Ciências ULisboa | CQB Centro de Química e Bioquímica

U LISBOA | UNIVERSIDADE DE LISBOA

Mestrado em Química, Métodos Avançados de Análise 2017/2018

Bloco de Ressonância Magnética Nuclear

Parte 2 – 14 de Novembro 2017

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Gab 8.5.49, Lab 8.5.55

Why performed ^{13}C NMR?

if ^{13}C carbon thirteen is 6400 times less sensitive than Proton ^{1}H

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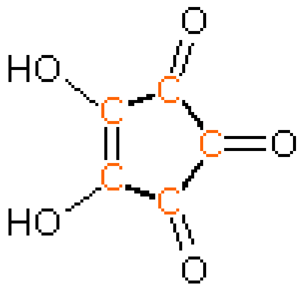
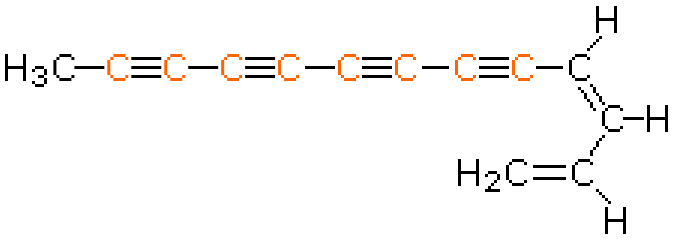
More samples amounts
More time experience
(more number of scans -ns ⇔ more FIDs)

Why to perform ^{13}C NMR?

- When significant portions of a molecule lack C-H bonds



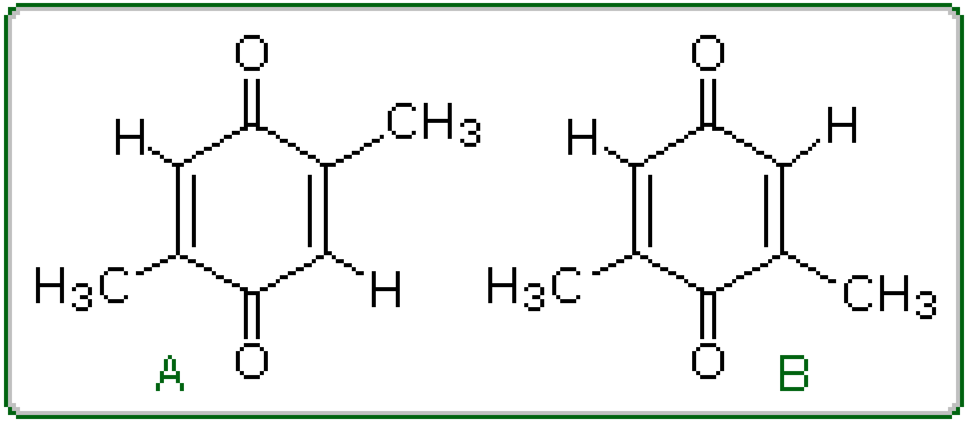
croconic acid



a polyacetylene from *Dahlia*

Why performed ^{13}C NMR?

- When compounds display similar proton nmr spectra and might be difficult to distinguish them by proton nmr alone

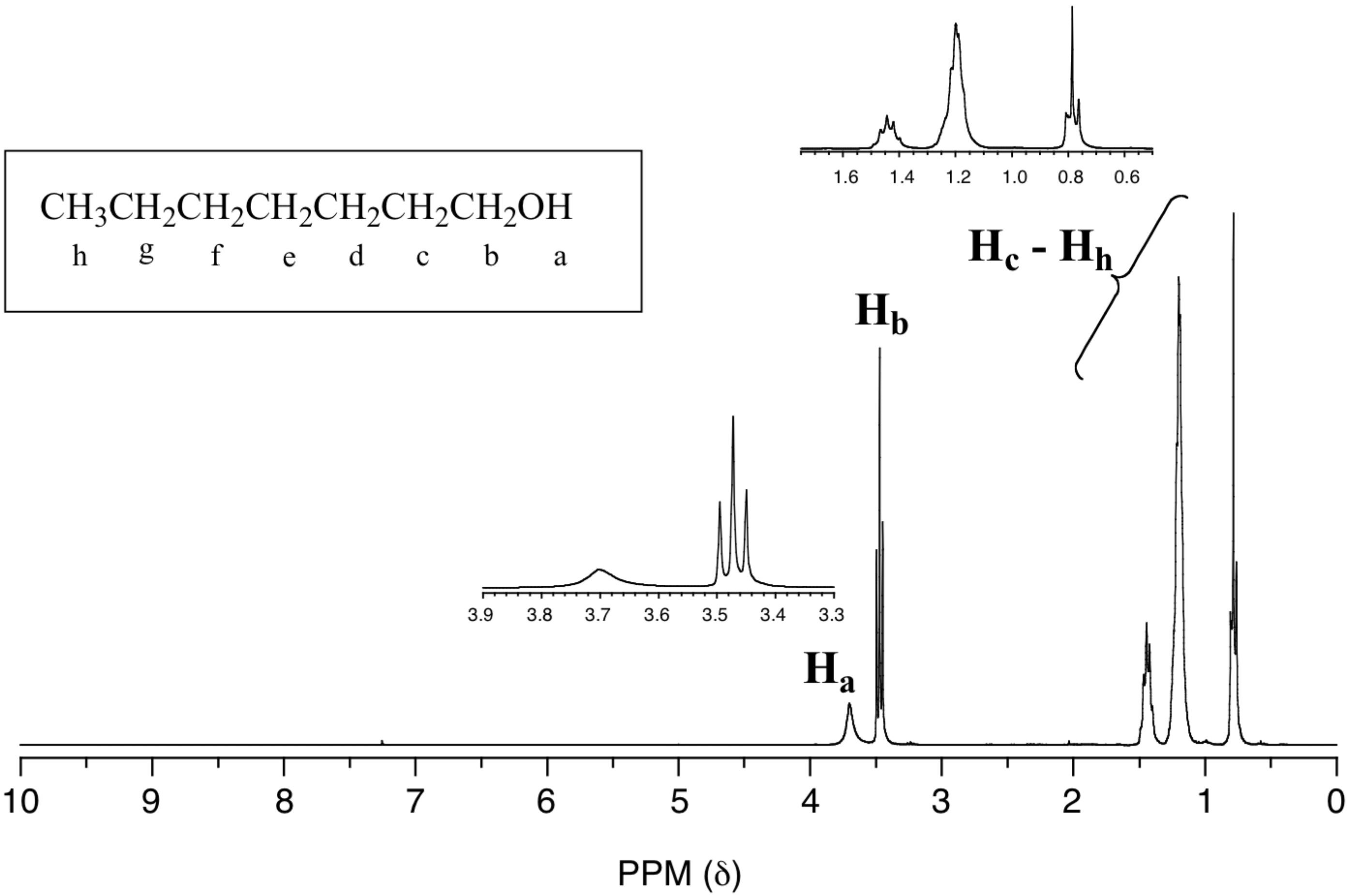


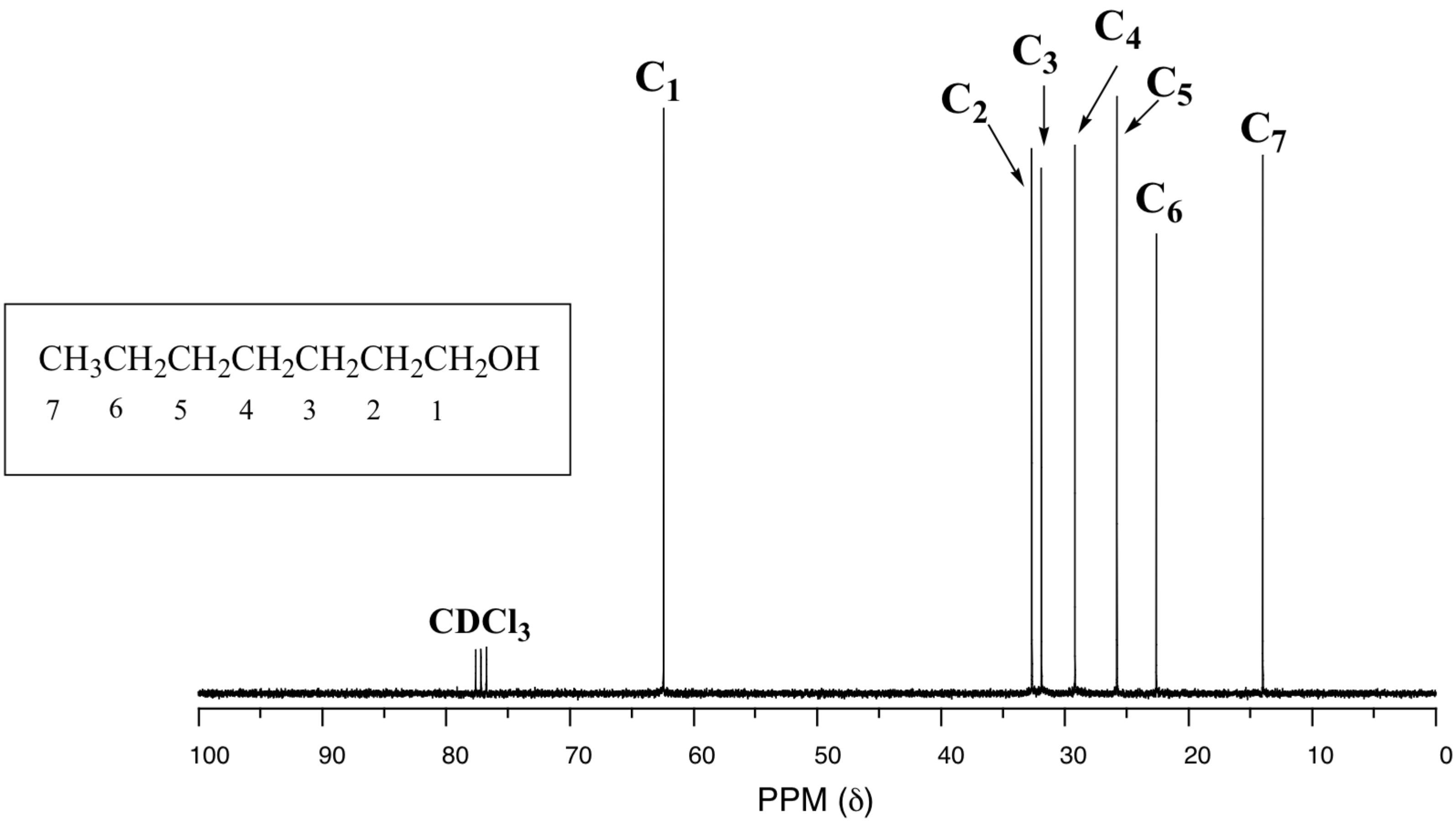
Why performed ^{13}C NMR?

- Carbons resonate from ~0-220 ppm relative to the TMS standard, as opposed to only ~0-12 ppm for protons

We can distinguish separate peaks for each carbon, even in a relatively large compound containing carbons in very similar environments, even if it is not possible to distinguish the corresponding protons

$$\underset{h}{CH_3}\underset{g}{CH_2}\underset{f}{CH_2}\underset{e}{CH_2}\underset{d}{CH_2}\underset{c}{CH_2}\underset{b}{CH_2}\underset{a}{OH}$$


CH3CH2CH2CH2CH2CH2CH2OH
h g f e d c b a
1.6 1.4 1.2 1.0 0.8 0.6
Hc - Hh
Hb
3.9 3.8 3.7 3.6 3.5 3.4 3.3
Ha
10 9 8 7 6 5 4 3 2 1 0
PPM (δ)



$CH_3CH_2CH_2CH_2CH_2CH_2CH_2OH$
7 6 5 4 3 2 1
C1
C2
C3
C4
C5
C6
C7
CDCl3
100
90
80
70
60
50
40
30
20
10
0
PPM (δ)

Interpretation of ^{13}C NMR spectra

Chemical shift

(depends on the chemical surrounding)

How many types of C are in the molecule

It's diaganostic of the chemical environment (shielding/deshilding)

Signal multiplicity (spliting)

(splitting due to the surrounding nuclei)

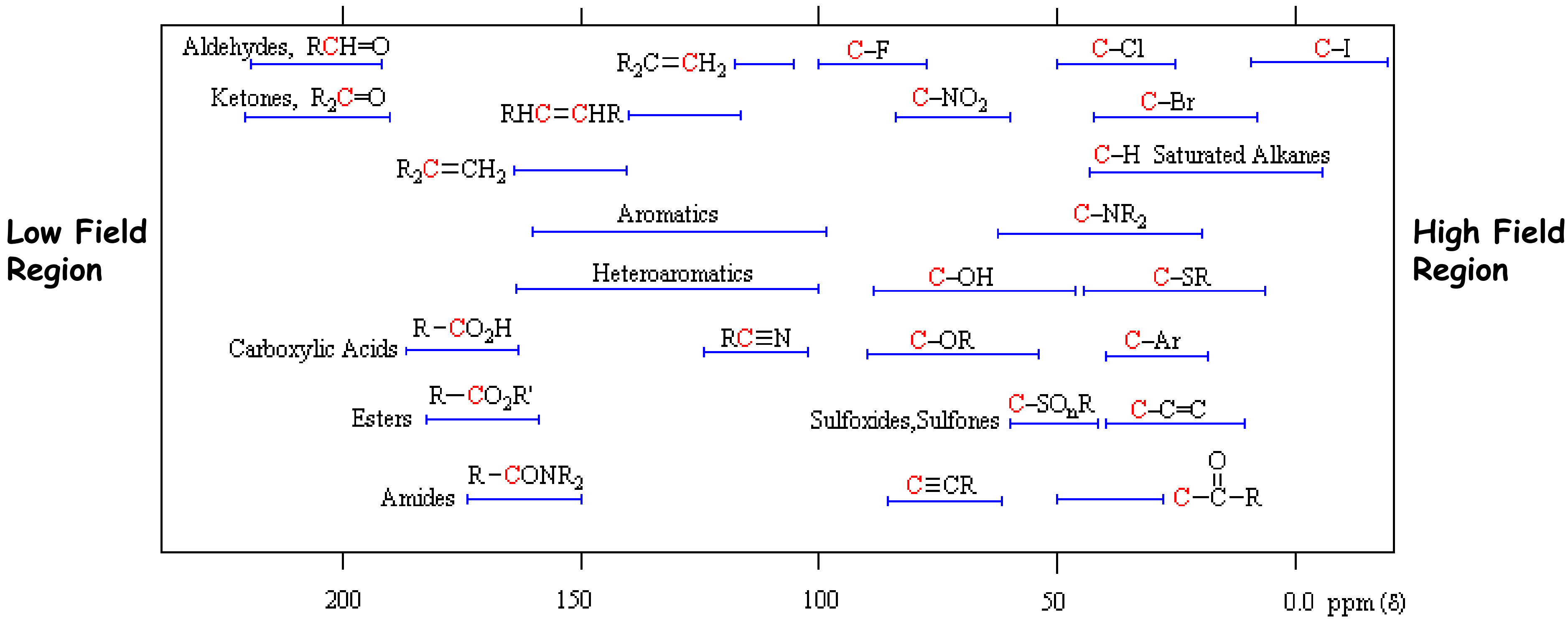
What are the surronding nucleus – connectivity in the molecule

Carbon types: C, CH, CH_2, CH_3

^{13}C NMR chemical shift depends on chemical surrounding

Structural information of molecules

^{13}C NMR Table – **Carbon chemical shift ranges**

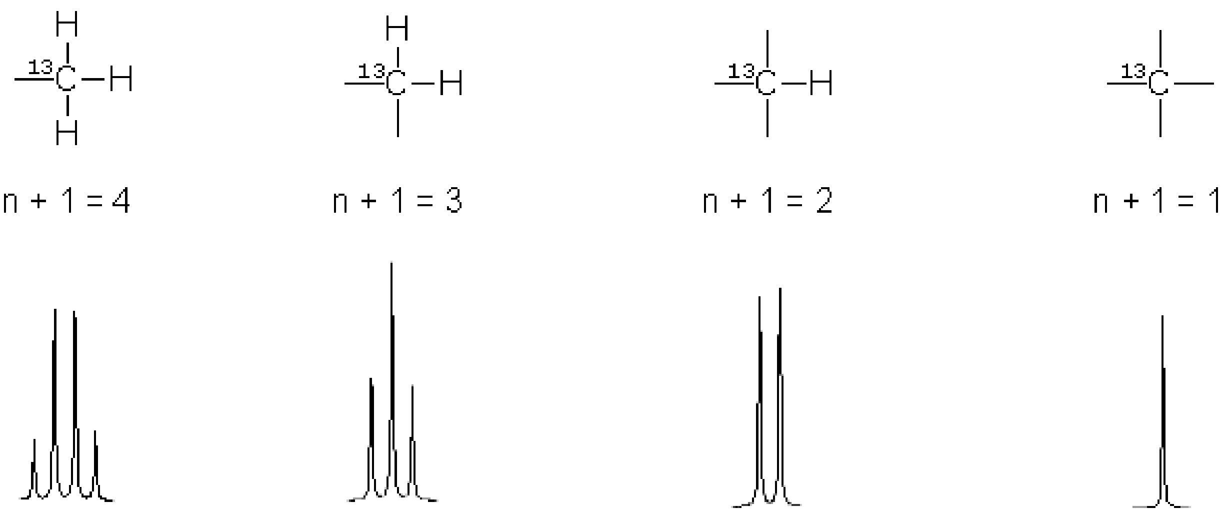


Carbon chemical shift range (220-0 ppm) ref TMS

Signal multiplicity - spin-spin coupling

- Homonuclear coupling – spin-spin coupling between neighboring carbons in not observed due the low natural abundance of ^{13}C nuclei (<1%)

- Heteronuclear coupling – spin-spin coupling between carbons and the hydrogens (n+1 rule) with J_{CH} 130 – 270 Hz



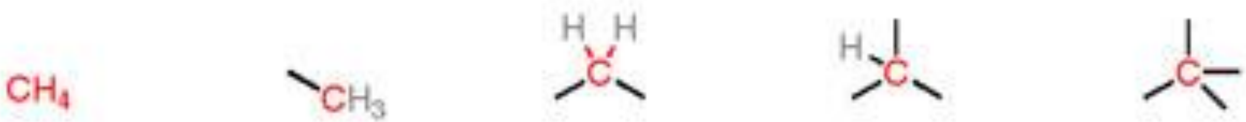
Information – distinguishing between a C, CH_3 , a CH_2 , and a CH groups

Carbon Types

Primary | Secondary | tertiary | Quaternary

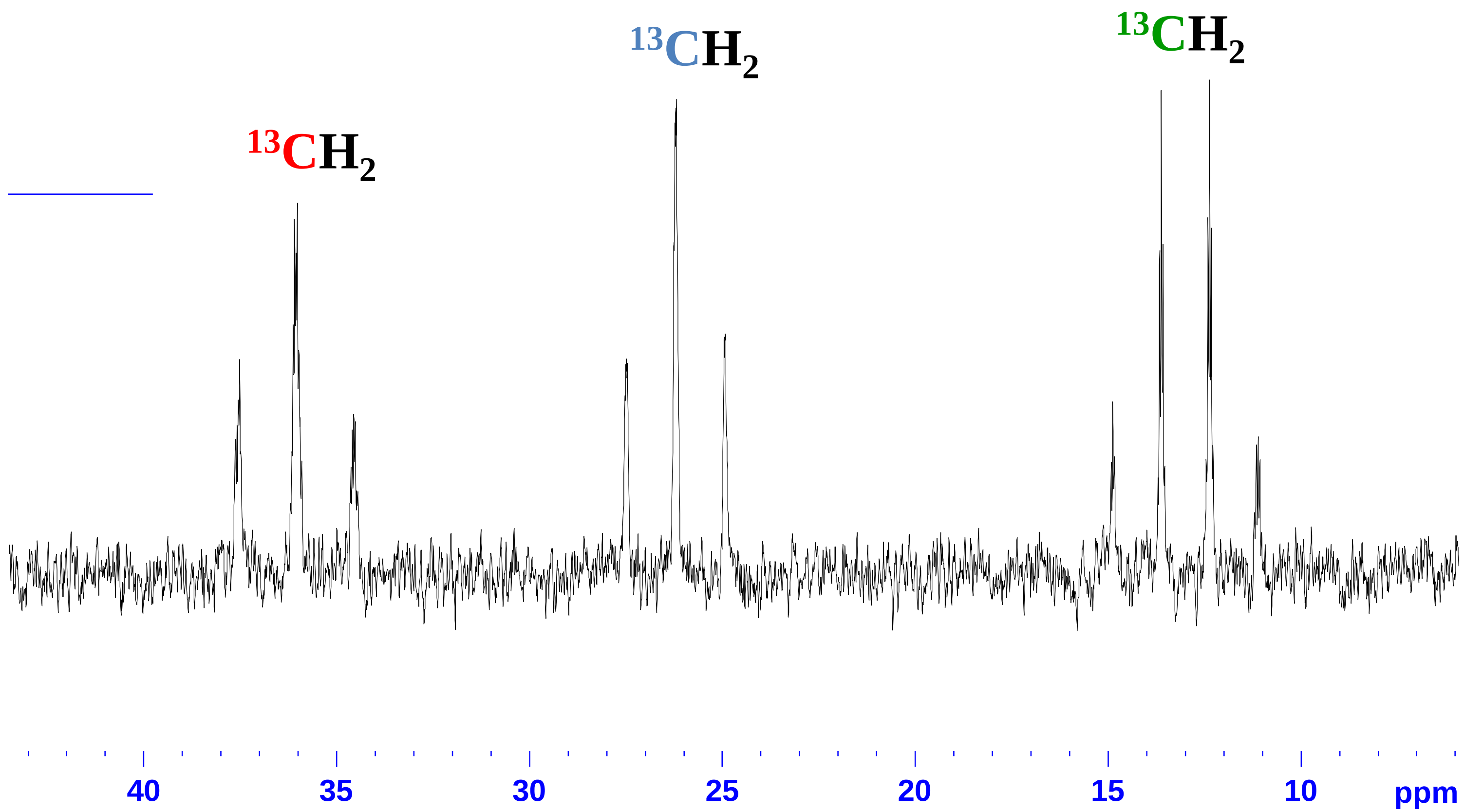
$n + 1 = 4$ | $n + 1 = 3$ | $n + 1 = 2$ | $n + 1 = 1$

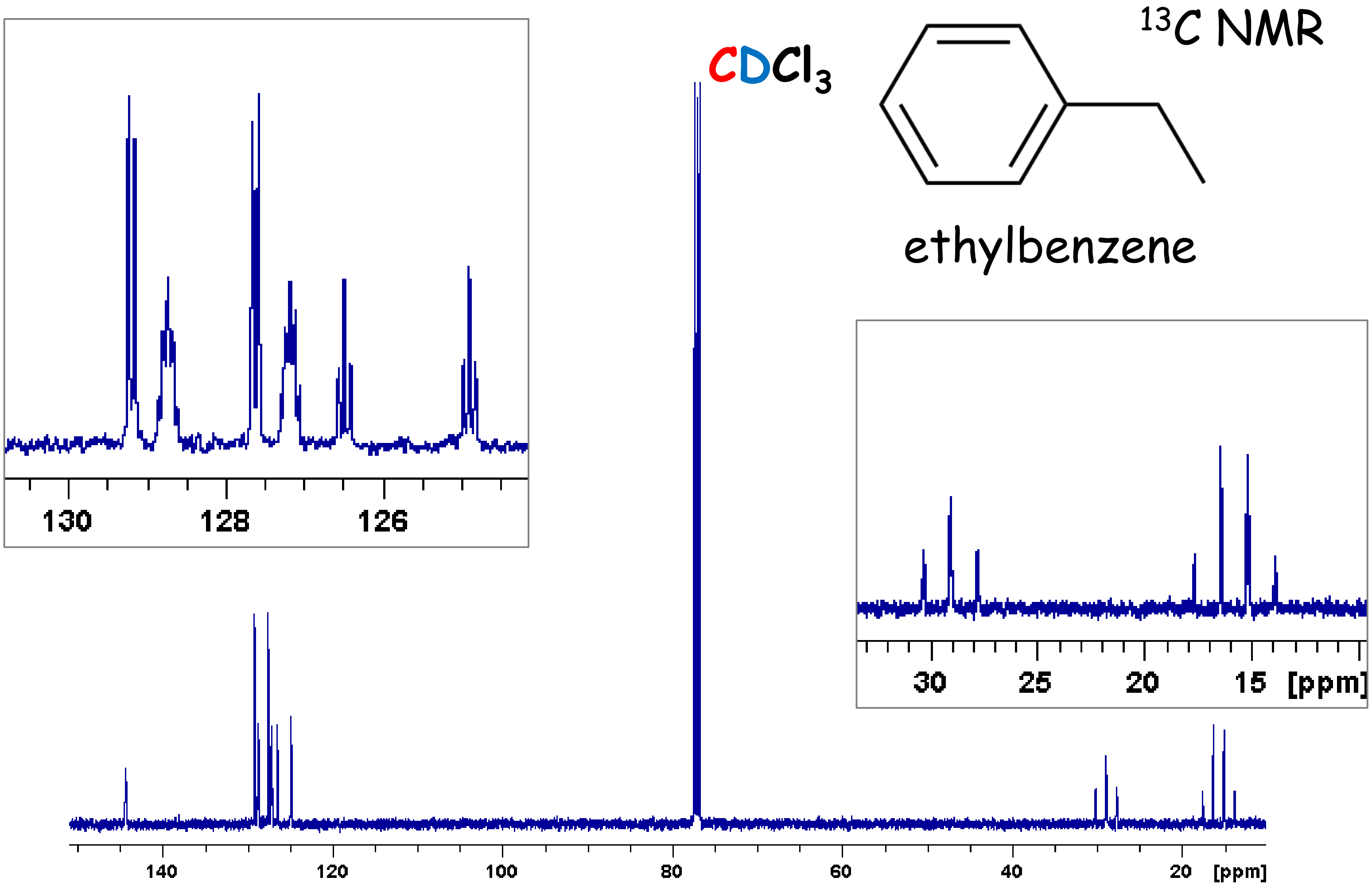
The name depends on the number of **carbons** attached (not hydrogens!)



0 carbons attached	*1 carbon attached*	*2 carbons attached*	*3 carbons attached*	*4 carbons attached*
Methane (unique)	Primary (1°) carbon	Secondary (2°) carbon	Tertiary (3°) carbon	Quaternary (4°) carbon

Bromobutane $BrCH_2CH_2CH_3$ ^{13}C NMR





Very complex!

Number spin states = 2 *I* +1 , I=1/2 (^{1}H, ^{13}C), 2 spin states
I=1 (2H deuterium D), 3 spin states

Signal Multiplicity= 2nI + 1

n	*I* = 1/2	*I* = 1	*I* = 3/2
0	1	1	1
1	1 1	1 1 1	1 1 1 1
2	1 2 1	1 2 3 2 1	1 2 3 4 3 2 1
3	1 3 3 1	1 3 6 7 6 3 1	1 3 6 10 12 12 10 6 3 1
4	1 4 6 4 1	1 4 10 16 19 16 10 4 1	1 4 10 20 31 40 44 40 31 20 10 4 1

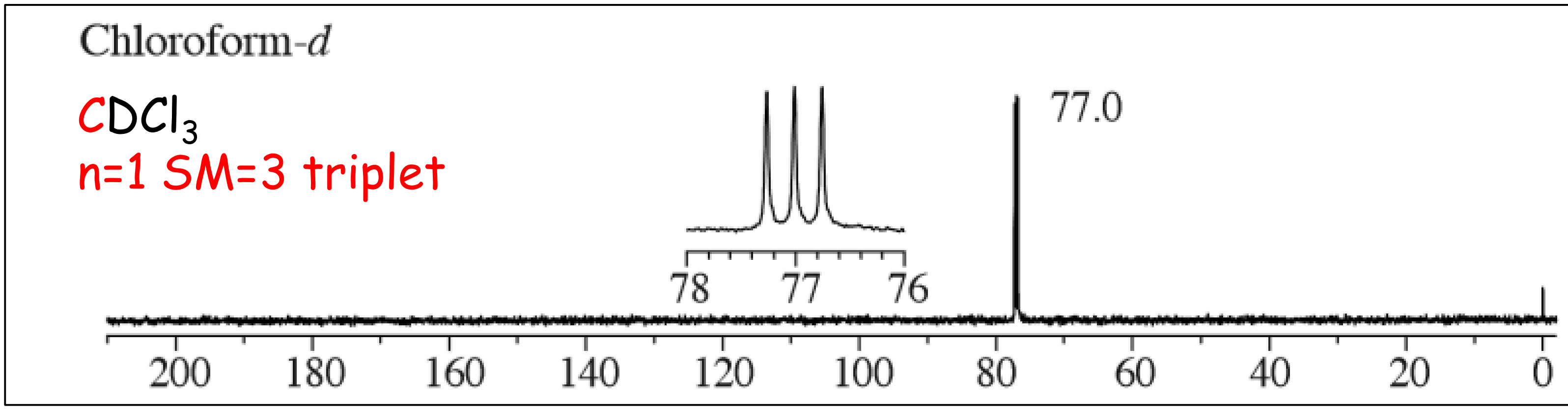
^{13}C Deuterated solvents, deuterium (D) I=1

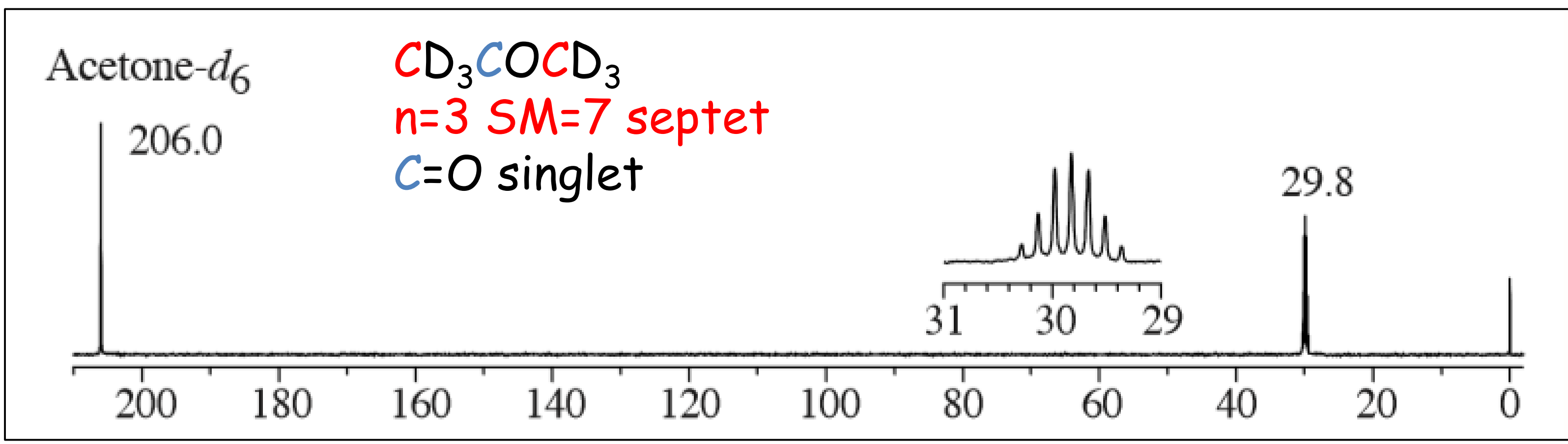
Signal Multiplicity= 2nI + 1

n	$I = 1$
0	1
1	1 1 1
2	1 2 3 2 1
3	1 3 6 7 6 3 1
4	1 4 10 16 19 16 10 4 1

Chloroform $CDCl_3$	n=1 triplet
Acetone CD_3COCD_3	n=3 septet and C=O singlet
Dimethyl sulfoxide CD_3SOCD_3	n=3 septet
Methanol CD_3OD	n=3 septet

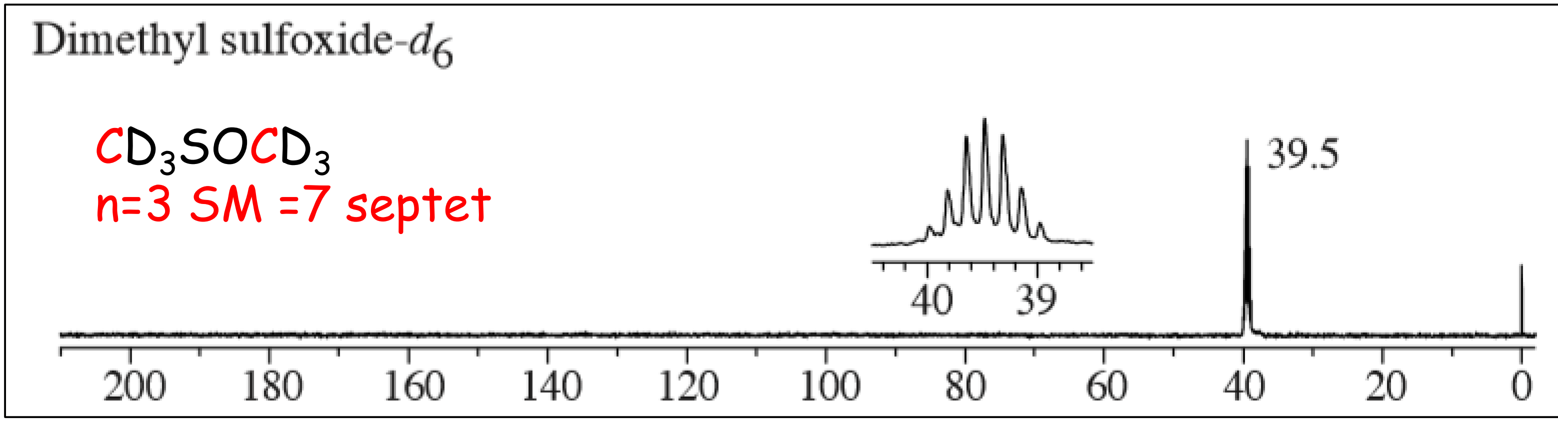
Signal Multiplicity= 2nI + 1 deuterium (D) I=1

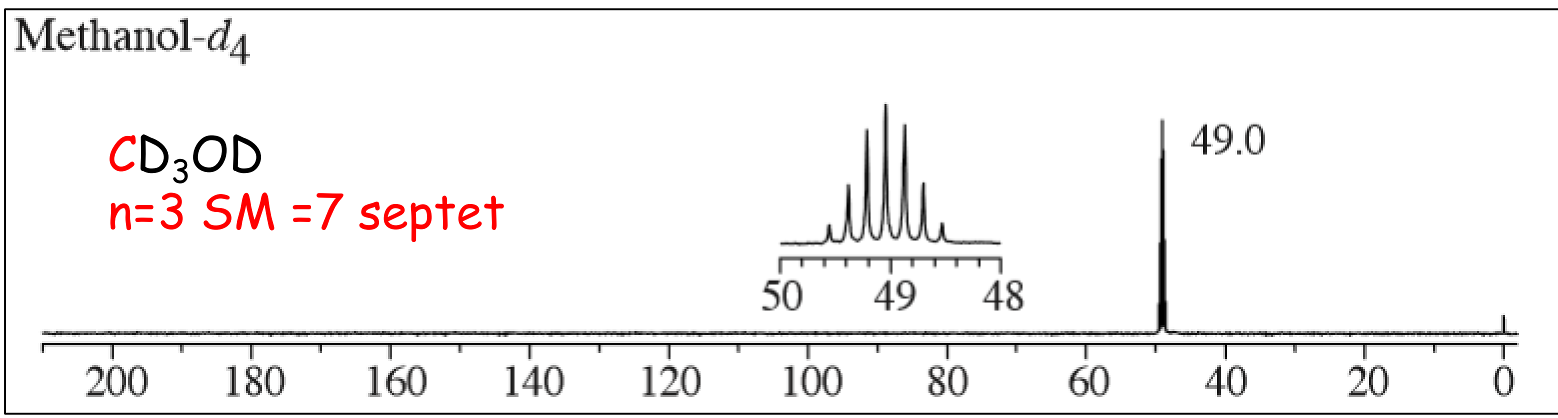




Structure Determination of Organic Compounds, Tables of Spectral Data, Pretsch, Buhlmann, Badertscher, 4Th edition, 2009

Signal Multiplicity= 2nI + 1 deuterium (D) I=1





Structure Determination of Organic Compounds, Tables of Spectral Data, Pretsch, Buhlmann, Badertscher, 4Th edition, 2009

How to remove all heteronuclear coupling ^{13}C-^{1}H?

Broadband decoupling ($^{13}C\ \{^{1}H\}$) - by irradiation of all protons resonance the spin-spin coupling (J_{CH}) is suppressed (decoupled), resulting in all signals appearing as singlets and with lagers intensities due to the **N**uclear **O**verhauser **E**nhancement (NOE)

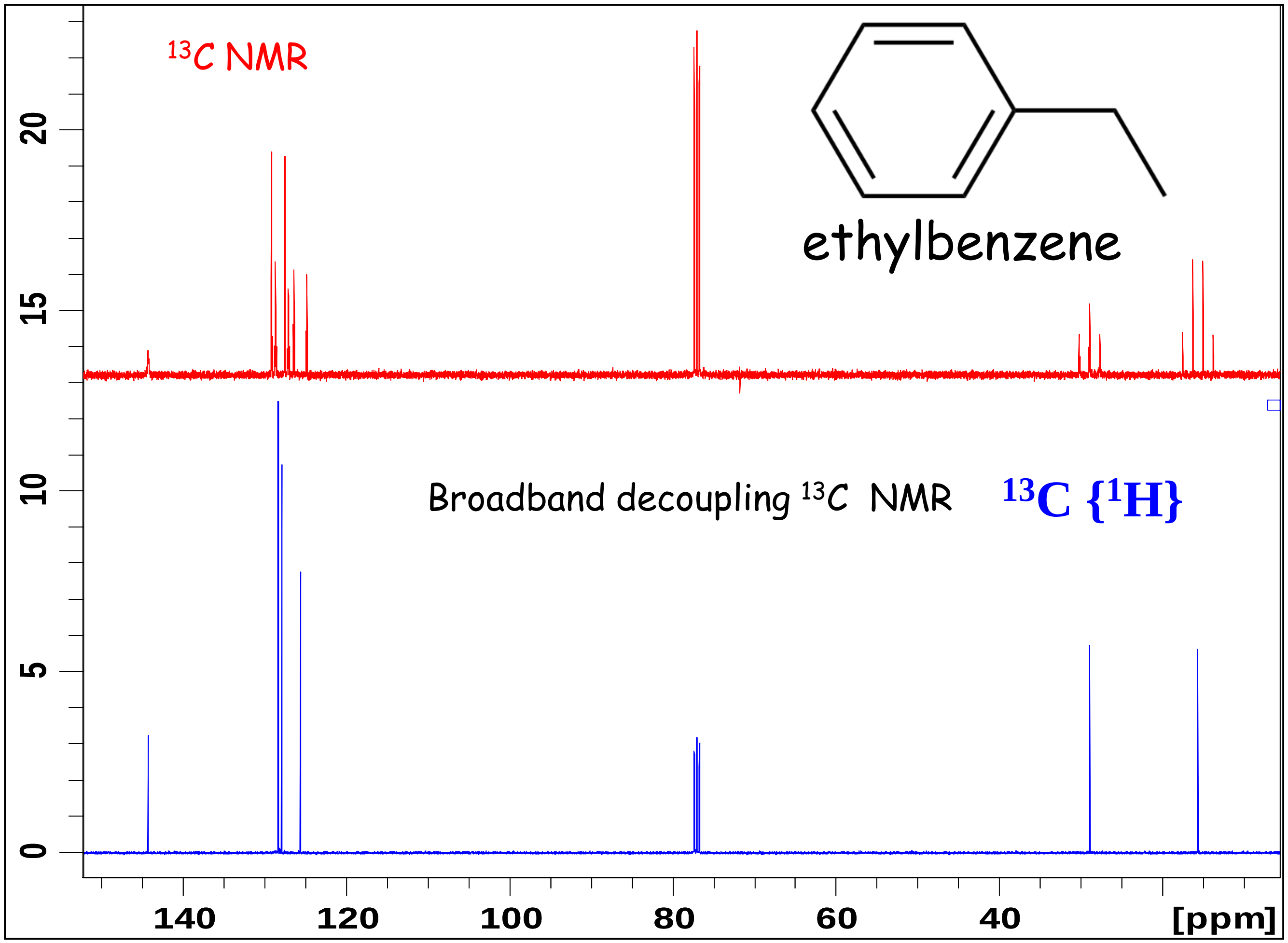
NOE -The irradiation of protons resonance increase the signal of attached carbon (4x more, due $\gamma\ ^{1}H/\gamma\ ^{13}C$ =~4)

Major number of protons => greater increase of intensity

⬇

Integration of ^{13}C signal area is not proportional to the number of carbon that give rise that resonance

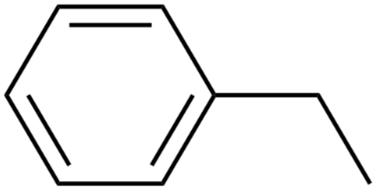
^{13}C-^{1}H decoupling simplify and facilitates the interpretation ^{13}C NMR

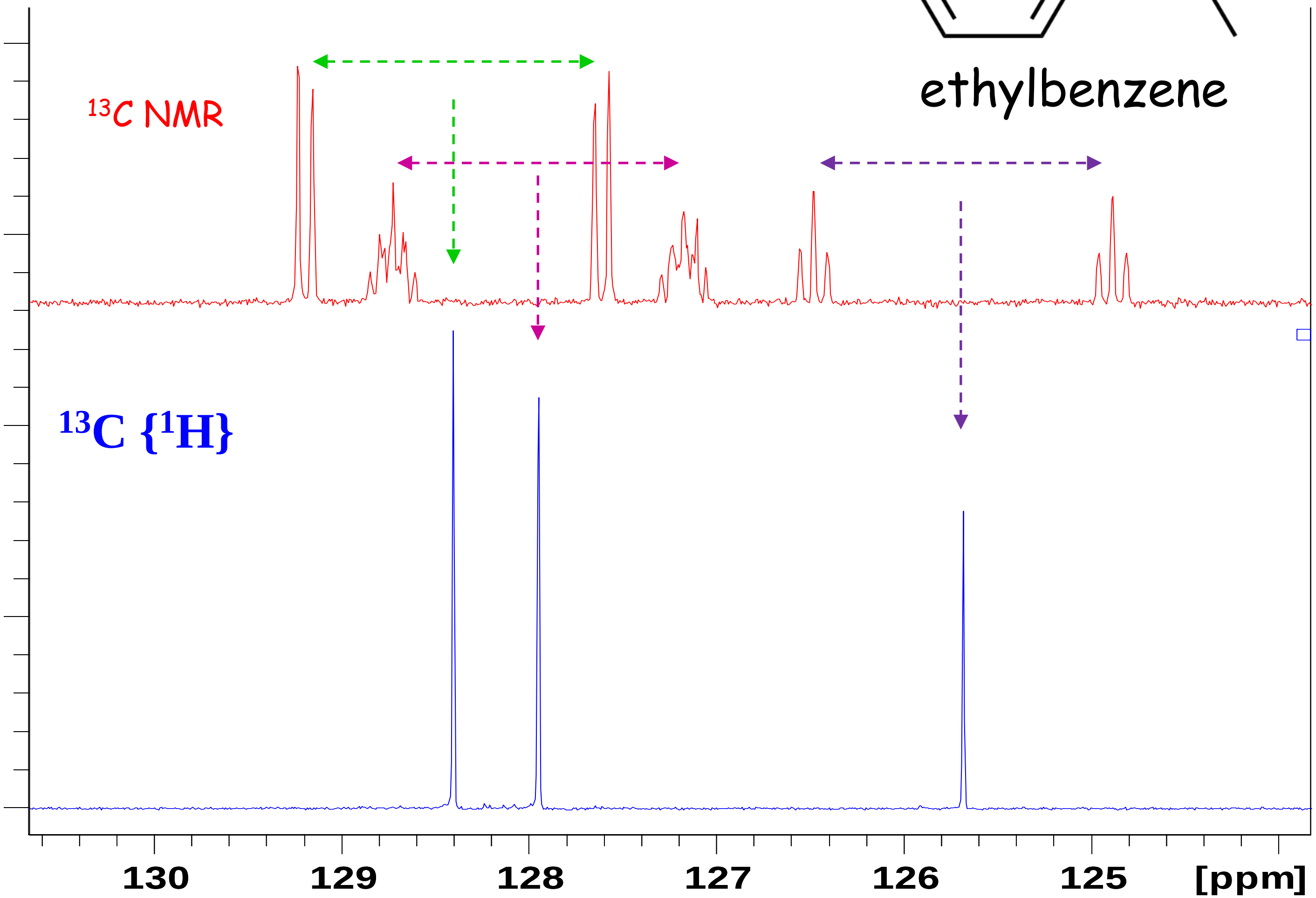


4x

1x

J_{CH} may be greater than the difference between the chemical shifts





- Disadvantage – in decoupling ^{13}C spectra the multiplicity signals is lost!

↓

the information about how many protons are coupled to each carbon is lost (C, CH, CH_2 and CH_3 carbons)

How to obtain the lost information (multiplicity)?

- DEPT - Distortionless Enhancement by Polarization Transfer
- APT – Attached Proton Test

Are multiplet detection technique

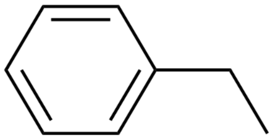
Multipulse ^{13}C NMR experiments - DEPT

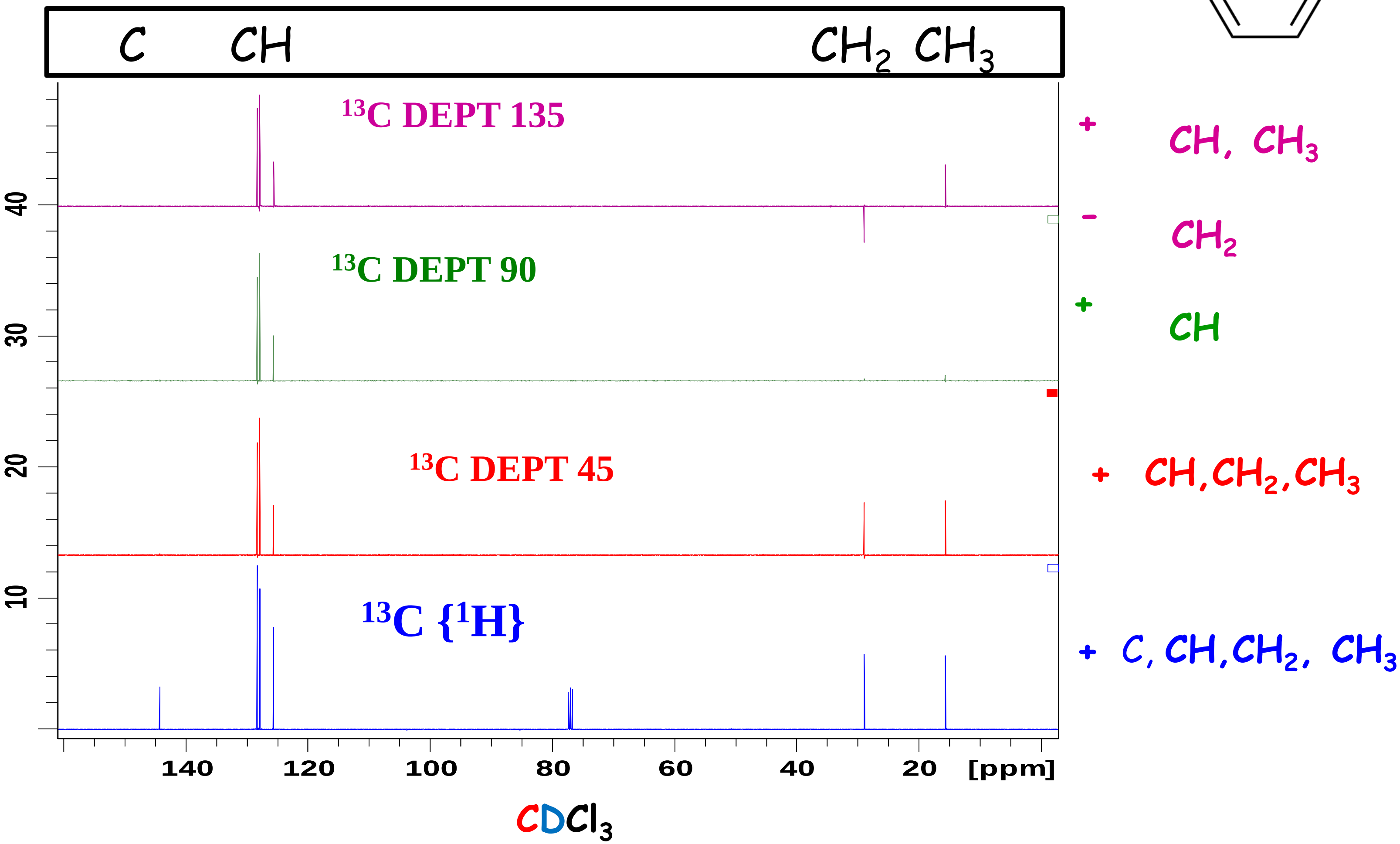
- It is a proton-carbon polarization transfer method
- Quaternary carbon atoms do not appear
- The intensity of carbon signals depends on ^{1}H pulse angle θ

- DEPT 45 (θ = 45) – show carbons that have attached hydrogen atoms (CH, CH_2, CH_3)
- DEPT 90 (θ = 90) – show carbons with a single attached hydrogen atom (CH)
- DEPT 135 (θ = 135) – distinguishes between carbon atoms based on their phasing
  CH and CH_3 carbon atoms are phased the same way (usually positive)
  CH_2 carbon atoms are phased the opposite way (usually negative)

Information – DEPT for distinguishing between a CH_3 , a CH_2 , and a CH groups

^{13}C NMR DEPT - ethylbenzene





Broadband decoupled	**DEPT-90**	**DEPT-135**
C, CH, CH_2, CH_3	CH	CH_3, CH are positive CH_2 is negative

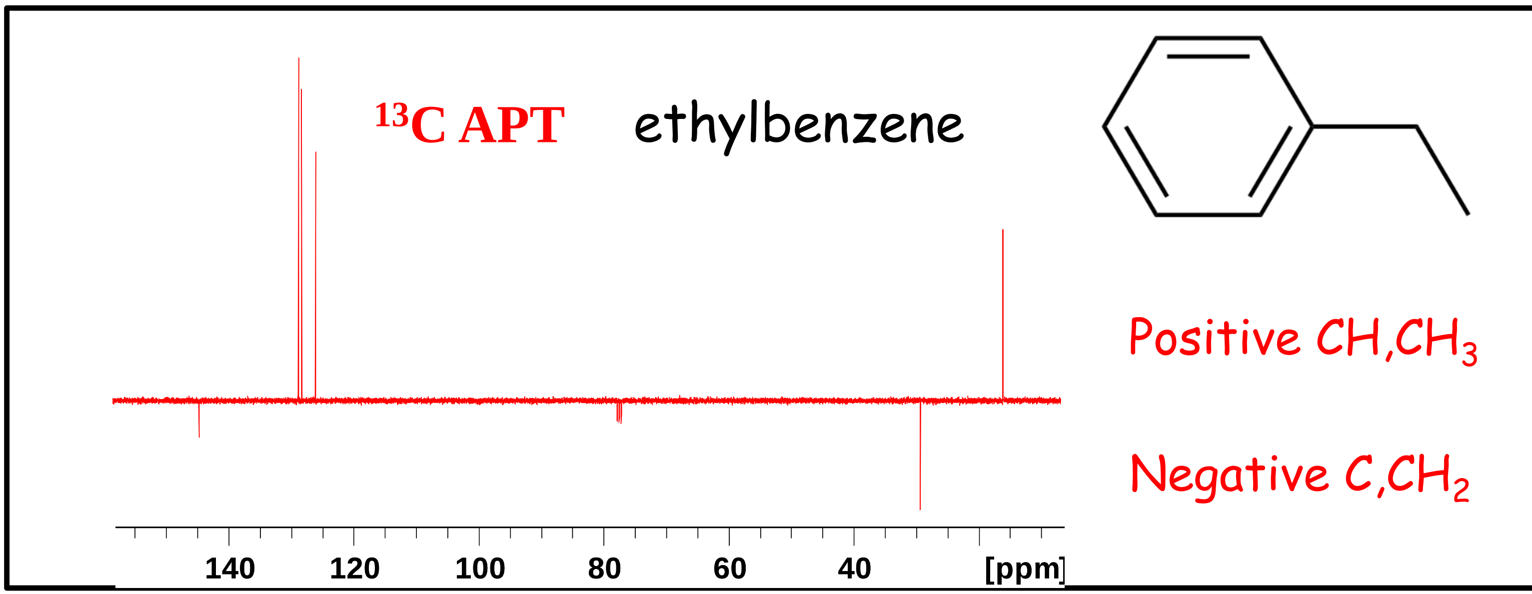
C	Subtract DEPT-135 from broadband decoupled
CH	DEPT-90
CH_2	Negative DEPT-135
CH_3	Subtract DEPT-90 from positive DEPT-135

Information – DEPT for distinguishing between a CH_3 , a CH_2 , and a CH groups

Multipulse ^{13}C NMR experiment - APT

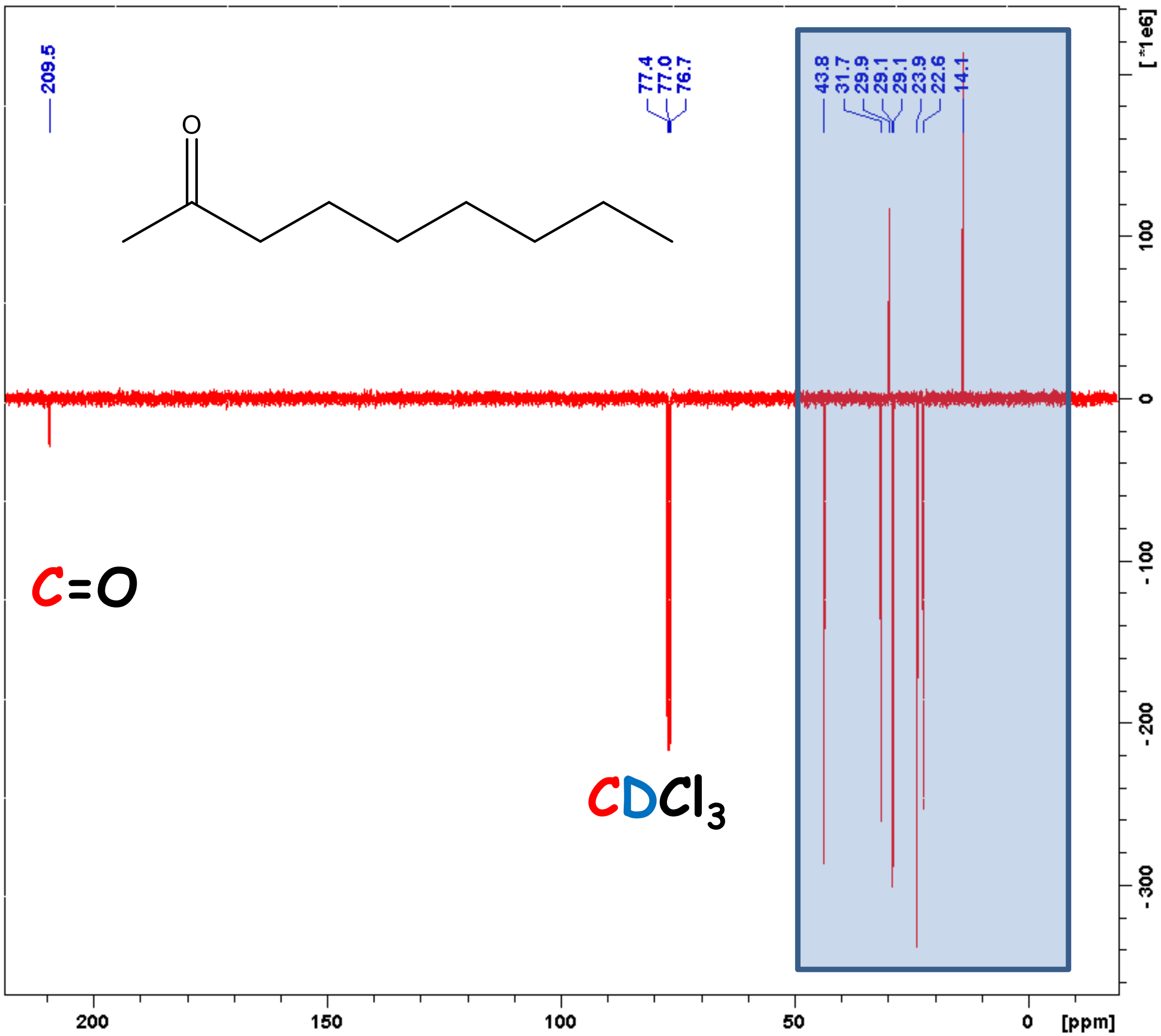
APT - Attached Proton Test

- It is a proton-carbon spin-echo experiment
- Quaternary carbon atoms are also observed

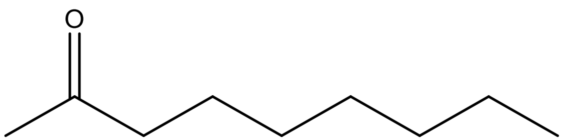


APT ^{13}C NMR Spectrum 2-nonanone

+ CH_3 CH
- CH_2 C

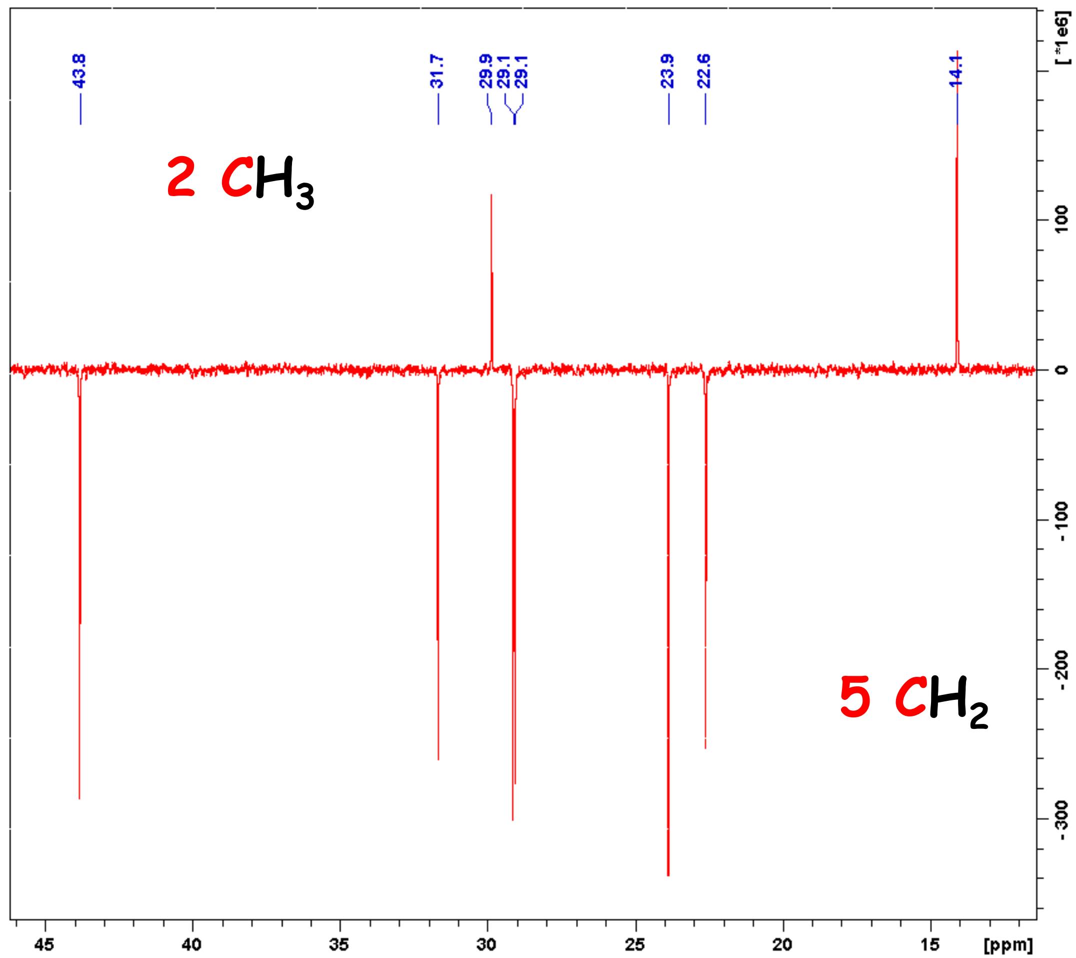


APT ^{13}C NMR Spectra



+ CH_3 CH

− CH_2 C



2D NMR

Two-dimensional nuclear magnetic resonance spectroscopy

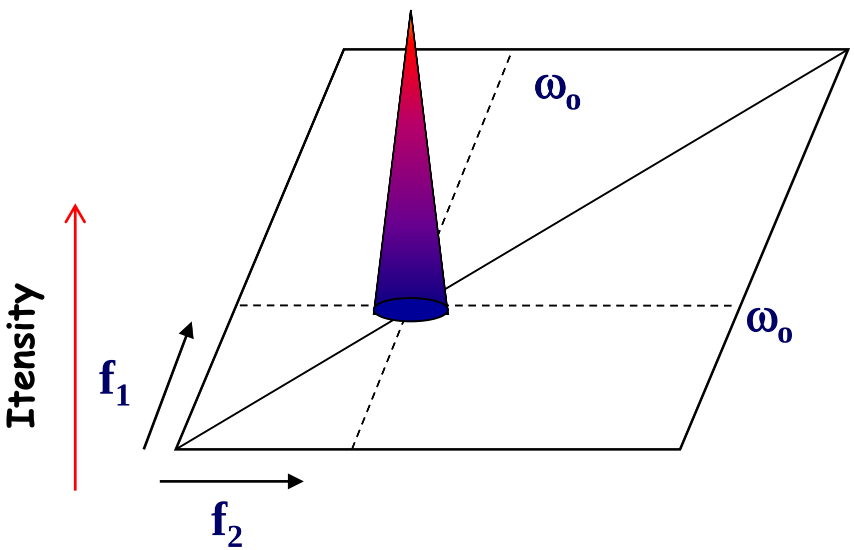
Homonuclear ^{1}H-^{1}H

- COSY – Corelation SpectroscopY
- TOCSY - TOtal Correlated SpectroscopY
- NOESY – Nuclear Overhauser Effect SpectroscopY

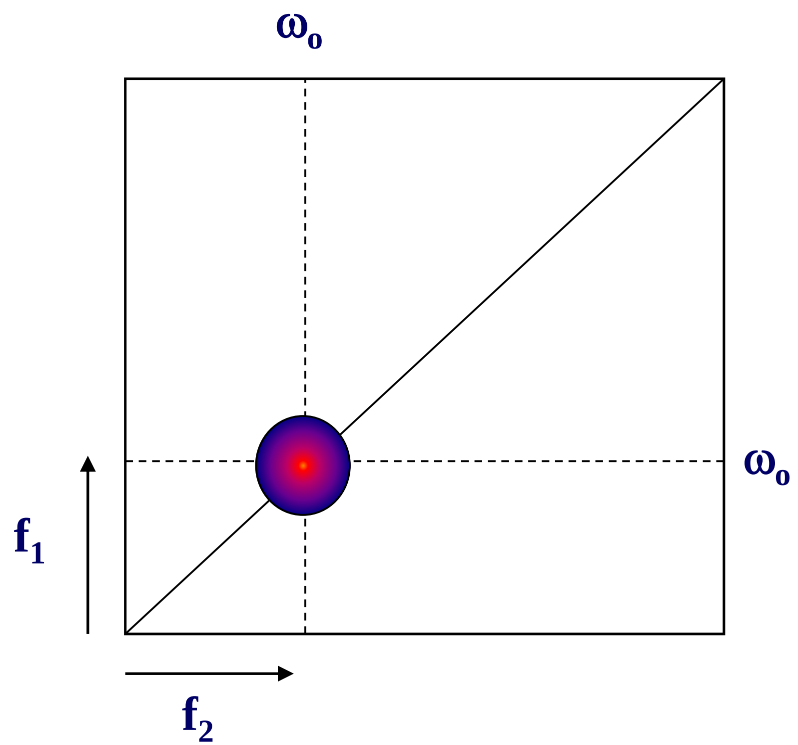
Heteronuclear ^{13}C-^{1}H

- HSQC – Heteronuclear Single-Quantum Correlation Spectroscopy
- HMQC - Heteronuclear Multiple-Quantum Correlation Sepectroscopy
- HMBC - Heteronuclear Multiple-Bond Correlation Spectroscopy
- HSQC-TOCSY
- HMQC-TOCSY

2D NMR Spectra



Tridimensional

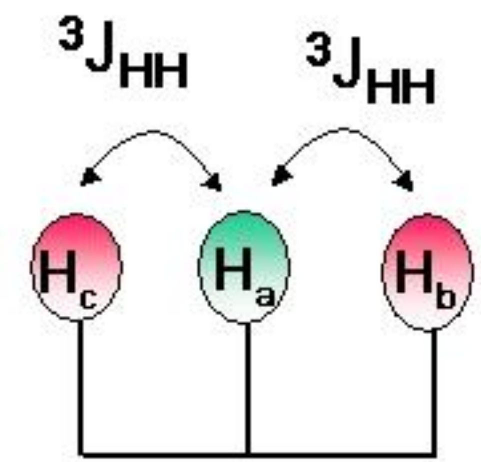


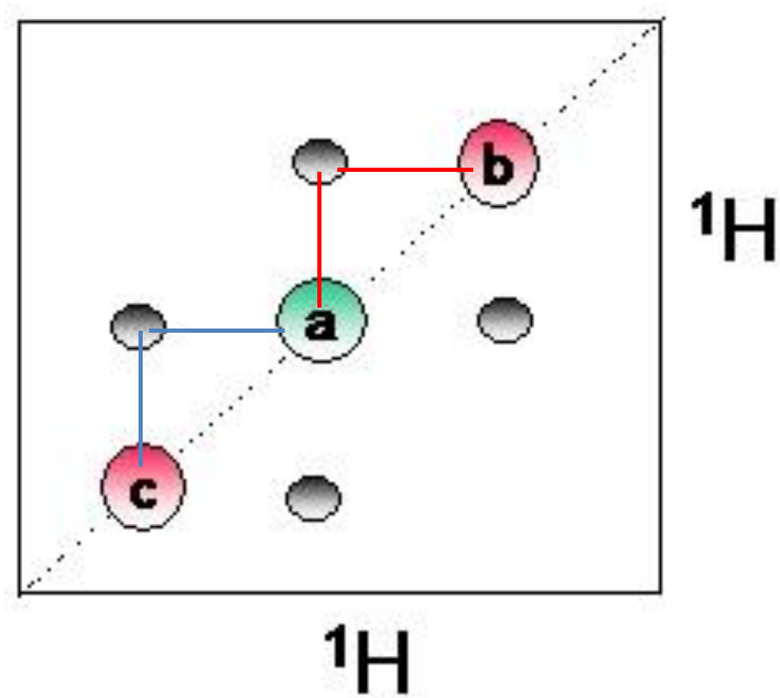
bidimensional

Homonuclear ^1H-^1H

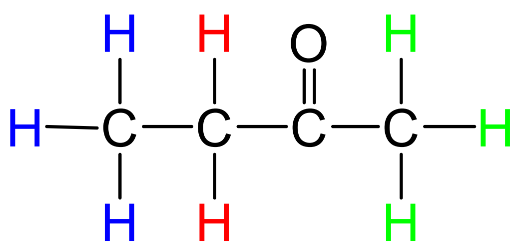
COSY - COrrelationSpectroscopY

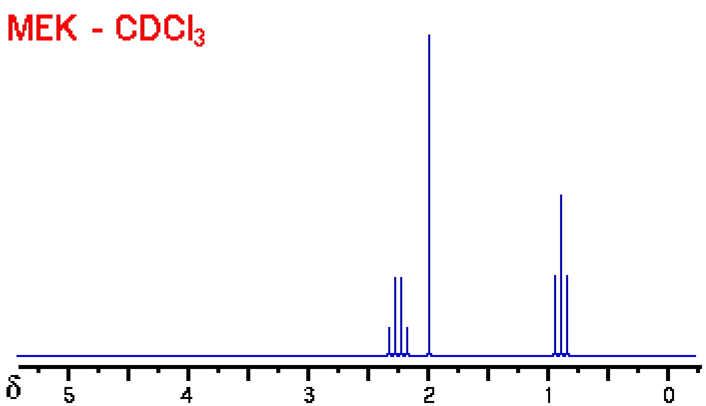
Information – what are the protons that have spin-spin coupling (H-H coupling through-bond)

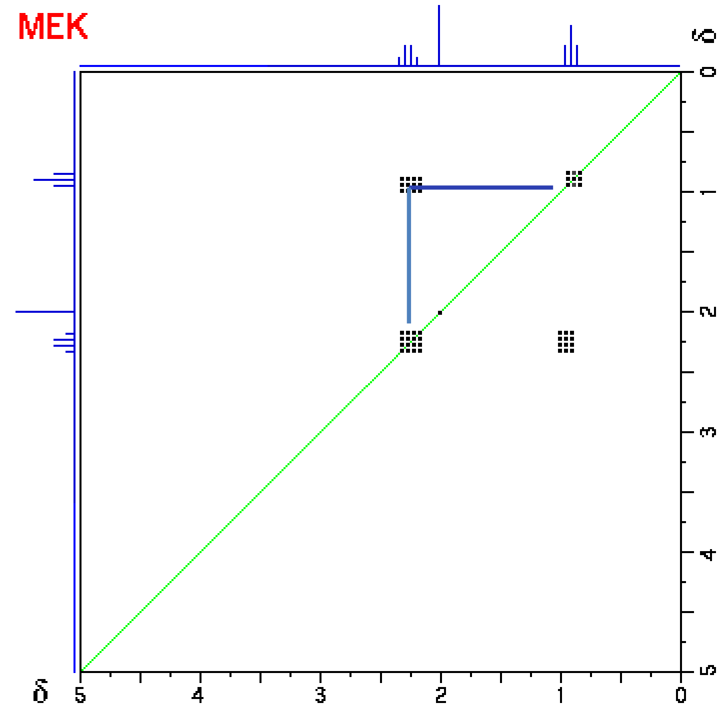


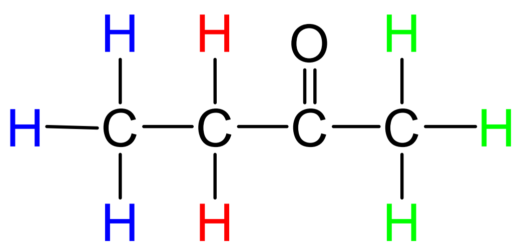


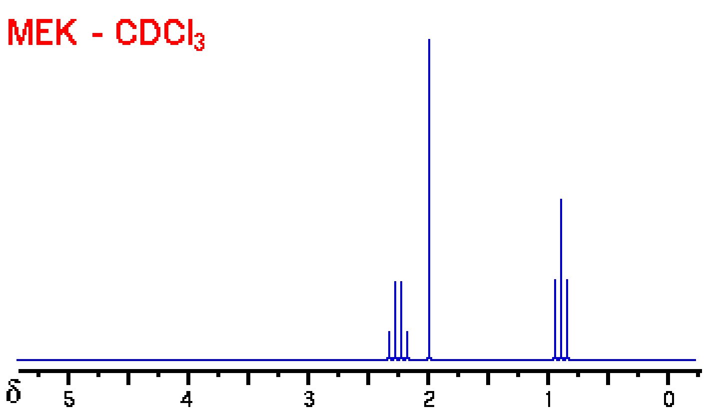
- Diagonal peaks are the protons seeing themselves = 1D proton spectrum
- Cross peaks exist only when there is *J* coupling between protons

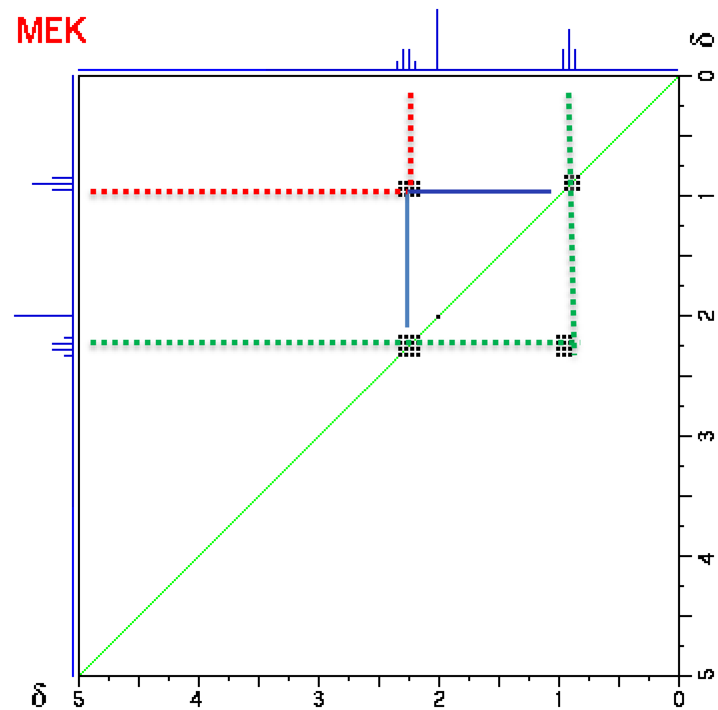



MEK - CDCl3
δ
5
4
3
2
1
0


MEK
δ
5
4
3
2
1
0


H
H
H
C
C
C
C
O
H
H
H
H
H

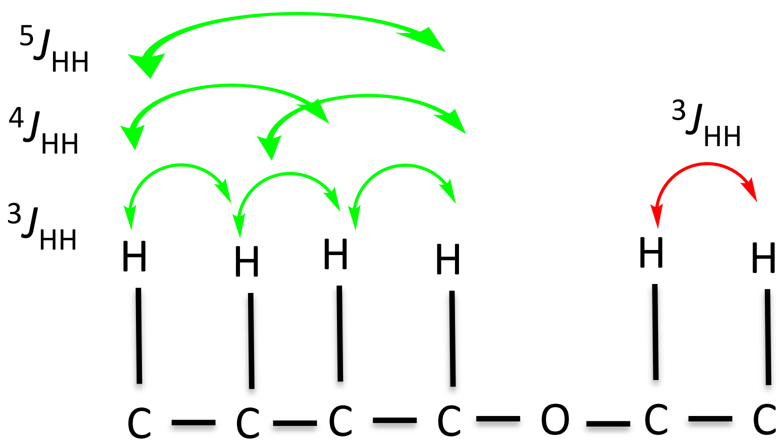

MEK - $CDCl_3$
δ
5
4
3
2
1
0

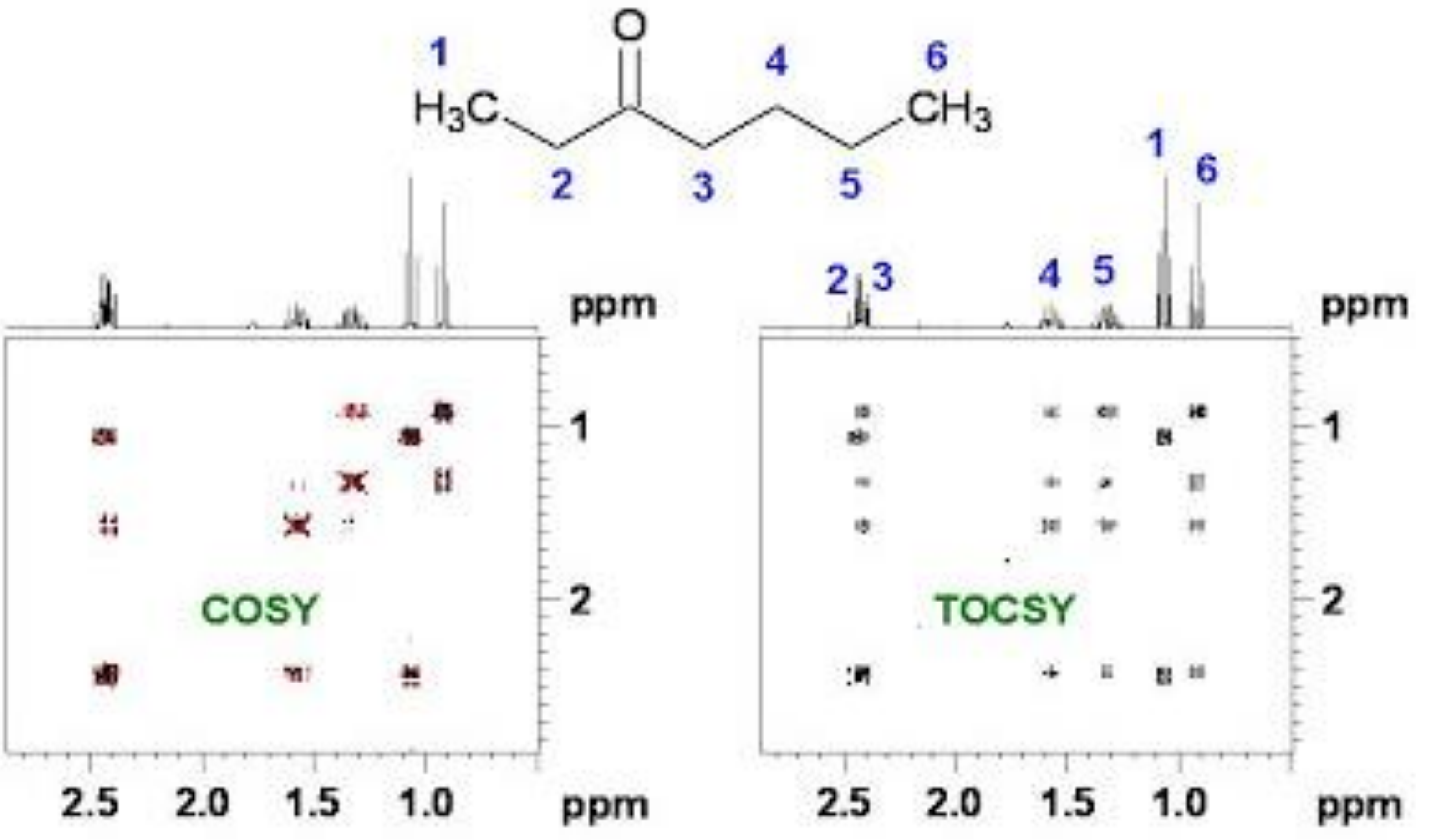

MEK
δ
5
4
3
2
1
0

Homonuclear ^{1}H-^{1}H

TOCSY- TOtal Correlated SpectroscopY

Information – what are the protons that have spin-spin coupling (H-H coupling through-bond) into groups (coupling networks)





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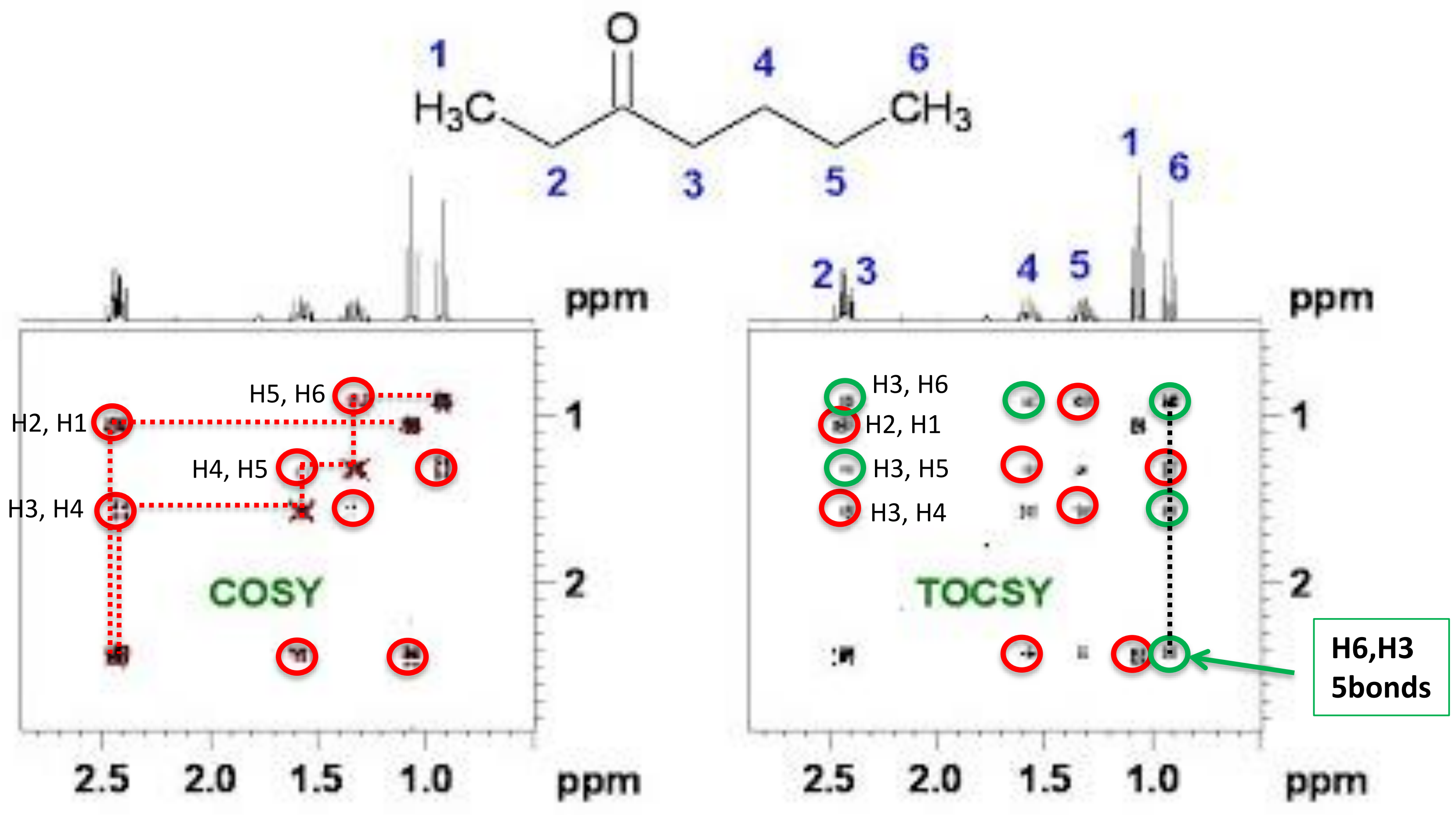

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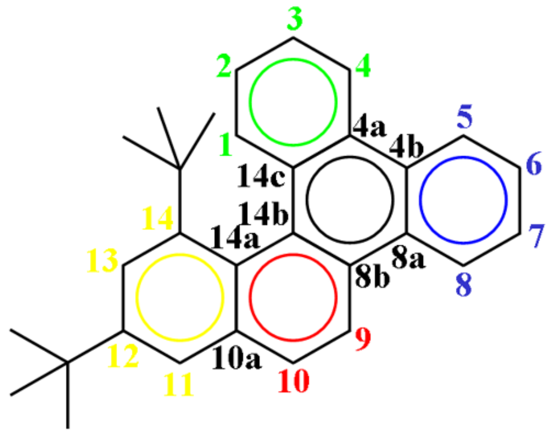
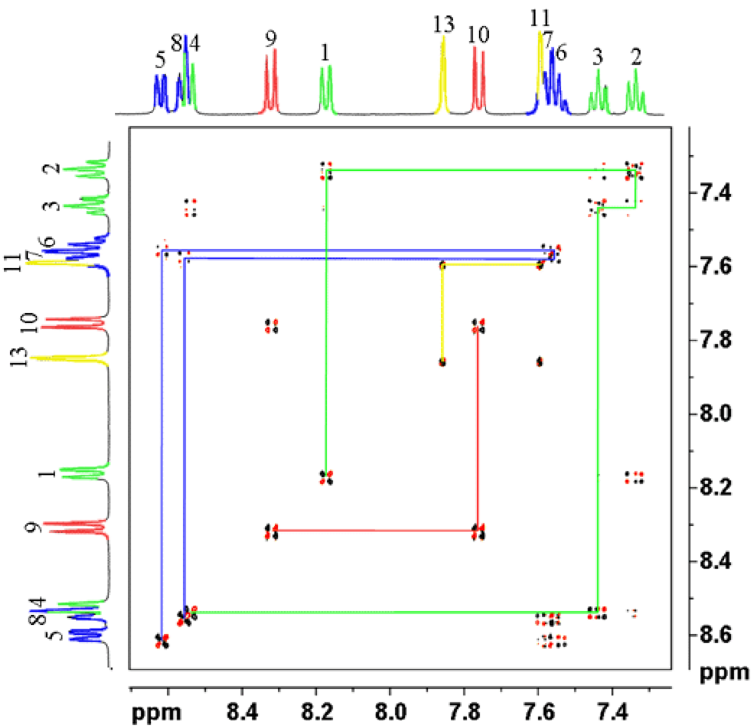
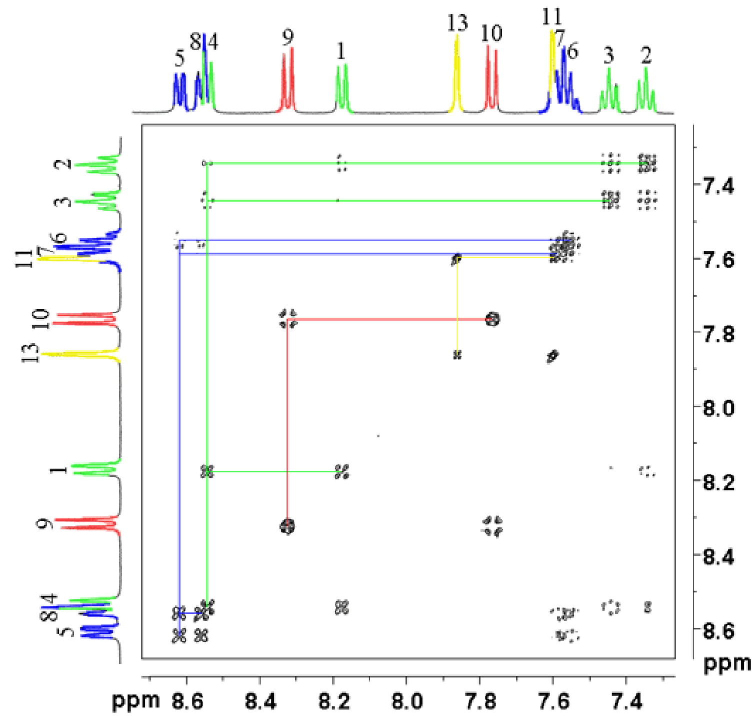


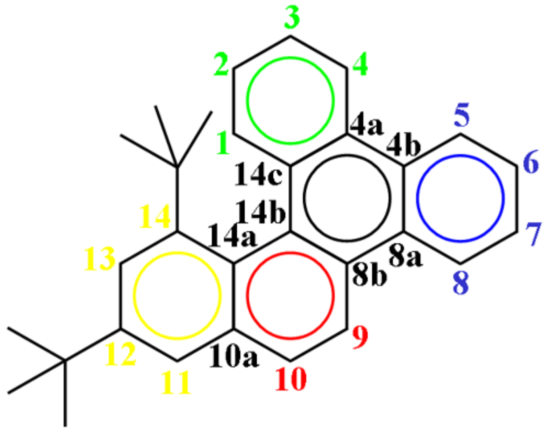
COSY



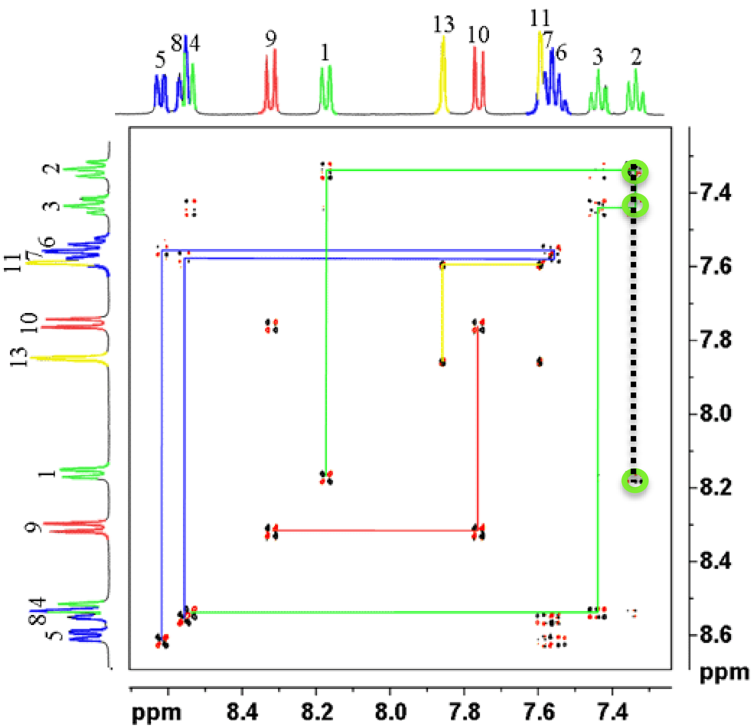
TOCSY



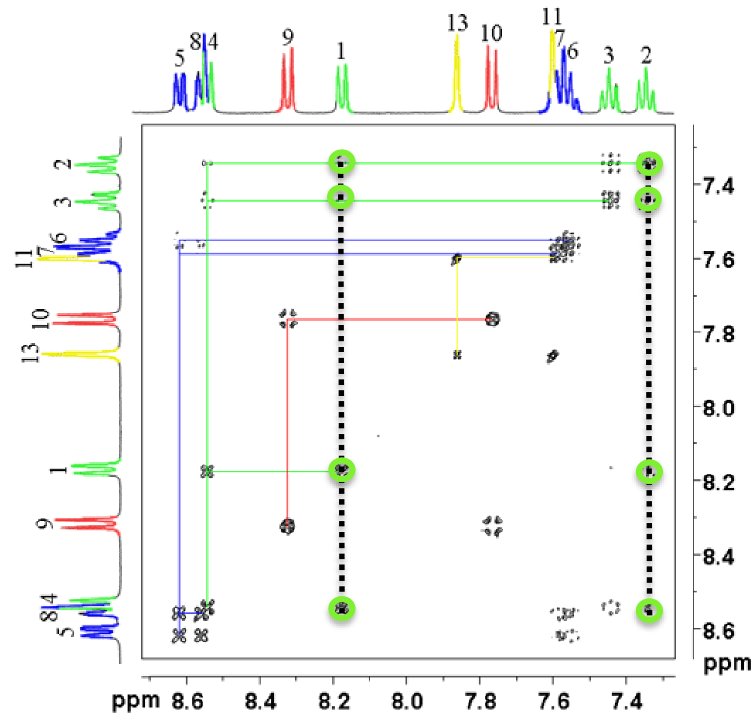
http://chem.ch.huji.ac.il/nmr/



COSY

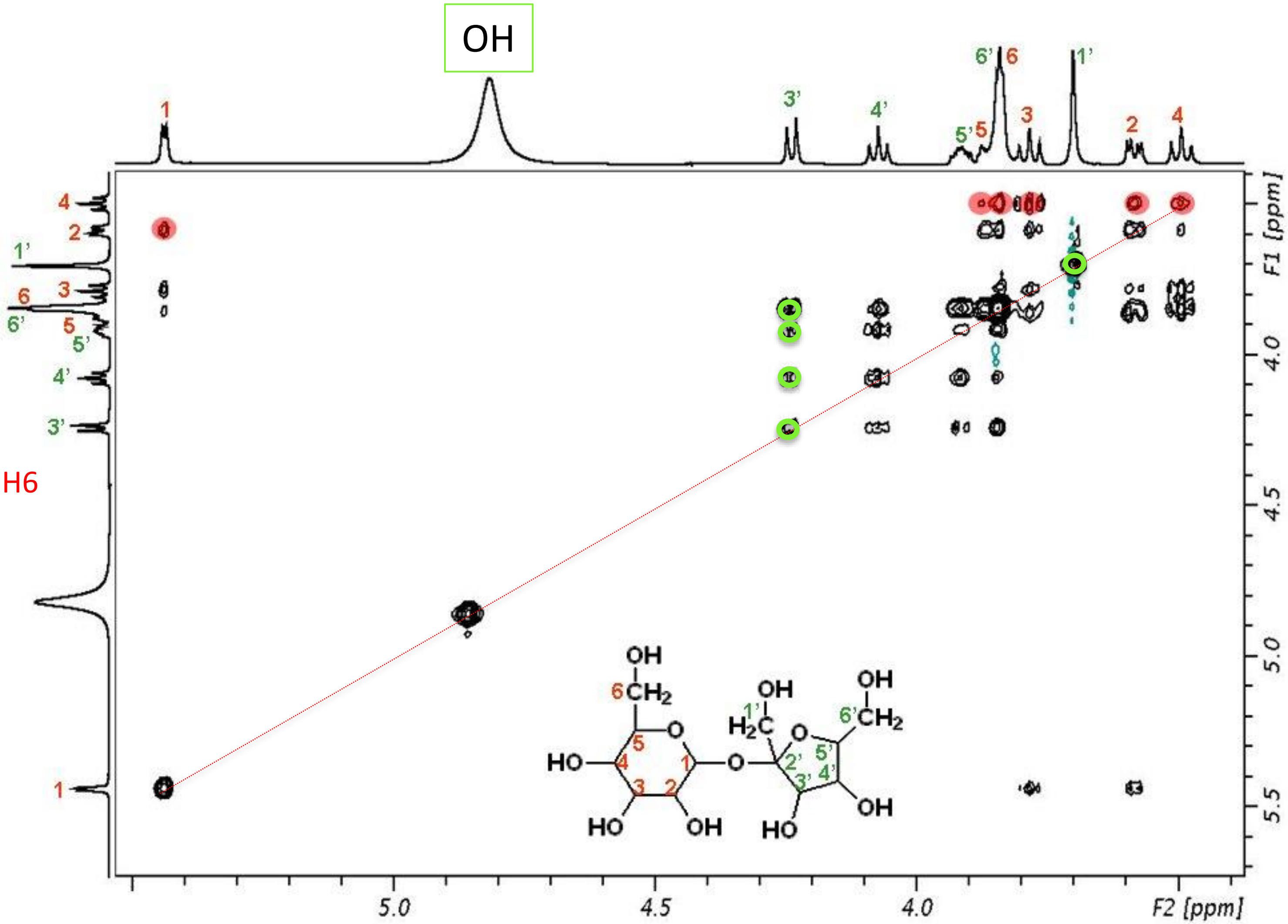


TOCSY



http://chem.ch.huji.ac.il/nmr/

TOCSY



3 *spin* groups:

H1,H2,H3,H4,H5,H6

H1'

H3', H4', H5', H6'

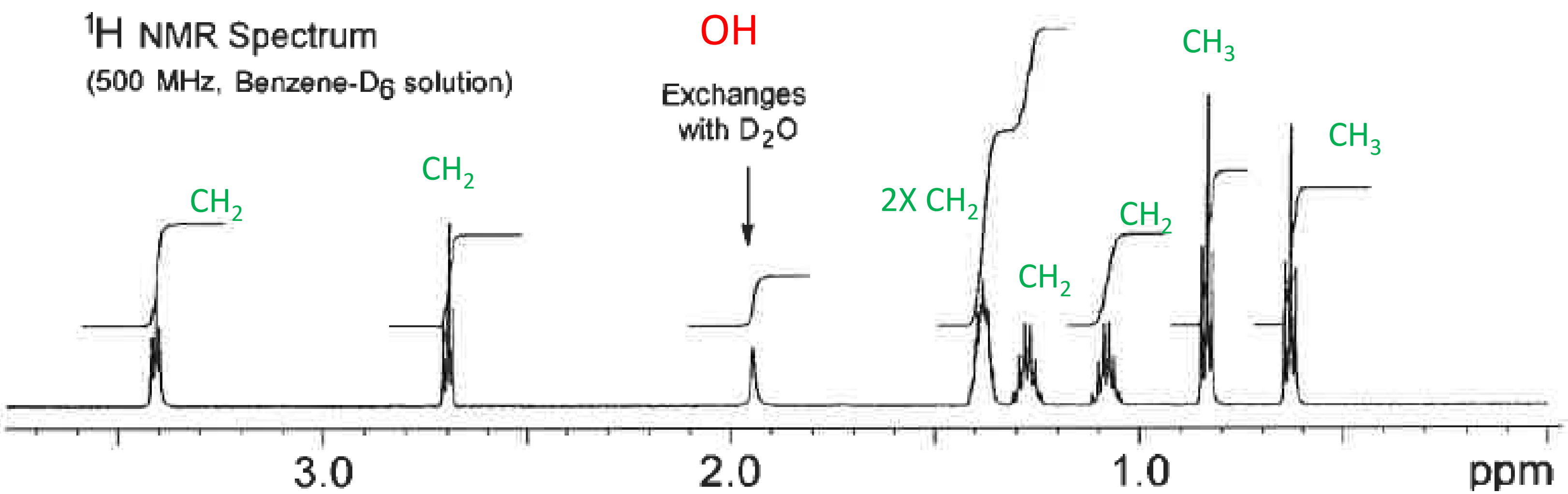
http://www.columbia.edu/cu/chemistry/groups/nmr/tocsy.JPG

1-iodobutane

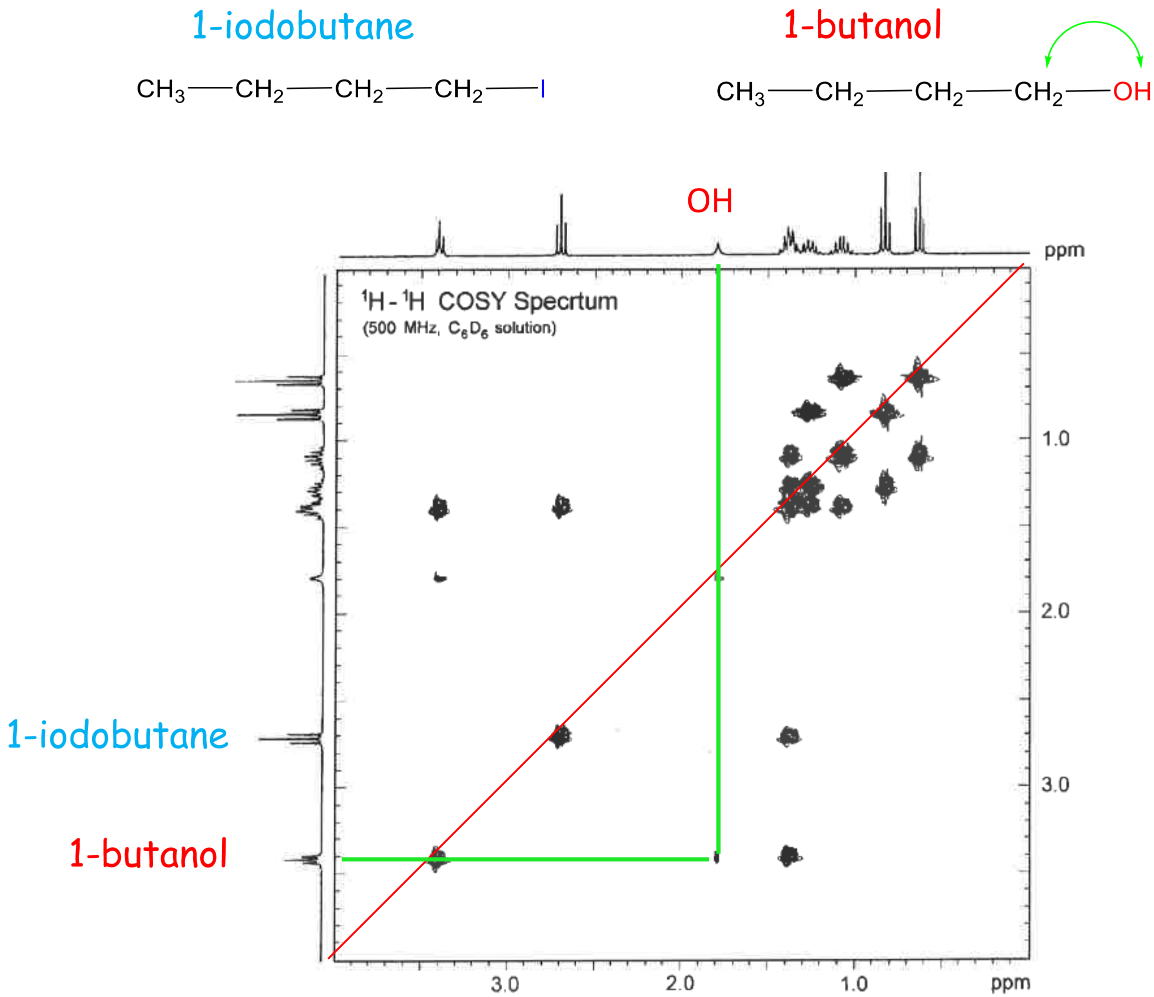
$CH_3—CH_2—CH_2—CH_2—I$

1-butanol

$CH_3—CH_2—CH_2—CH_2—OH$



Organic Structures from Spectra, Field, Sternhell, Kalman, 4th edition, 2007



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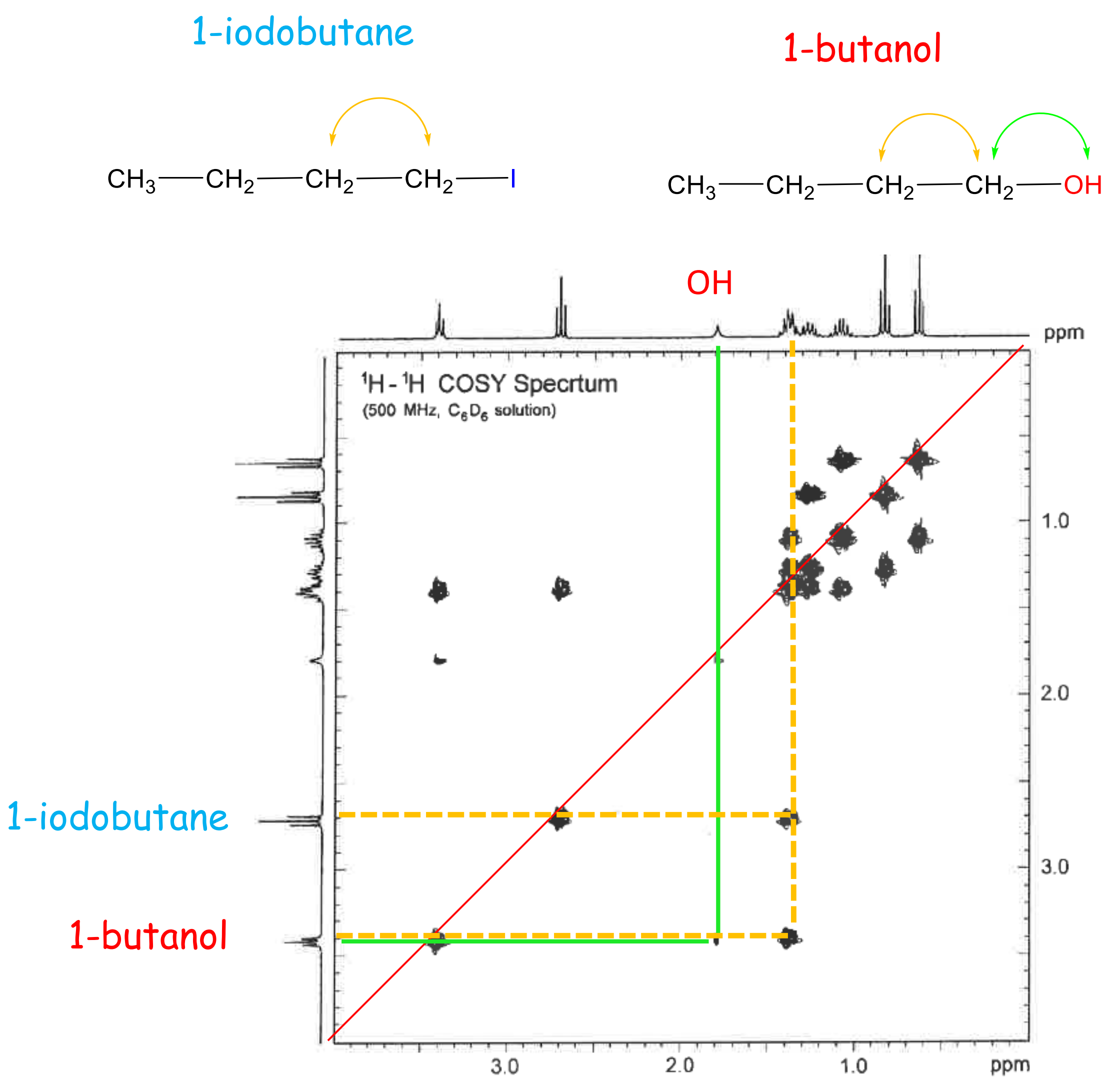

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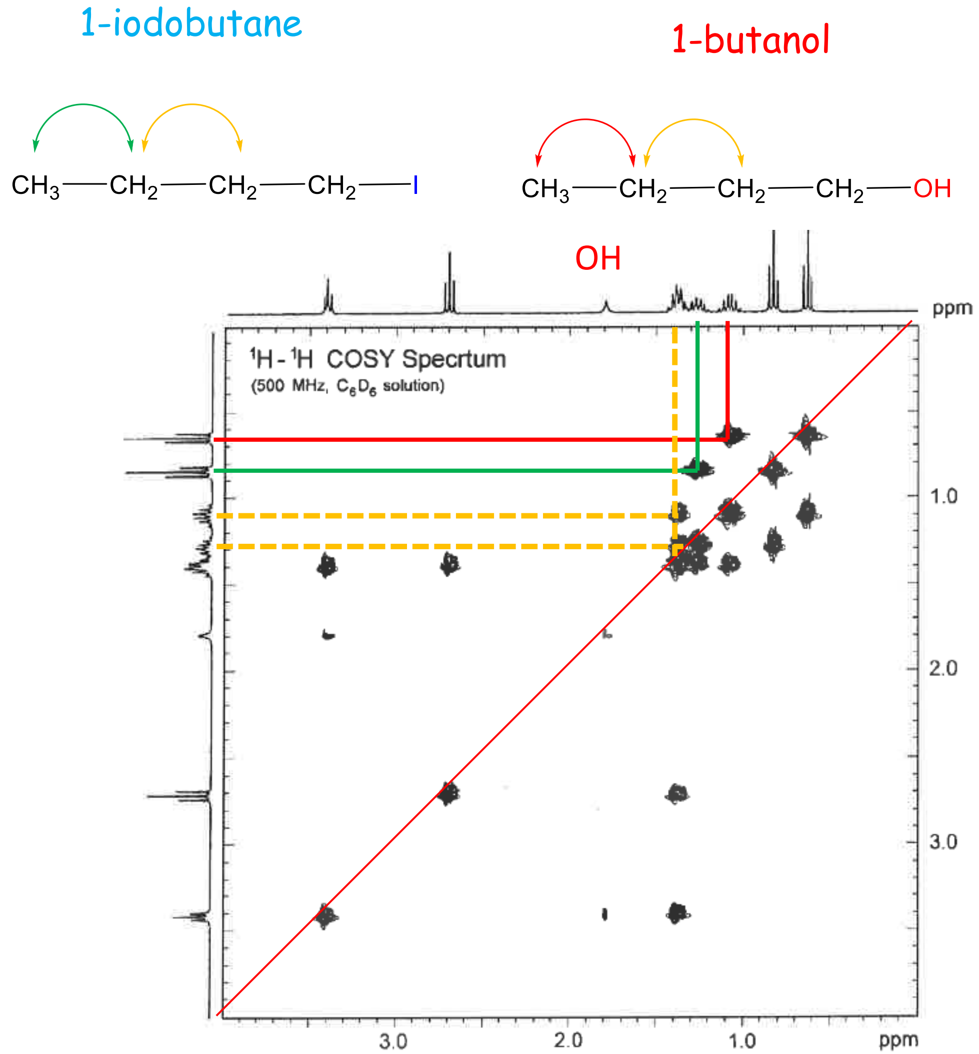

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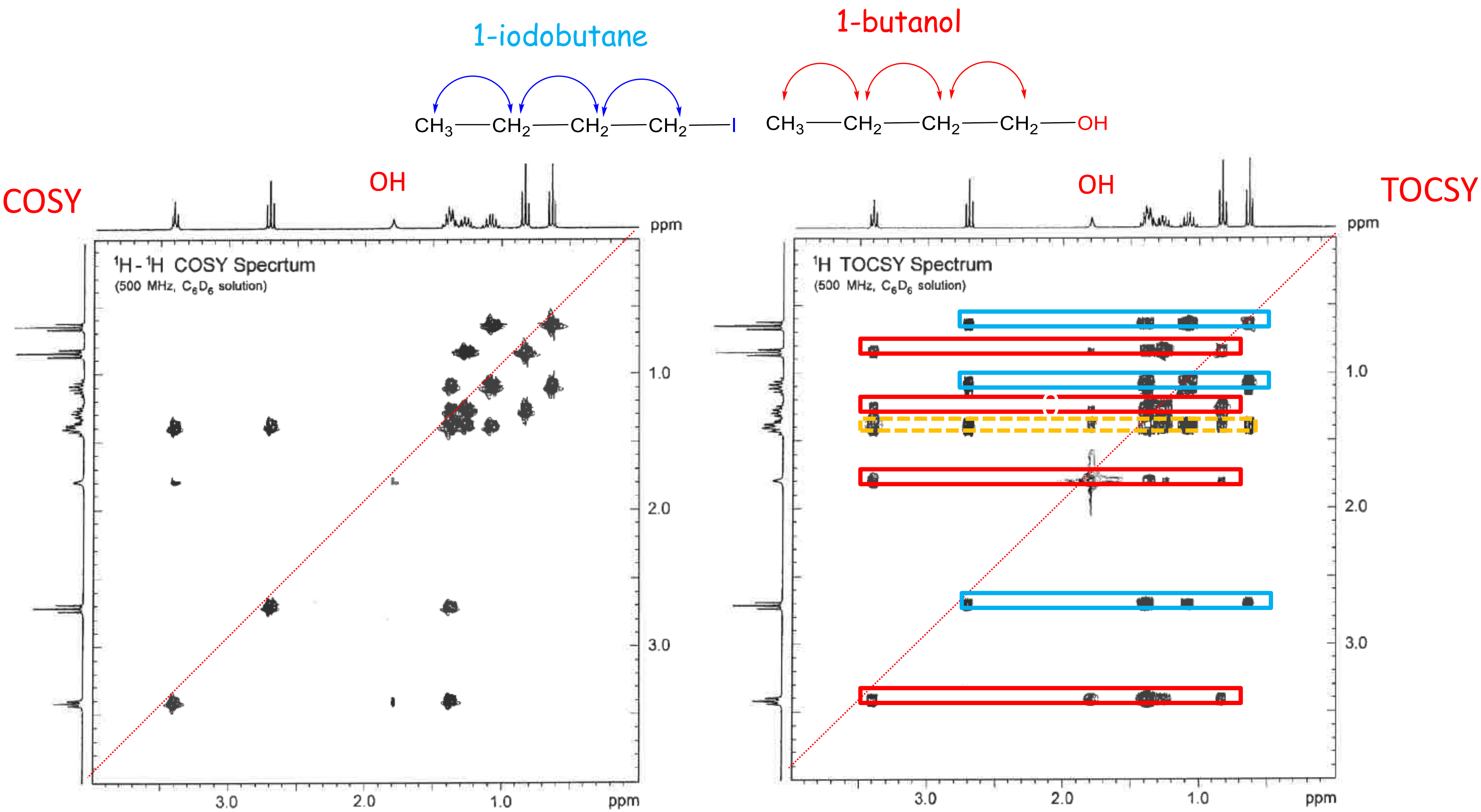
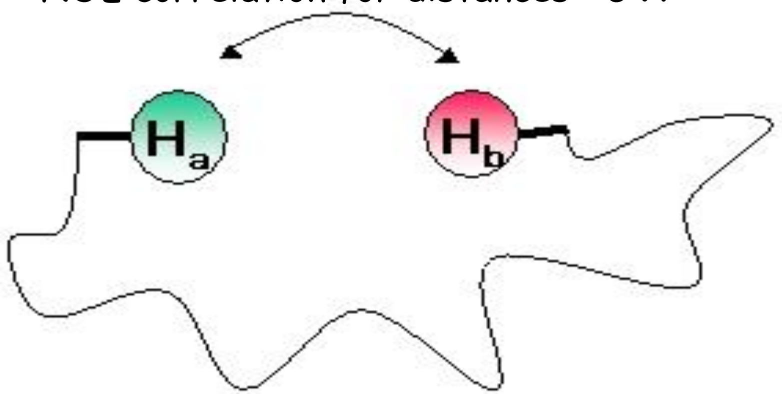


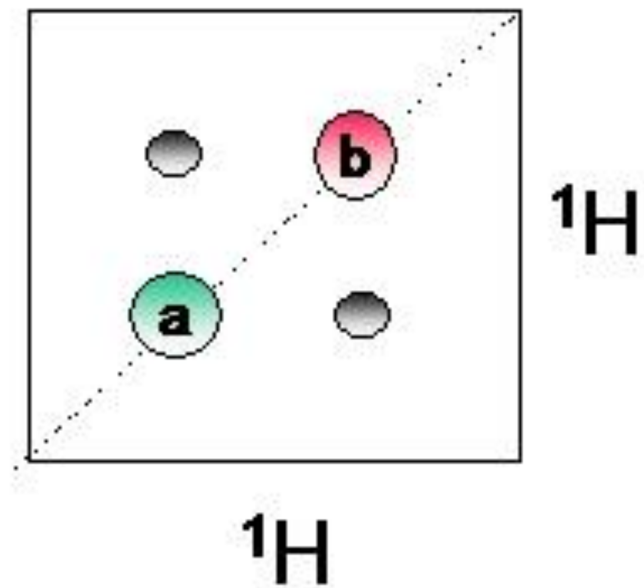
Organic Structures from Spectra, Field, Sternhell, Kalman, 4th edition, 2007

Homonuclear ^1H-^1H

NOESY - Nuclear Overhauser Effect SpectroscopY

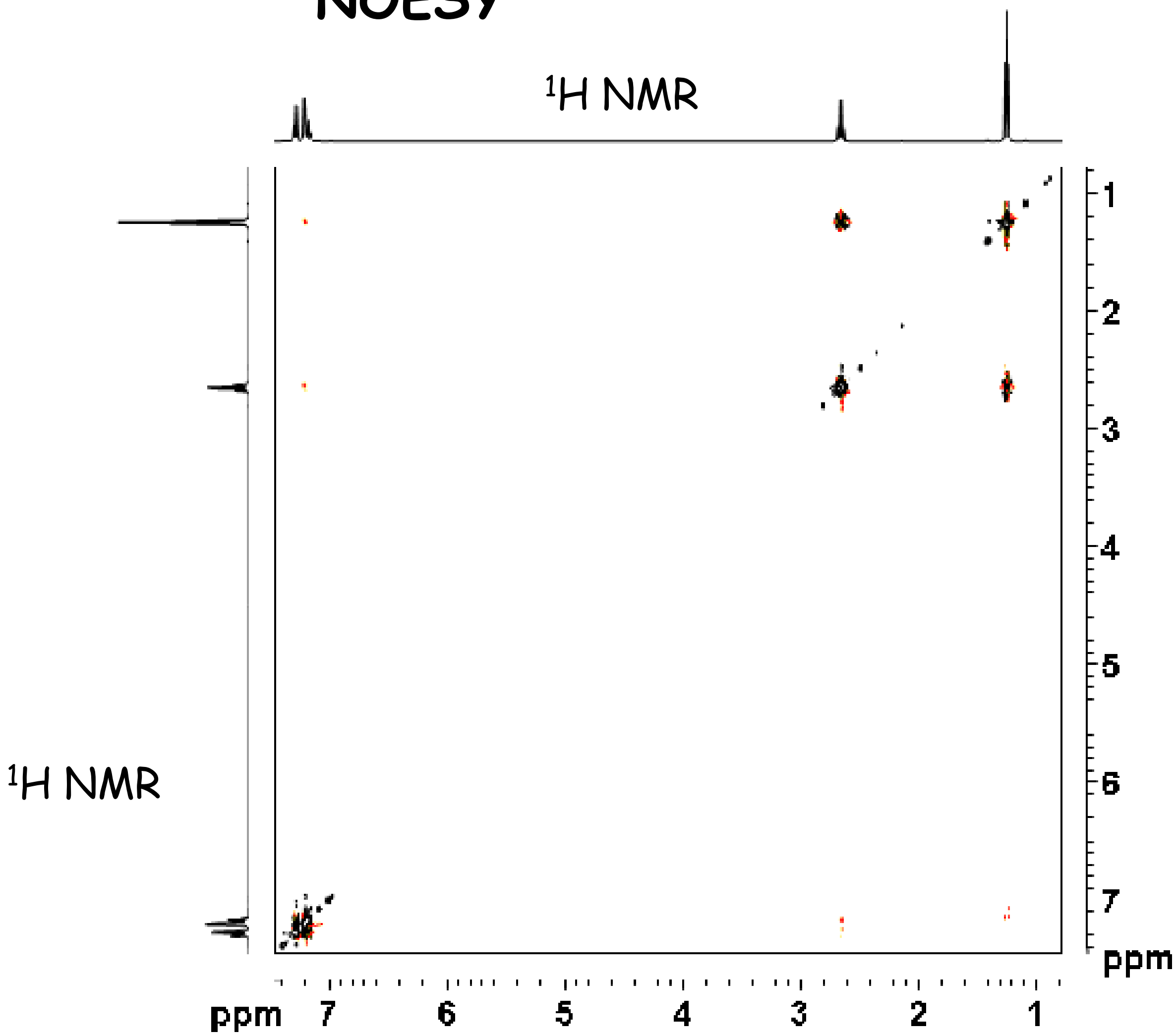
Information – what are the protons that are close in space, distances < 5 Å (H-H coupling trough-space)

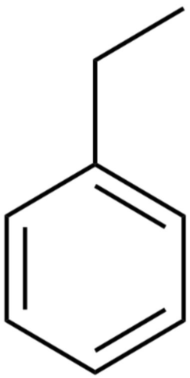




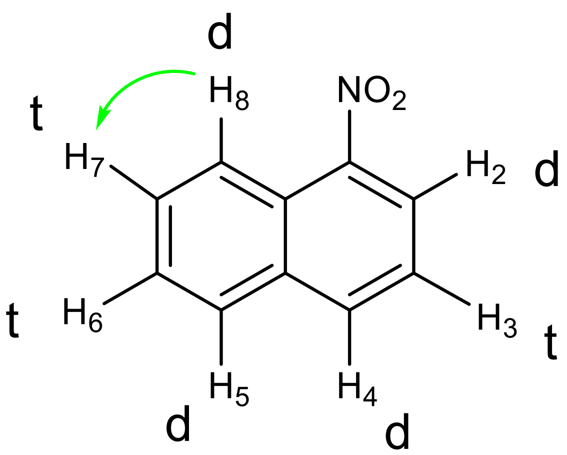
- Diagonal peaks are the protons seeing themselves 1D proton spectrum
- Cross peaks exist only when there is NOE between protons (connect resonances from H that are spacially close)

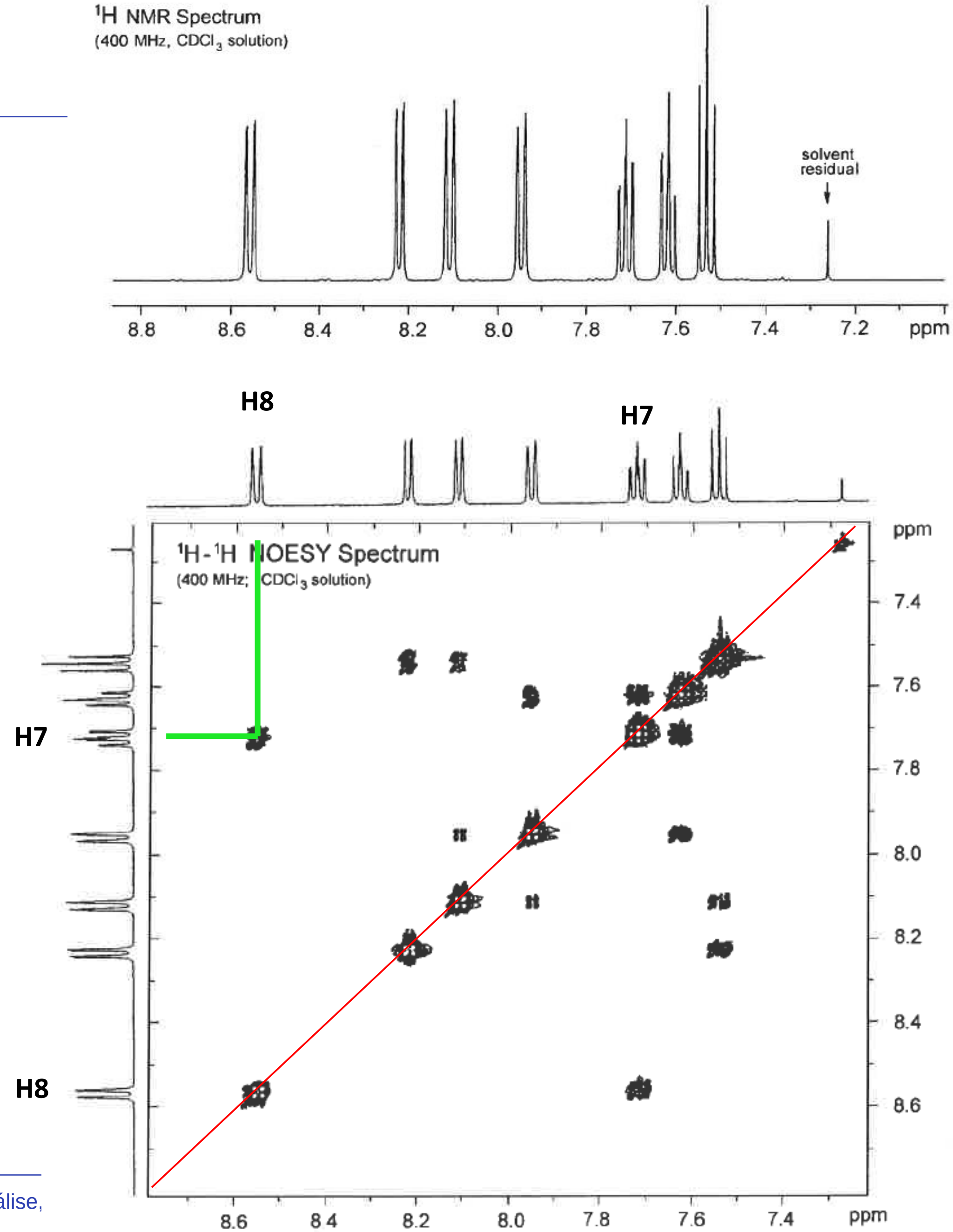
NOESY





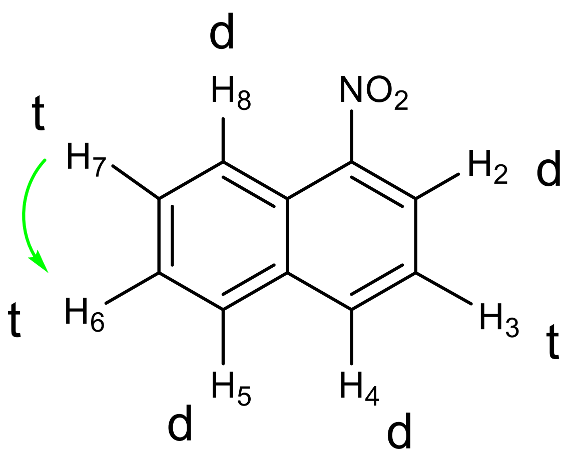
http://chem.ch.huji.ac.il/nmr/

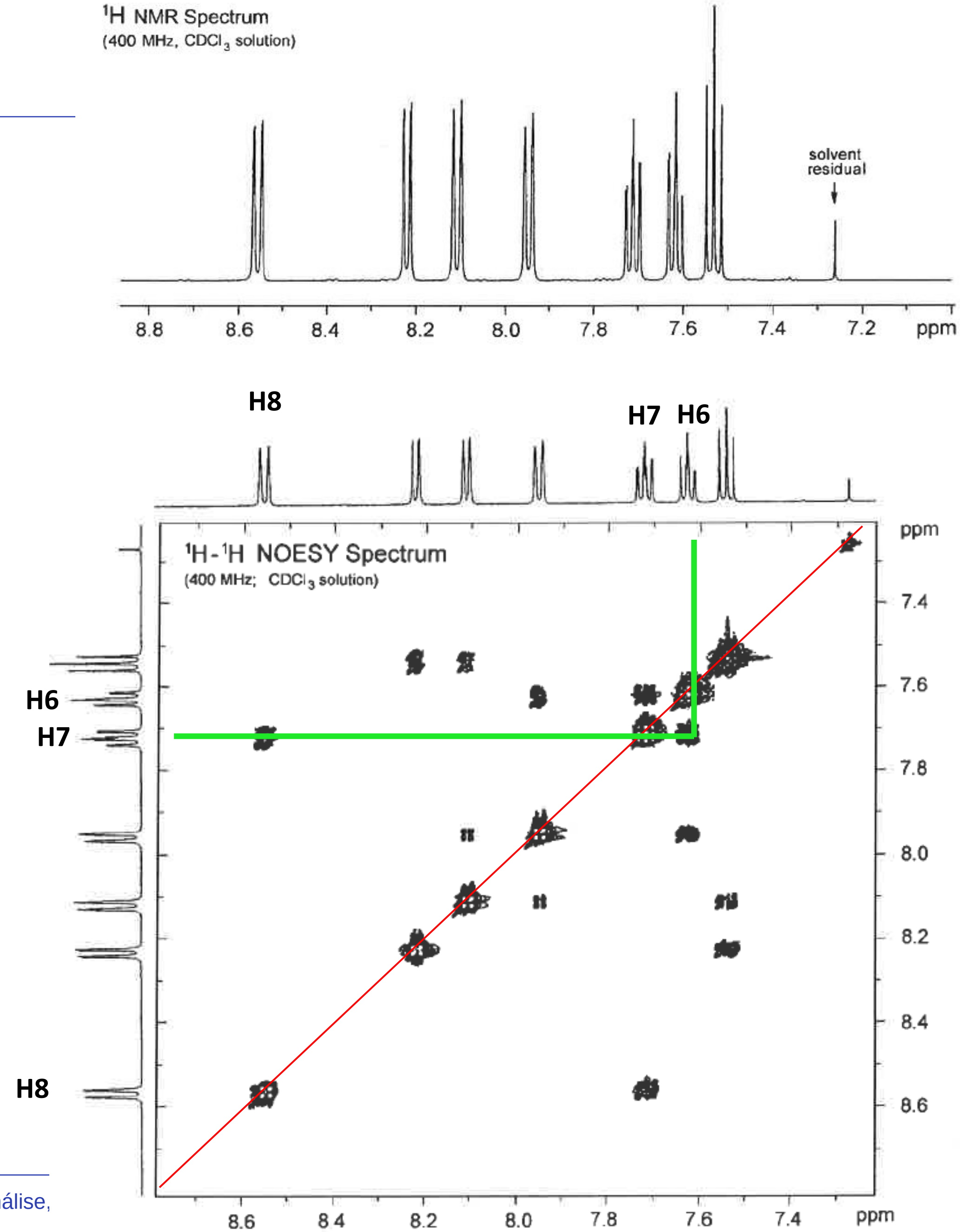


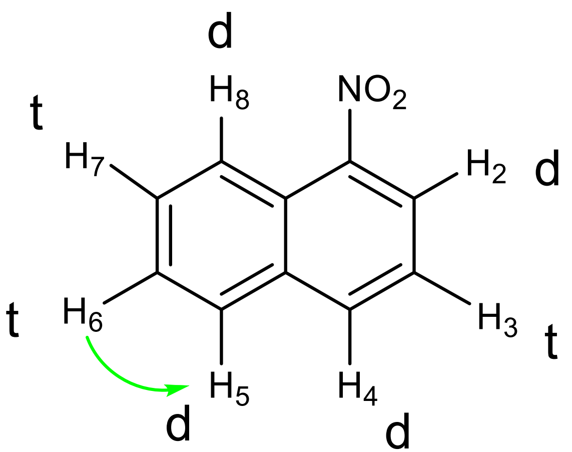


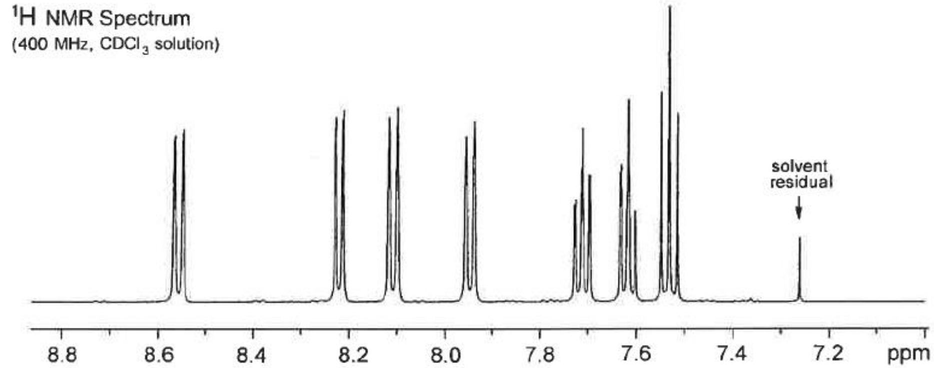
Organic Structures from Spectra,
Field, Sternhell, Kalman, 4th edition, 2007

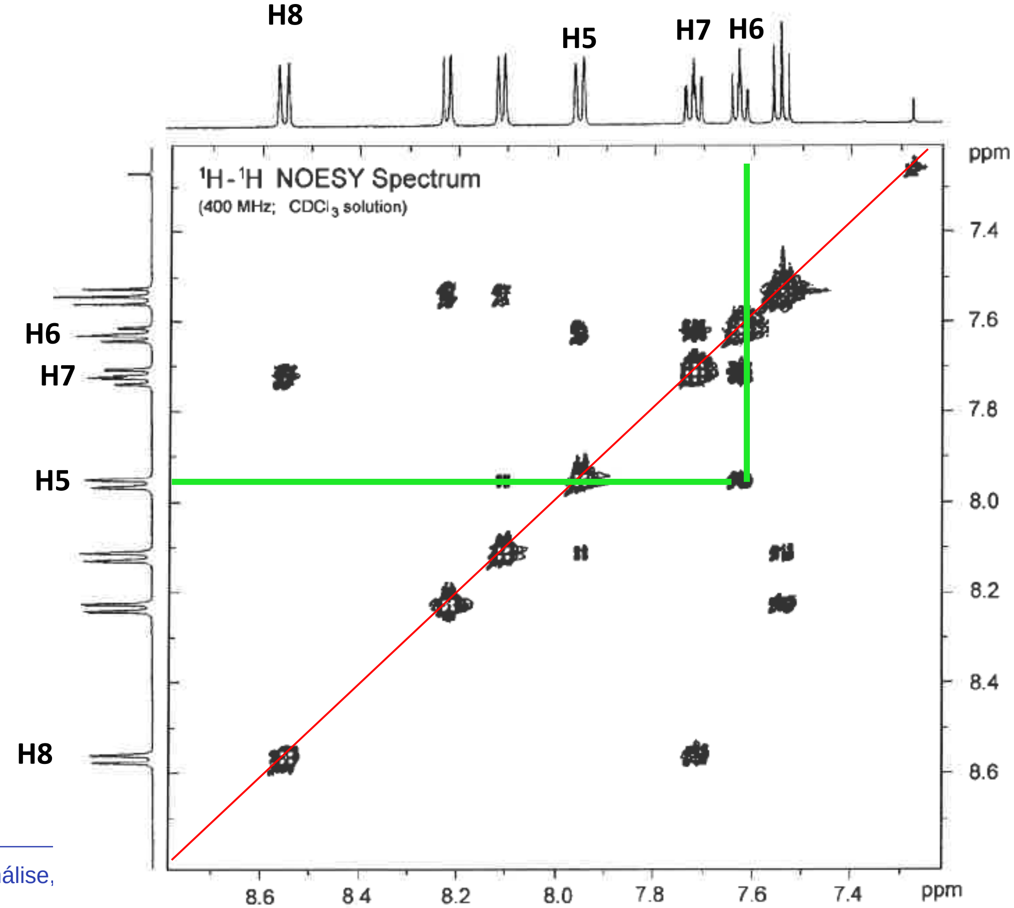
Ciências ULisboa



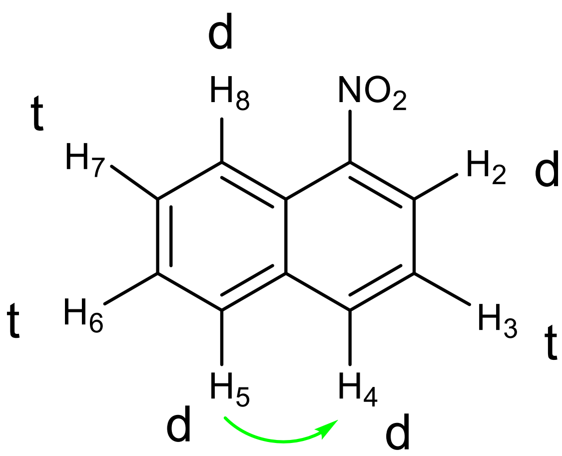


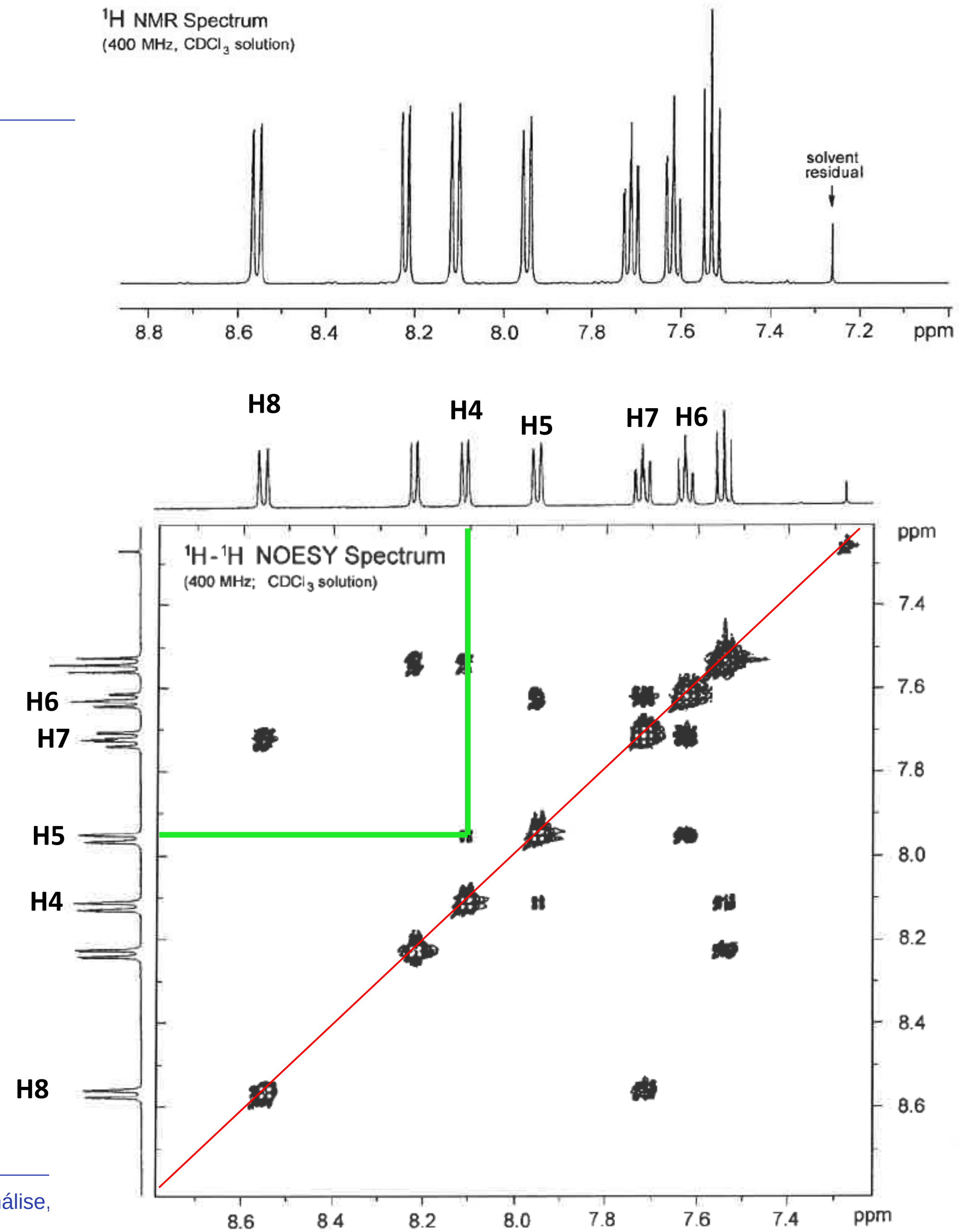

d
H8
NO2
t
H7
H2
d
t
H6
H3
t
H5
H4
d
d


1H NMR Spectrum
(400 MHz, CDCl3 solution)
solvent residual
8.8
8.6
8.4
8.2
8.0
7.8
7.6
7.4
7.2
ppm

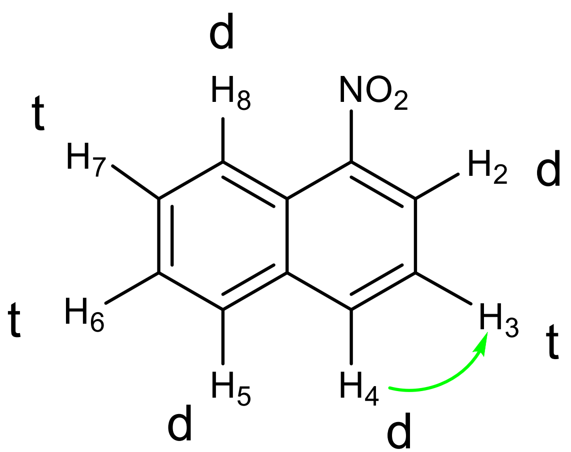

H8
H5
H7
H6
1H-1H NOESY Spectrum
(400 MHz; CDCl3 solution)
H6
H7
H5
H8
ppm
7.4
7.6
7.8
8.0
8.2
8.4
8.6
8.6
8.4
8.2
8.0
7.8
7.6
7.4
ppm

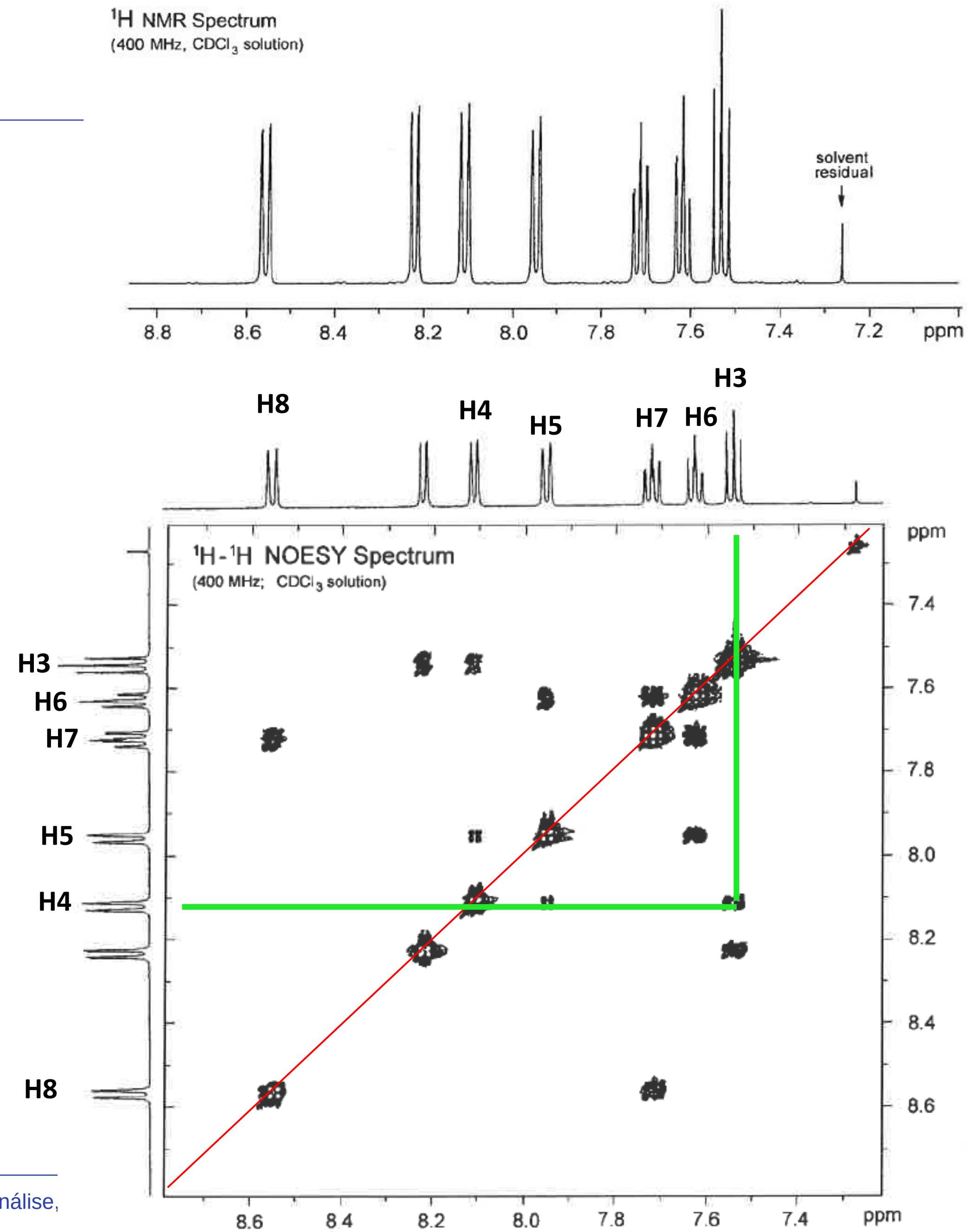
Ciências ULisboa

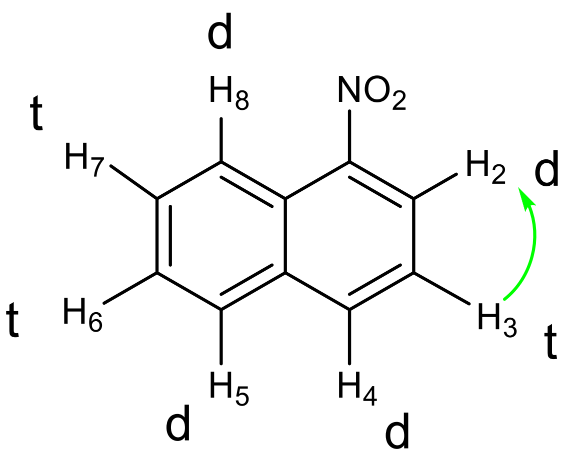




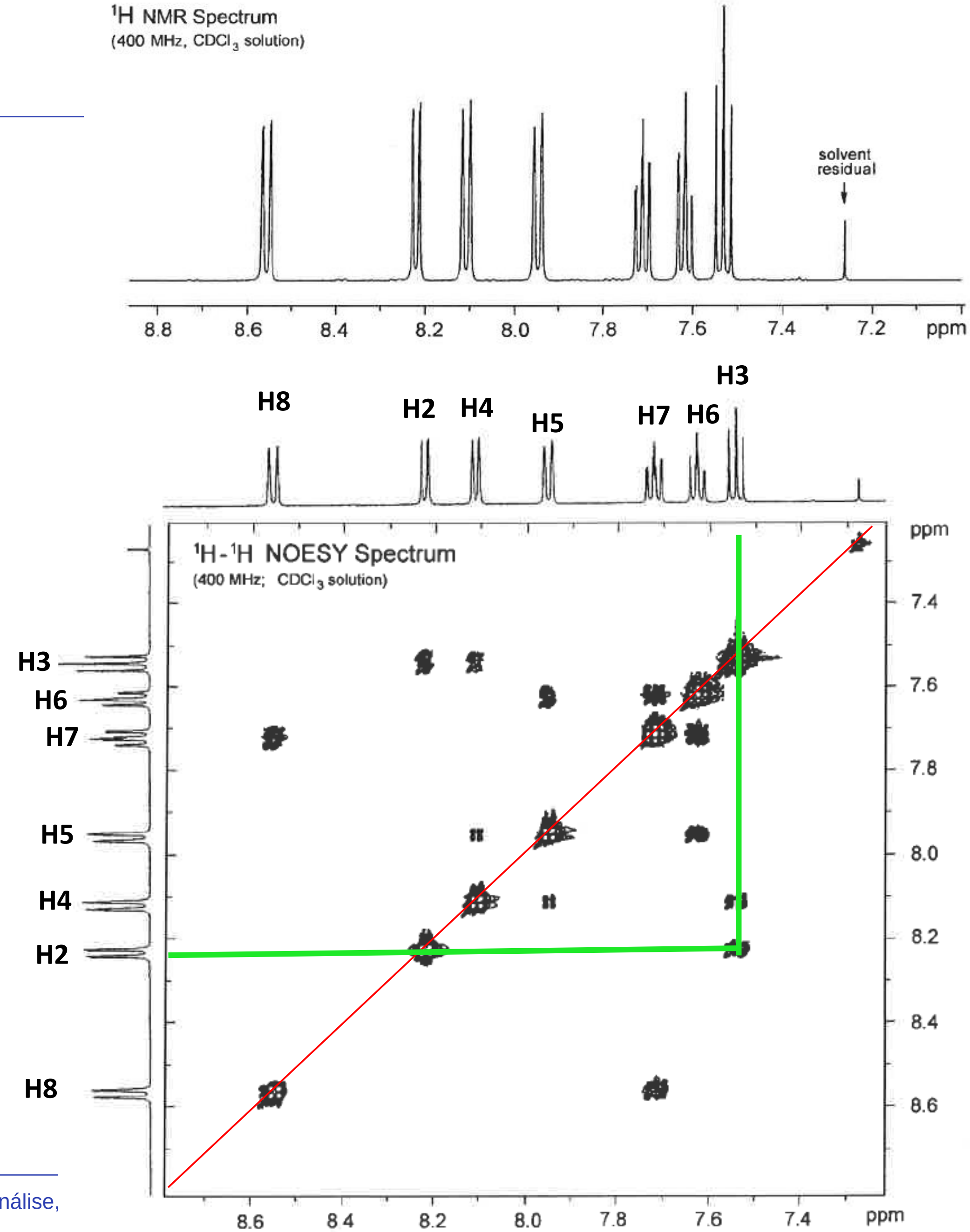
Ciências ULisboa





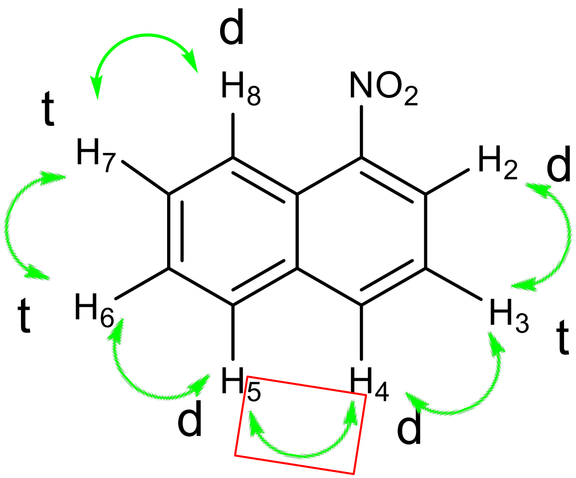


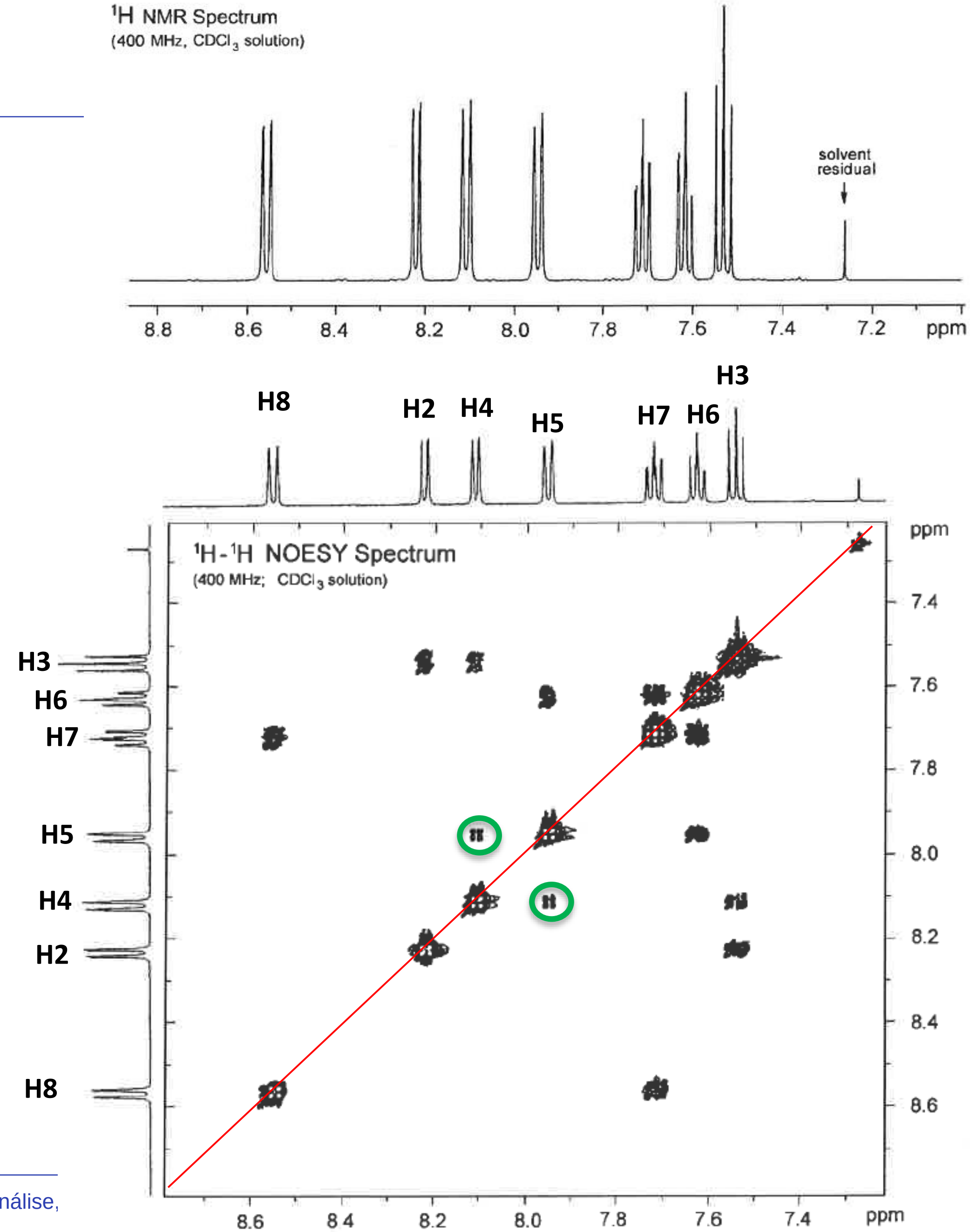
d
H8
NO2
t
H7
H2
d
t
H6
H3
t
H5
H4
d
d

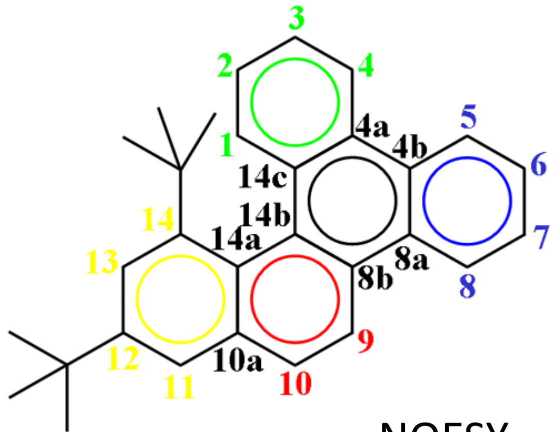


1H NMR Spectrum
(400 MHz, CDCl3 solution)
solvent residual
8.8
8.6
8.4
8.2
8.0
7.8
7.6
7.4
7.2
ppm
H8
H2
H4
H5
H7
H6
H3
1H-1H NOESY Spectrum
(400 MHz; CDCl3 solution)
H3
H6
H7
H5
H4
H2
H8
ppm
7.4
7.6
7.8
8.0
8.2
8.4
8.6
8.6
8.4
8.2
8.0
7.8
7.6
7.4
ppm

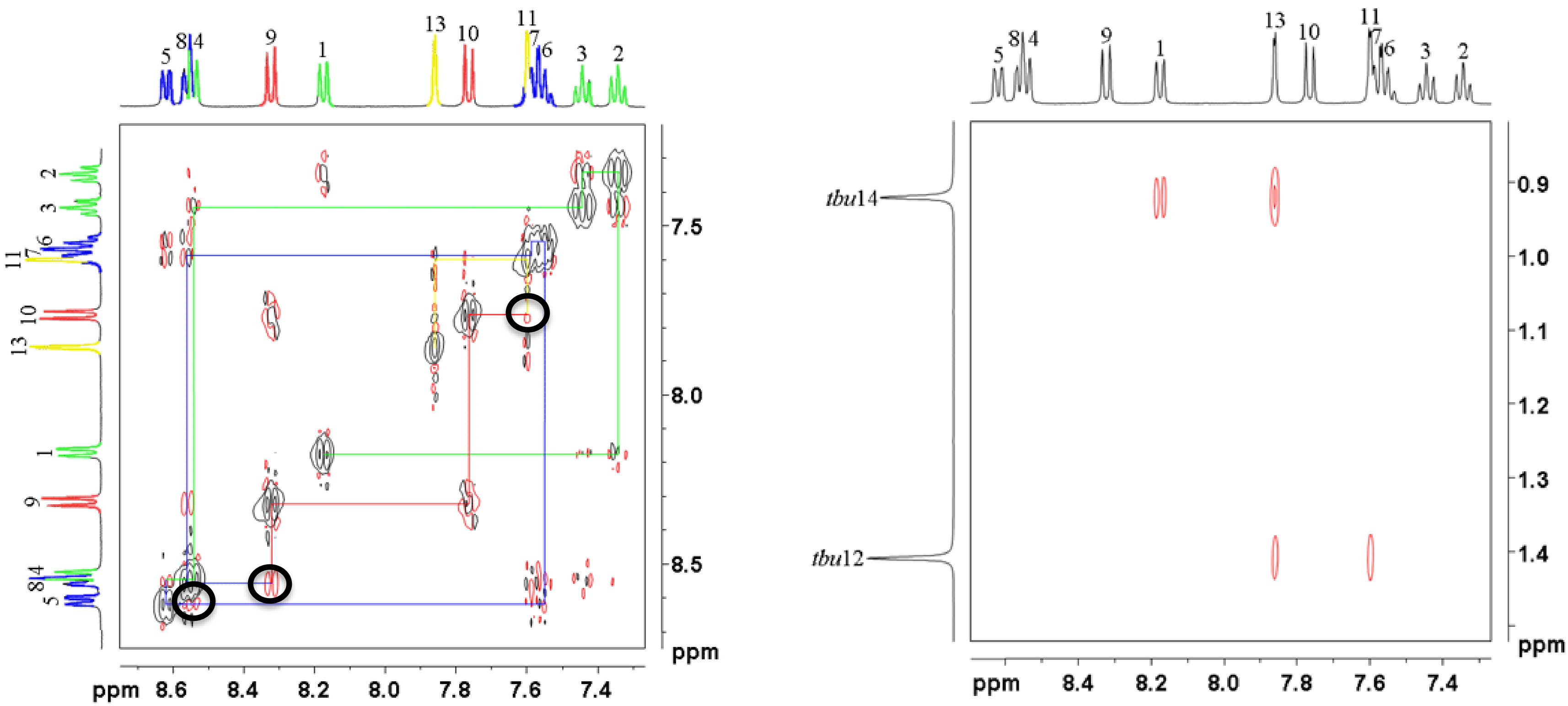
Ciências ULisboa







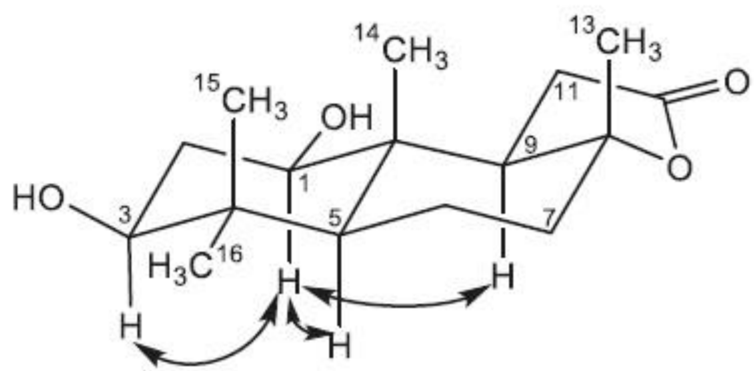
NOESY



http://chem.ch.huji.ac.il/nmr/

NOESY

H-1 *ax* or eq?



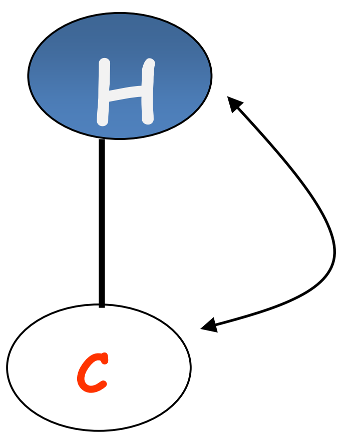
http://www.scielo.org.mx/img/revistas/rmcf/v45n4/a9f3.jpg

Heteronuclear ^{13}C-^{1}H

HSQC – **H**eteronuclear **S**ingle-**Q**uantum **C**orrelation Spectroscopy
HMQC - **H**eteronuclear **M**ultiple-**Q**uantum **C**orrelation Sepectroscopy

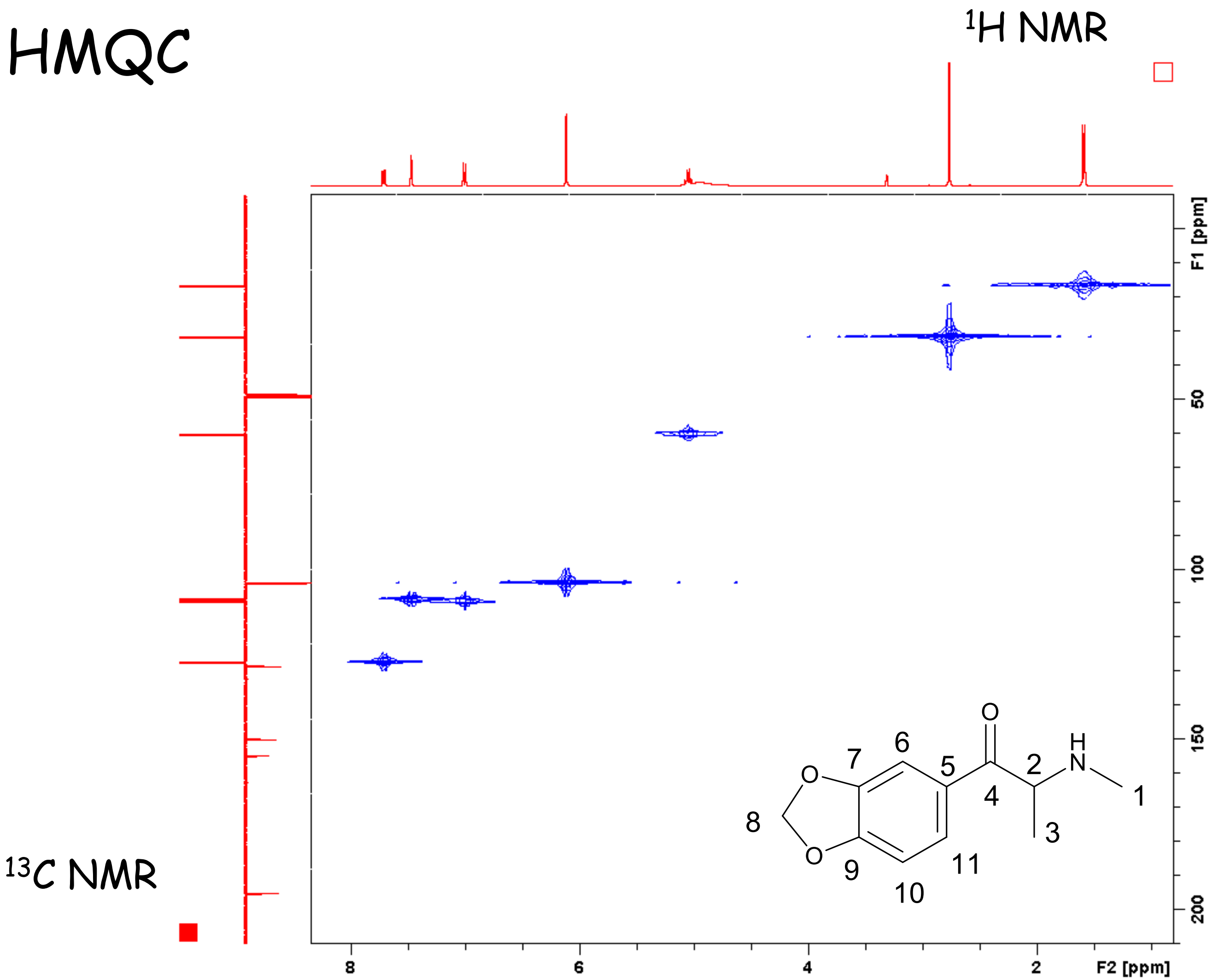
Information – what are the proton directly attached to a specific carbon (one bond correlation C-H)

<> ^{1}J C-H



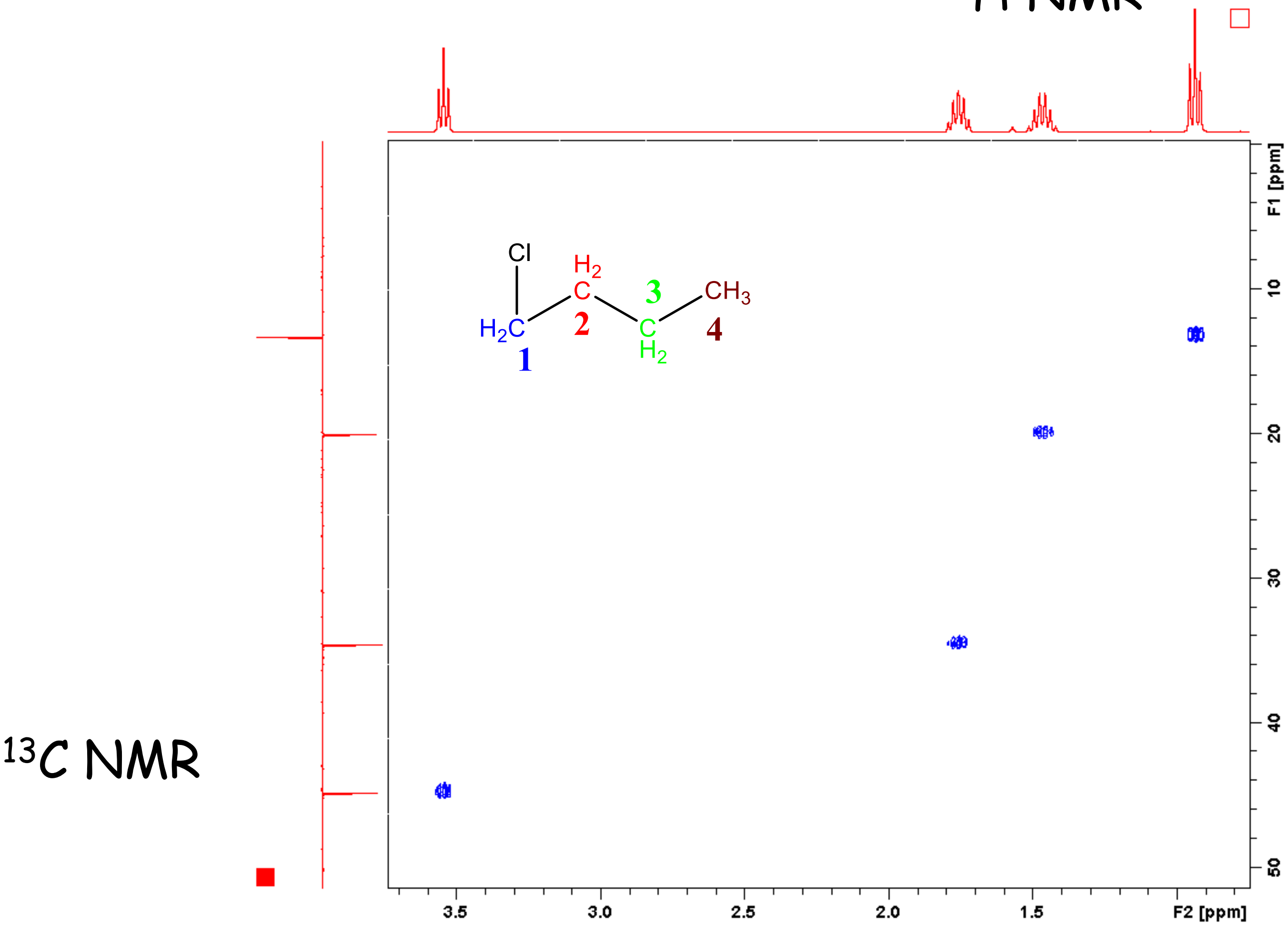
- Cross peaks exist only when there is a C-H bond (^{1}J C-H)

HMQC

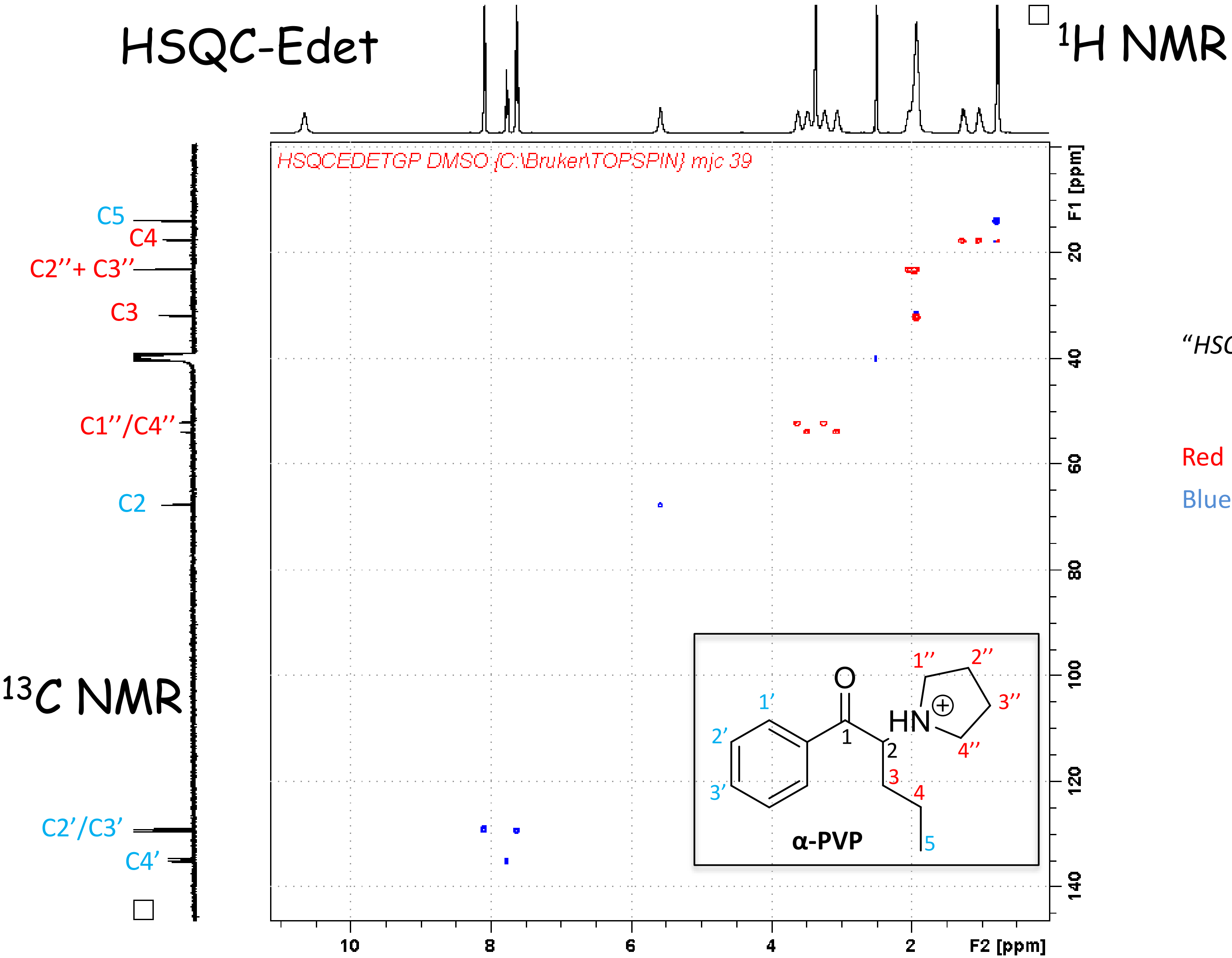


^{1}H NMR
^{13}C NMR
F1 [ppm]
50
100
150
200
F2 [ppm]
8
6
4
2
O
H
N
1
2
3
4
5
6
7
8
9
10
11
O
O

HSQC


^{1}H NMR
^{13}C NMR
Cl
H_2C
H_2 C
C H_2
CH_3
1
2
3
4
F1 [ppm]
10
20
30
40
50
3.5
3.0
2.5
2.0
1.5
F2 [ppm]

HSQC-Edet

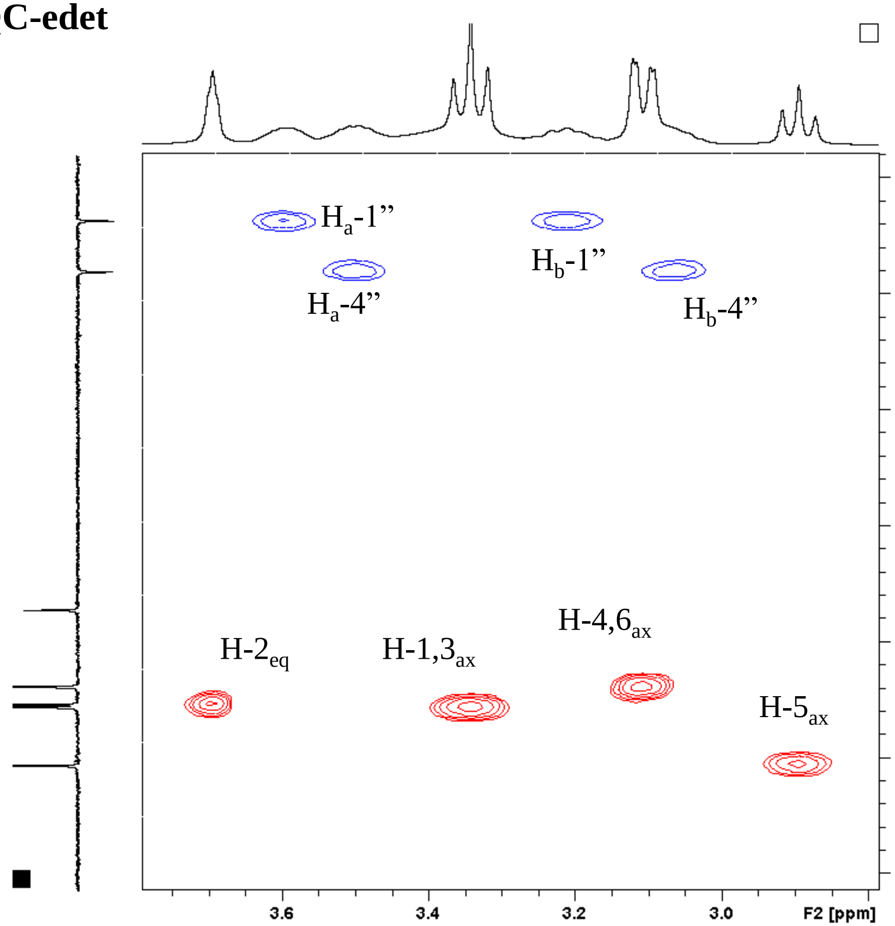


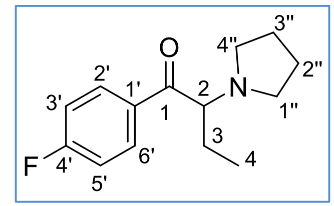
"HSQC-Dept135"

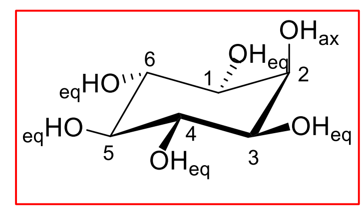
Red (negative)- CH_2

Blue (negative)- CH or CH_3

HSQC-edet







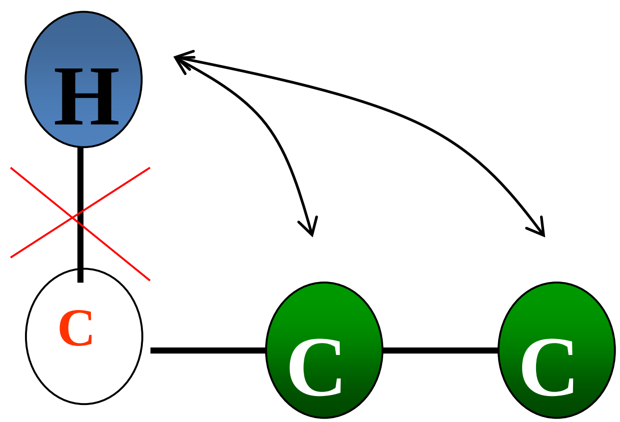
H. Gaspar et al, *4F-PBP , a new substance of abuse: structural characterization and purity NMR profiling*, ***Forensic. Science International*** **2015**, (252) 168

Heteronuclear ^{13}C-^{1}H

HMBC - Heteronuclear **M**ultiple **B**ond Quantum

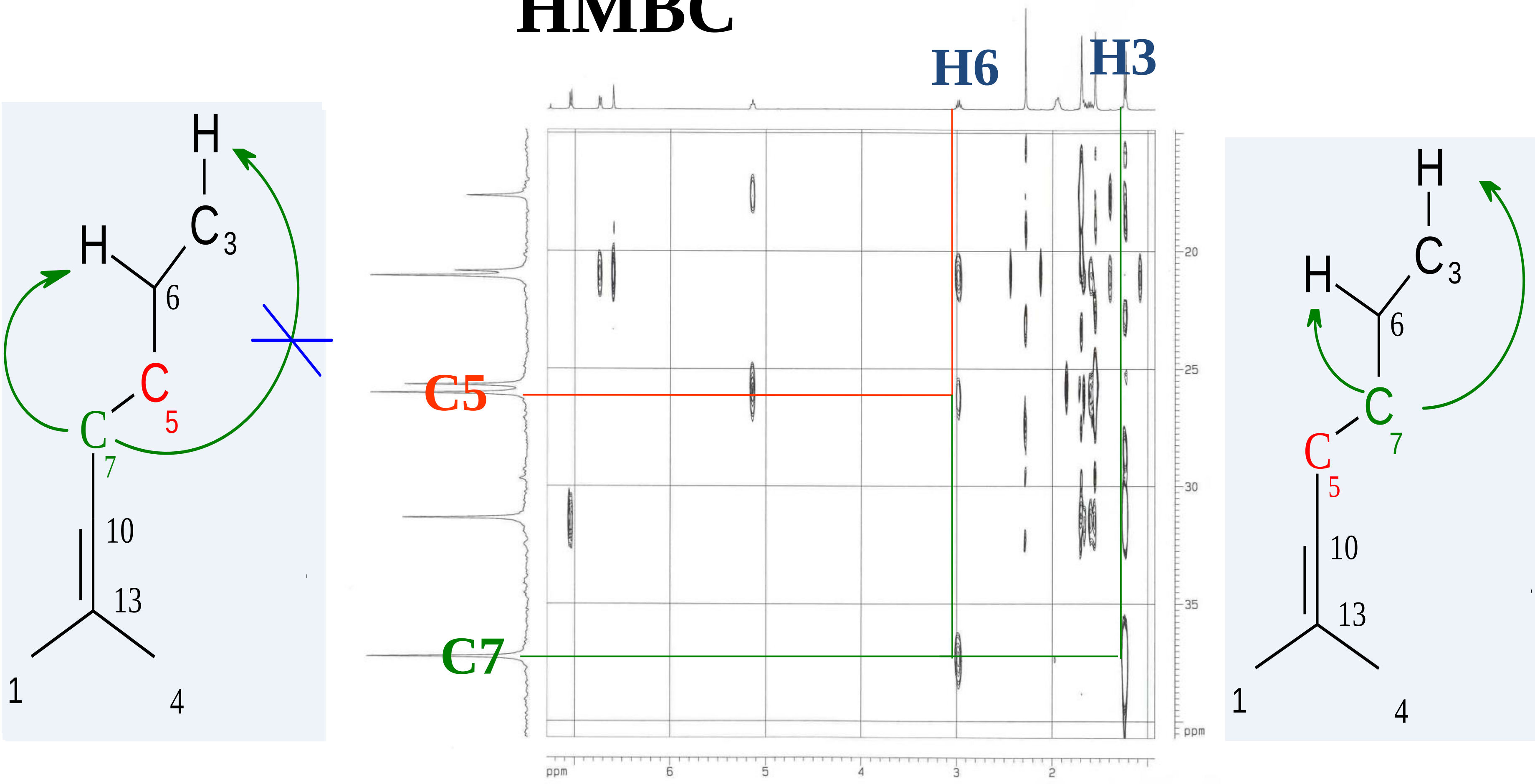
Information – what are the protons that correlate to carbons at 2 and 3 bonds distance (long range coupling)

<> ^{2}J C-H and ^{3}J C-H

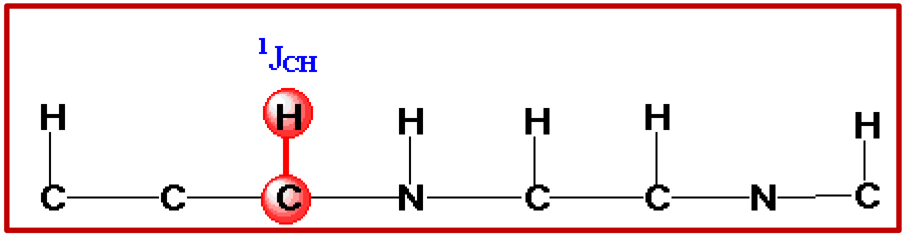


- Cross peaks exist only when there is a proton-carbon correlation (^{2}J or ^{3}J)

HMBC

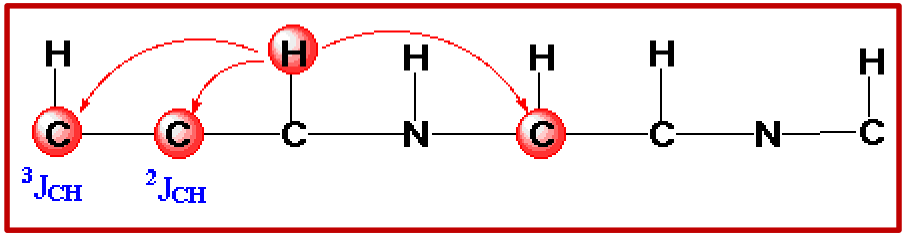


Heteronuclear ^{13}C-^{1}H



HMQC
HSQC

The correlations is used to establish C-H connectivity
(How many types of **C**, **CH**, **CH_2**, **CH_3**) ⇔ also from DEPT



HMBC

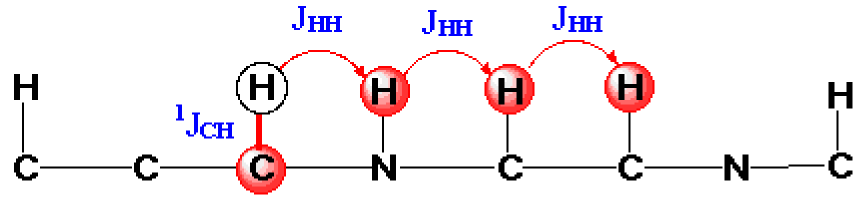
The correlations is used to establish C-C connectivity

Heteronuclear ^{13}C-^{1}H

HSQC-TOCSY or HMQC-TCOSY

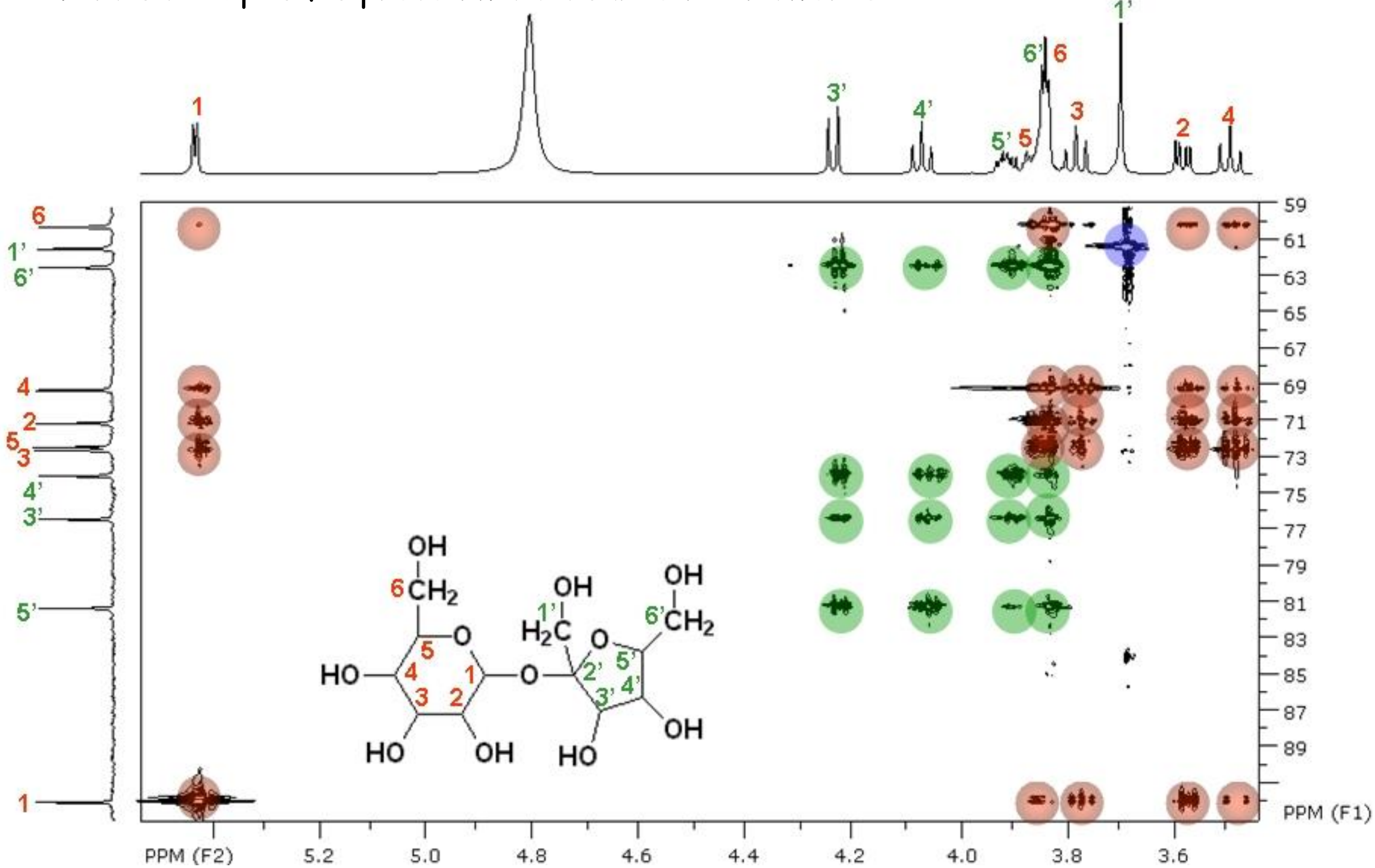
2D TOCSY that has been resolved into the carbon dimension

Information – Proton "coupling networks" with a specific carbone.



Horizontal - Proton "coupling networks" with a specific carbone

Vertical – Specific proton with a carbone "networks"



http://www.columbia.edu/cu/chemistry/groups/nmr/hsqctocsy4.jpg