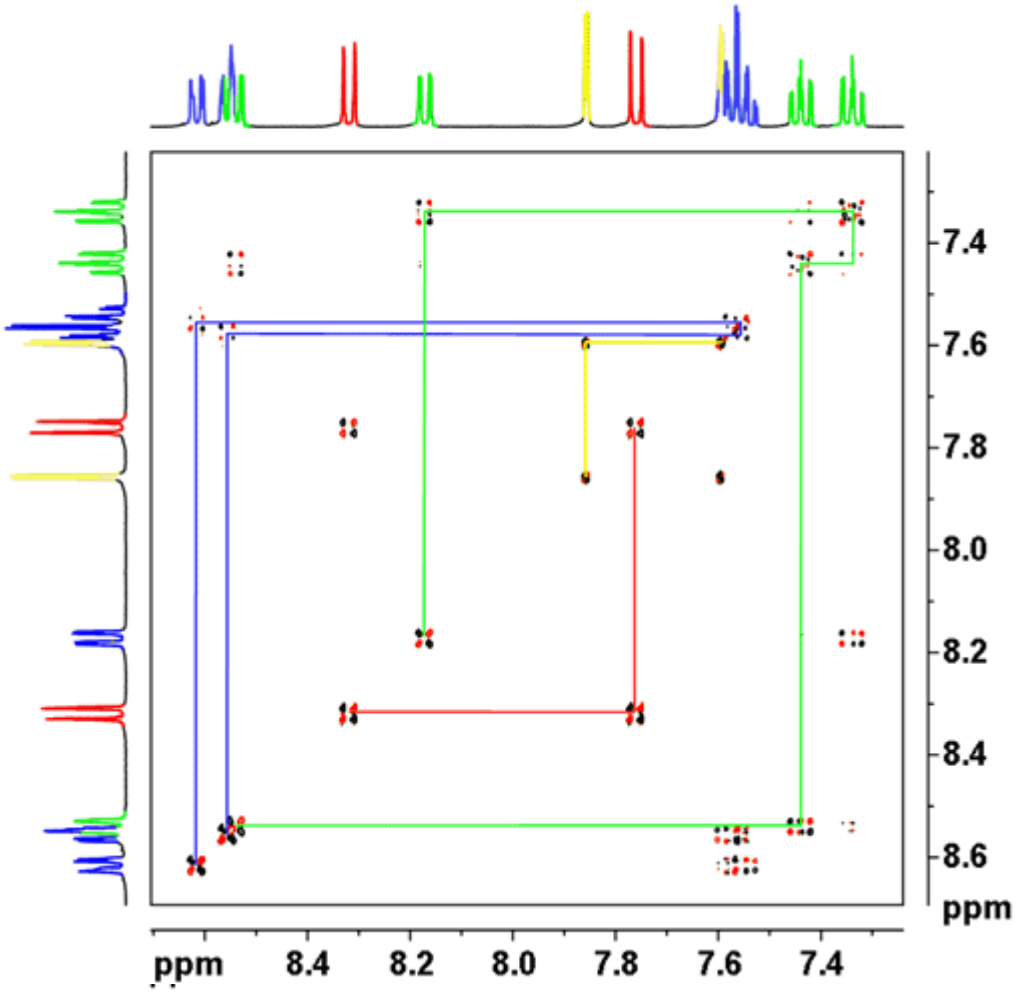
COSY example:12,14-ditbutylbenzo[g]chrysene





Division into coupling networks

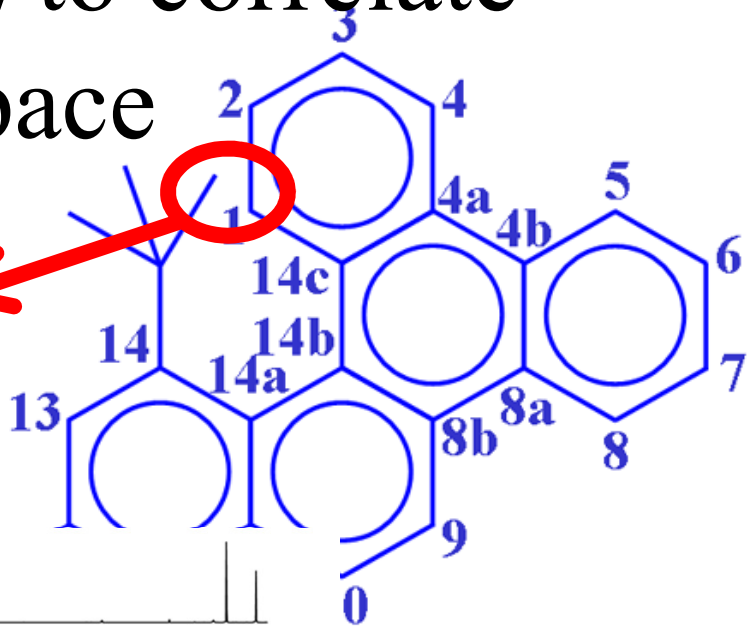




Nuclear Overhauser effect (NOE)

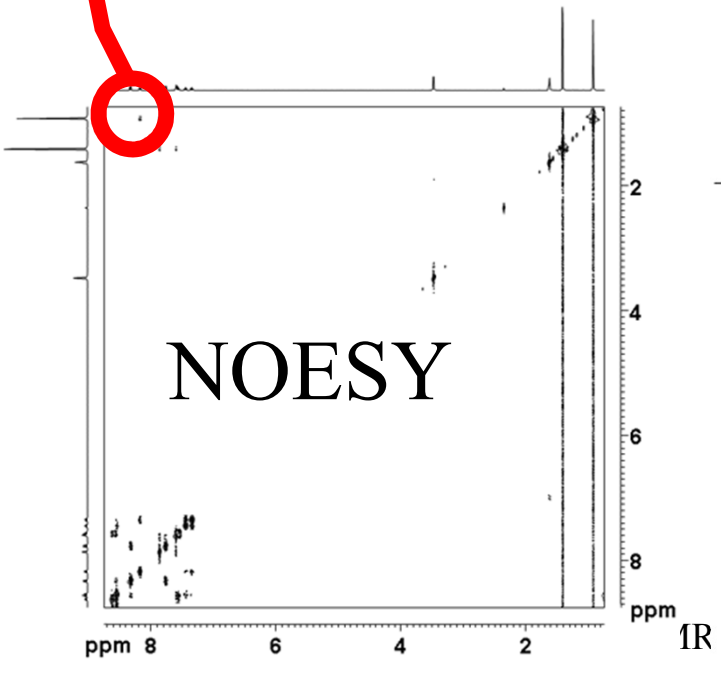
- When a specific nucleus is magnetically excited and its neighbor is at equilibrium, relaxation occurs between the two nuclei.
- The smaller the physical distance between the equilibrium nucleus and the excited nucleus, the bigger the expected change.
- This dependence can be used to estimate the distance between nuclei.
- NOE is used to observe through space correlations in NOESY and ROESY experiments.

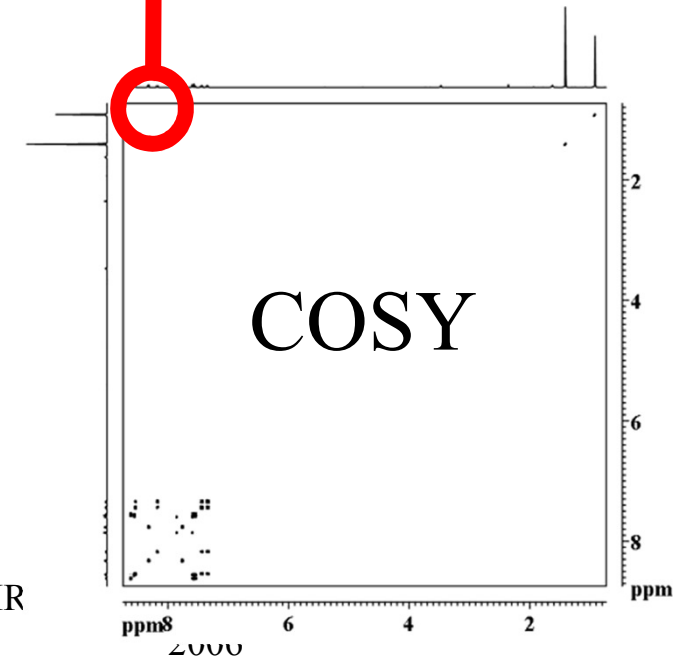
NOESY uses NOE to correlate through space

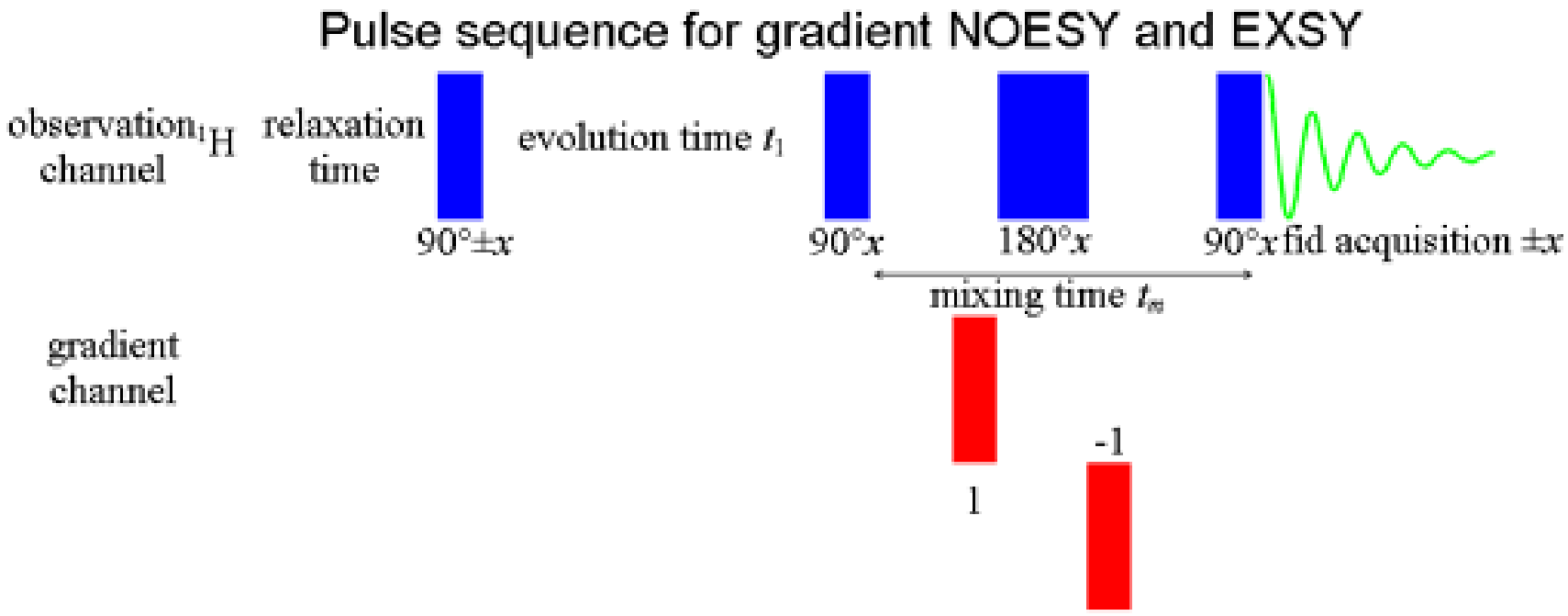


Physically close so strong NOE

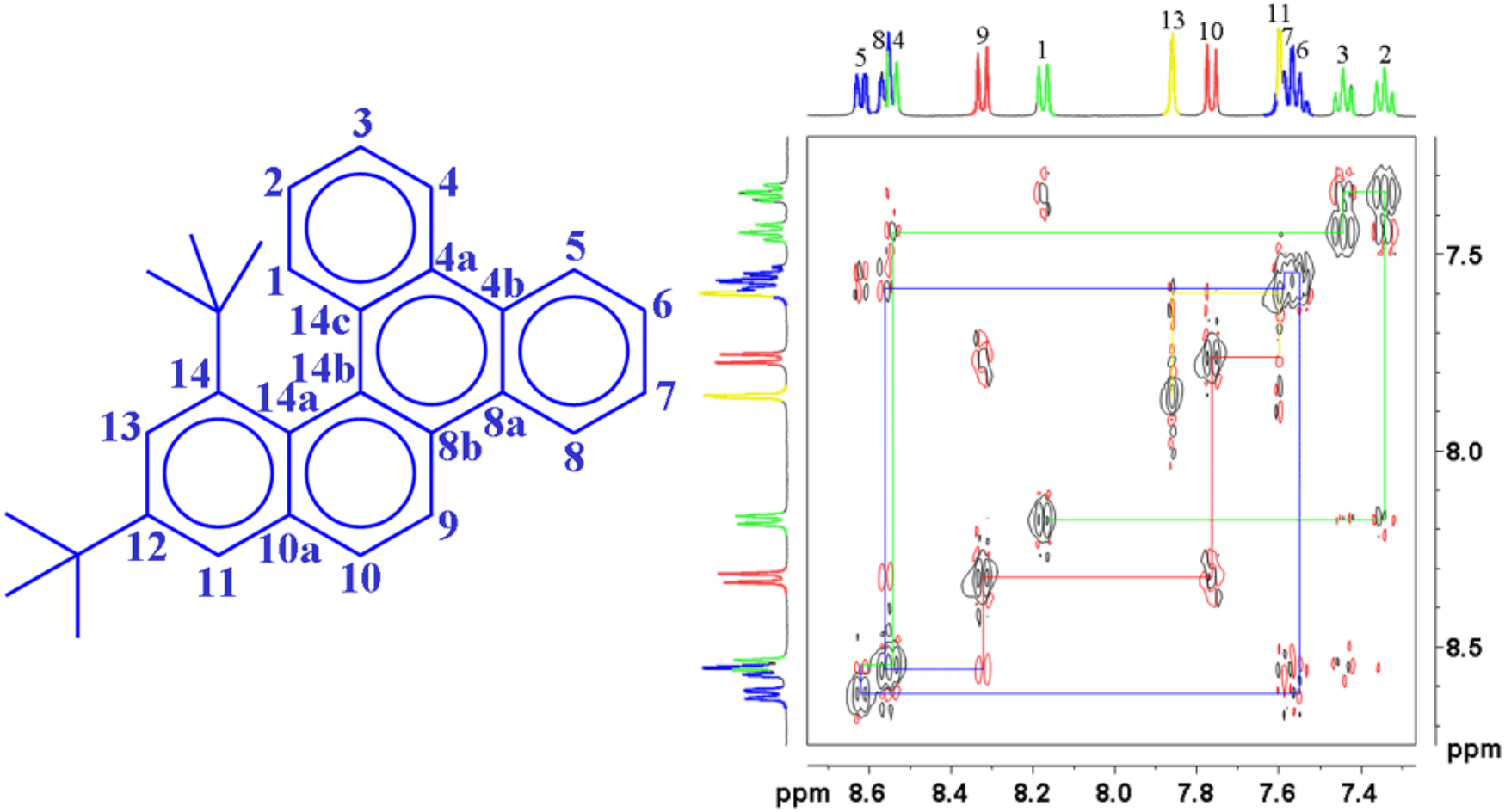
but 6-bonds away so no COSY





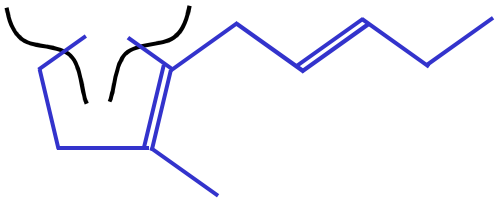

Pulse sequence for gradient NOESY and EXSY
observation ^{1}H channel
relaxation time
evolution time t_1
90°±x
90°x
180°x
90°x
fid acquisition ±x
mixing time t_m
gradient channel
1
-1

Aromaric region of the 2D NOESY spectrum of 12,14-ditbutylbenzo[g]chrysene showing connectivity and separation into four color-coded proton groups

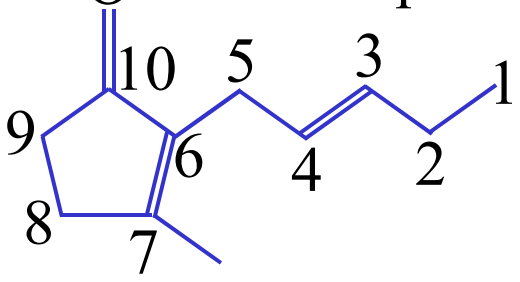


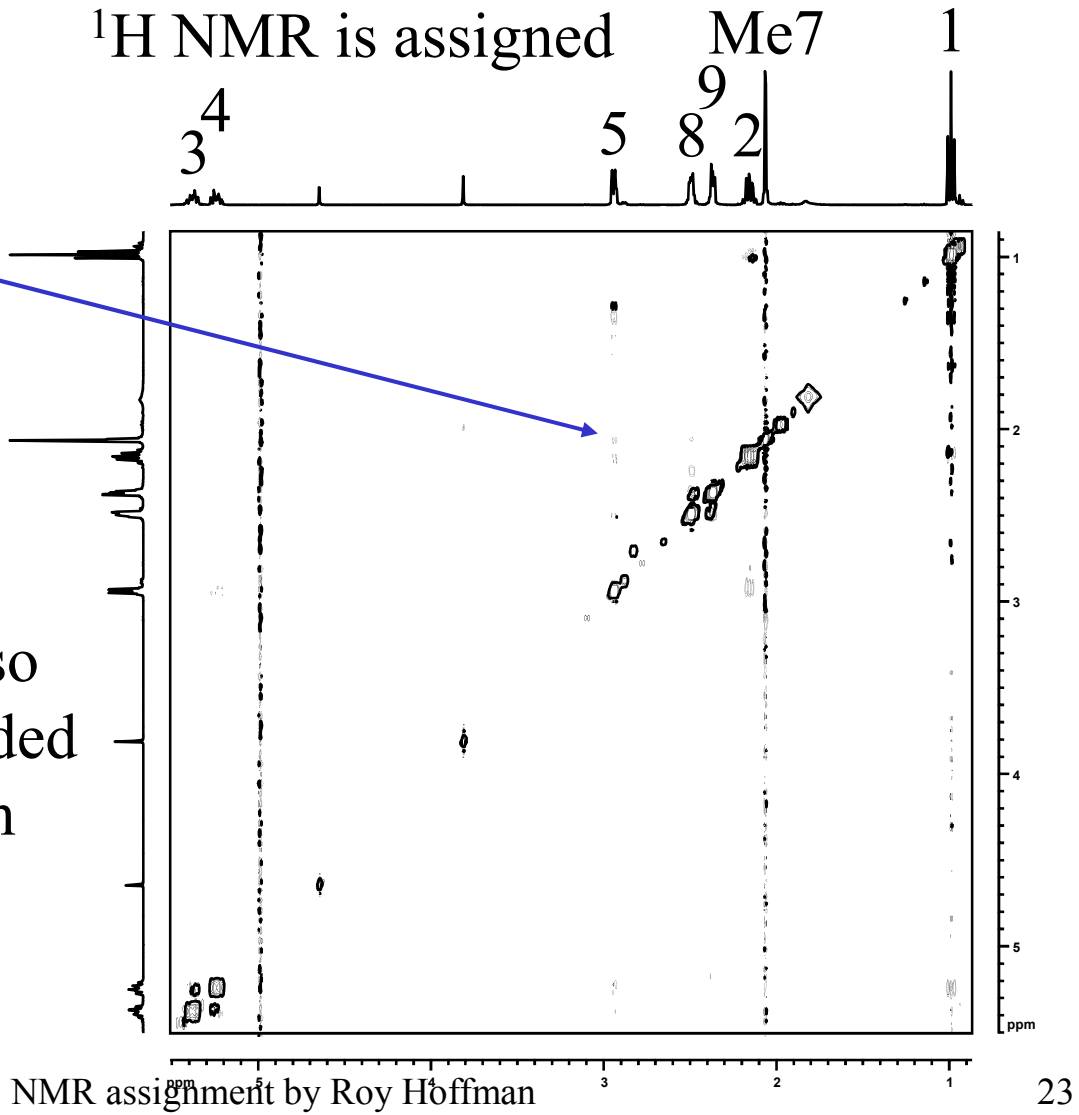
NOESY of the unknown compound

Correlation between the two networks

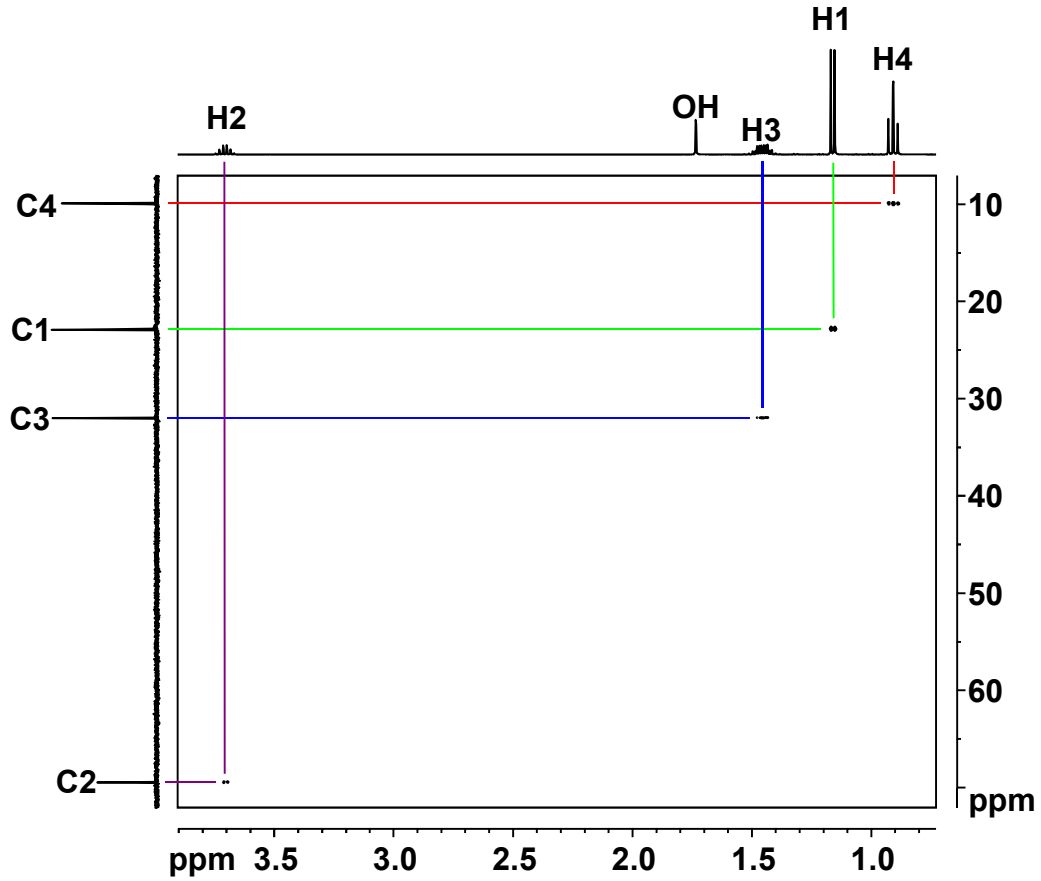


A ring is required so close with C=O needed to fit ^{13}C spectrum

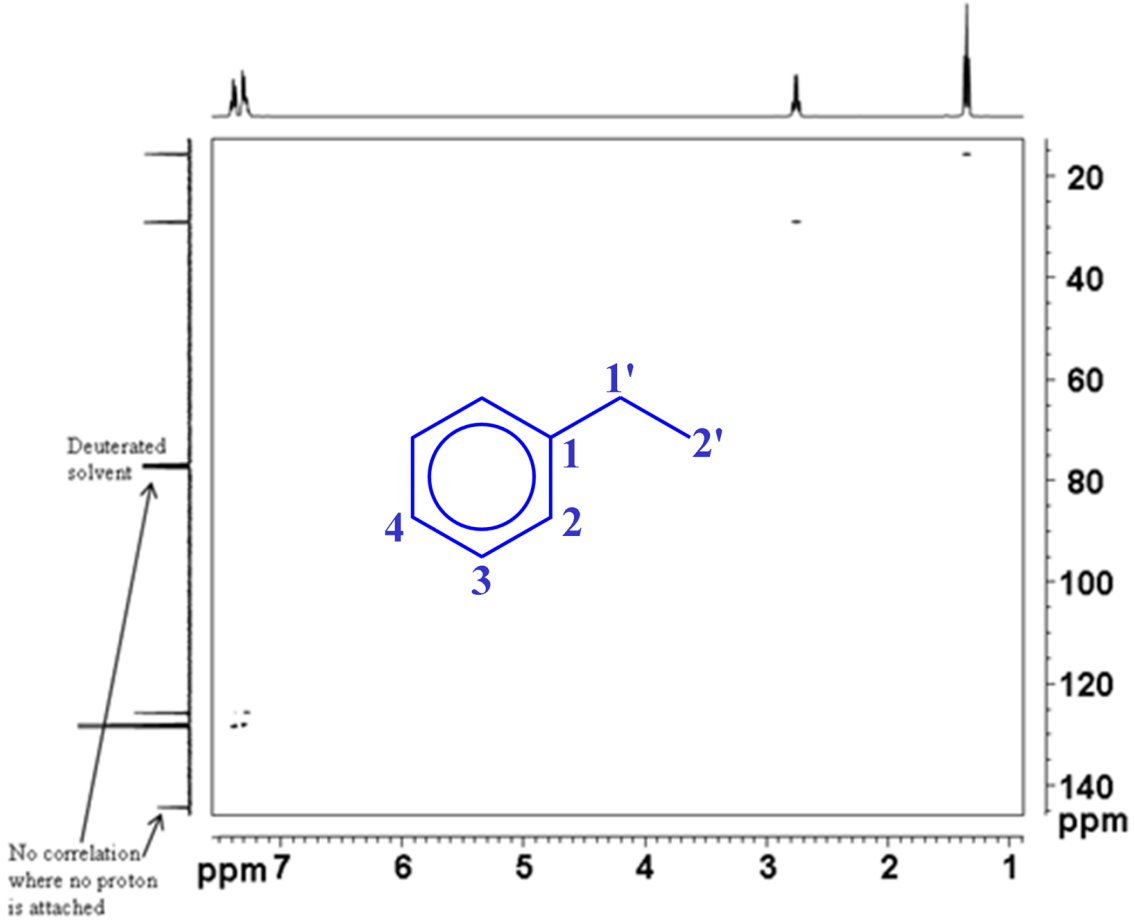


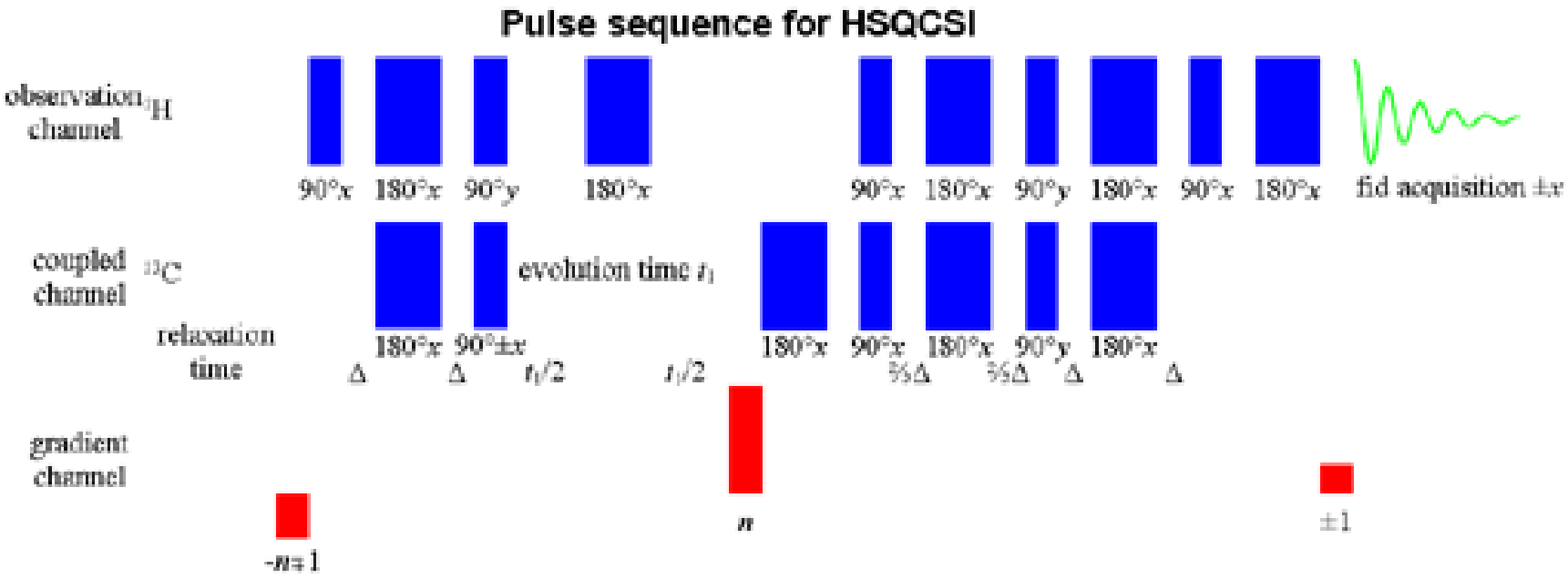


One-bond ^{1}H-^{13}C correlation (HSQCSI) of butan-2-ol



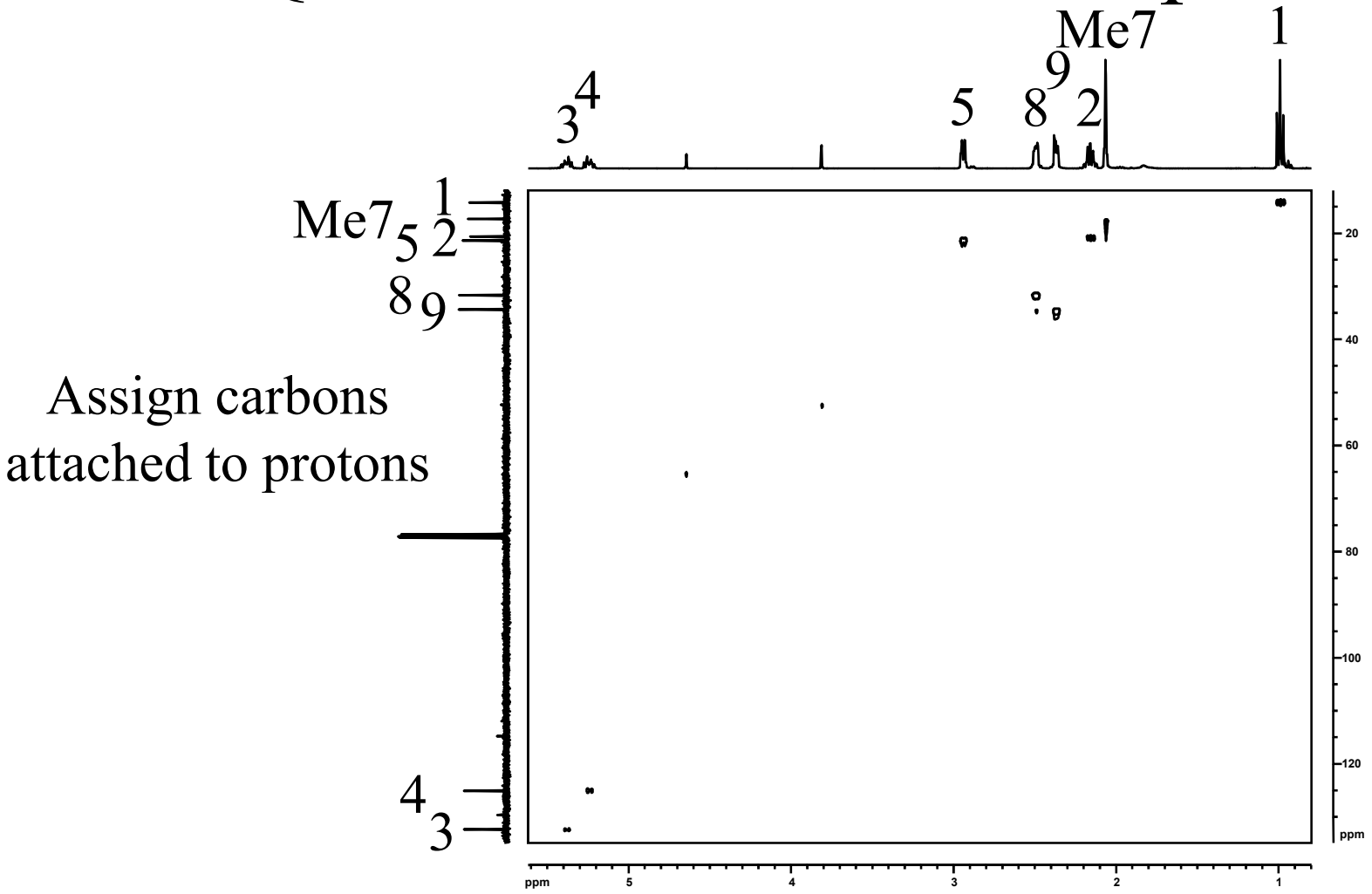
One-bond ^{1}H-^{13}C correlation (HSQCSI) of ethylbenzene




Pulse sequence for HSQCSI
observation ^{1}H channel
90°x 180°x 90°y 180°x 90°x 180°x 90°y 180°x 90°x 180°x fid acquisition ±x
coupled ^{13}C channel
evolution time t_1
relaxation time
180°x 90°±x 180°x 90°x 180°x 90°y 180°x
Δ Δ t_1/2 t_1/2 ⅗Δ ⅗Δ Δ Δ
gradient channel
-n+1
n
±1

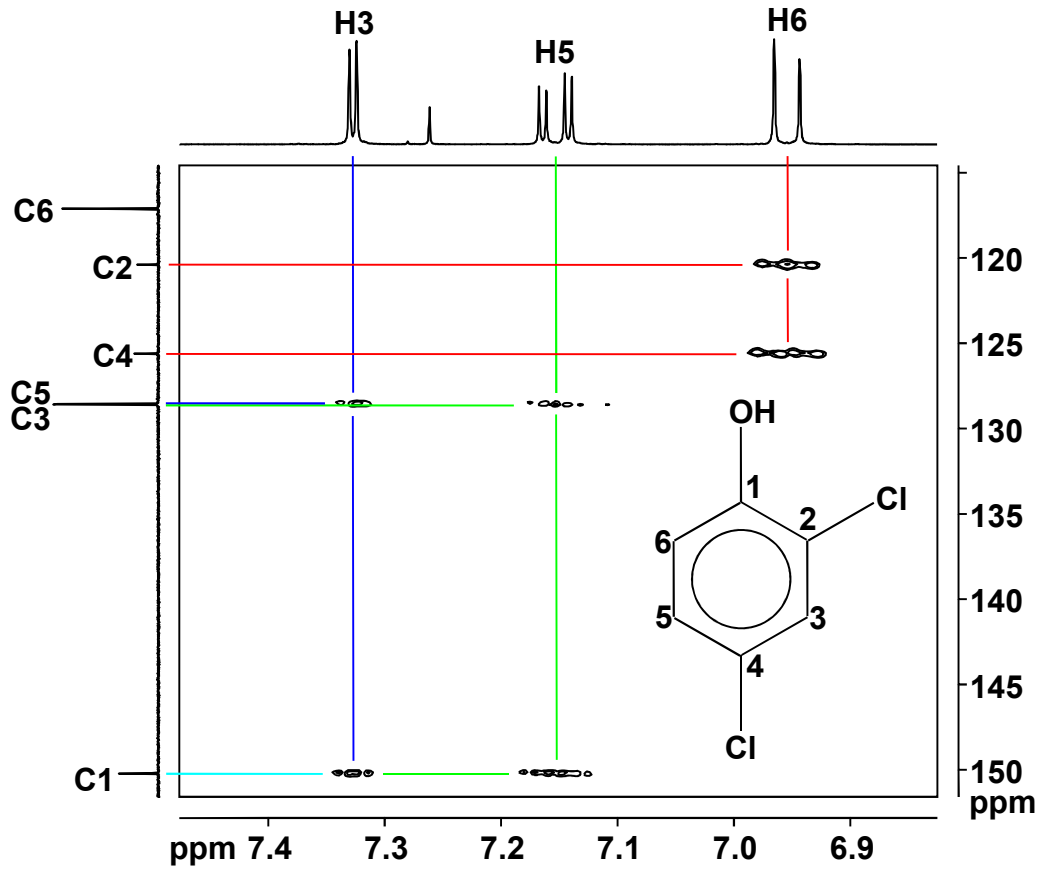
$n = 3.98$

HSQC of the unknown compound



Assign carbons attached to protons

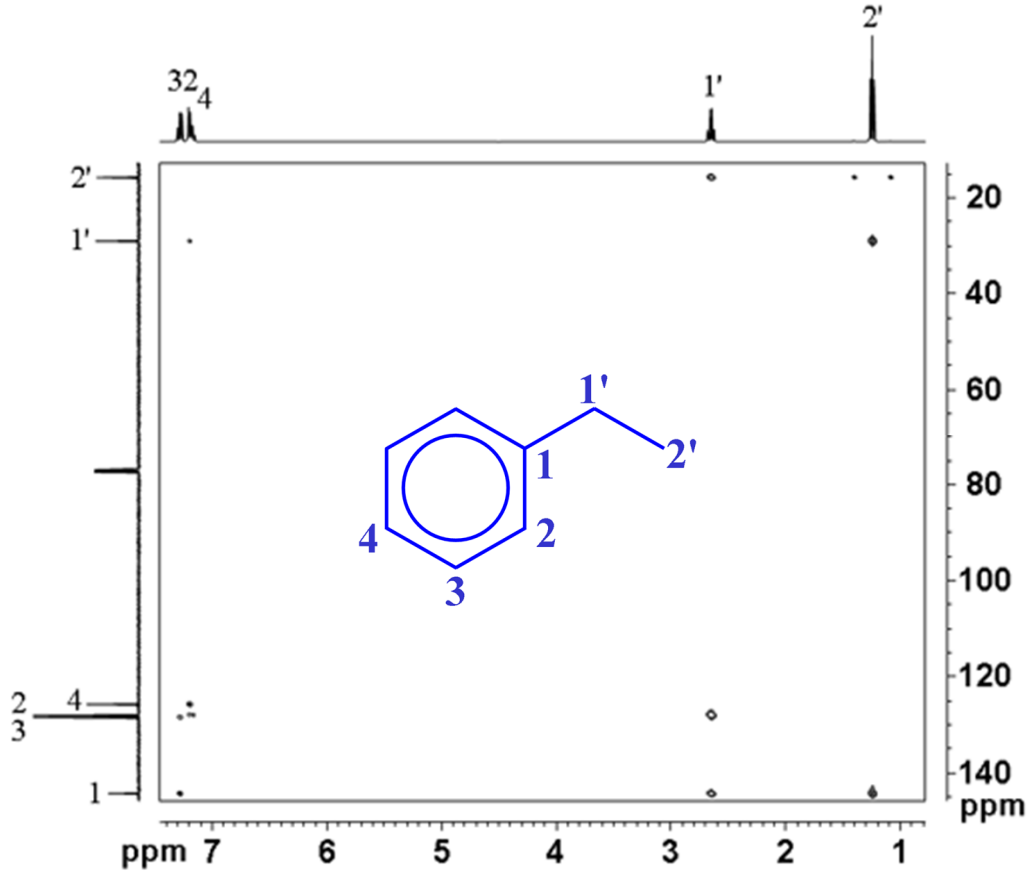
Multi-bond ^{1}H-^{13}C correlation (HMBC) of 2,4-dichlorophenol



Correlation table

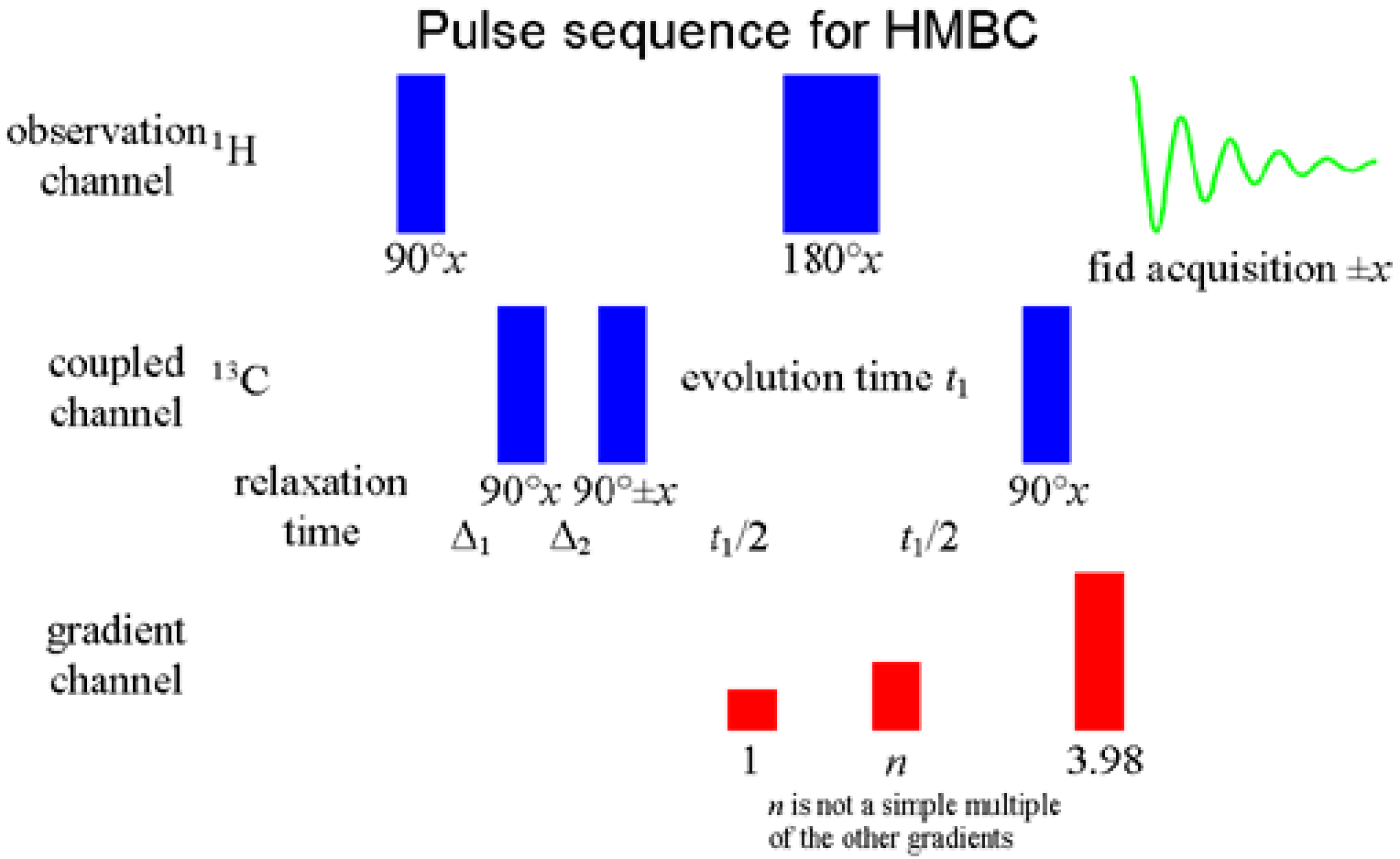
C\H	3	5	6
6			
2			3
4			3
5	3		
3		3	
1	3	3	

Multi-bond ^{1}H-^{13}C correlation (HMBC) of ethylbenzene



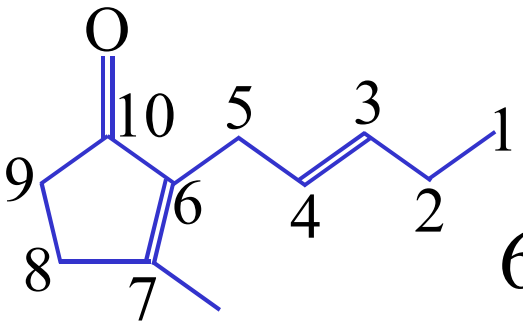
Correlation table

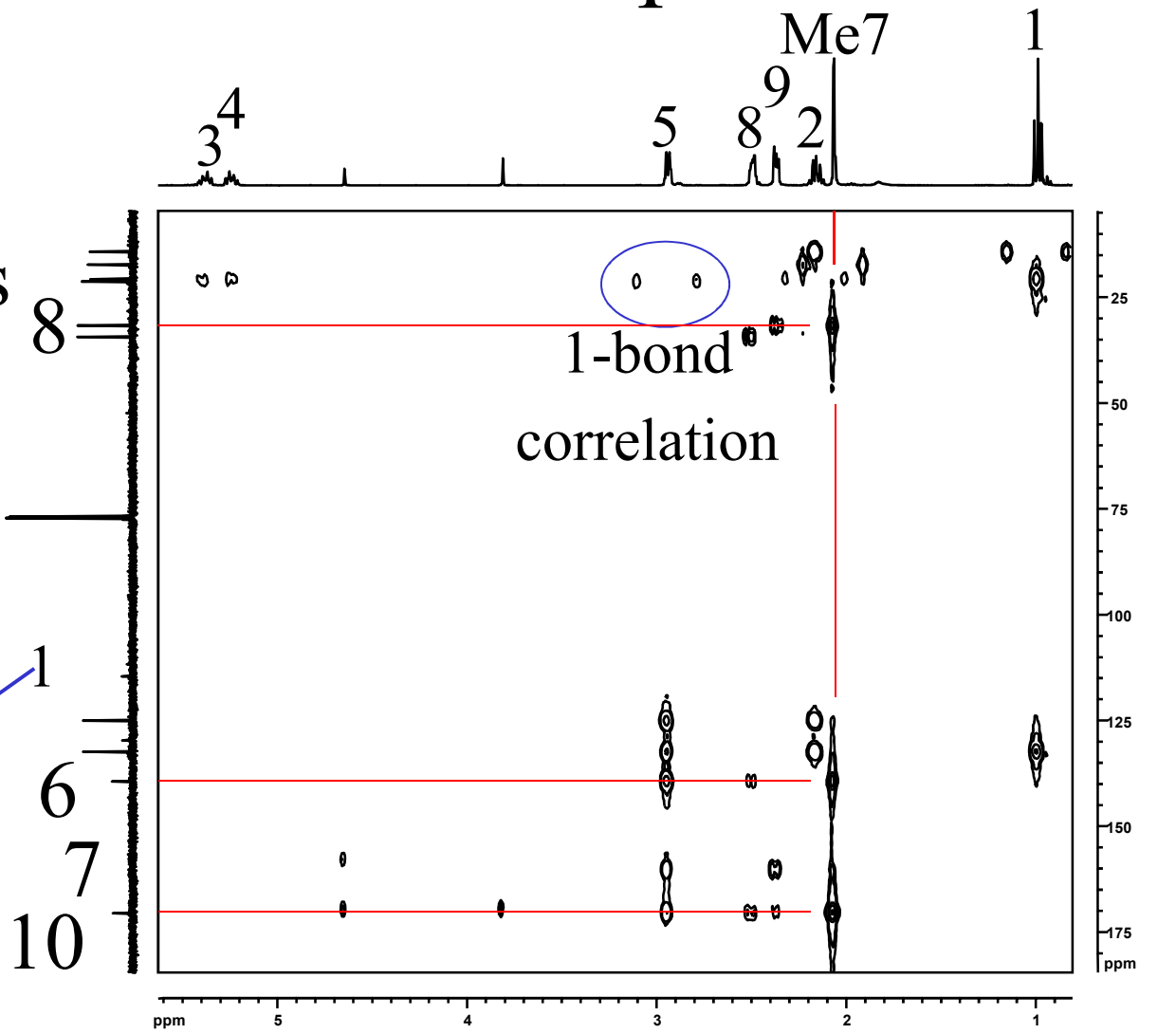
C\H	1'	2'	2	3	4
1'		2			
2'	2	1			
1	2	3		3	
2	3		3		3
3				3	
4			3		


Pulse sequence for HMBC
observation channel ^{1}H
90°x
180°x
fid acquisition ±x
coupled channel ^{13}C
evolution time t_1
relaxation time
90°x
90°±x
90°x
Δ_1
Δ_2
$t_1/2$
$t_1/2$
gradient channel
1
n
3.98
n is not a simple multiple of the other gradients

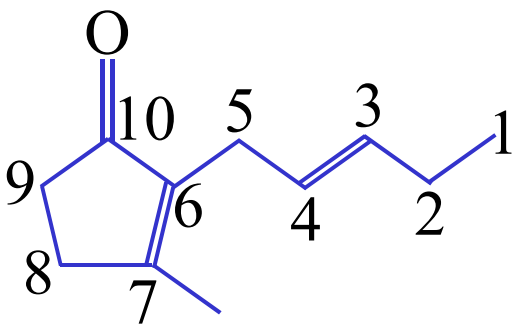
HMBC to assign other carbons of the unknown compound

- 3-bond correlations strongest



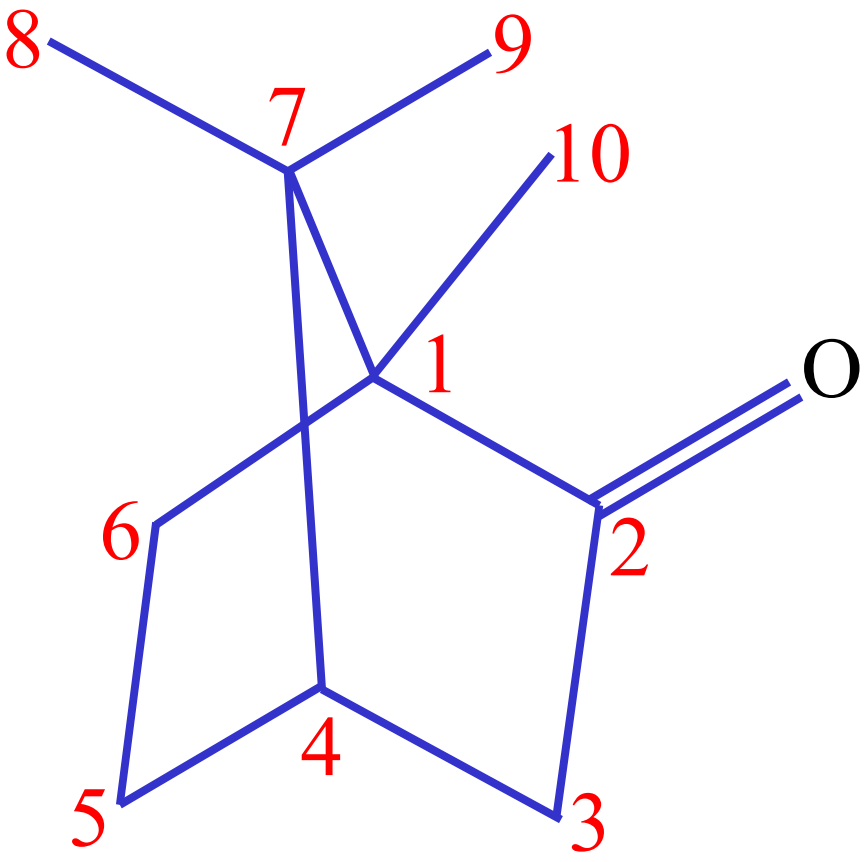


Use correlation table

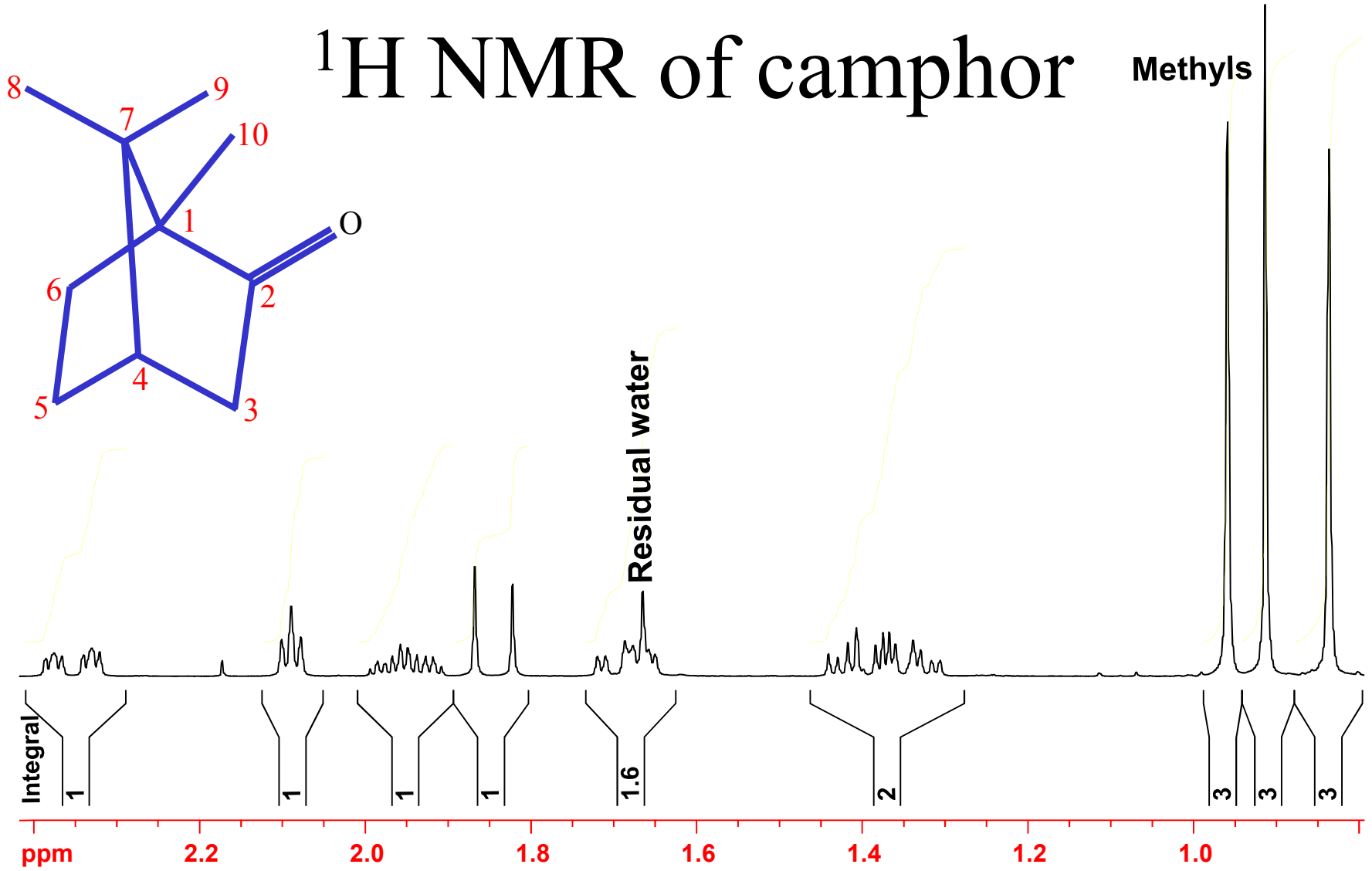


	H1	H2	H3	H4	H5	Me7	H8	H9
C1		2						
C2	2			3				
C3	3	2			3			
C4		3			2			
C5			3					
C6					2	3	3	
C7					3			3
Me7								
C8						3		2
C9							2	
C10					3	4	3	

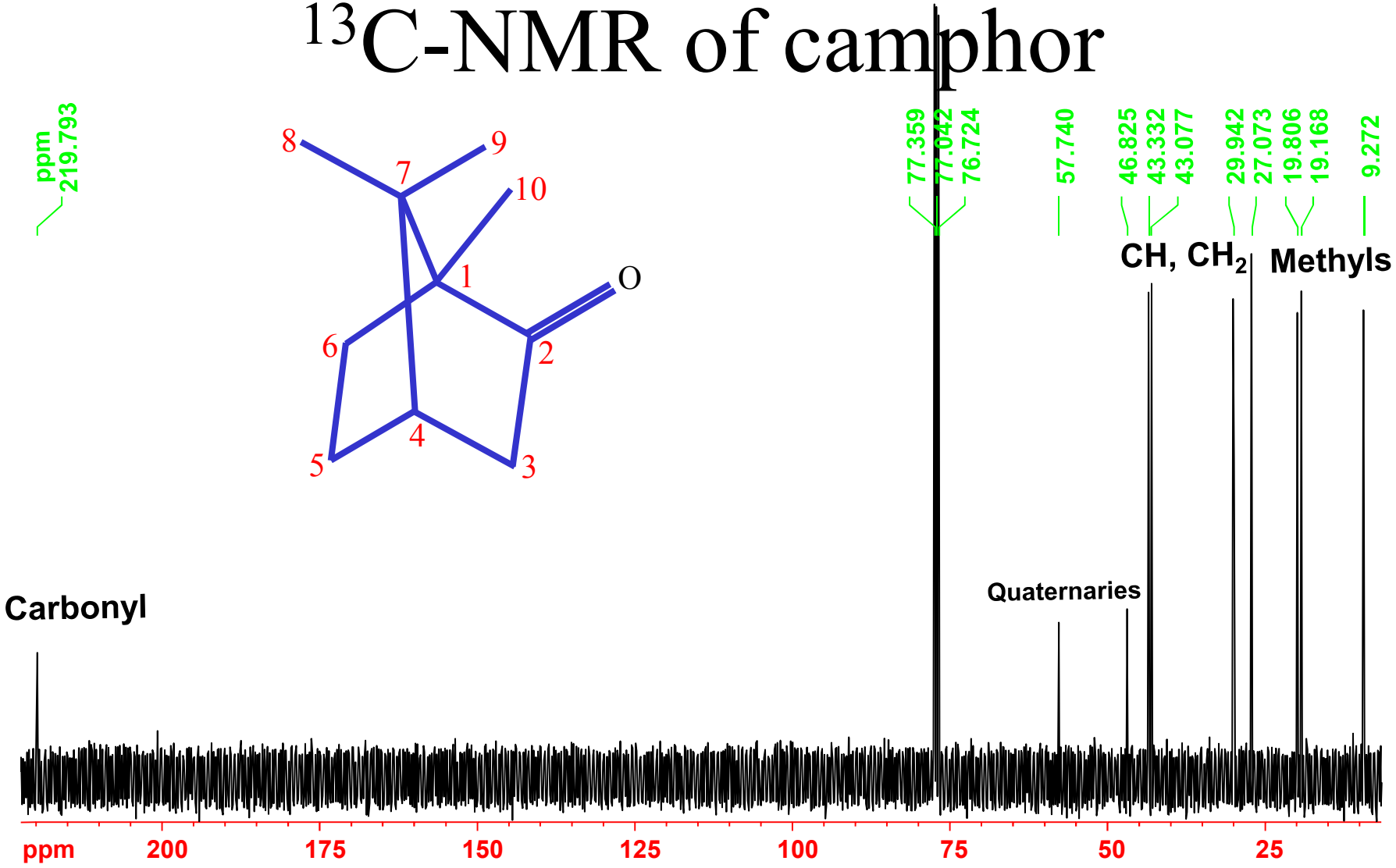
Assignment example camphor



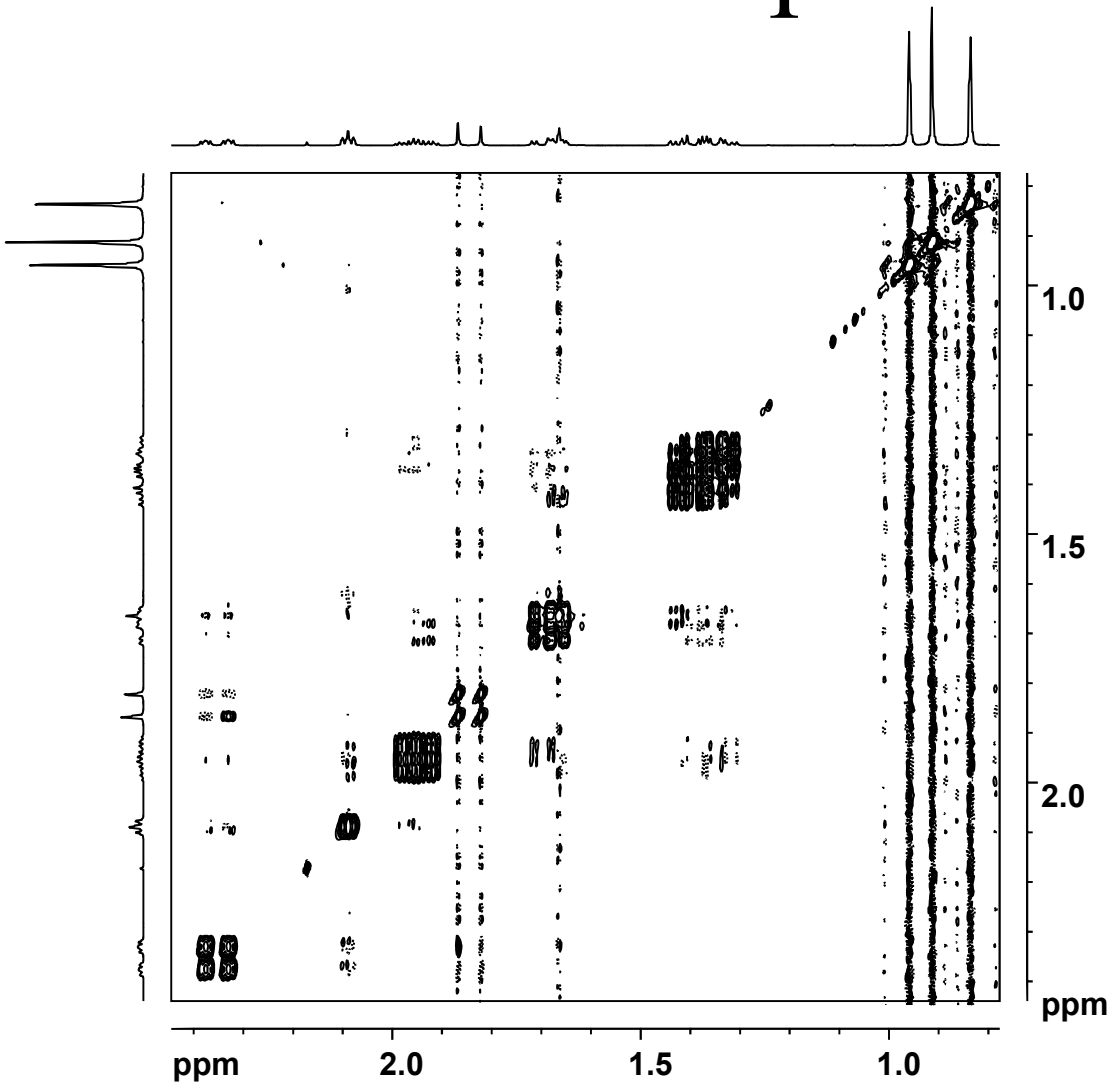
^{1}H NMR of camphor



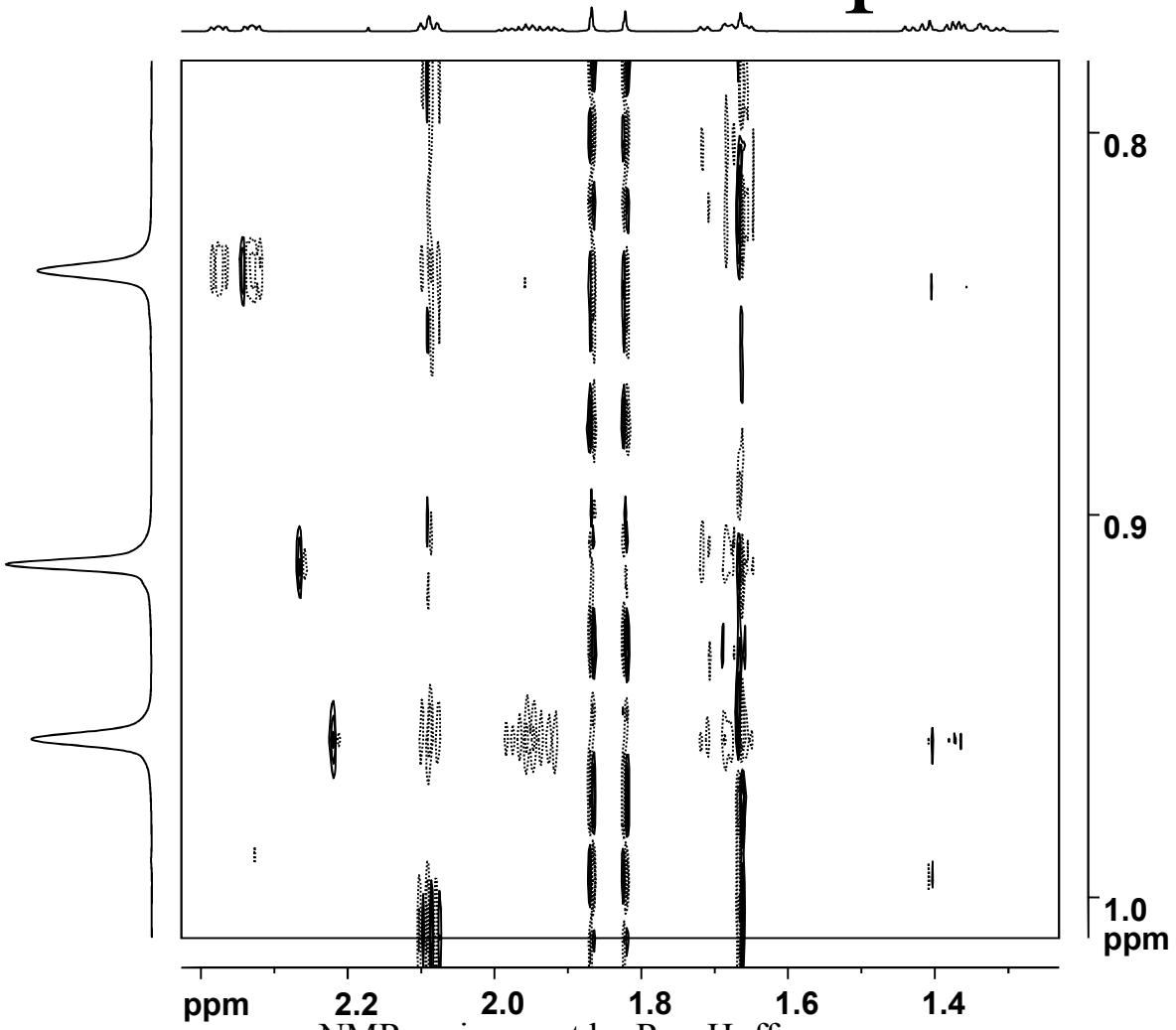
^{13}C-NMR of camphor



NOESY of camphor

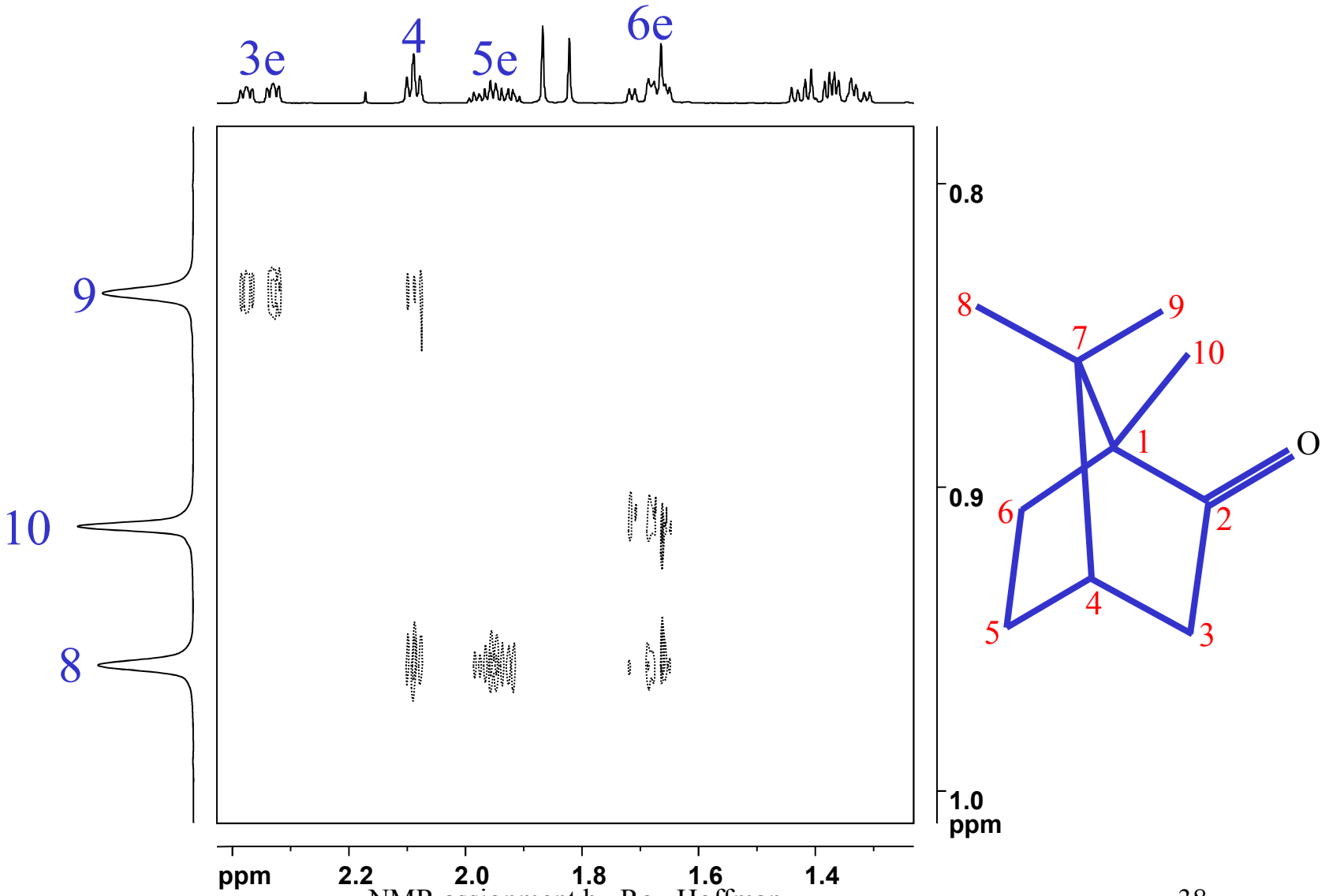


NOESY of camphor



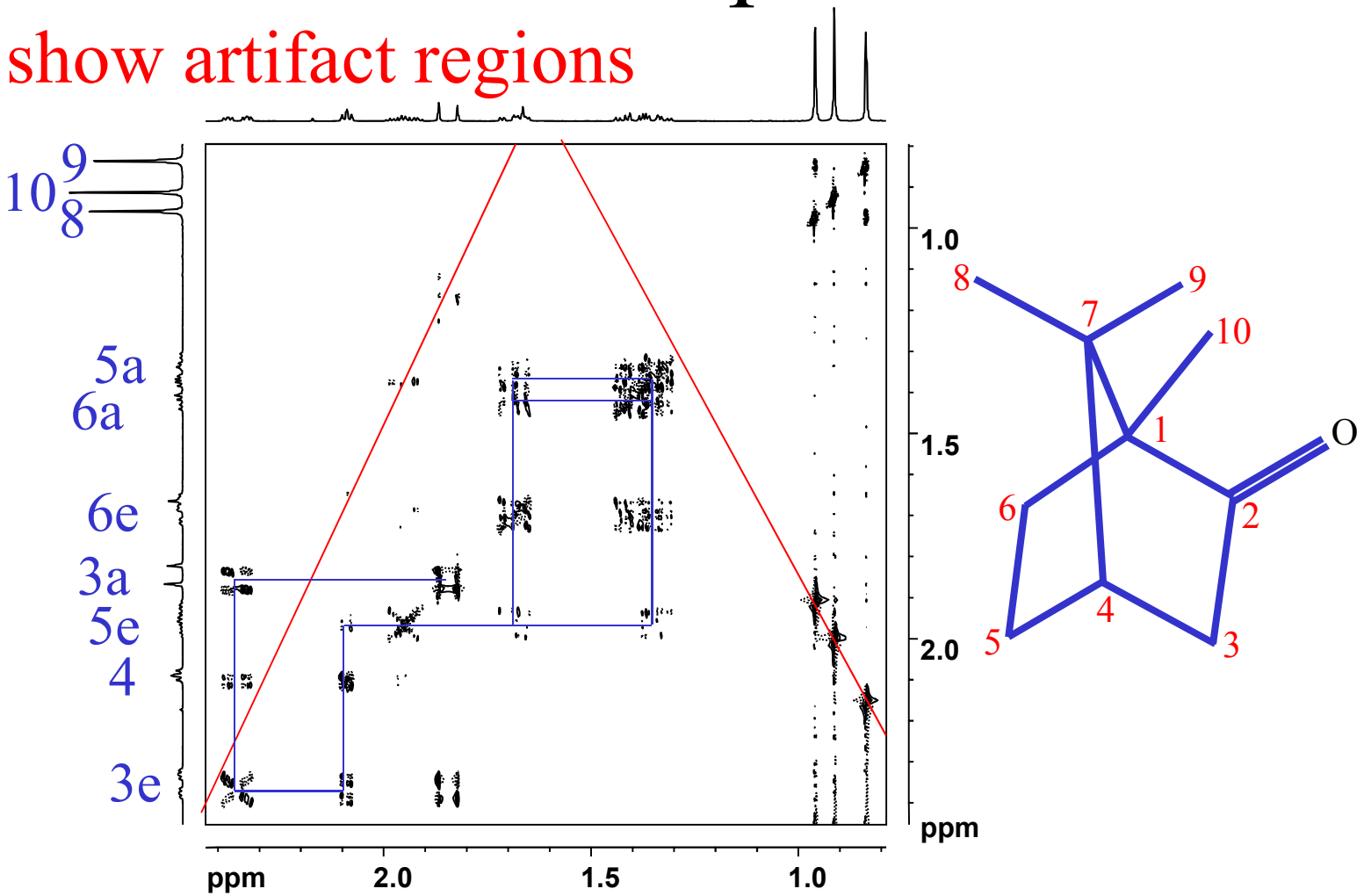
NMR assignment by Roy Hoffman
2006

NOESY of camphor

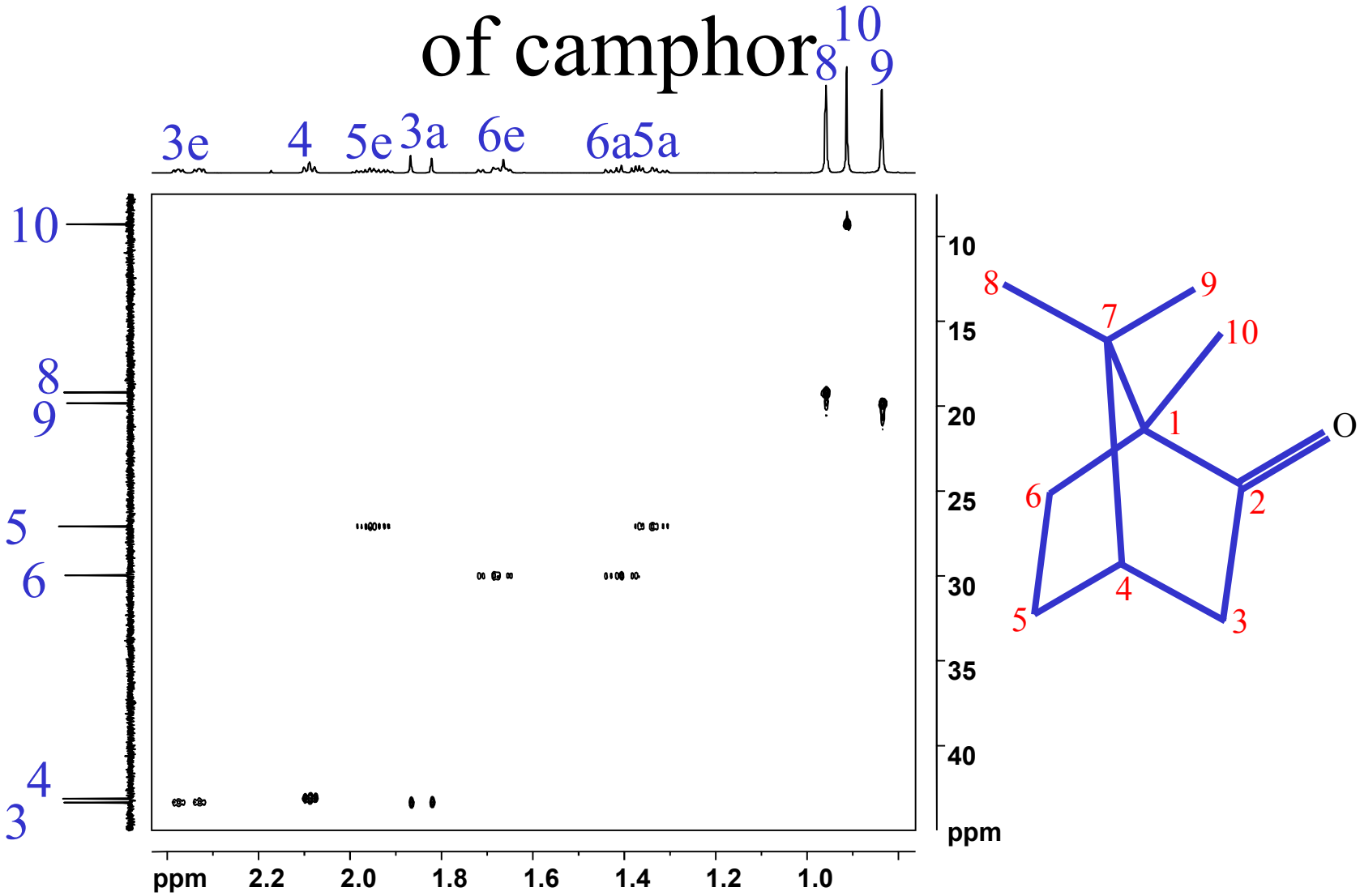


COSY of camphor

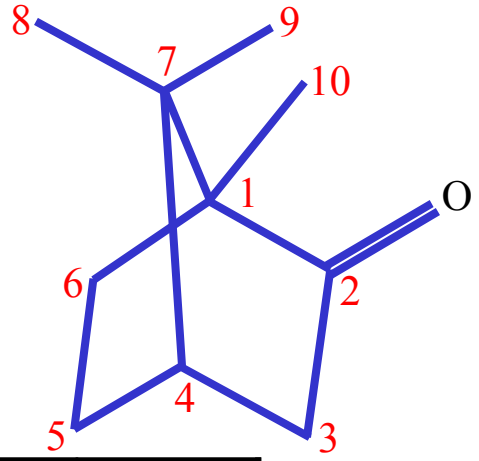
Red lines show artifact regions



One-bond ^{1}H-^{13}C correlation HSQC of camphor

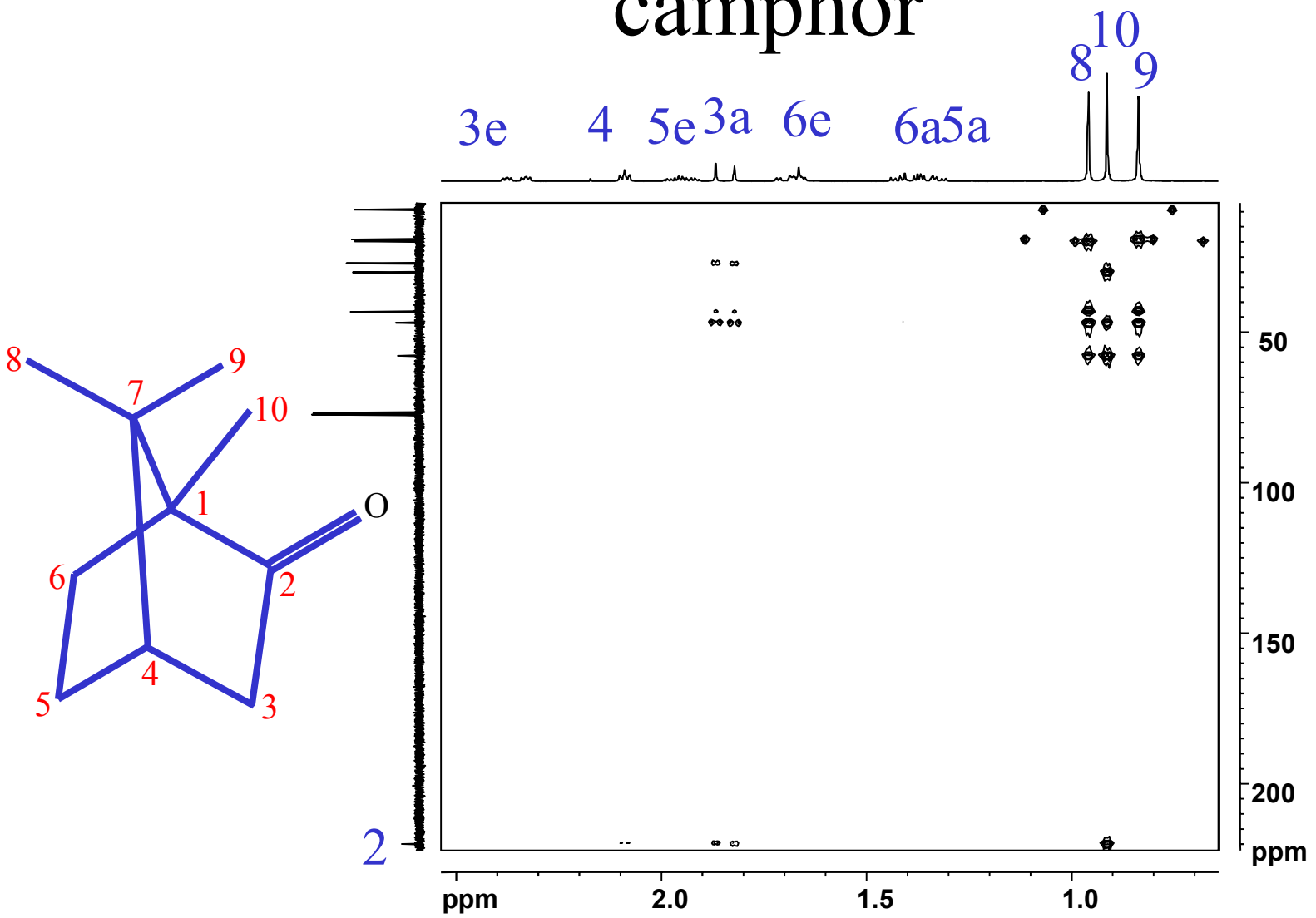


Multi-bond correlation HMBC of camphor

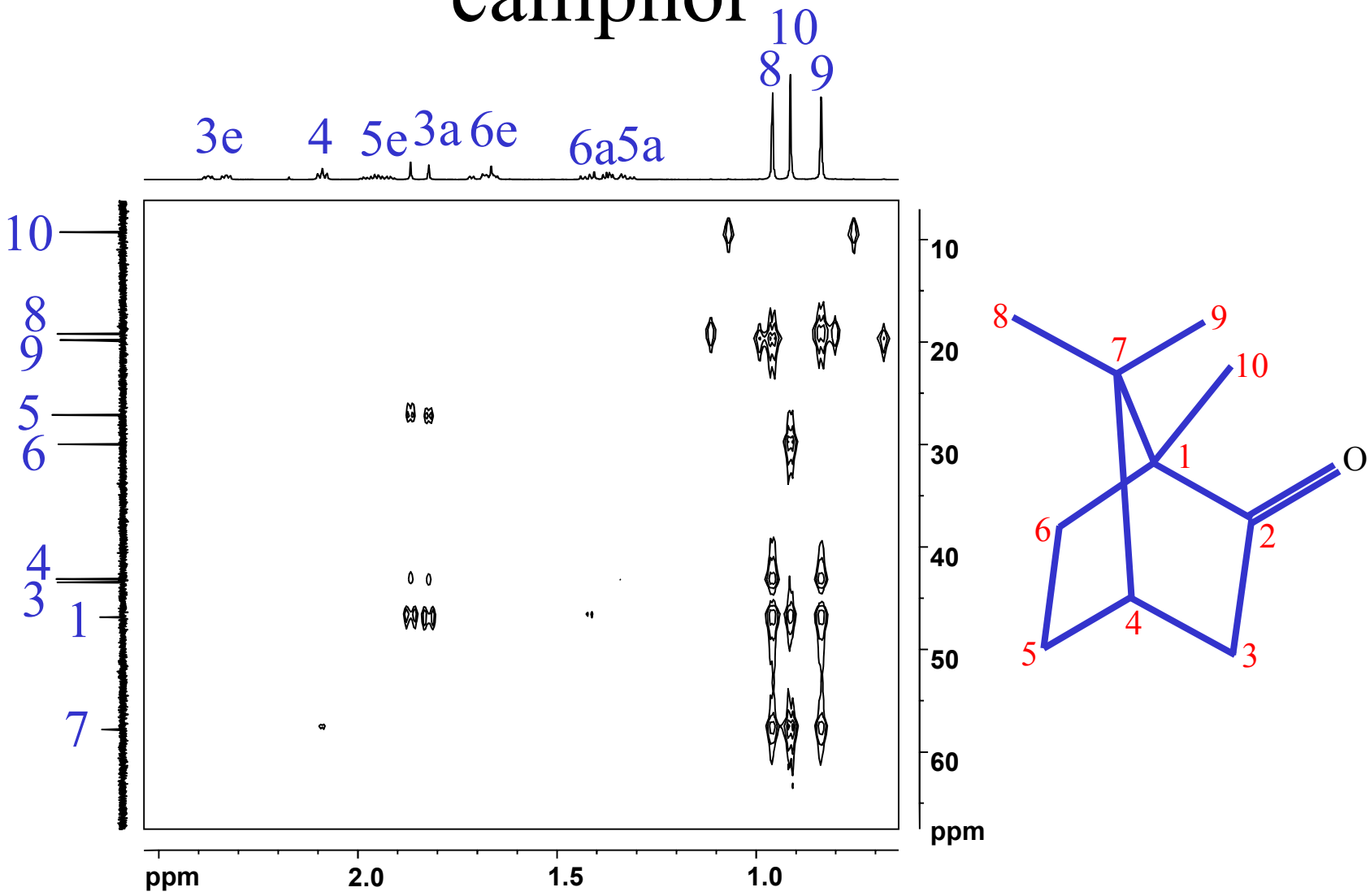


	H3	H4	H5	H6	H8	H9	H10
C1	3			2	3	3	2
C2							
C3							
C4	2				3	3	
C5	3						
C6							3
C7		2			2	2	3
C8					1	3	
C9					3	1	
C10							1

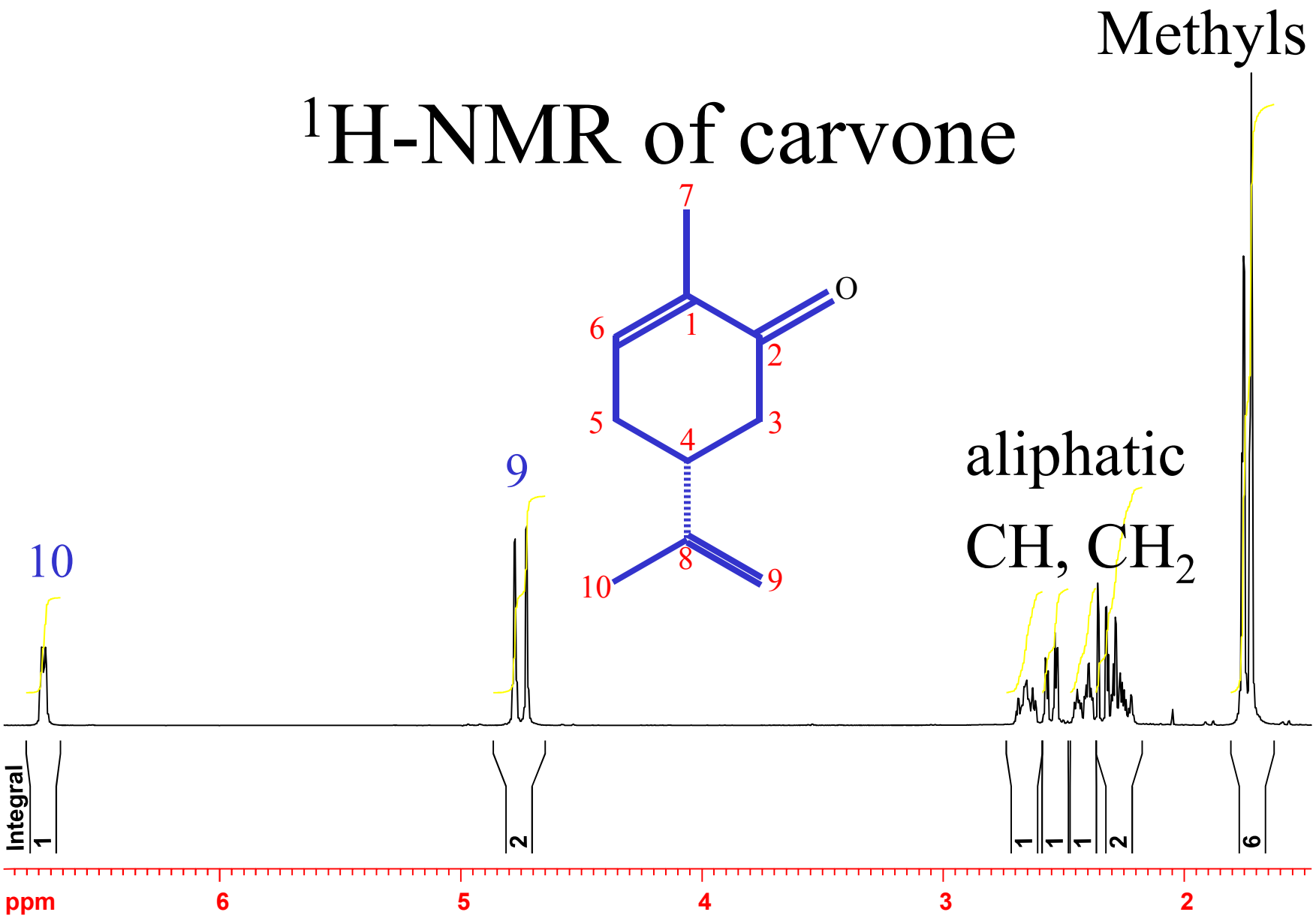
Multi-bond correlation HMBC of camphor



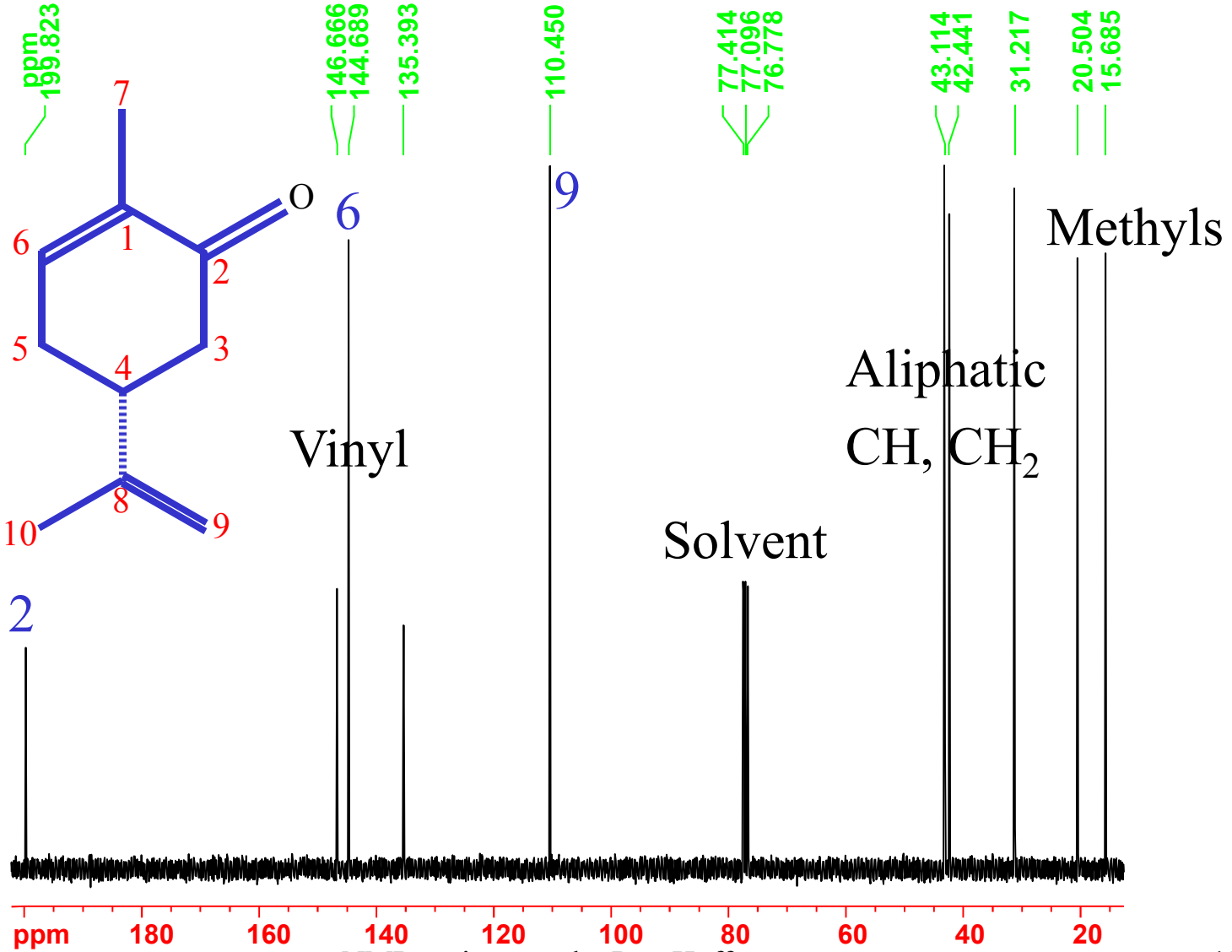
Multi-bond correlation HMBC of camphor



^{1}H-NMR of carvone

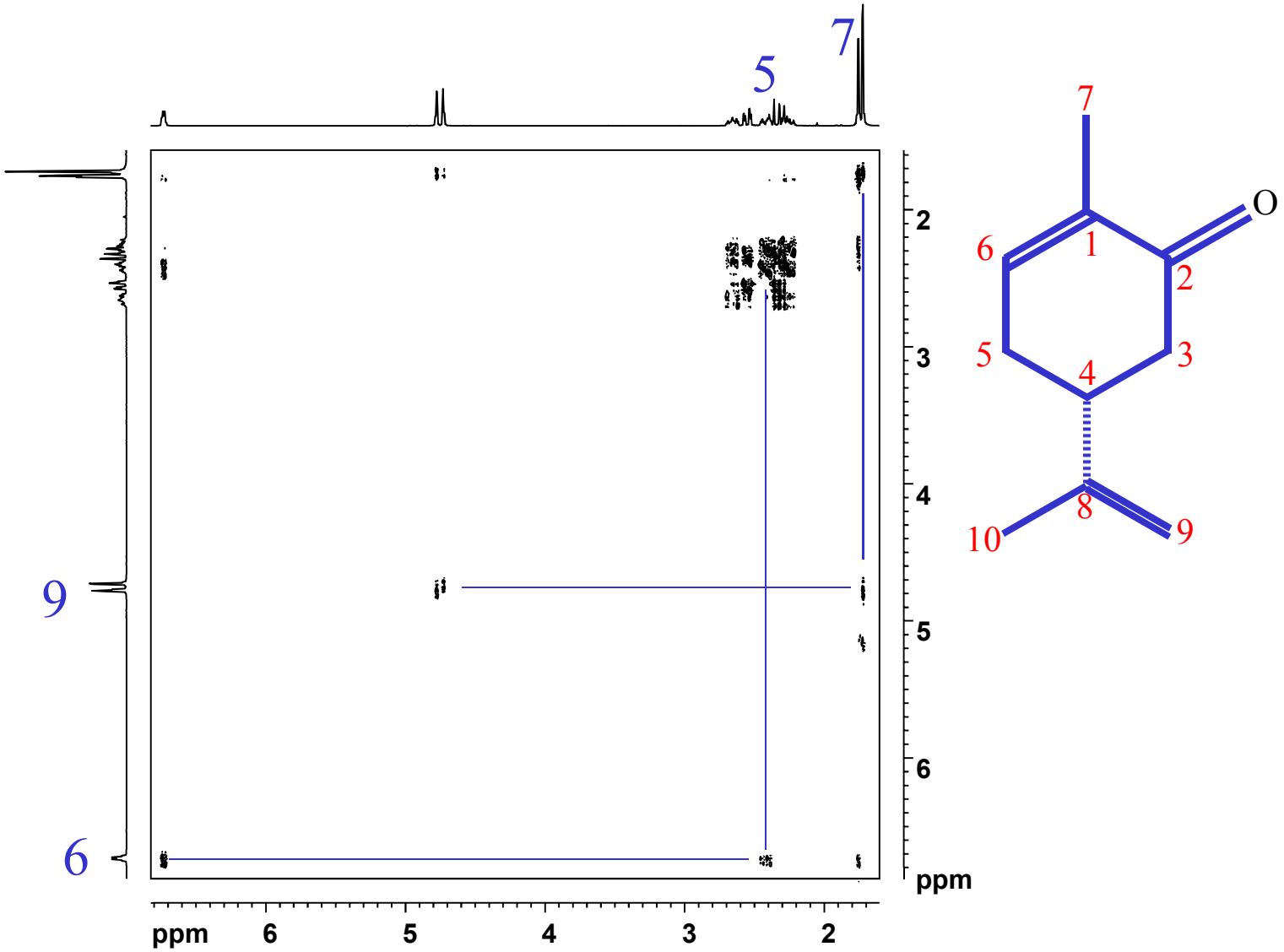


^{13}C-NMR of carvone

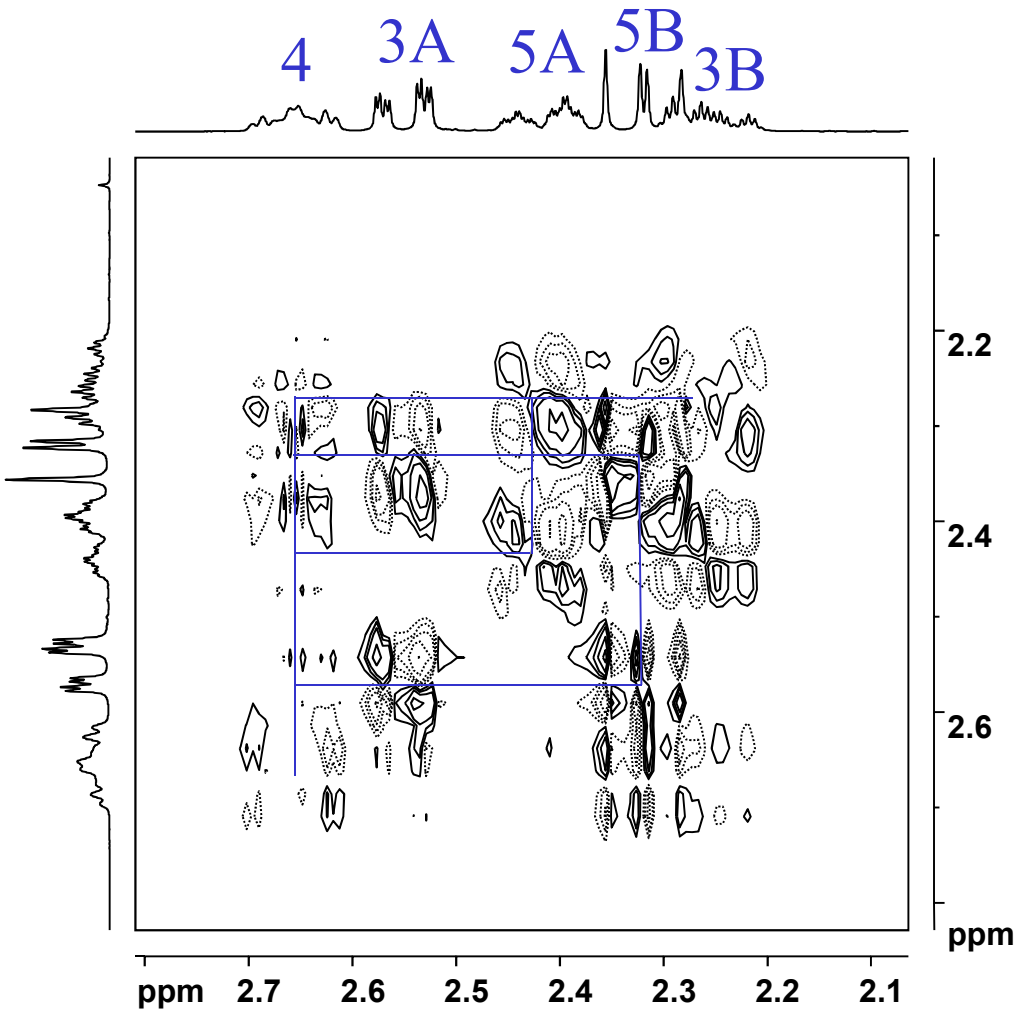


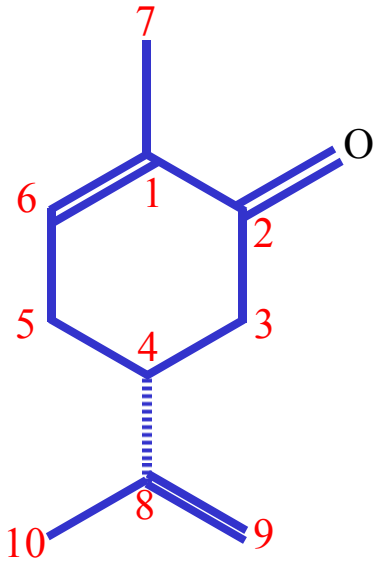
NMR assignment by Roy Hoffman
2006

COSY of carvone 10

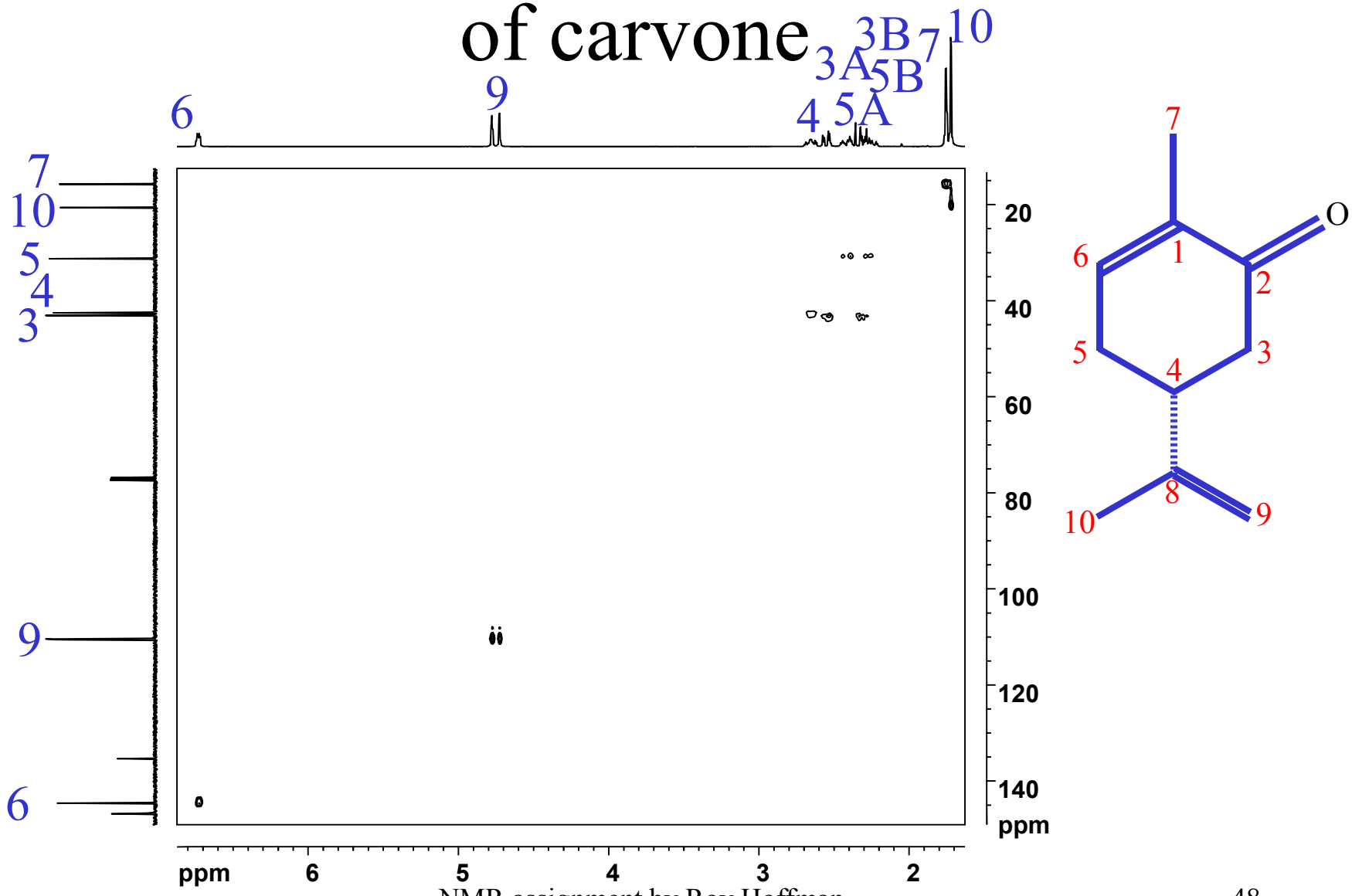


COSY of carvone

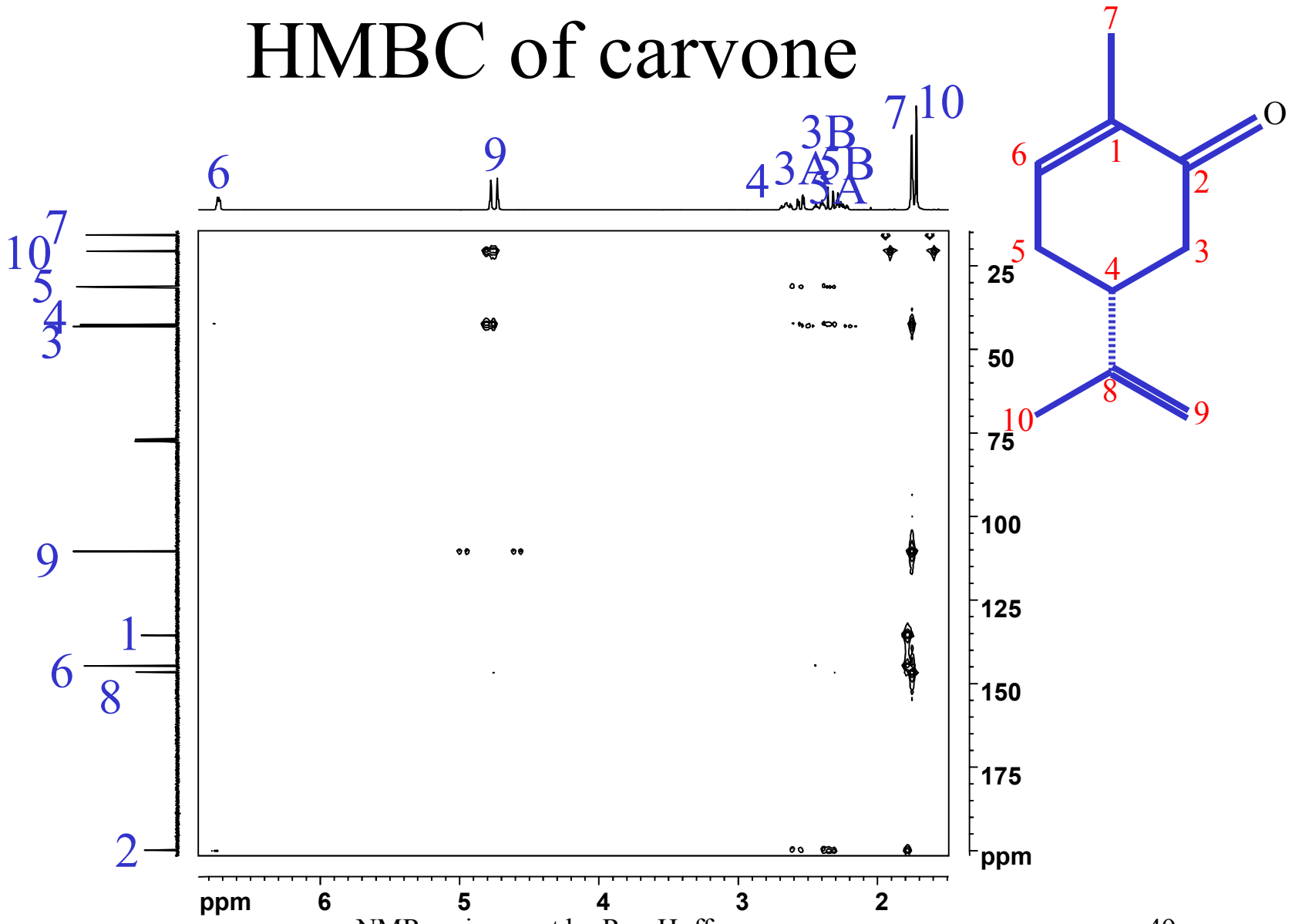




One-bond ^{1}H-^{13}C correlation HSQC of carvone



Multi-bond ^{1}H-^{13}C correlation
HMBC of carvone



Multi-bond correlation HMBC of carvone

	H3	H4	H5	H6	H7	H9	H10
C1					2		
C2	2			3	3		
C3			3				
C4						3	3
C5	3						
C6							
C7					1		
C8							2
C9						1	3
C10						3	1