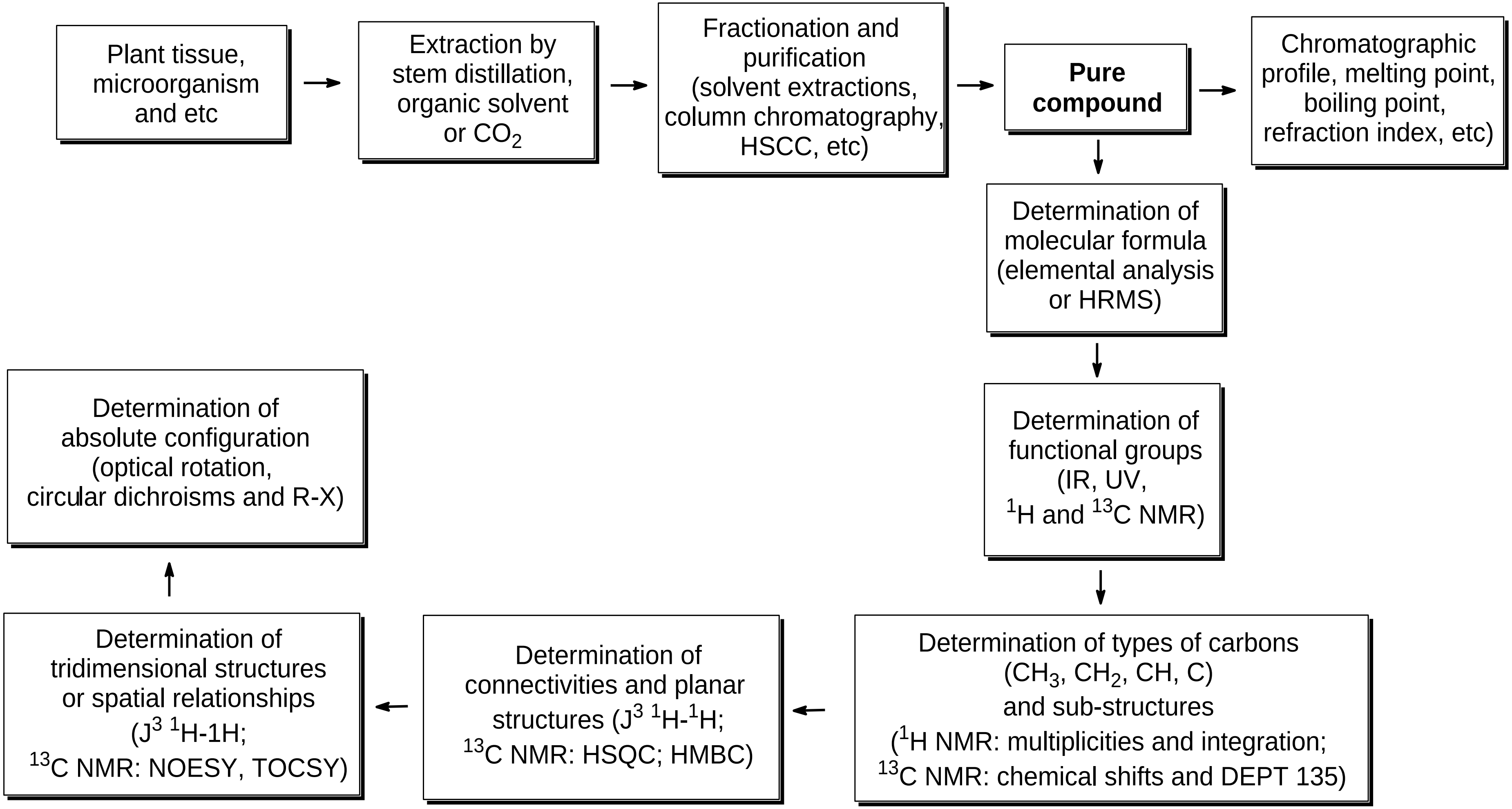
QFL-0314

19/11/19

Ressonância Magnética Nuclear

General scheme for structural elucidation of natural compounds



Plant tissue, microorganism and etc

Extraction by stem distillation, organic solvent or CO_2

Fractionation and purification (solvent extractions, column chromatography, HSCC, etc)

Pure compound

Chromatographic profile, melting point, boiling point, refraction index, etc)

Determination of molecular formula (elemental analysis or HRMS)

Determination of functional groups (IR, UV, ^{1}H and ^{13}C NMR)

Determination of types of carbons (CH_3, CH_2, CH, C) and sub-structures (^{1}H NMR: multiplicities and integration; ^{13}C NMR: chemical shifts and DEPT 135)

Determination of connectivities and planar structures (J^3 ^{1}H-^{1}H; ^{13}C NMR: HSQC; HMBC)

Determination of tridimensional structures or spatial relationships (J^3 ^{1}H-1H; ^{13}C NMR: NOESY, TOCSY)

Determination of absolute configuration (optical rotation, circular dichroisms and R-X)

Why to learn NMR?

Organic chemistry

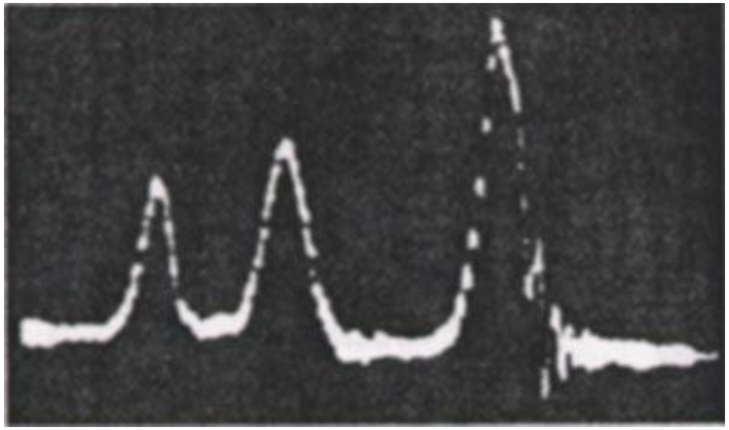
- Structural (chemical) elucidation.
- Natural product chemistry.
- Synthetic organic chemistry.
- Study of dynamic processes and kinetics.
- Study of equilibrium (chemical or structural).
- Structural (three-dimensional) studies.

Biochemistry and medicine

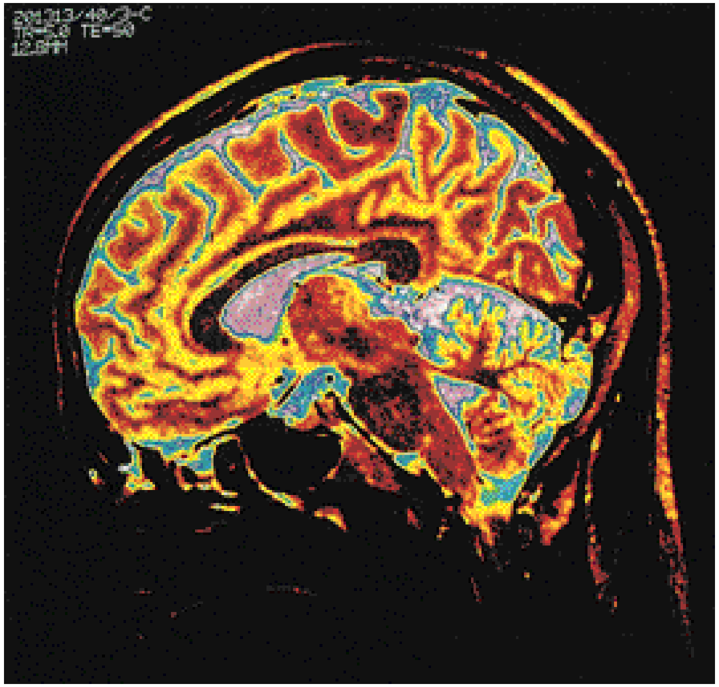
- Protein, protein/DNA complexes.
- Polysaccharides.
- Drug design and structure activity relationships.
- MRI (magnetic resonance imaging)

Nuclear magnetic resonance

1952
^{1}H NMR spectrum of EtOH



1980
First body MRI
(magnetic resonance imaging)



X-ray Crystallography


William Bragg


Lawrence Bragg

Physics, 1915

Ultramicroscope


Richard Zsigmondy

Chemistry, 1925

Nuclear Magnetic Resonance


Felix Bloch


E. M. Purcell

Physics, 1952

Phase Contrast Microscope


Frits Zernike

Physics, 1953

Large-Array Radio Telescopes


Martin Ryle

Physics, 1974

Electron Microscope


Ernst Ruska

Physics, 1986

Scanning Tunneling Microscope


Gerd Binnig


Heinrich Rohrer

Physics, 1986

Computer Assisted Tomography


Allan Cormack


Godfrey Hounsfield

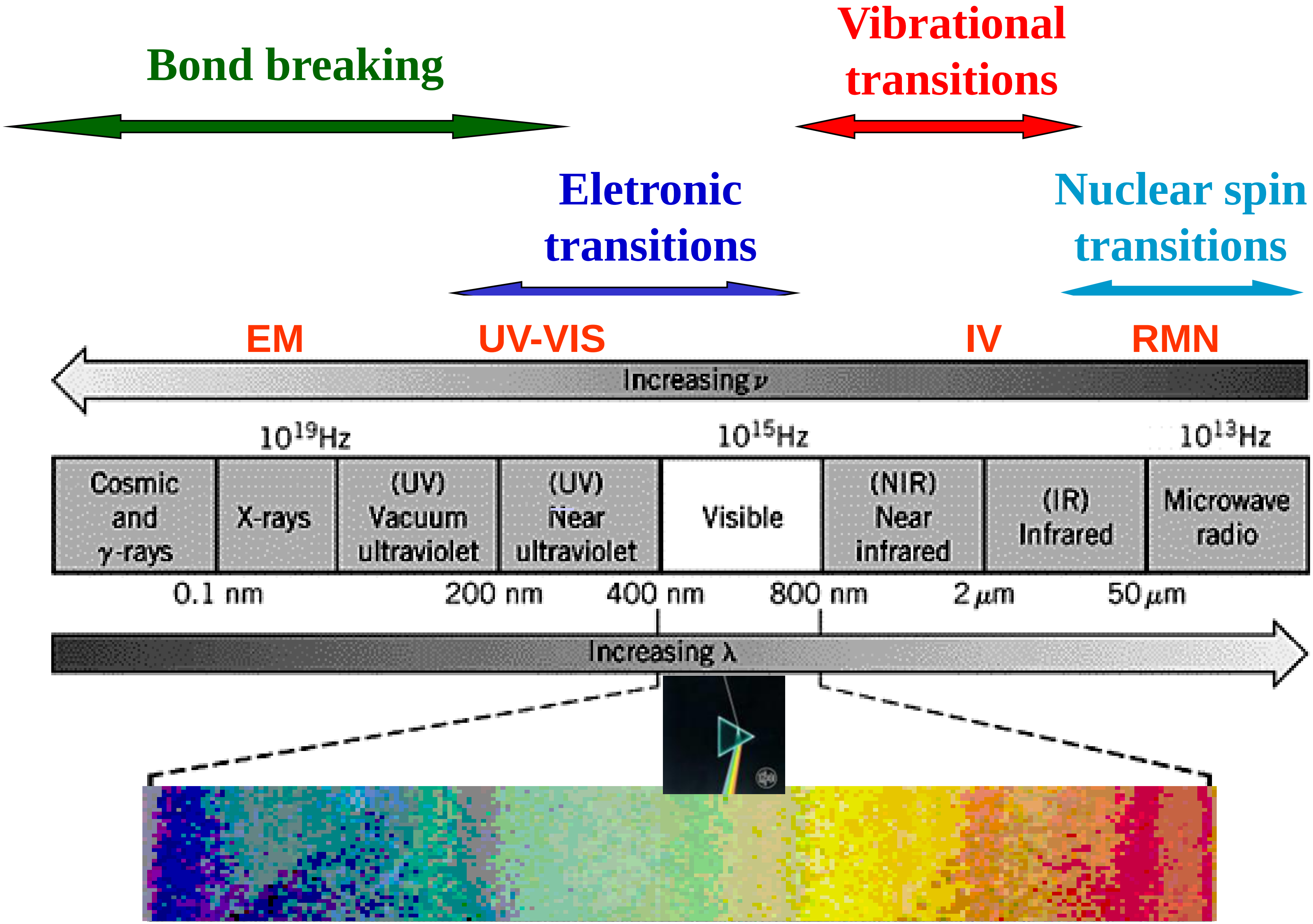
Physiology or Medicine, 1979

Magnetic Resonance Imaging


Paul Lauterbur


Peter Mansfield

Physiology or Medicine, 2003



Bond breaking

Vibrational transitions

Eletronic transitions

Nuclear spin transitions

EM

UV-VIS

IV

RMN

Quantic number for common nuclei

^{1}H and ^{13}C isotopes have spins and are ubiquitous (*) in organic compounds

Element	^{1}H	^{2}H	^{12}C	^{13}C	^{14}N	^{16}O	^{17}O	^{19}F
Nuclear Spin Quantum No (I)	1/2	1	0	1/2	1	0	5/2	1/2
No. of Spin States	2	3	0	2	3	0	6	2

Atomic mass	1	2	12	13	14	16	17	19
Atomic number	1	1	6	6	7	8	8	9

*Ubiquitous: onipresente

Nuclear Magnetic Resonance

Nuclear spin

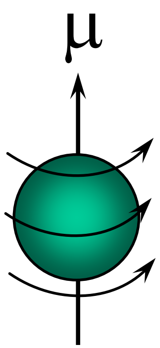
$$\mu = \gamma I \hbar$$

μ - magnetic moment

γ - gyromagnetic constant

I - spin quantum number

$\hbar$ - Planck's constant

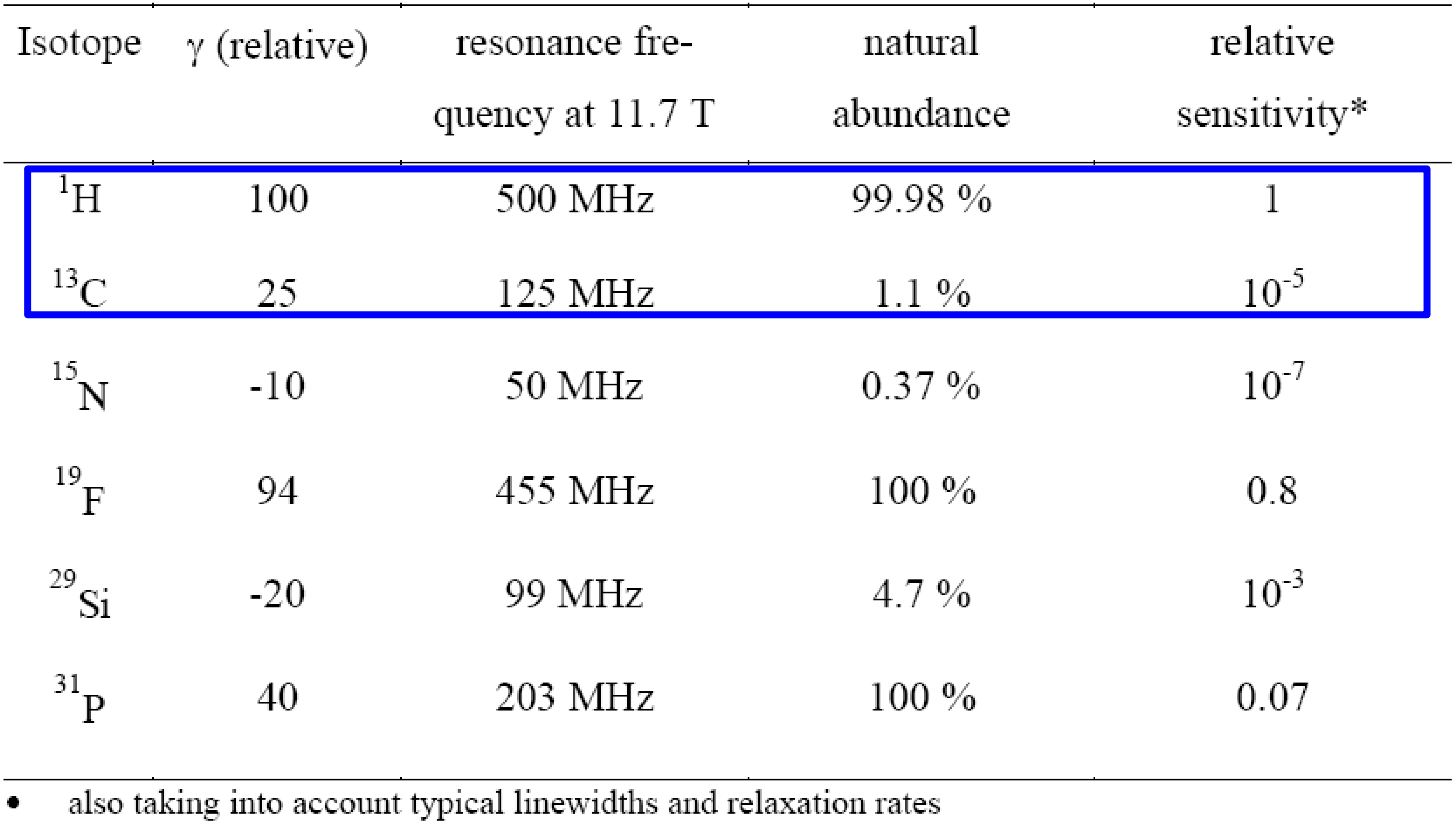


I is a property of the nucleus

Mass #	Atomic #	I
Odd	Even or odd	1/2, 3/2, 5/2,…
Even	Even	0
Even	Odd	1, 2, 3

Odd: impar
Even: par

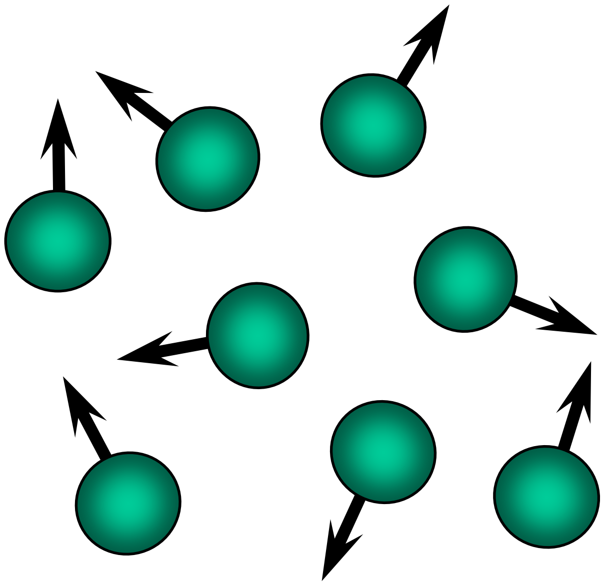
Some characteristics of NMR active nuclei



Isotope	γ (relative)	resonance frequency at 11.7 T	natural abundance	relative sensitivity*
^{1}H	100	500 MHz	99.98 %	1
^{13}C	25	125 MHz	1.1 %	10^{-5}
^{15}N	-10	50 MHz	0.37 %	10^{-7}
^{19}F	94	455 MHz	100 %	0.8
^{29}Si	-20	99 MHz	4.7 %	10^{-3}
^{31}P	40	203 MHz	100 %	0.07

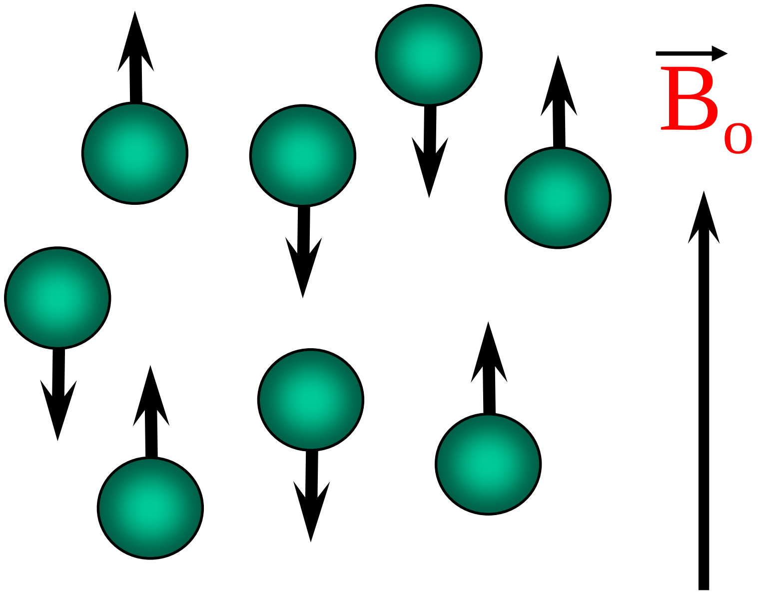
- also taking into account typical linewidths and relaxation rates

Ensemble of Nuclear Spins



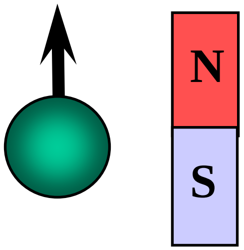
$\vec{B}_o = 0$

Randomly oriented



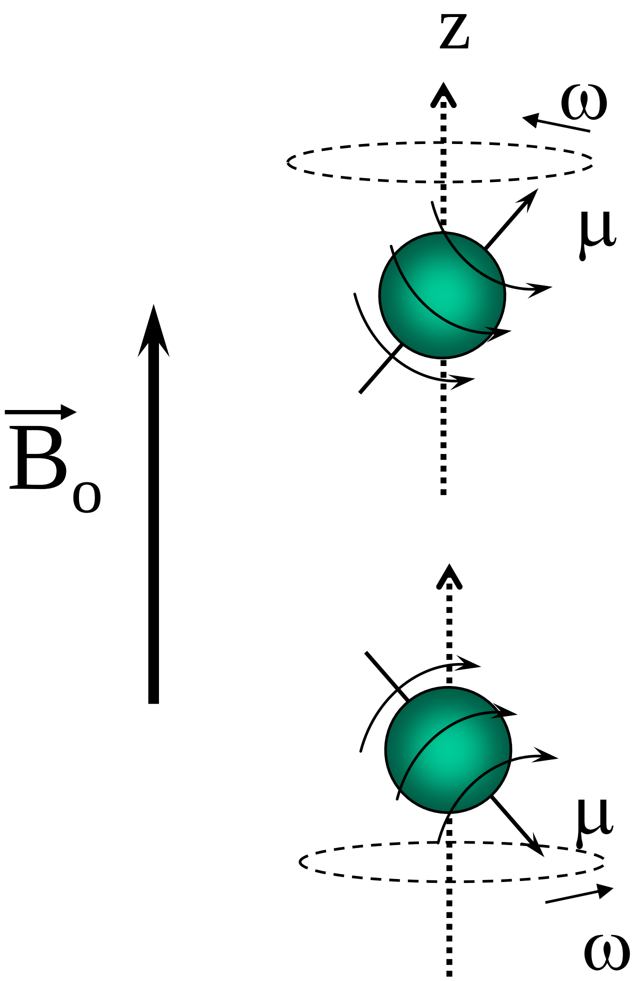
$\vec{B}_o > 0$

Highly oriented



Each nucleus behaves like a bar magnet.

Apply an external magnetic field



$$\boldsymbol{\omega = \gamma B_o = \nu/2\pi}$$

ω - resonance frequency
in radians per second,
(Larmor frequency)

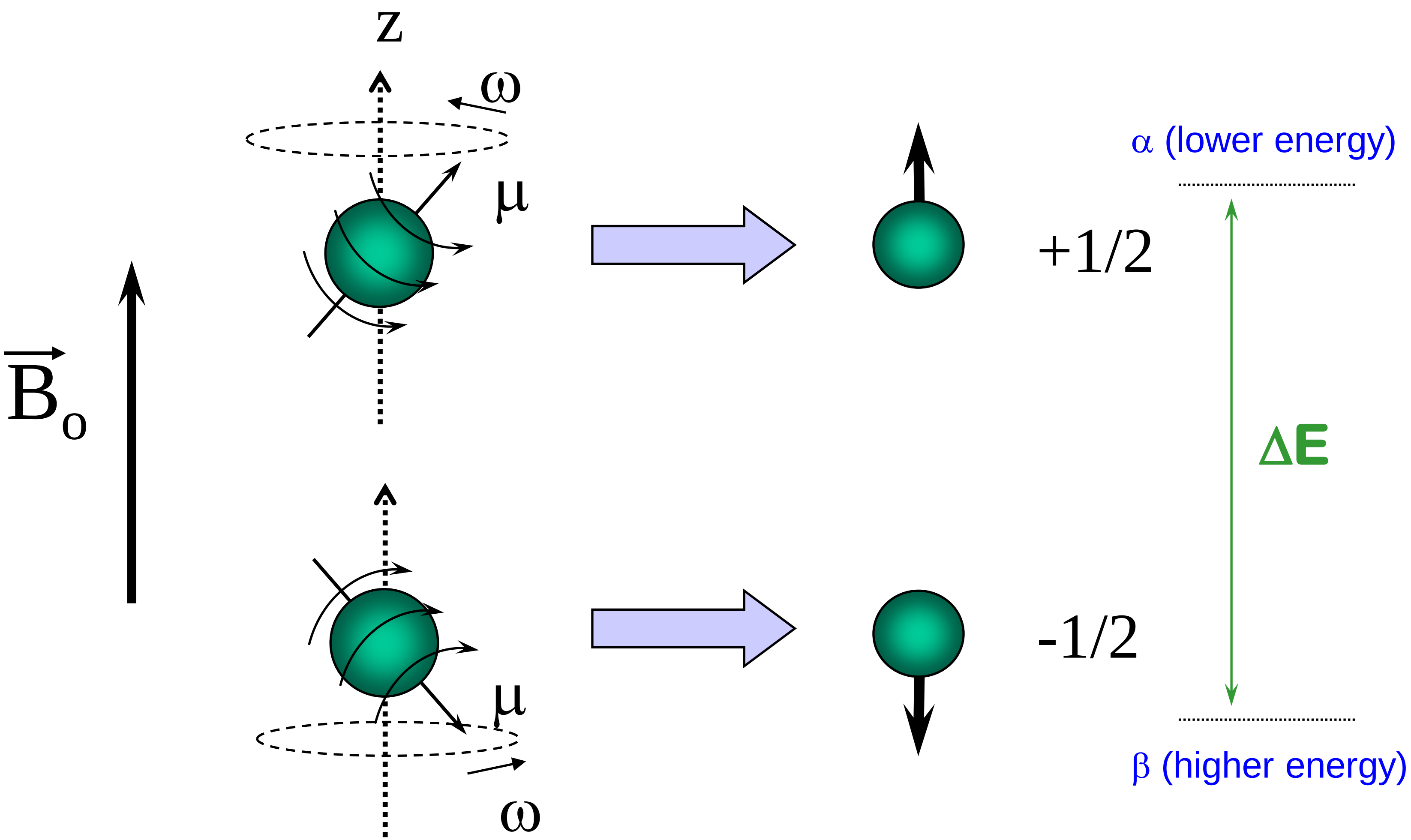
ν - resonance frequency
in cycles per second, Hz

γ - gyromagnetic ratio

B_o - external magnetic
field (the magnet)

Spin 1/2 nuclei will have two orientations in a magnetic field +1/2 and -1/2 (2I + 1)

Net magnetic moment



$+1/2$

$-1/2$

α (lower energy)

ΔE

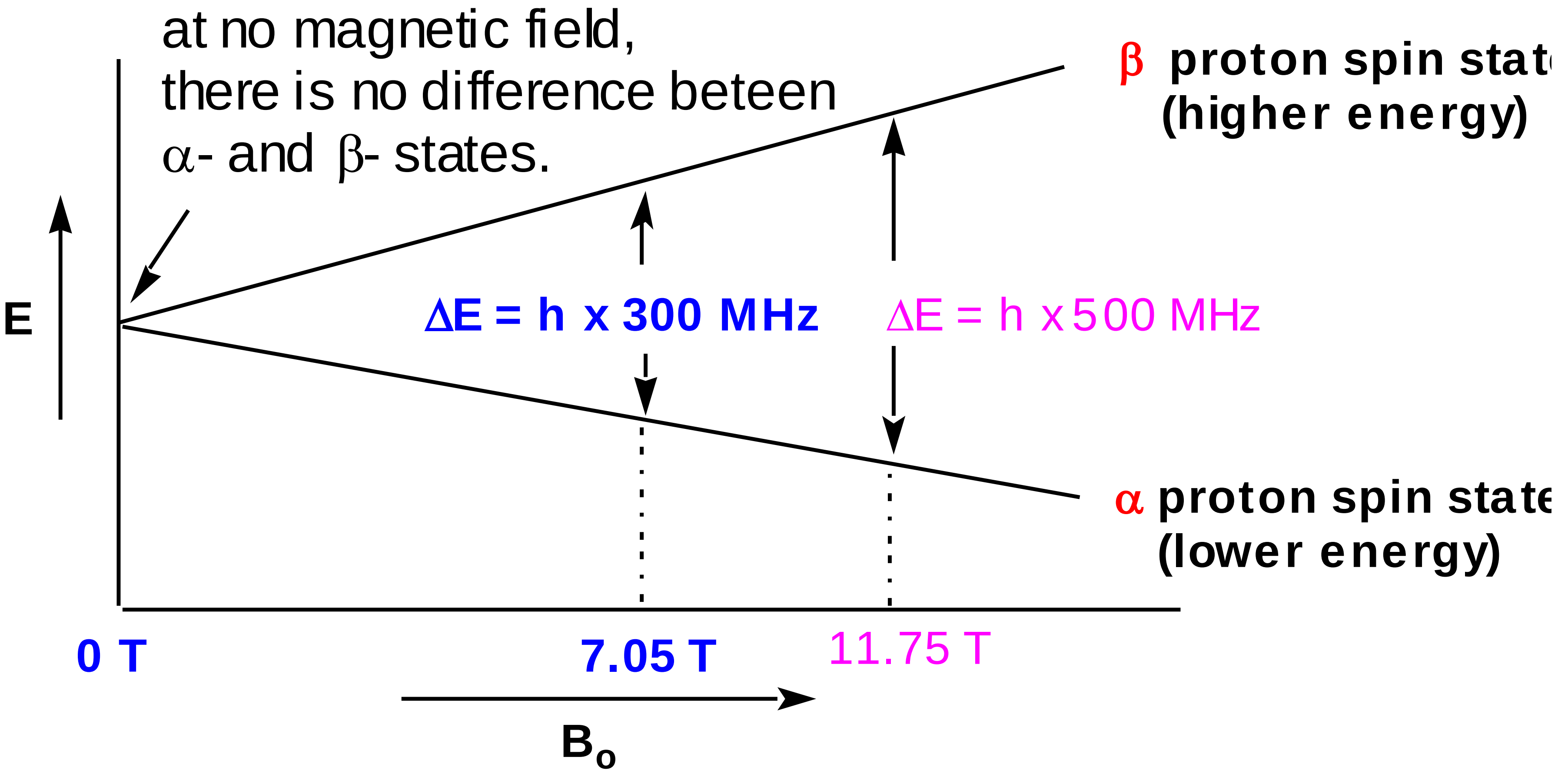
β (higher energy)

Variation of ^{1}H excess nuclei with operating frequency

---

frequency	excess nuclei
60	9
80	12
100	16
200	32
300	48
600	96

Graphical relationship between magnetic field (Bo) and frequency (ν) for ^{1}H NMR absorptions



at no magnetic field,
there is no difference beteen
α- and β- states.

β proton spin stat
(higher energy)

ΔE = h x 300 MHz

ΔE = h x 500 MHz

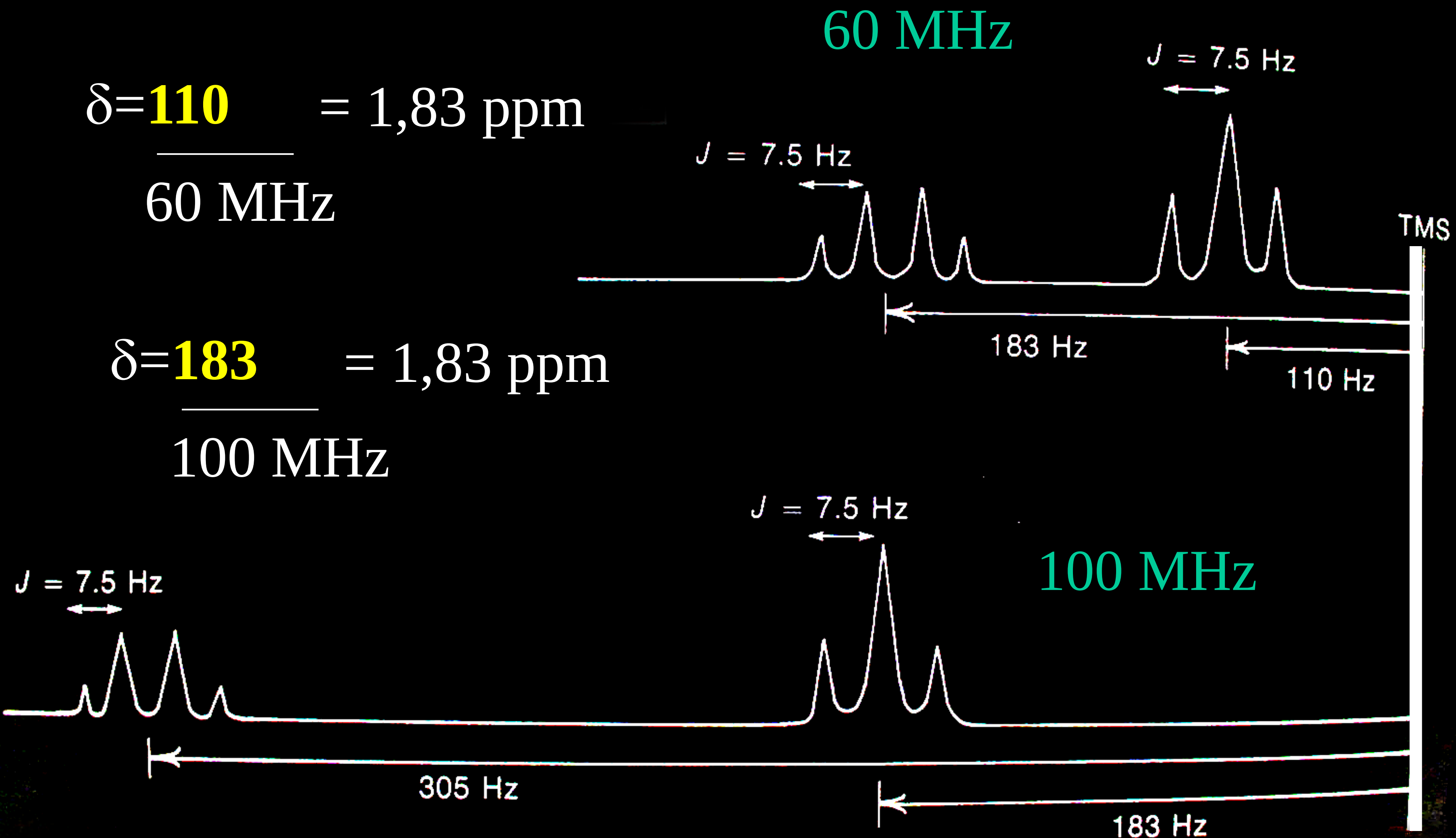
α proton spin state
(lower energy)

The unities for NMR are given in ppm instead of Hertz

(otherwise there is no way to compare chemical shifts obtained from diferents equipments)

$$\delta = \frac{\mathbf{110}}{60\ \text{MHz}} = 1{,}83\ \text{ppm}$$

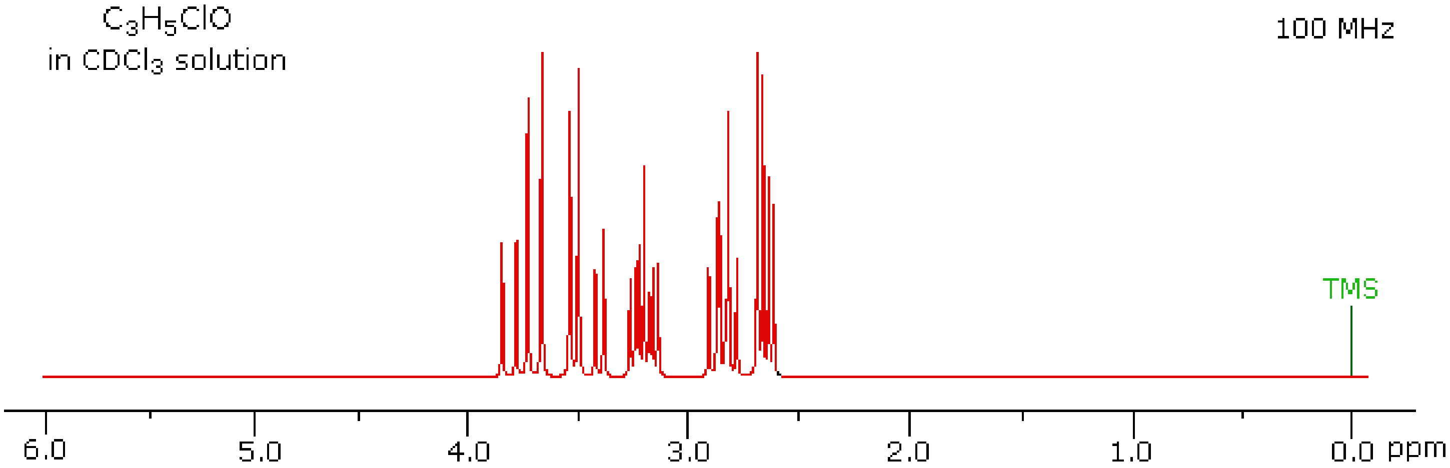
$$\delta = \frac{\mathbf{183}}{100\ \text{MHz}} = 1{,}83\ \text{ppm}$$



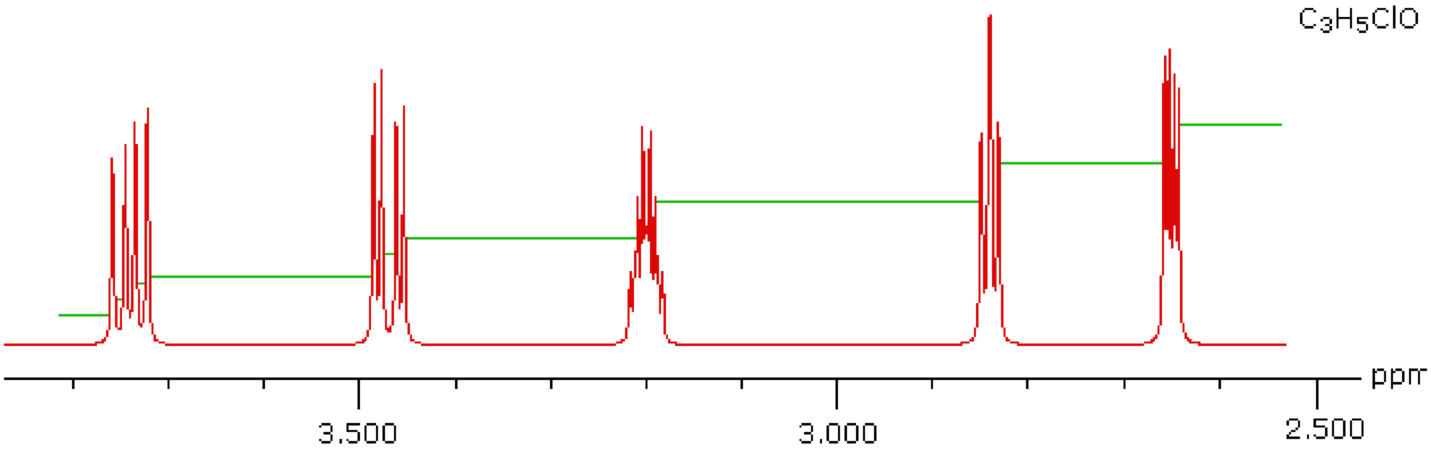
Effect of resolution in separation of signals in ^{1}H NMR absorptions

2-(chloromethyl)oxirane

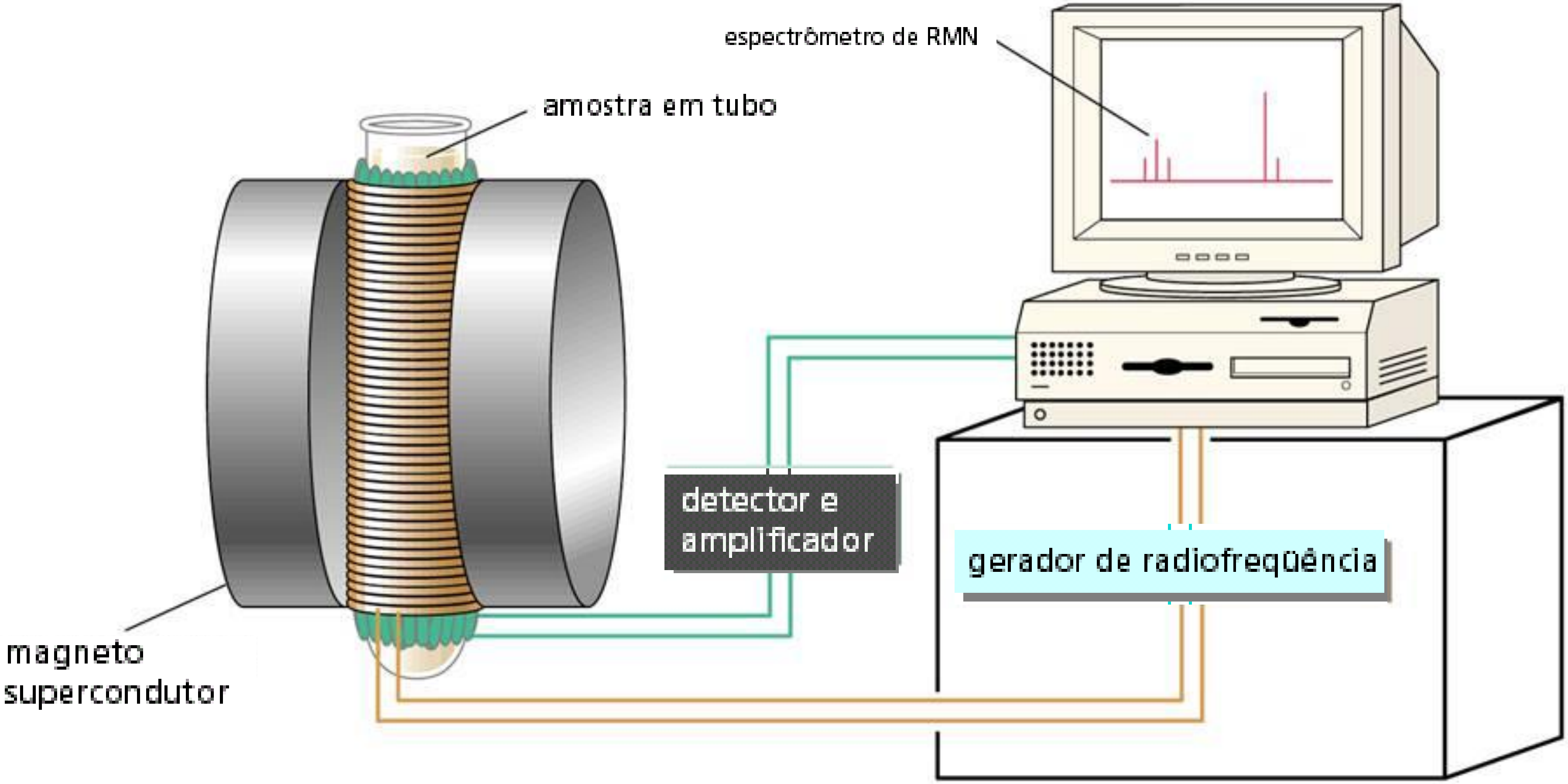
100 MHz



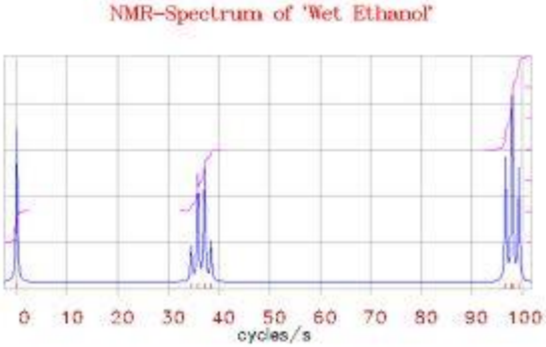
500 MHz



Esquema de um Espectrômetro de RMN



1961 Shoolery introduces the Varian A-60 high-resolution spectrometer.



RMN no IQUSP

1970: EM 360 Varian; T-60 Varian

1980: AC 80 e AC 200 Bruker

1990: DPX 300 Bruker;

1995: Bruker DRX 500;

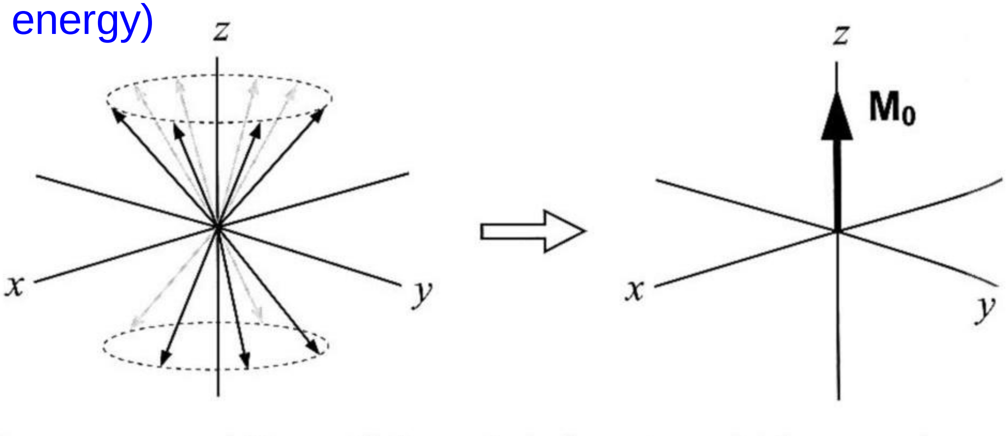
2013: Bruker 800 MHz

The vector model for NMR

Mo= vetor resultante
(magnetização macroscópica)

α (lower energy)

Magnetic
field

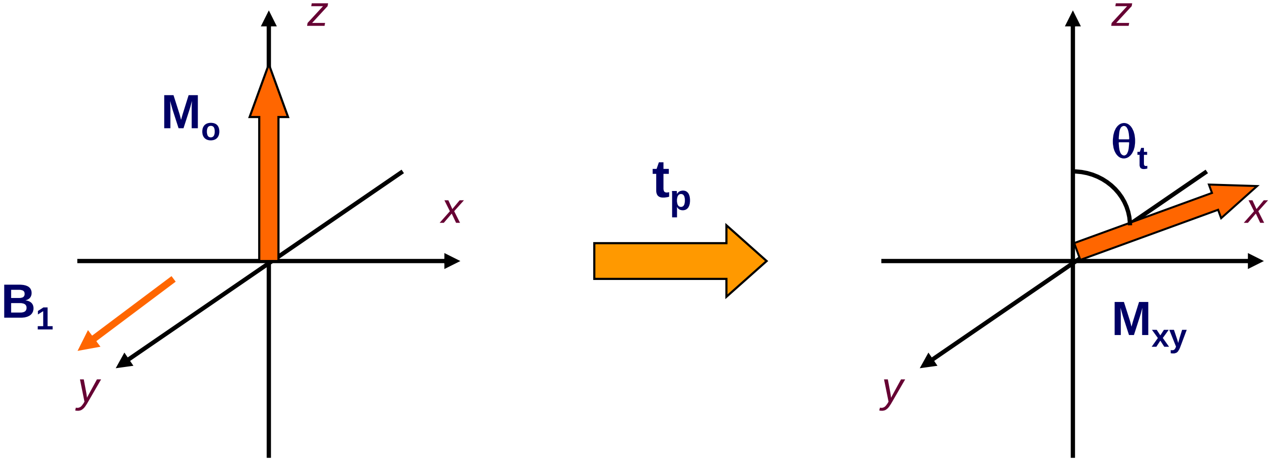


β (higher energy)

There are many nuclei in one NMR sample, in the vector model the many spins are represented by a single vector which is the bulk magnetization or average.

The small population difference between the ground and the excited states causes the bulk magnetization to be pointed to +z axis.

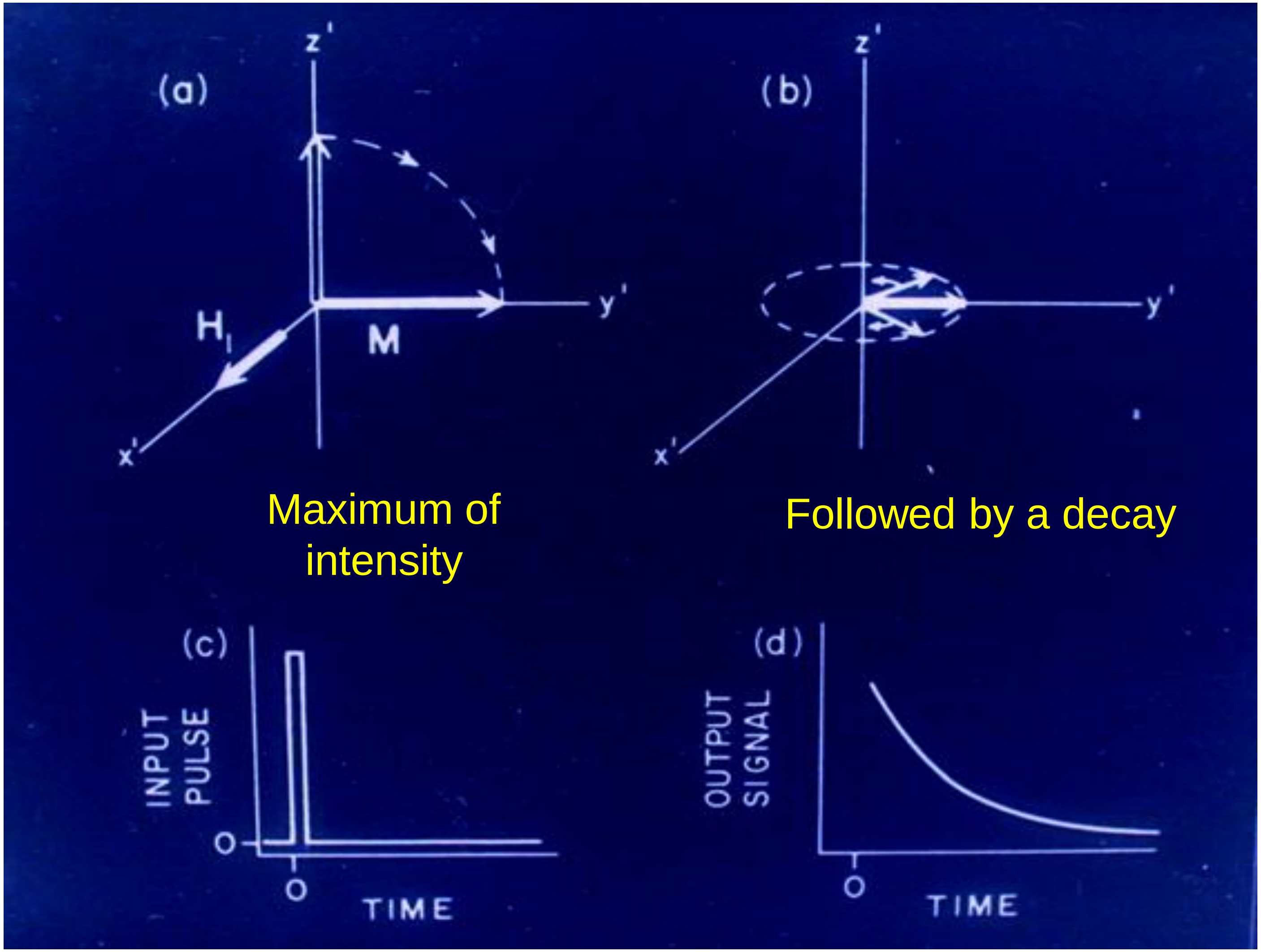
Pulse widths and tip angles



$$\theta_t = \gamma * t_p * B_1$$

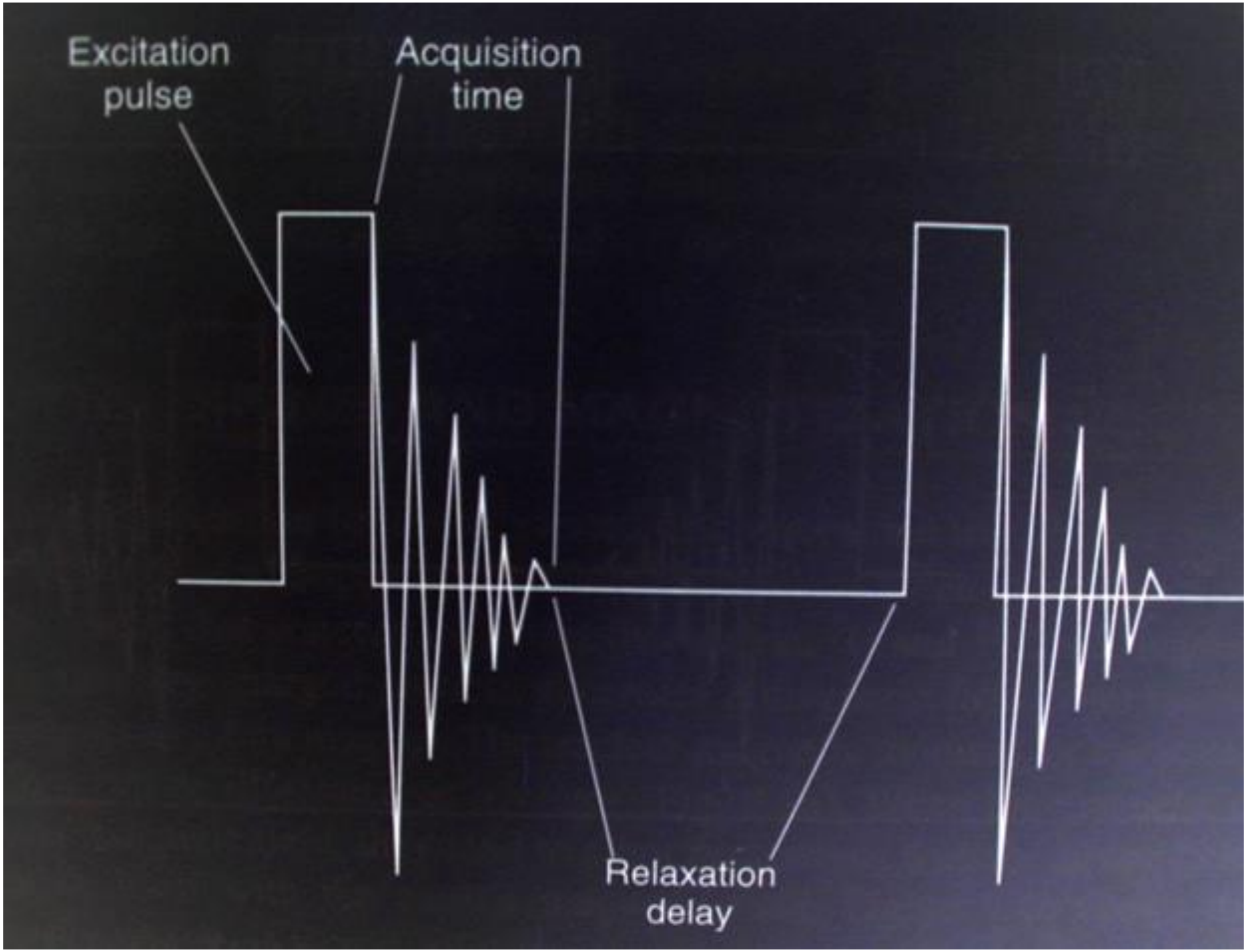
- As the pulse width (tp) for a certain flip angle will depend on the instrument (**B_1**), we will therefore refer to them in terms of the rotation we want to obtain of the magnetization. Thus, we will have **π / 4** (**45**), **π / 2** (**90**), **π** (**180**), etc., etc., pulses

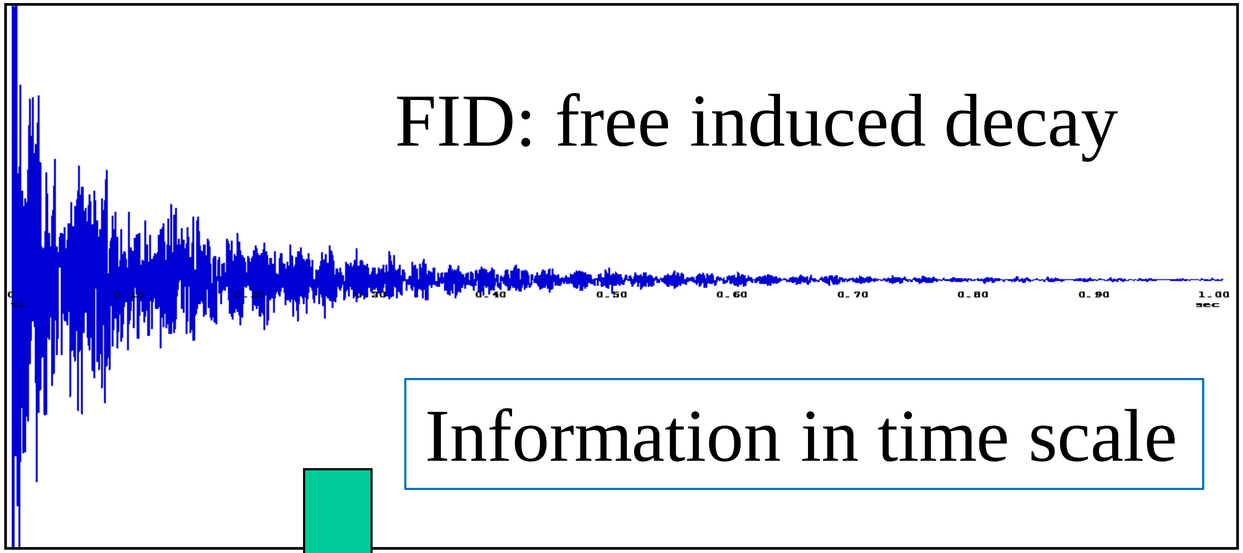
Pulse of 90^o (π/2)



Maximum of intensity

Followed by a decay


Excitation pulse
Acquisition time
Relaxation delay

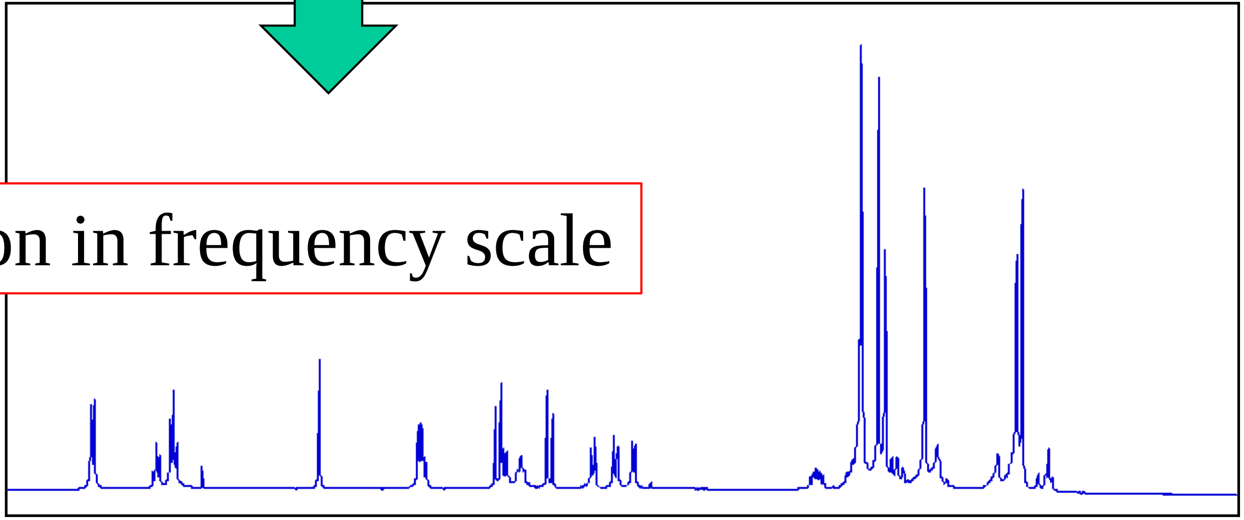


FID: free induced decay

Information in time scale

FT

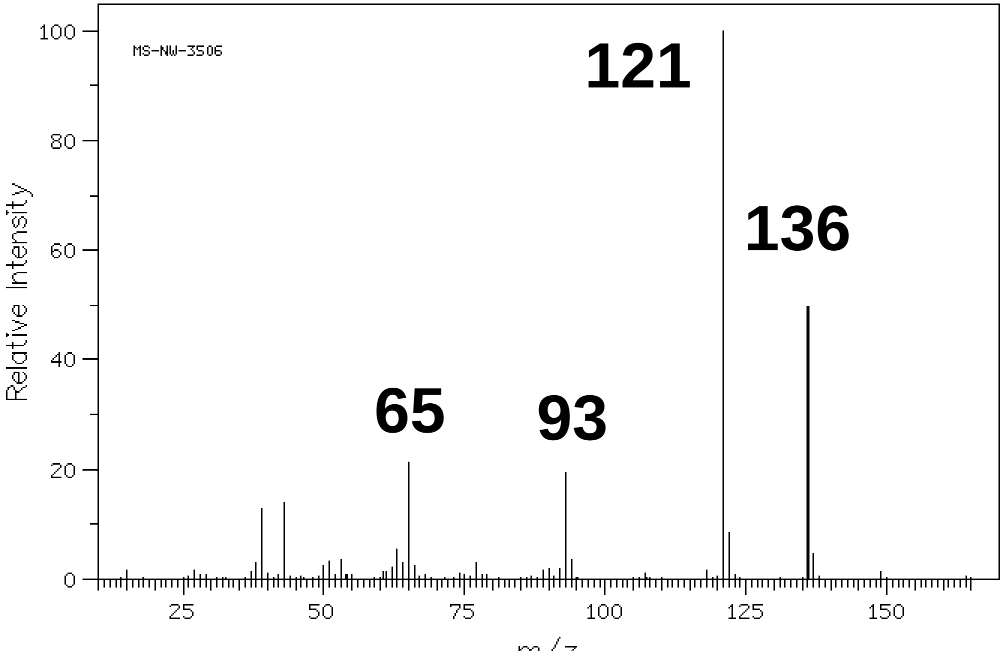
$$S(\omega) = \int_{-\infty}^{\infty} S(t)\, e^{-i\omega t}\, dt$$

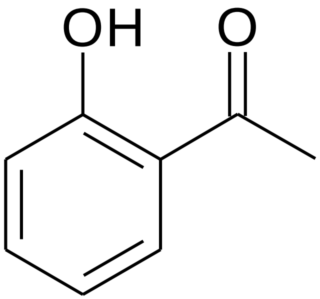


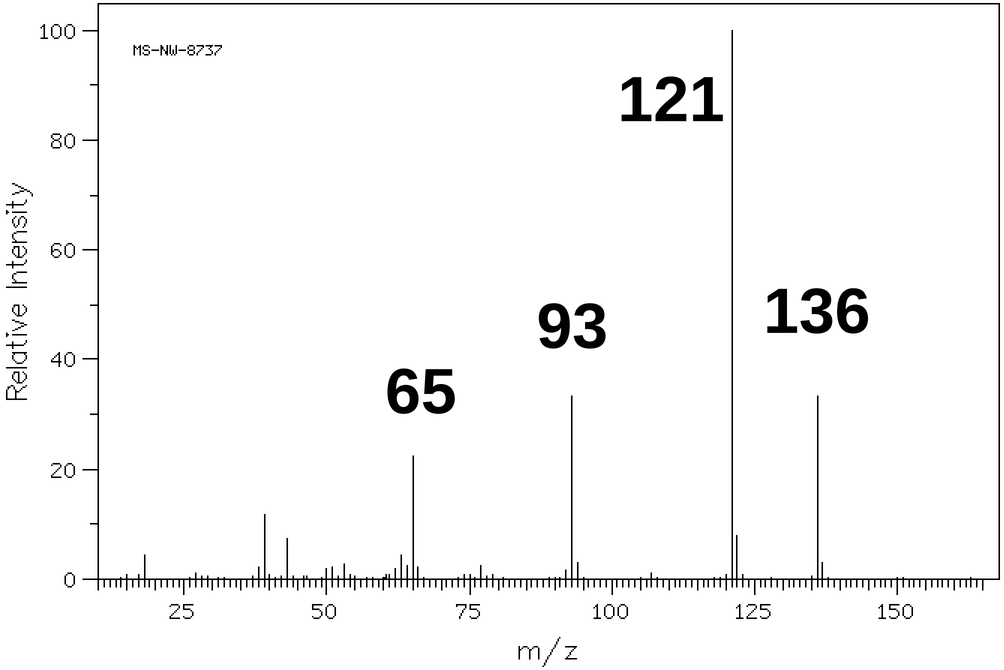
Information in frequency scale

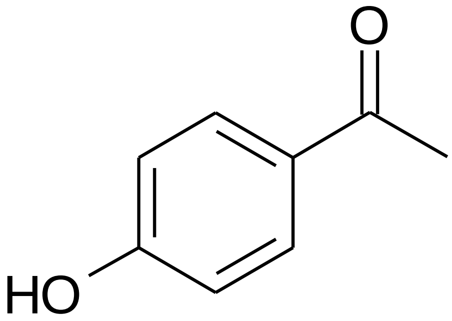
The application of NMR in structural elucidation

$C_8H_8O_2$







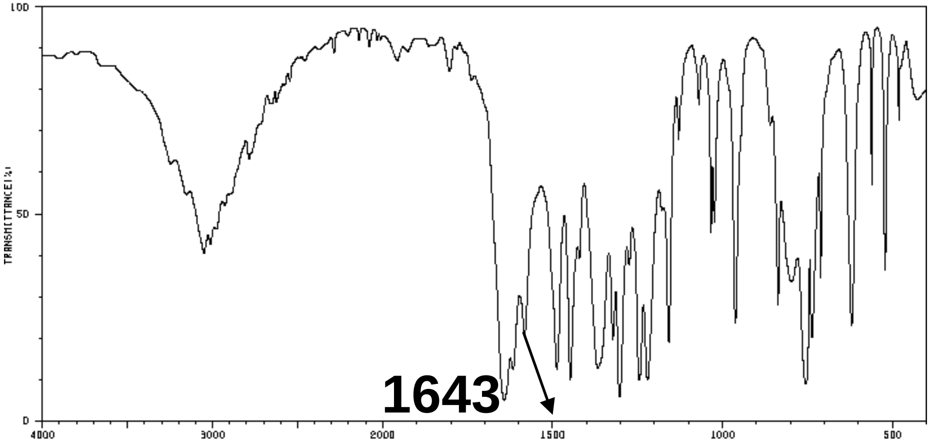


MS does not allow differentiation between isomers !!!

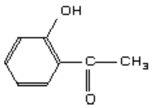
HIT-NO=1154	SCORE= ( )	SDBS-NO=724	IR-NIDA-20623 : LIQUID FILM

2'-HYDROXYACETOPHENONE

$C_8H_8O_2$



3061	38	1617	12	1303	6	1069	74	766	8
3011	41	1582	20	1275	36	1034	43	737	19
2786	80	1488	12	1245	9	1023	46	712	33
2773	62	1448	9	1221	9	963	22	621	22
2650	74	1421	37	1173	49	860	68	562	55
2823	74	1368	12	1158	18	837	26	522	35
1643	4	1323	18	1129	66	797	32	482	70

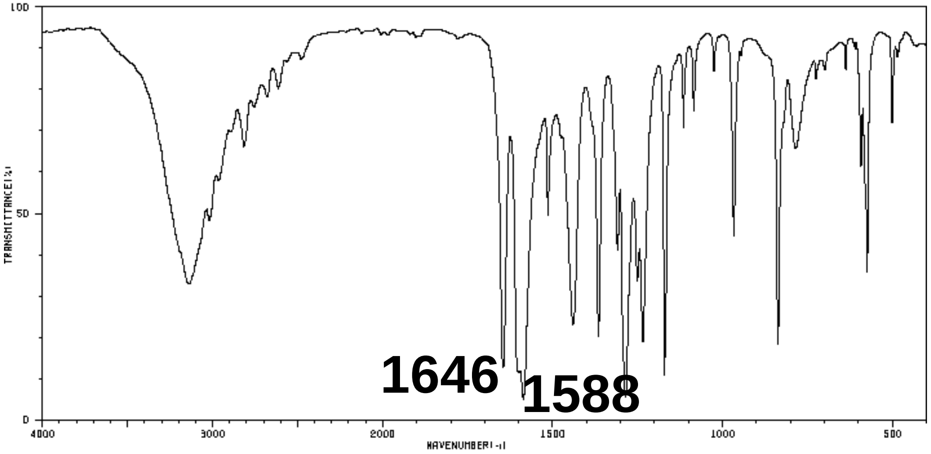


Hydrogen bonding lower the frequency of carbonyl Vibration.

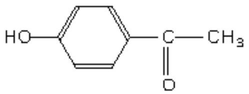
HIT-NO=1223	SCORE= ( )	SDBS-NO=1008	IR-NIDA-63404 : KBR DISC

4'-HYDROXYACETOPHENONE

$C_8H_8O_2$

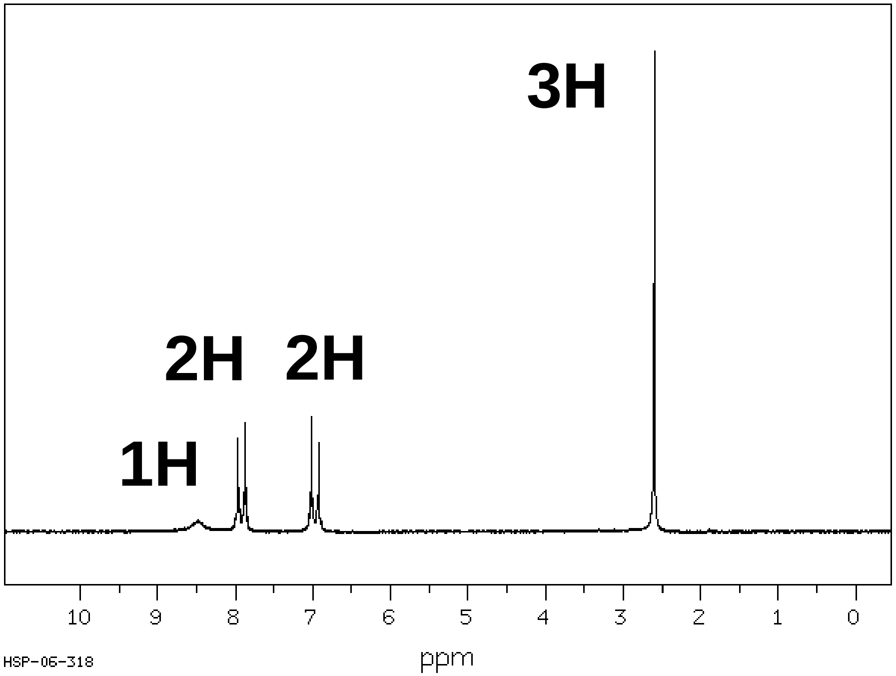


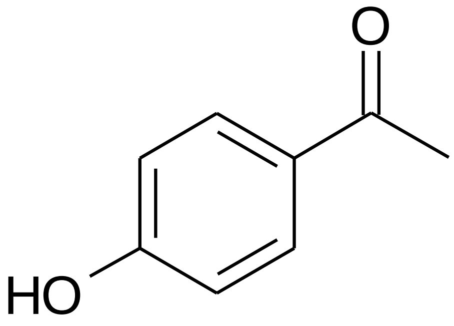
3147	32	2677	74	1441	22	1116	68	726	79
3135	32	2614	77	1366	19	1085	72	699	81
3117	34	2480	84	1309	39	1025	81	637	61
3019	46	1646	12	1286	6	967	43	693	68
2963	55	1600	11	1251	32	945	84	581	55
2816	84	1588	4	1235	18	837	17	575	34
2766	72	1614	47	1171	10	786	64	602	70

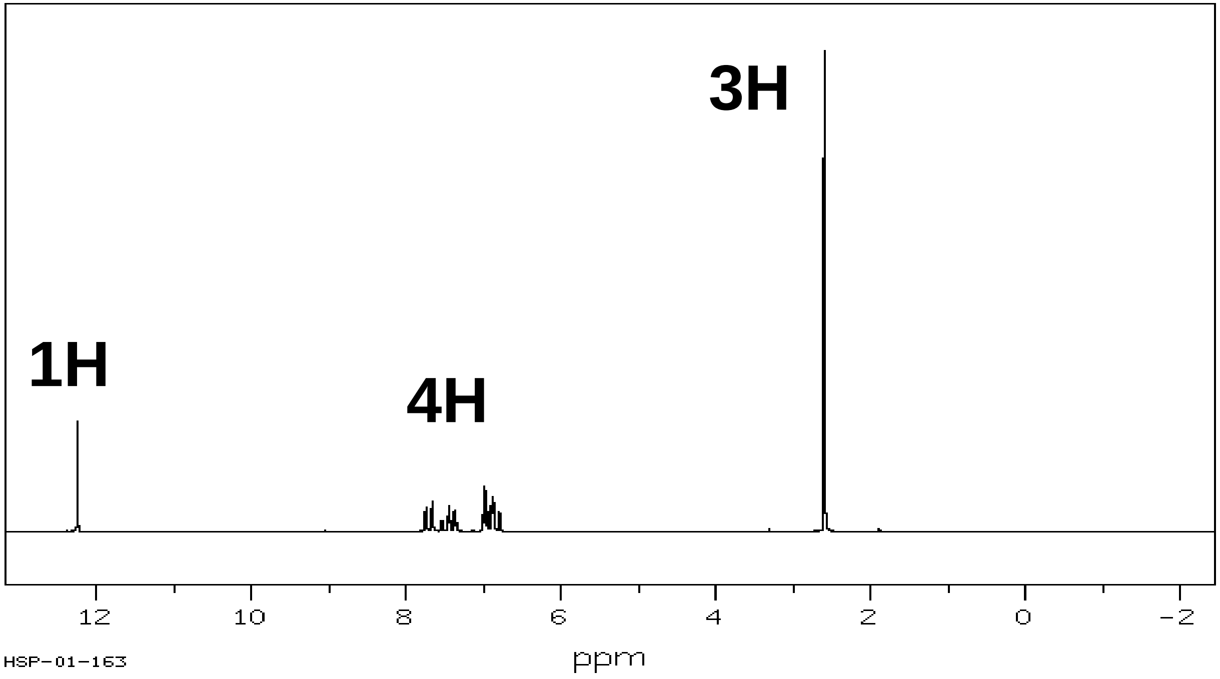


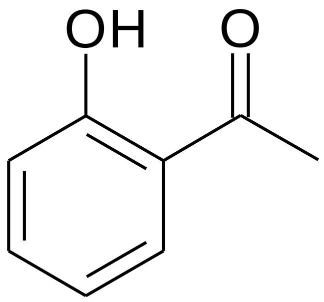
IR can differentiate Between these Two isomers

Espectro de RMN de ^{1}H









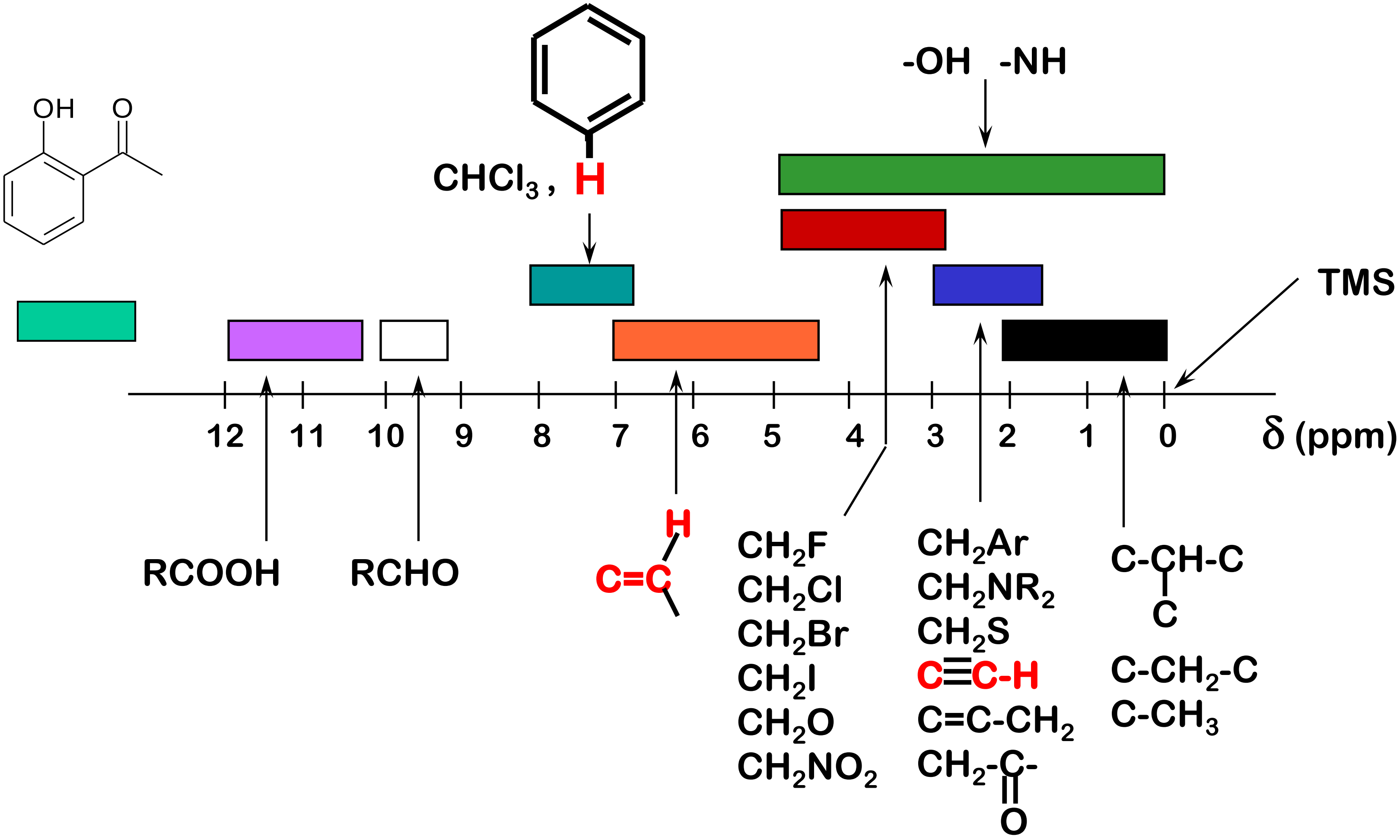
Informações obtidas de um espectro de RMN de ^{1}H:

1) Deslocamentos químicos (δ - ppm);

2) Integração (nº relativo de Hs);

3) Constantes de acoplamento (J em Hertz);

Δδ em RMN para diversos Hs



$CHCl_3$, **H**

-OH -NH

TMS

RCOOH

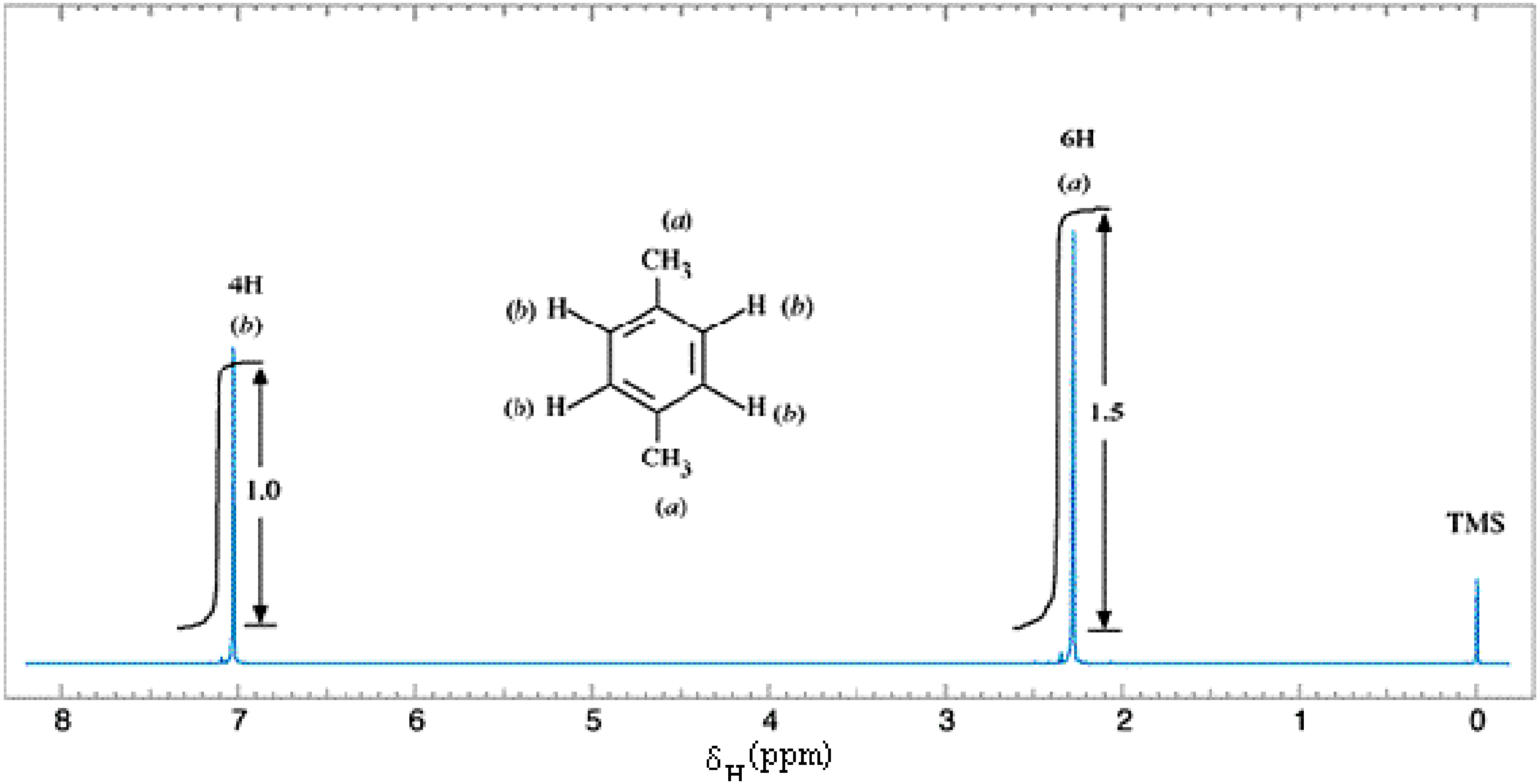
RCHO

H
C=C

CH_2F
CH_2Cl
CH_2Br
CH_2I
CH_2O
CH_2NO_2

CH_2Ar
CH_2NR_2
CH_2S
$C\equiv C\text{-}H$
$C{=}C\text{-}CH_2$
$CH_2\text{-}\underset{\text{O}}{\underset{\|}{\text{C}}}\text{-}$

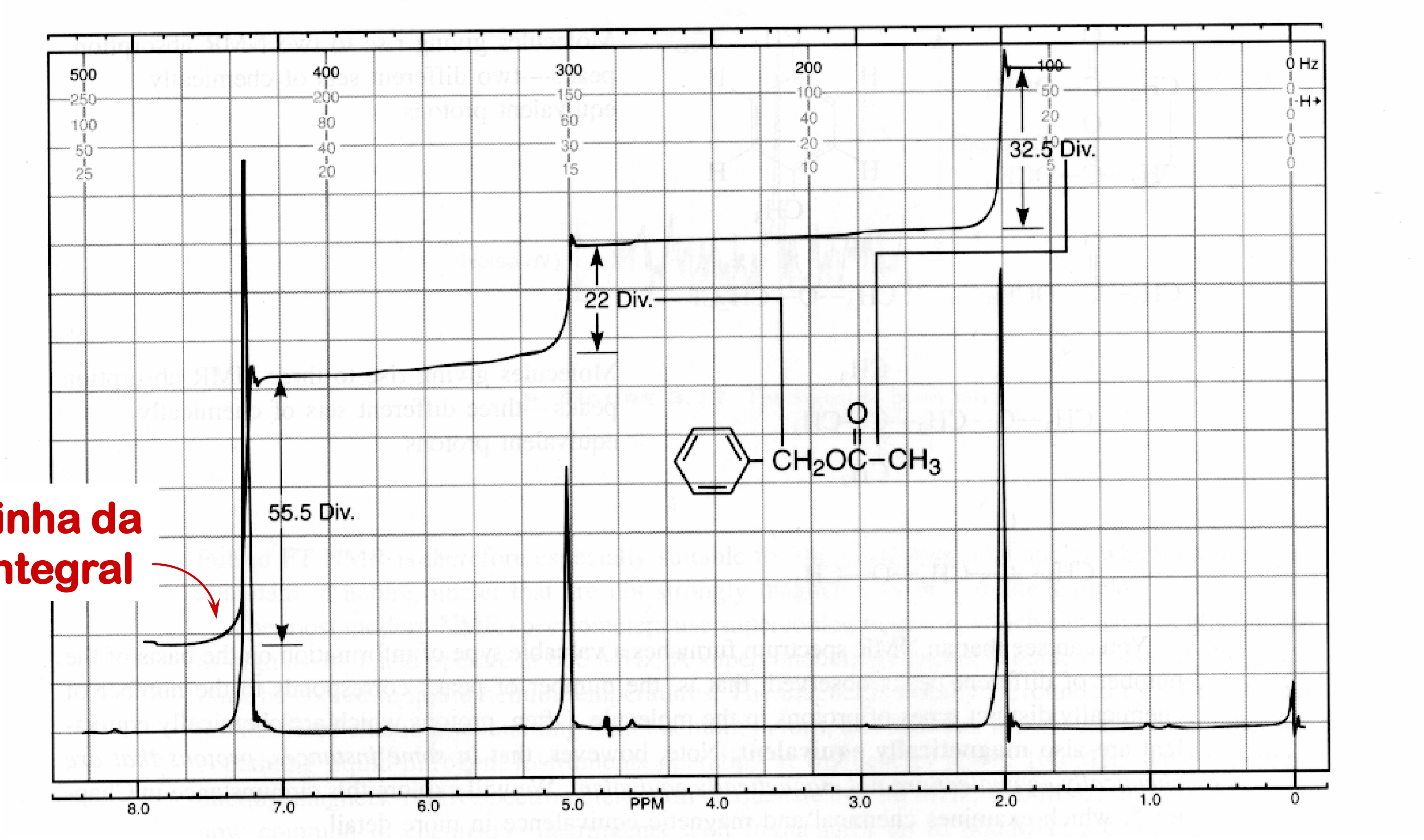
$\text{C-}\underset{\text{C}}{\underset{|}{\text{CH}}}\text{-C}$
$C\text{-}CH_2\text{-}C$
$C\text{-}CH_3$



Integração:

A área dos sinais é proporcional ao número de Hs!!!

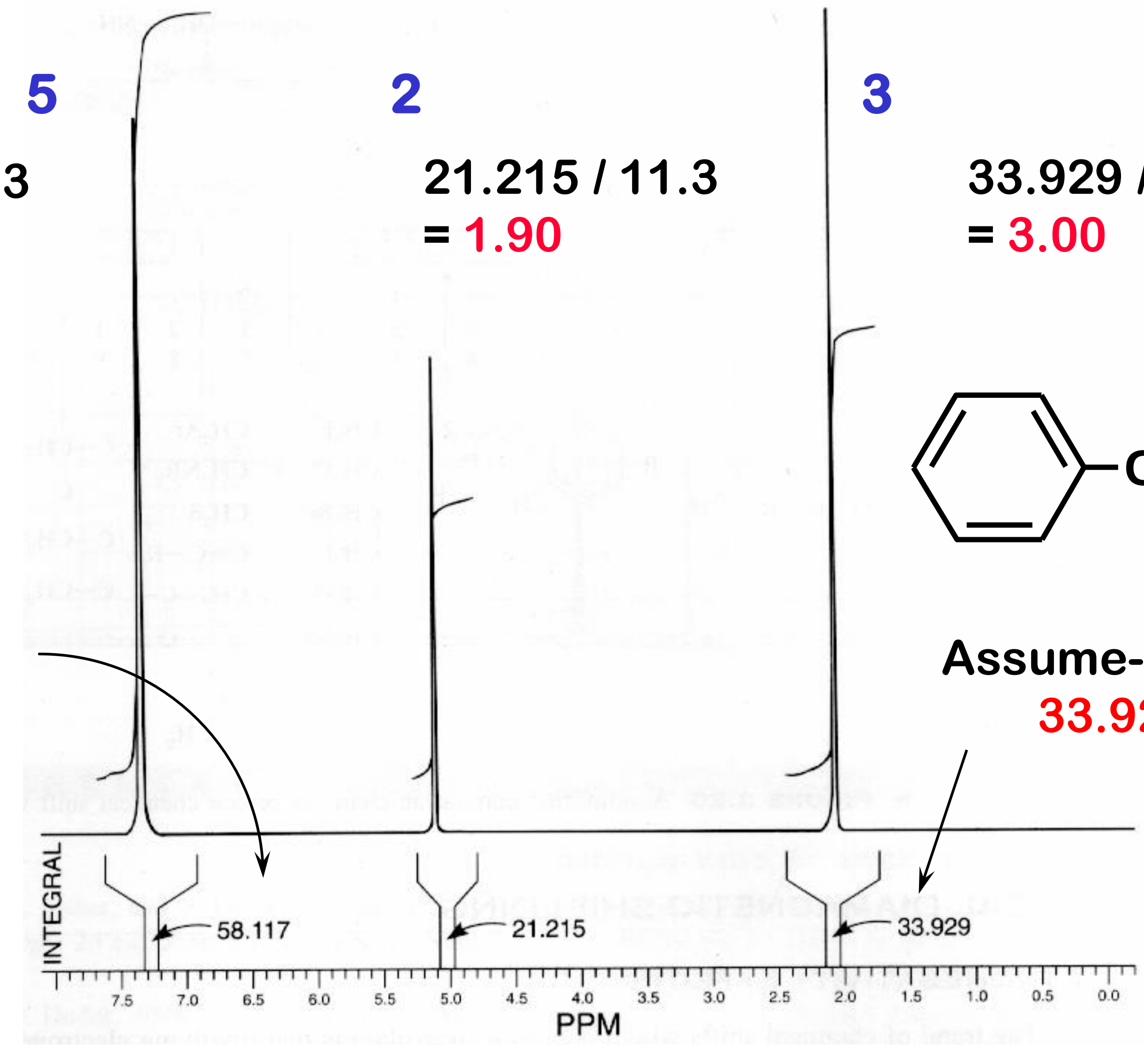
Acetato de benzila



Linha da integral

55 : 22 : 33 = 5 : 2 : 3

Acetato de benzila



5

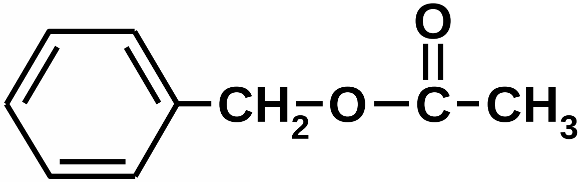
58.117 / 11.3
= 5.14

2

21.215 / 11.3
= 1.90

3

33.929 / 11.3
= 3.00



Assume-se que seja CH_3

33.929 / 3 = 11.3

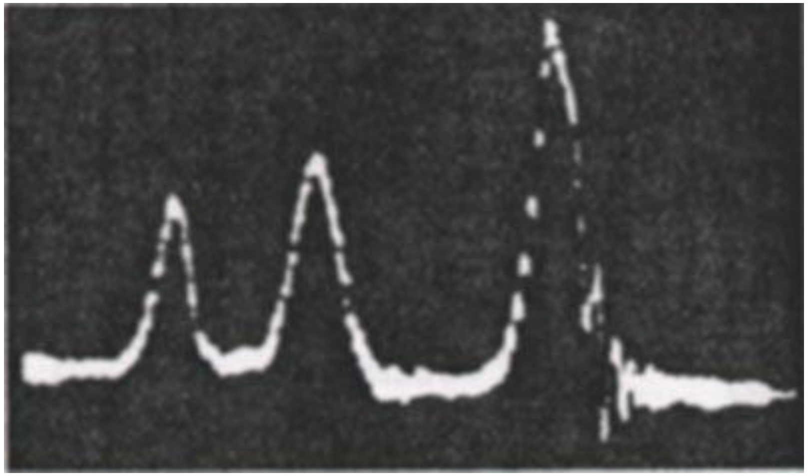
Informações obtidas de um espectro de RMN ^{1}H:

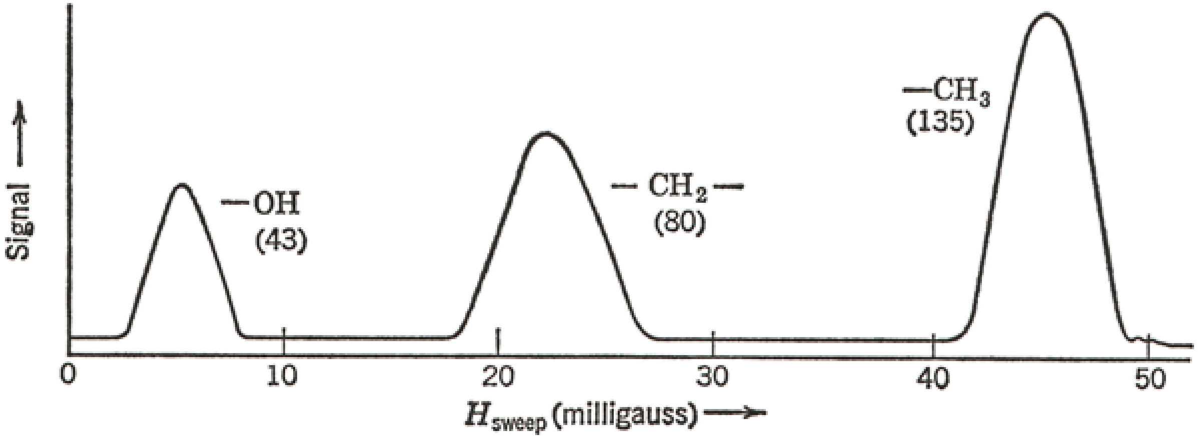
1) **Deslocamentos químicos (δ – ppm);**

2) **Integração (nº relativo de H);**

3) **Constantes de acoplamento (J em Hertz).**

The first NMR spectrum of ethanol, taken at Stanford University in 1951 (*Varian Associates, Inc.*)





^{1}H NMR spectrum of ethanol

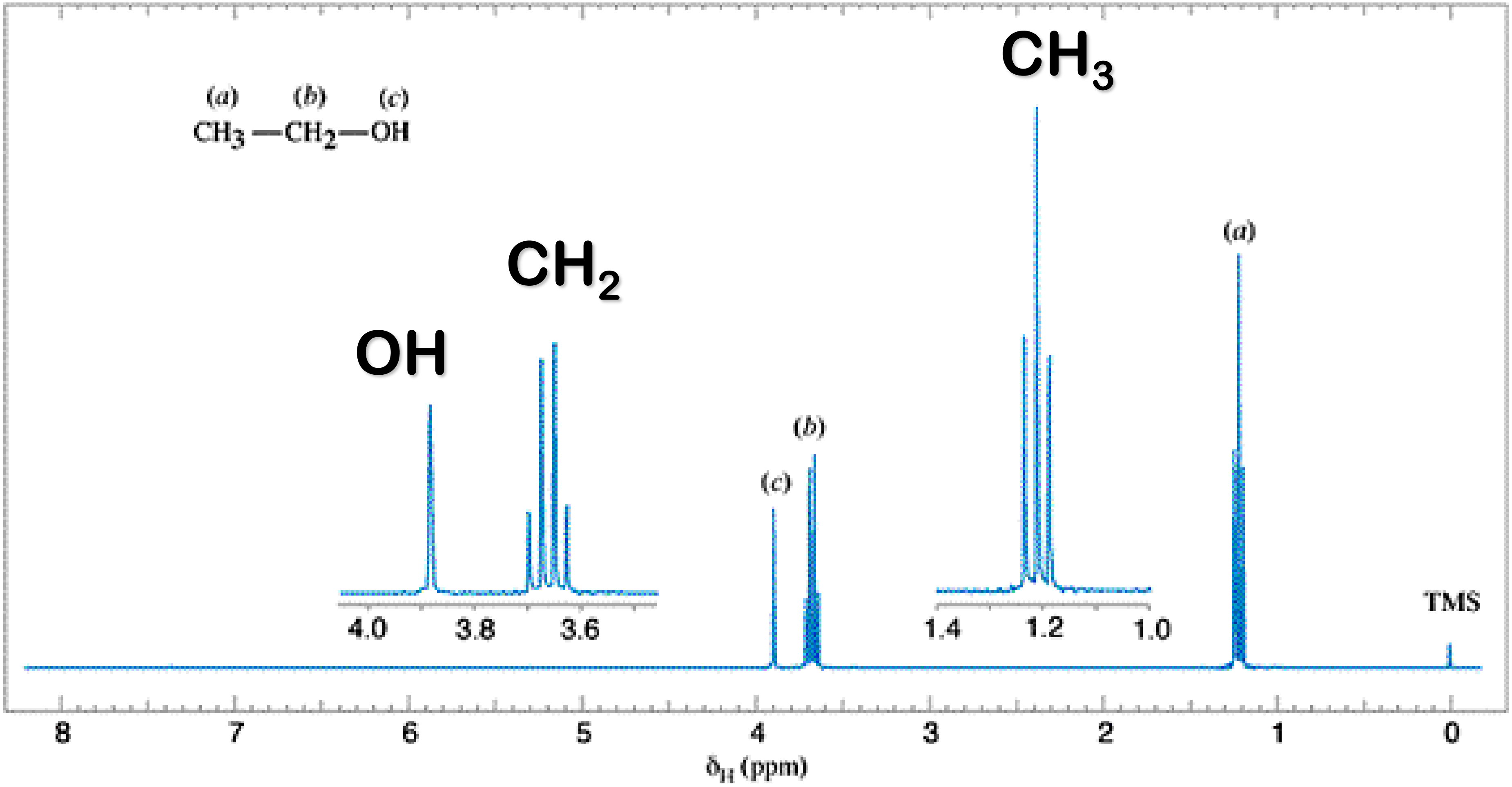
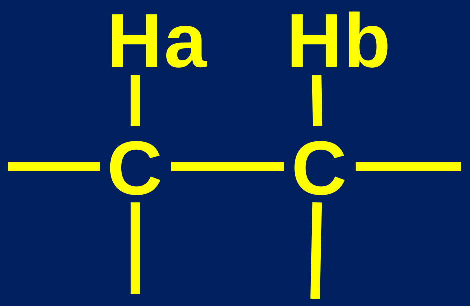
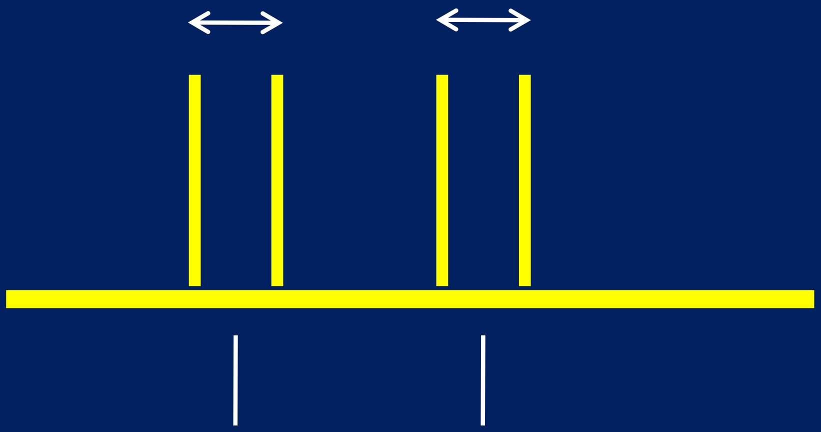


FIGURE 13.31 The 300 MHz ^{1}H NMR spectrum of ordinary ethanol. Expansions of the signals are shown in the offset plots.

Acoplamento mais simples para diferentes Hs:



Jab = Jba



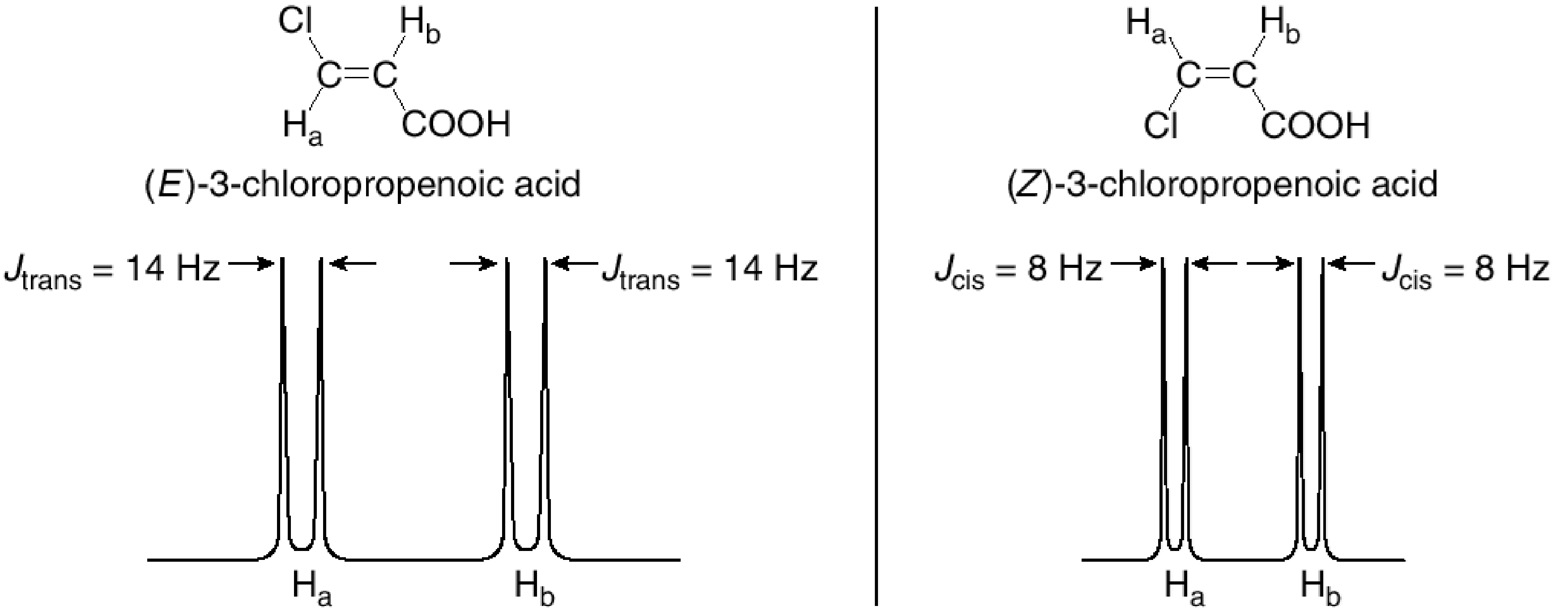
Deslocamentos químicos

Hb Hb

Nuclear Magnetic Resonance Spectroscopy

^{1}H NMR—Spin-Spin Splitting (coupling constant)

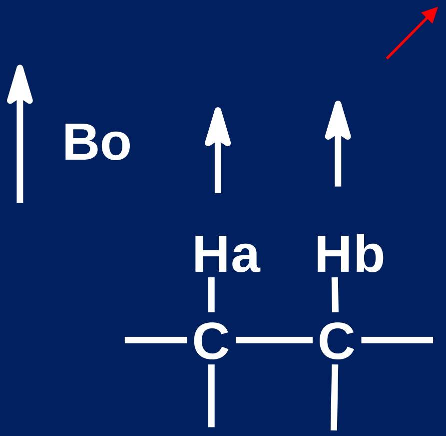
(um determinado sistema de spins apresenta o mesmo valor de J)



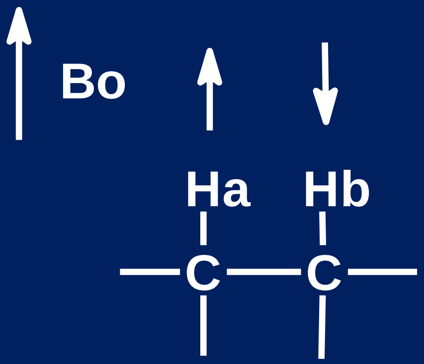
Although both (*E*)- and (*Z*)-3-chloropropenoic acid show two doublets in their ^{1}H NMR spectra for their alkenyl protons, $J_{trans} > J_{cis}$.

Acoplamento mais simples:

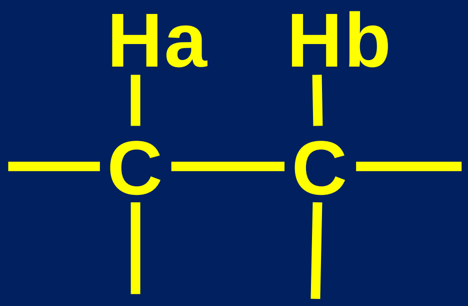
~50% em α ou β

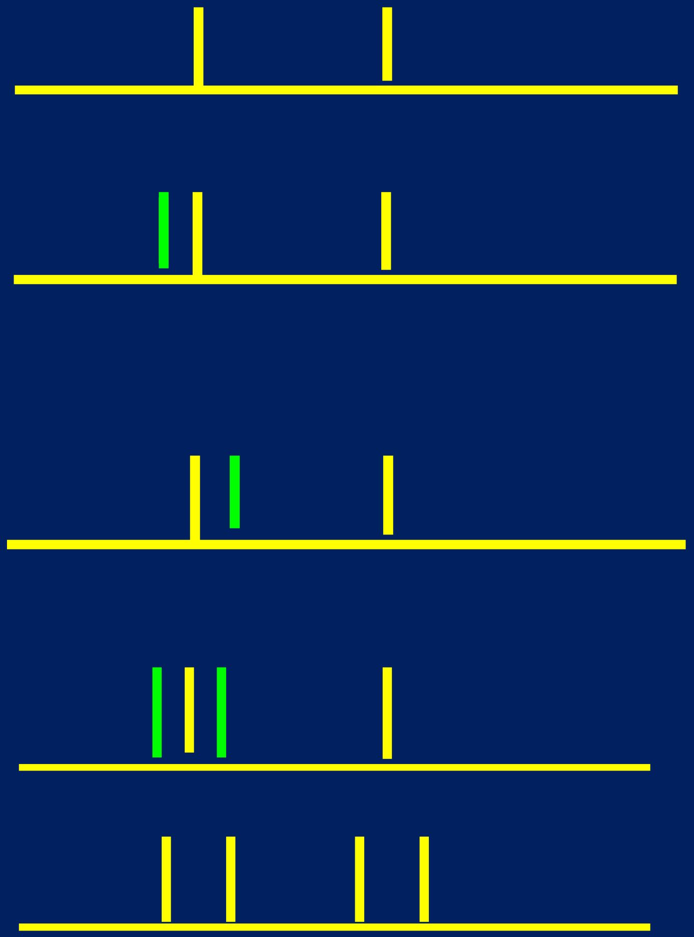


υHa=Bo + BHb

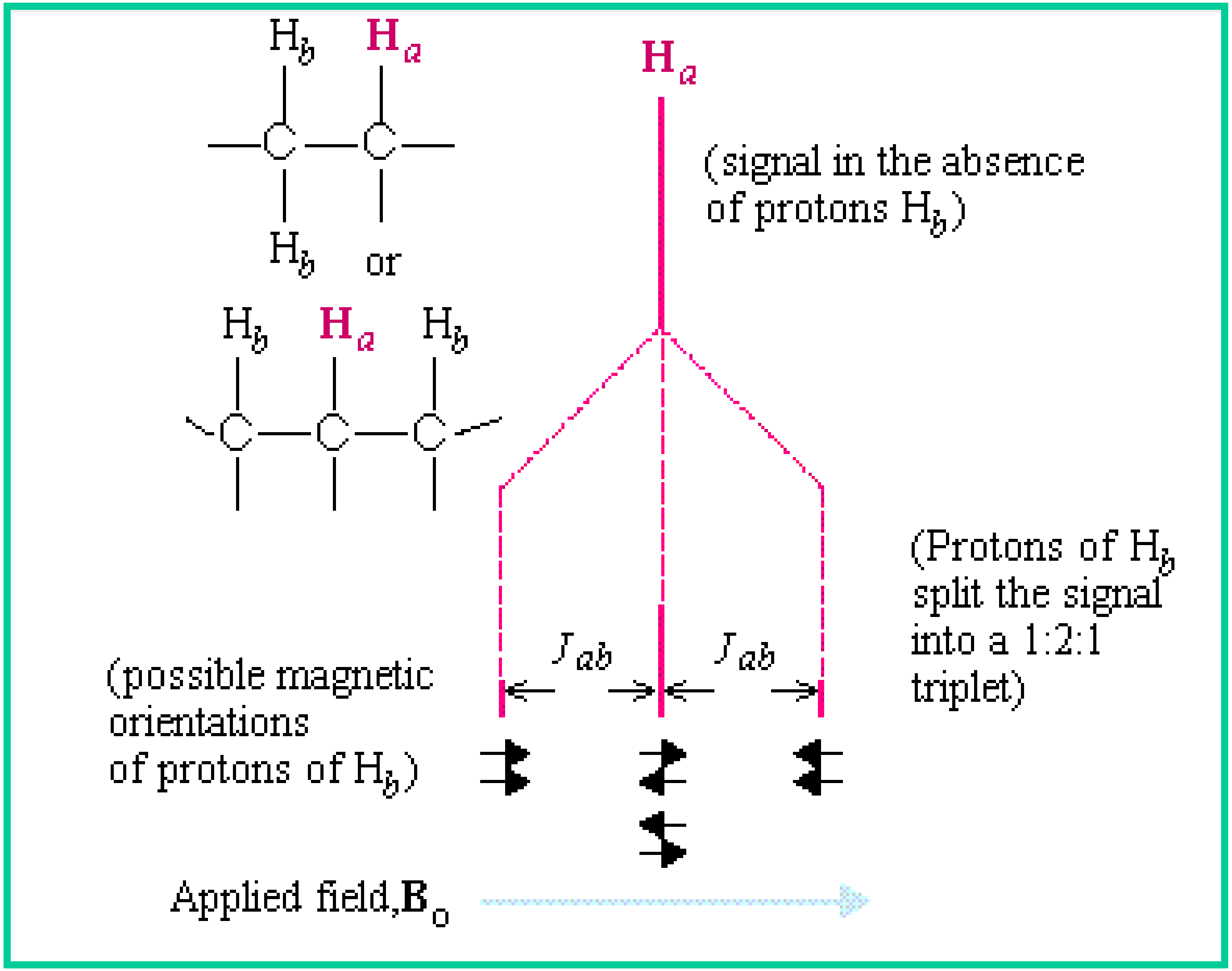


υHa=Bo - BHb





*J*ab = *J*ba



(signal in the absence of protons H$_b$)

(Protons of H$_b$ split the signal into a 1:2:1 triplet)

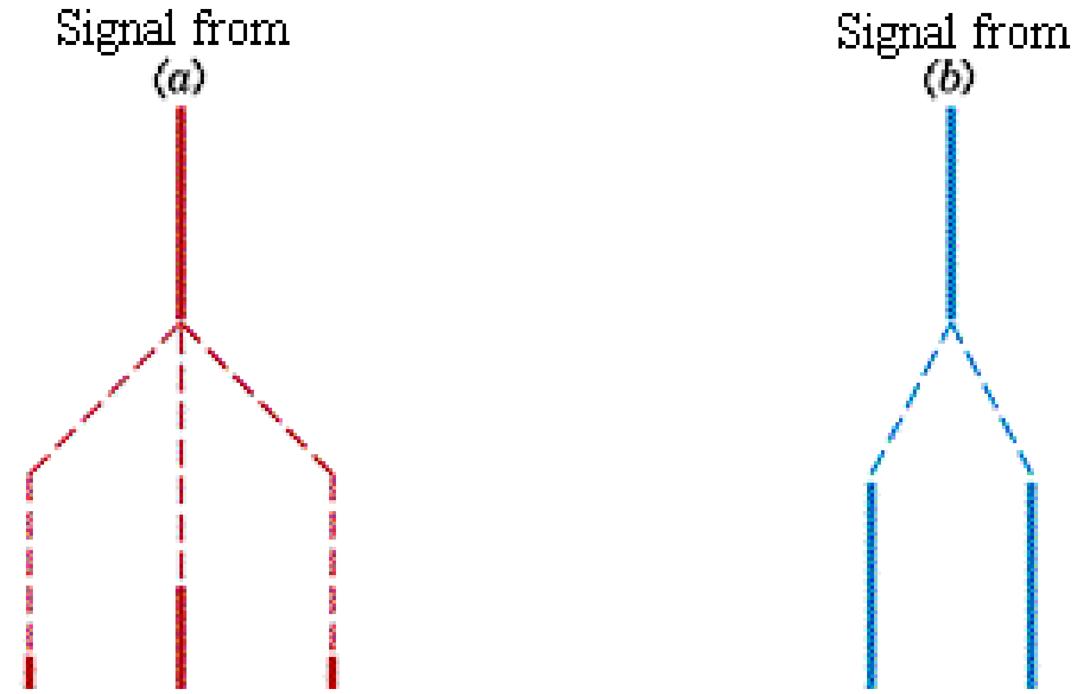
(possible magnetic orientations of protons of H$_b$)

Applied field, $\mathbf{B}_o$

(*a*) (*b*)

$CHCl_2CH_2Cl$

1,1,2-Trichloroethane



Split into a triplet by the two (*b*) proton

Split into a doublet by the (*a*) proton

1H

2H

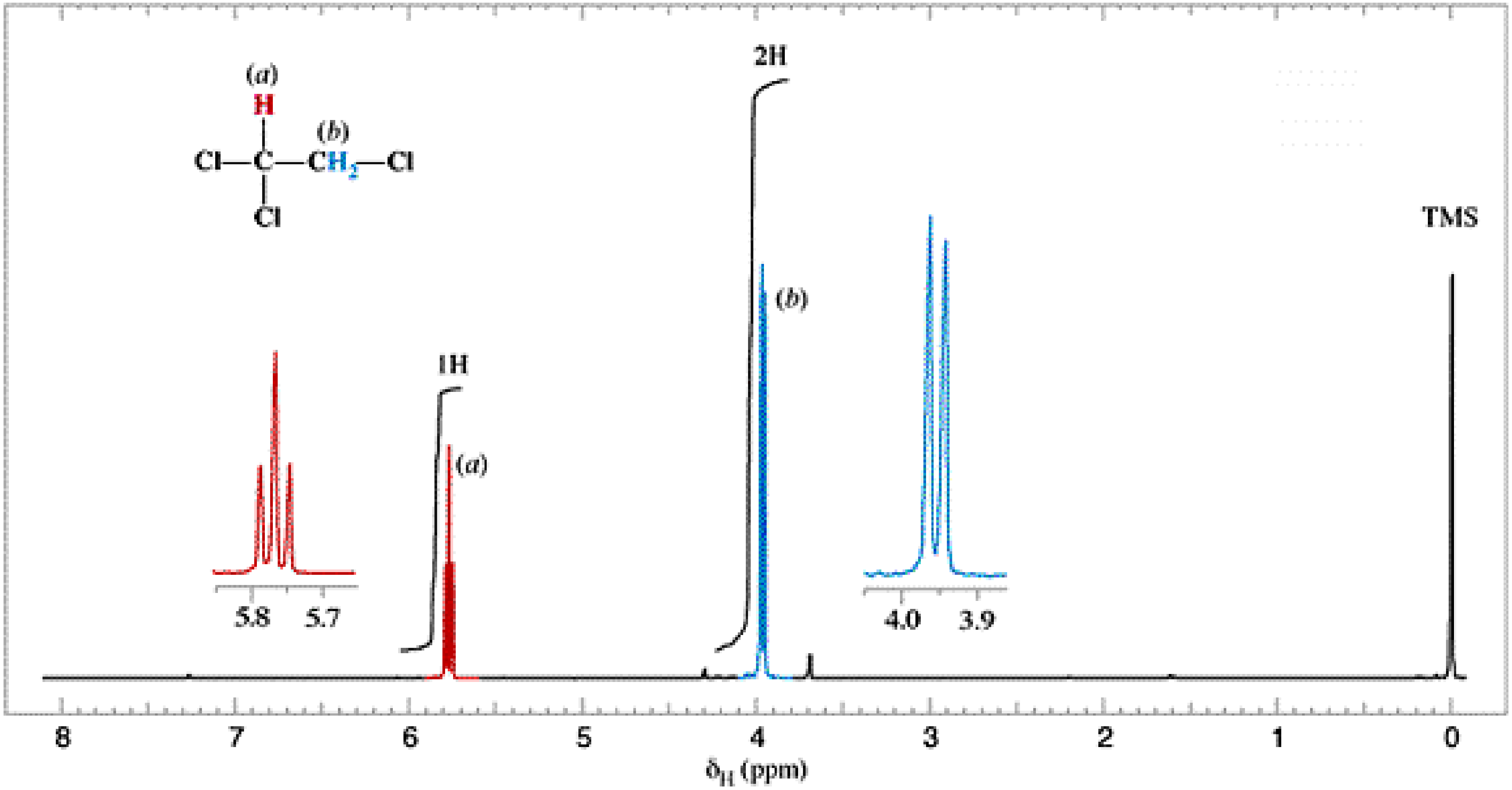
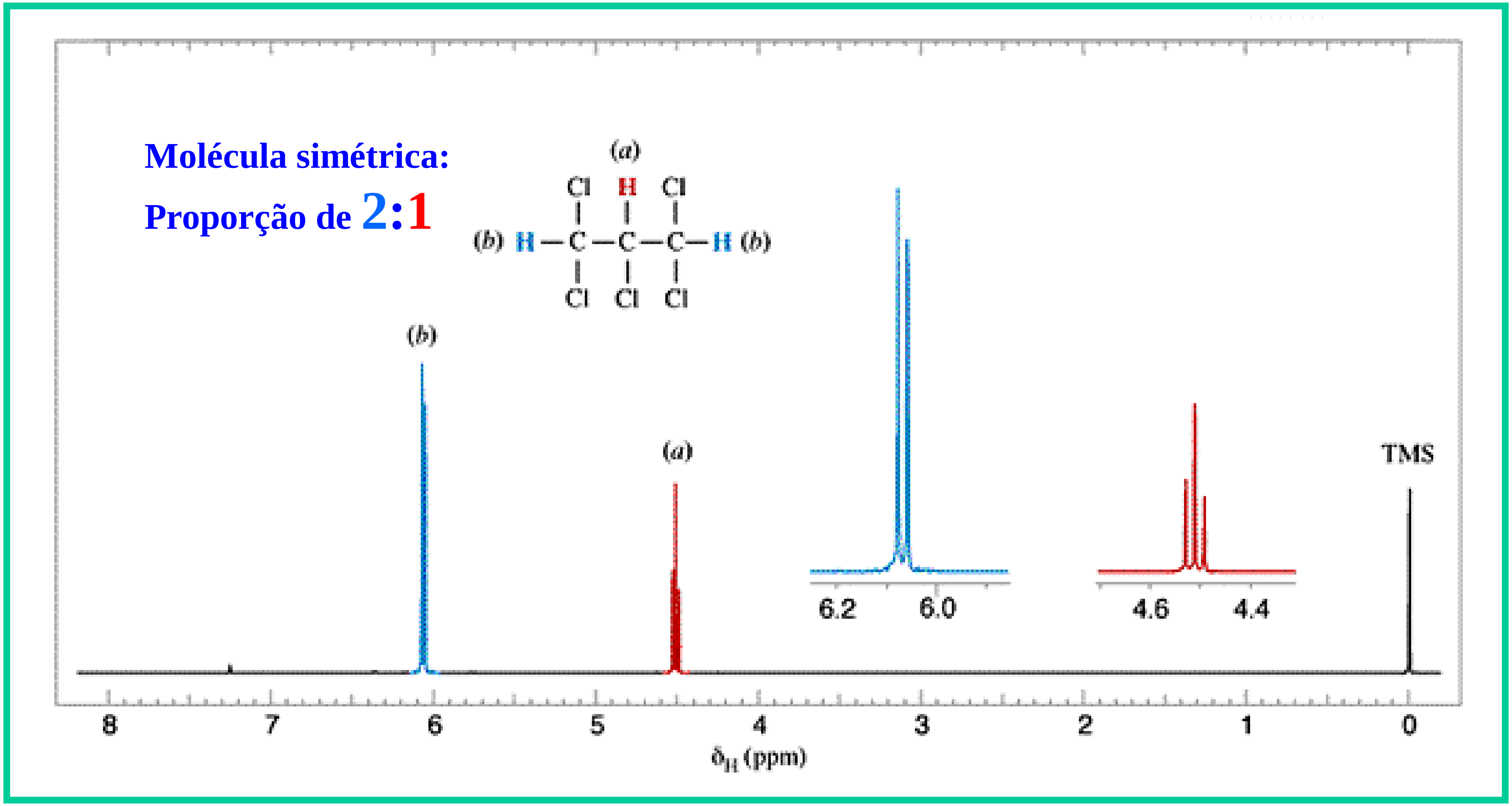


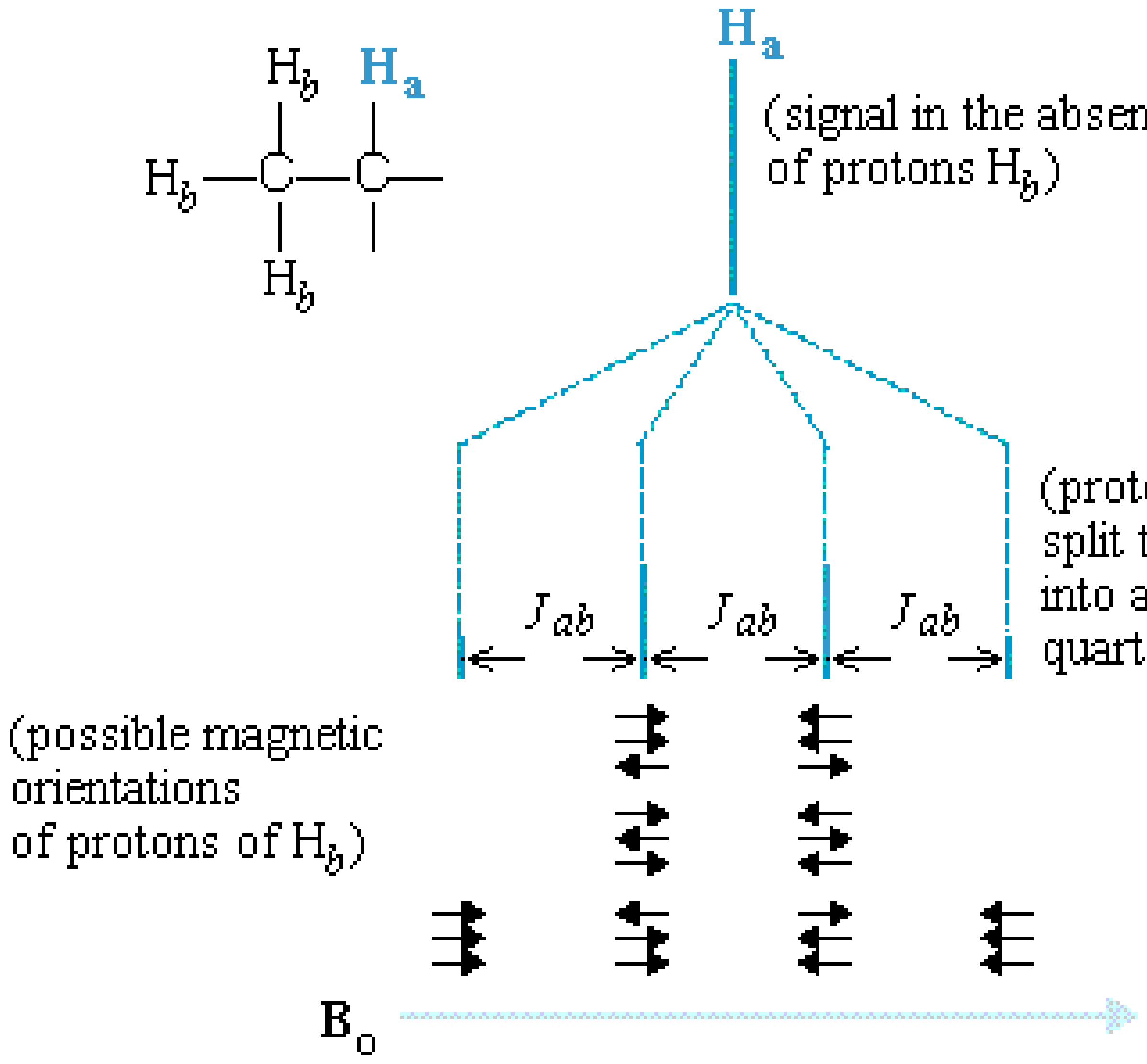
FIGURE 13.12 The 300 MHz proton 1H NMR spectrum of 1,1,2-trichloroethane. Expansions of the signals are shown in the offset plots.

Molécula simétrica:

Proporção de 2:1



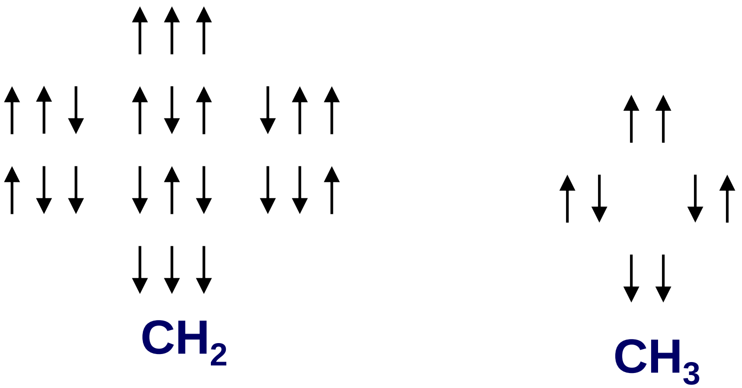
1,1,2,3,3-pentacloropropano

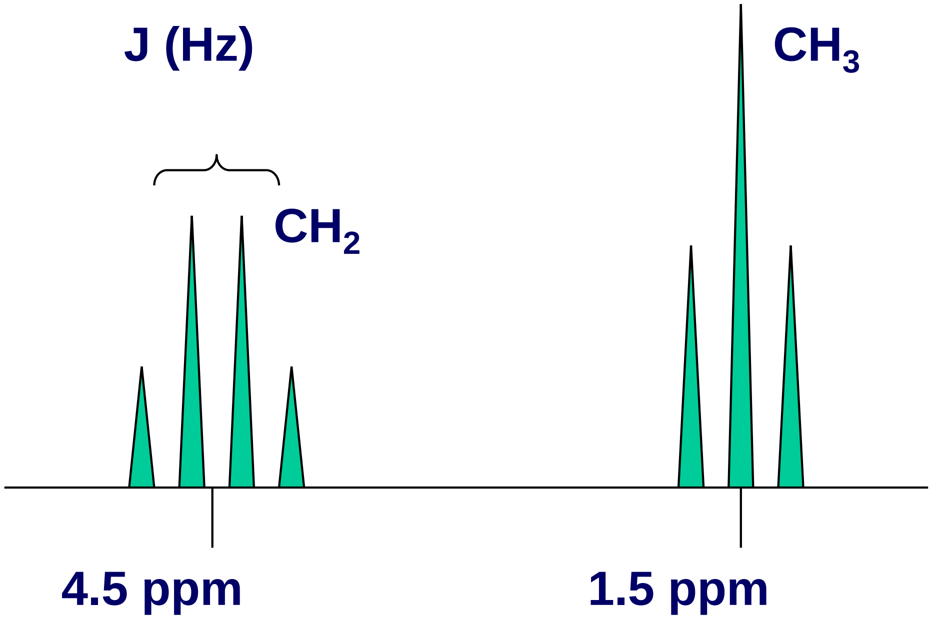


(signal in the absence of protons H_b)

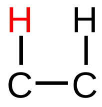
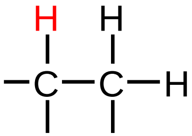
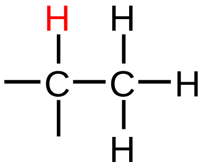
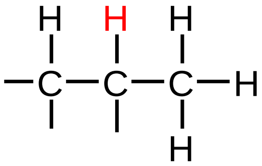
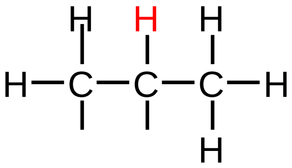
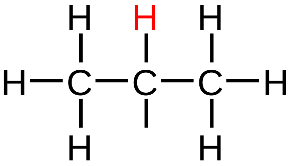
(protons of H_b split the signal into a 1:3:3:1 quartet)

(possible magnetic orientations of protons of H_b)


CH_2
CH_3

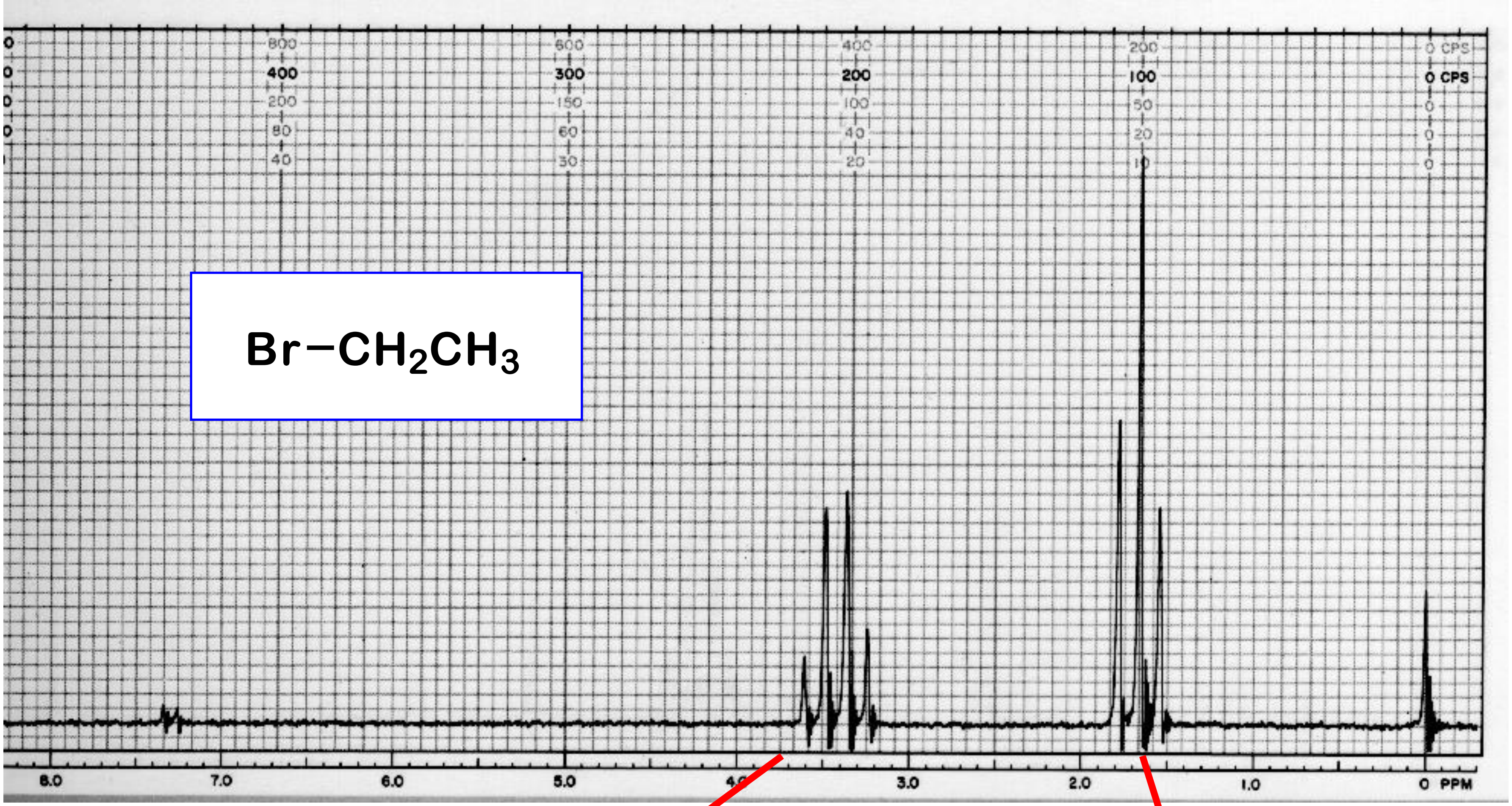

J (Hz)
CH_2
CH_3
4.5 ppm
1.5 ppm

Expected signal multiplicities for a given hydrogen

no. of neighbors	relative intensities	pattern	example
0	1	singlet (s)	
1	1 1	doublet (d)	
2	1 2 1	triplet (t)	
3	1 3 3 1	quartet (q)	
4	1 4 6 4 1	pentet	
5	1 5 10 10 5 1	sextet	
6	1 6 15 20 15 6 1	septet	

Pascal triangle

Bromoetano

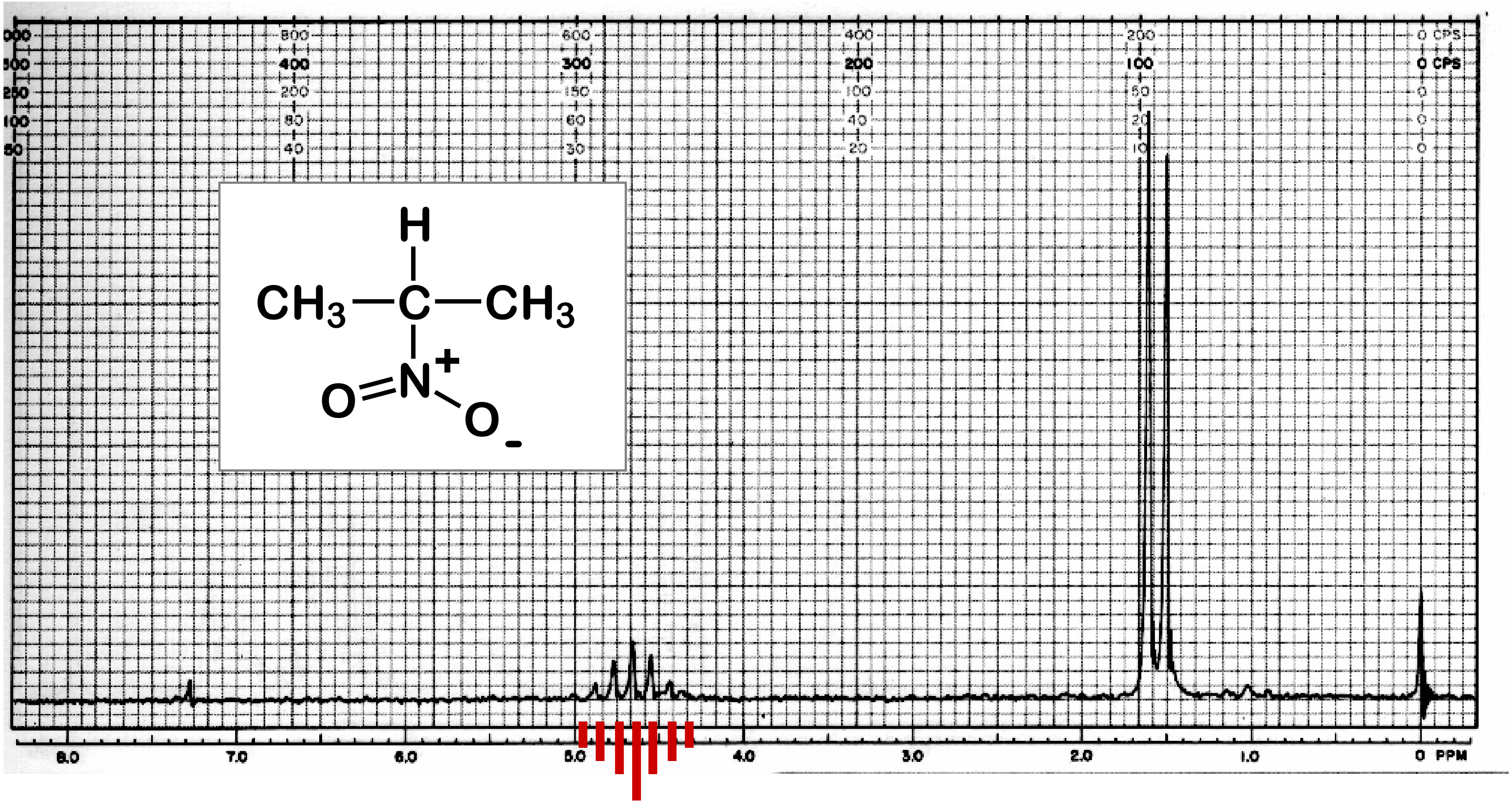


$Br-CH_2CH_3$

CH$_2$: acopla com CH$_3$
Resultando em um quadrupleto

CH$_3$: acopla com CH$_2$
Resultando em um tripleto

2-Nitropropano

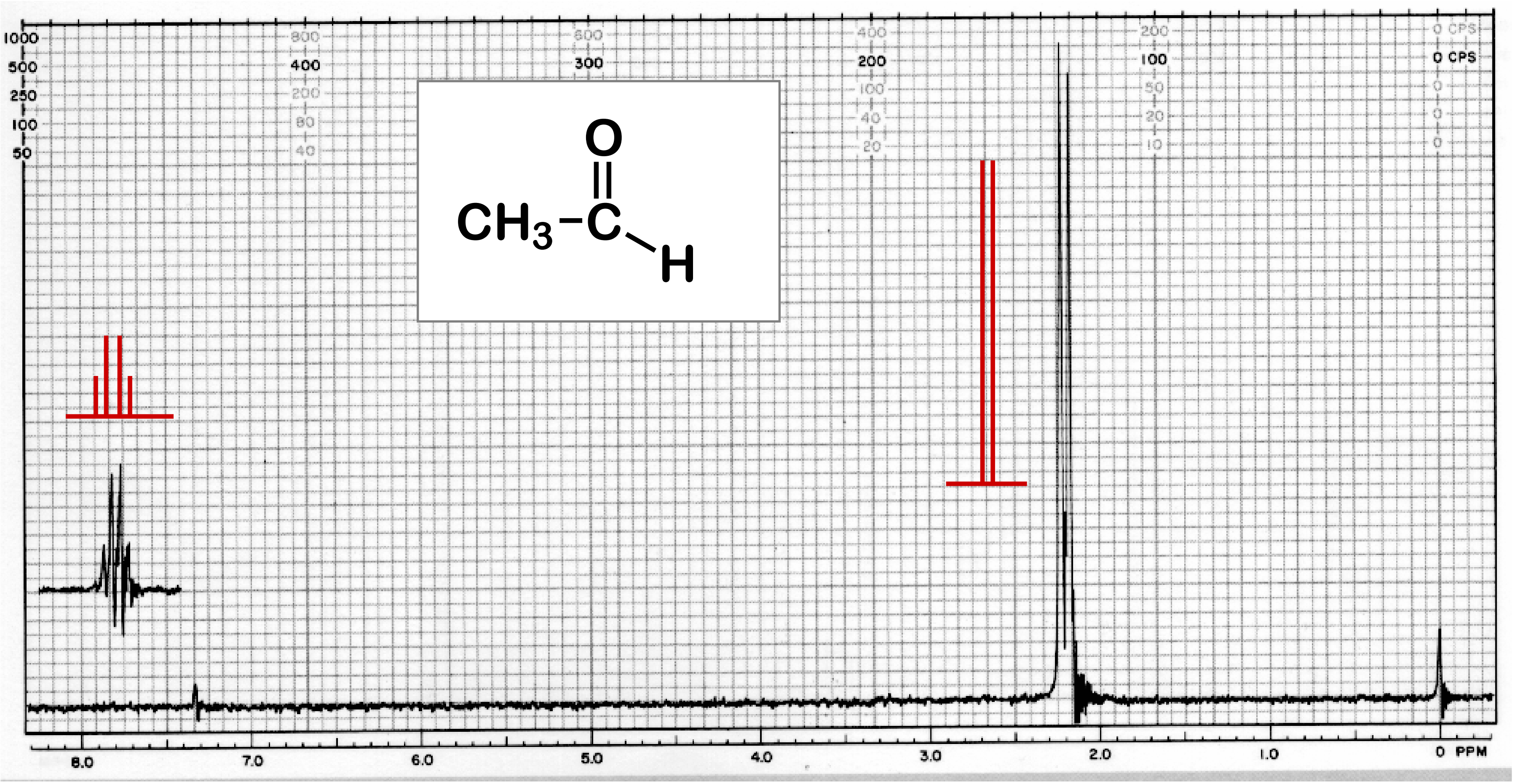


1:6:15:20:16:6:1

septupleto

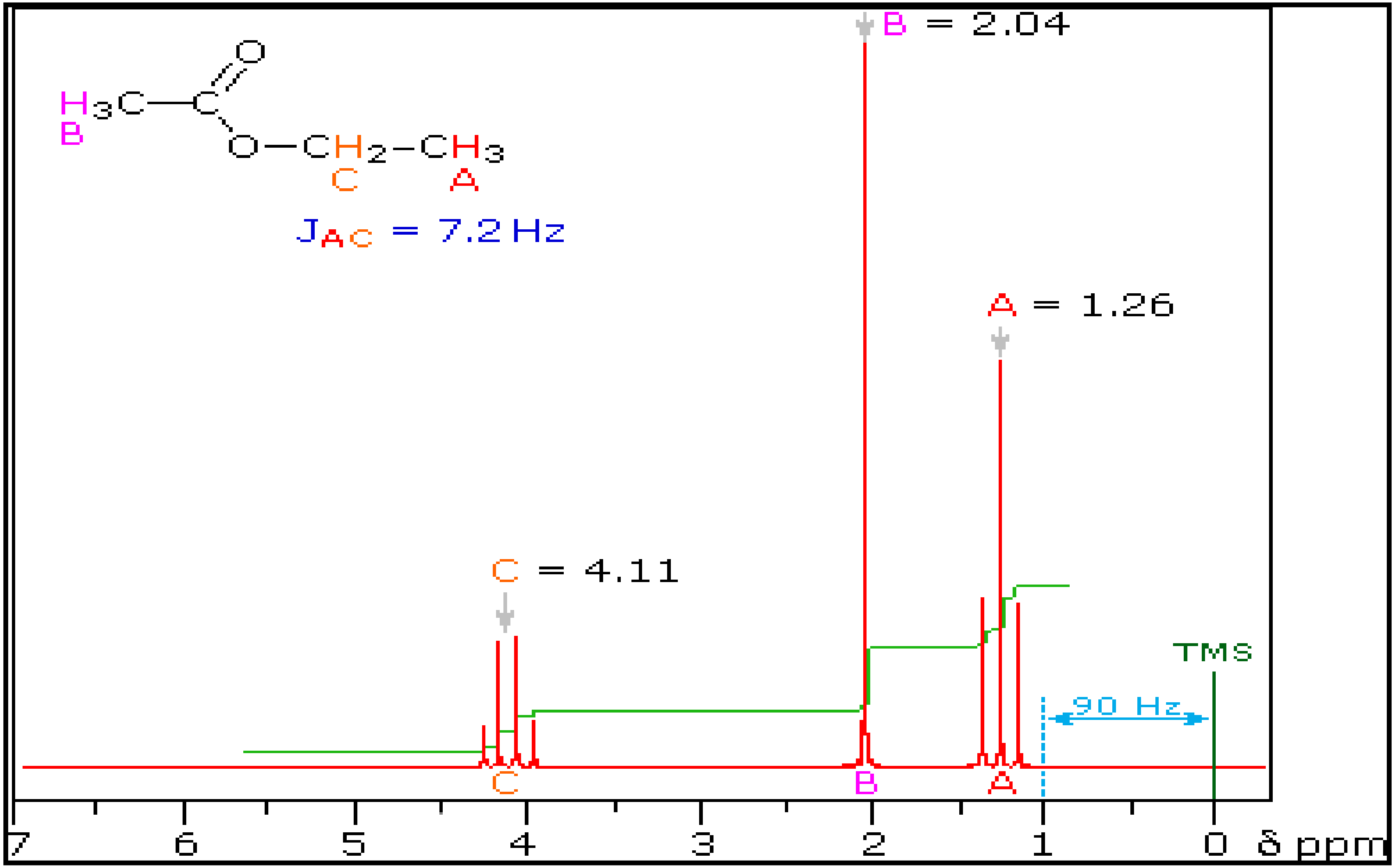
in higher multiplets the outer peaks are often nearly lost in the baseline

Acetaldeído

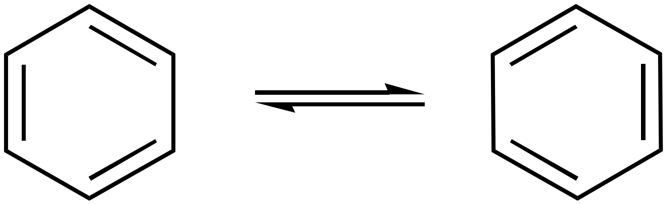


offset = 2.0 ppm

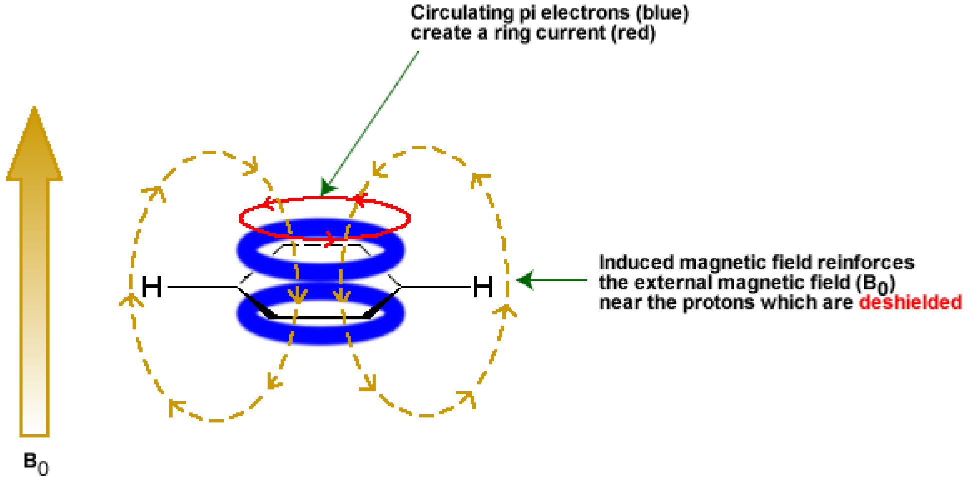
Acetato de etila



NMR spectrum of benzene

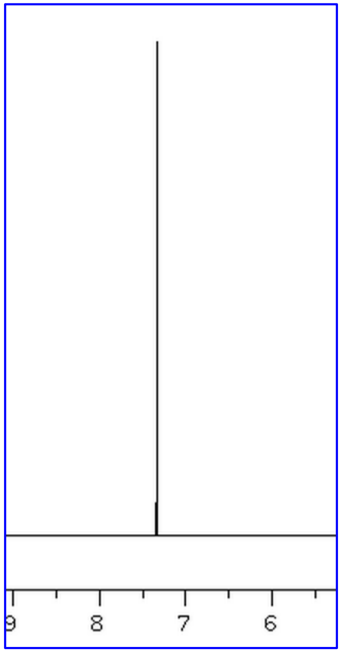


One single signal at 7.2 δ

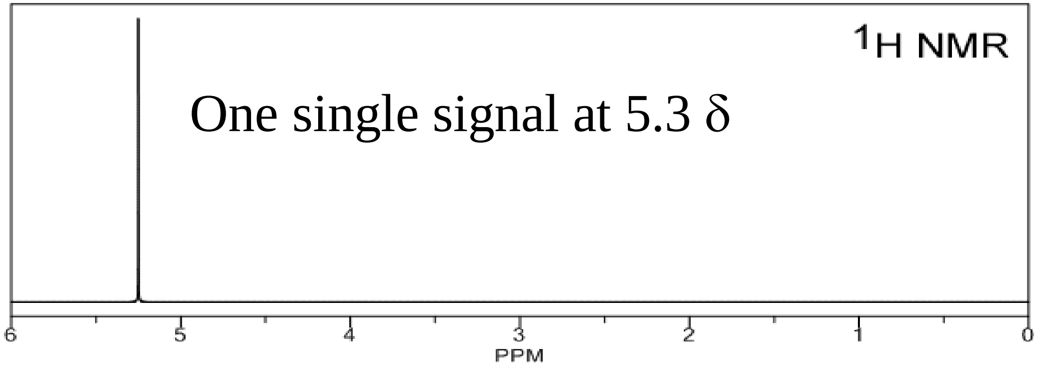


Circulating pi electrons (blue) create a ring current (red)

Induced magnetic field reinforces the external magnetic field (B_0) near the protons which are deshielded



^{1}H NMR spectrum of benzene

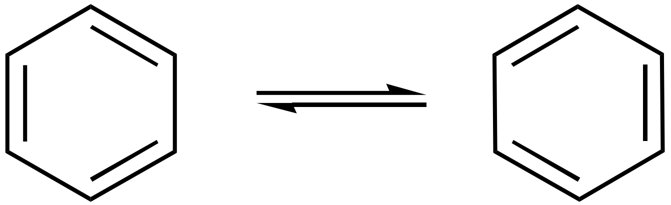


One single signal at 5.3 δ

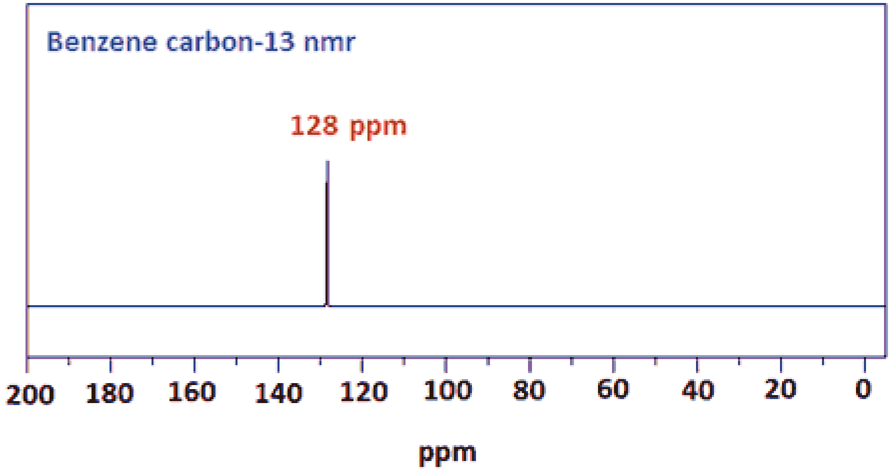
^{1}H NMR spectrum of ethylene

$H_2C=CH_2$

^{13}C NMR spectrum of benzene

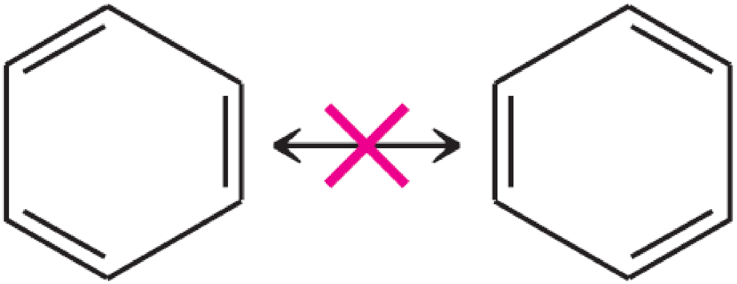


One signal at 128 δ

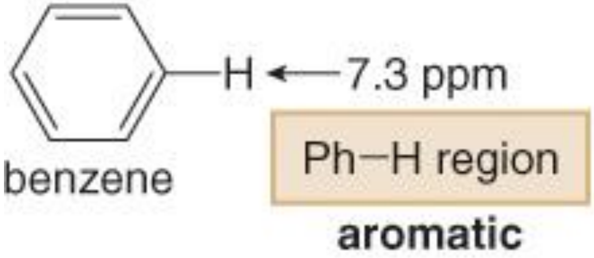


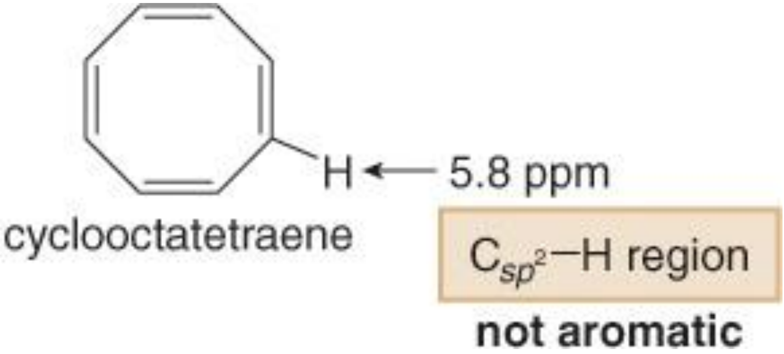
^{13}C NMR spectrum of benzene

Not compatible with:

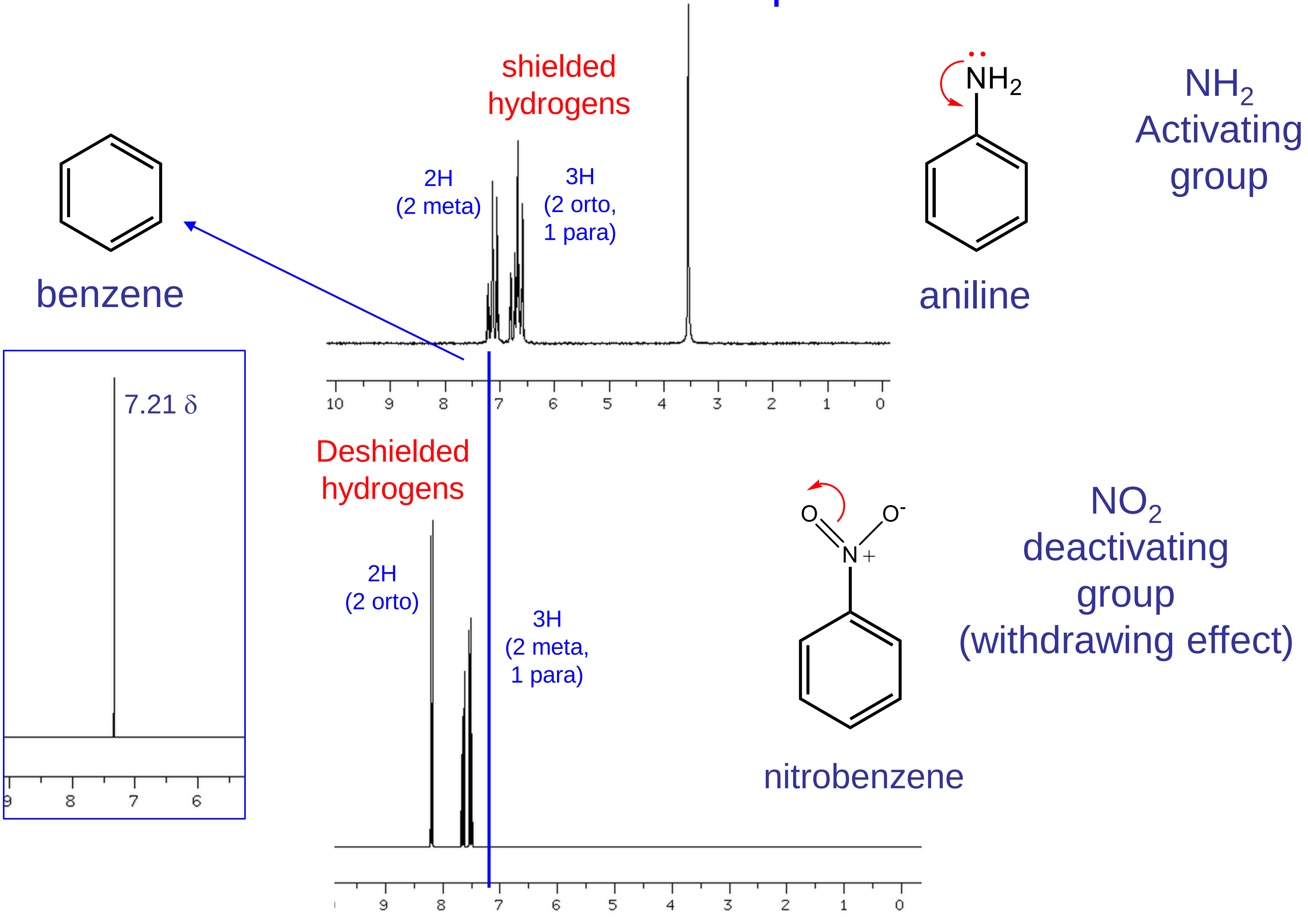


- **^{1}H NMR spectroscopy readily indicates whether a compound is aromatic.**
- **The protons on *sp*^{2} hybridized carbons in aromatic hydrocarbons are highly deshielded and absorb at 6.5-8 ppm, whereas hydrocarbons that are not aromatic absorb at 4.5-6 ppm.**





Substituent Effects on aromatic compounds: ^1H NMR data



shielded hydrogens

2H
(2 meta)

3H
(2 orto,
1 para)

aniline

NH_2
Activating
group

benzene

7.21 δ

Deshielded hydrogens

2H
(2 orto)

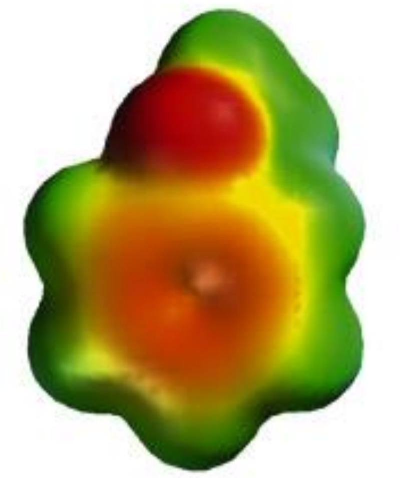
3H
(2 meta,
1 para)

nitrobenzene

NO_2
deactivating
group
(withdrawing effect)

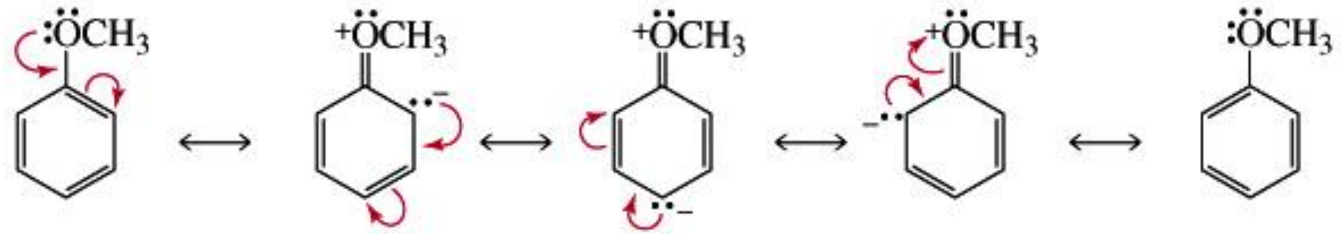
NMR spectra of benzenoid compounds

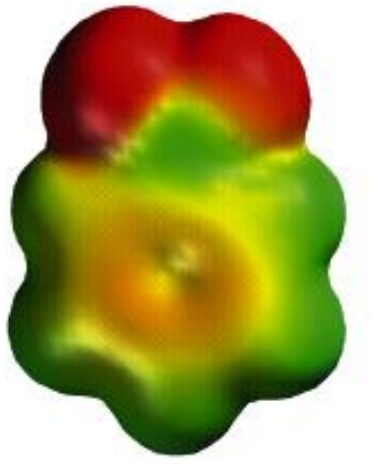
Efeito de Substituintes: (ii) Efeitos de Ressonância



anisole

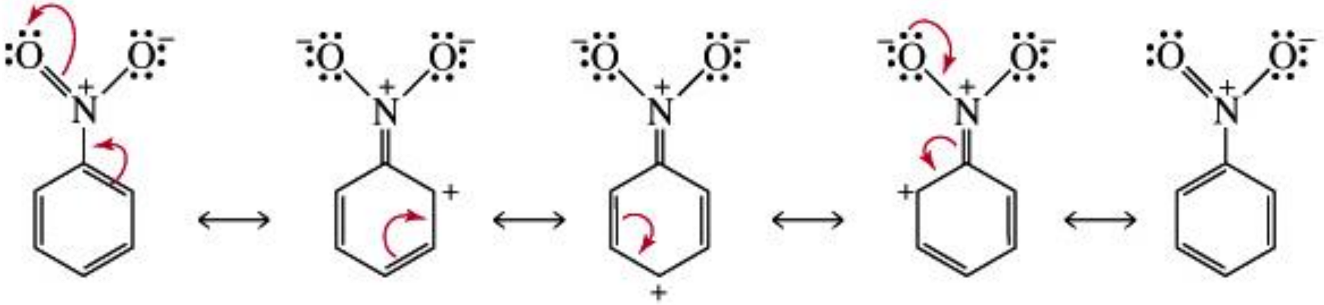
Doação de elétrons pelo efeito de ressonância (mesomérico)



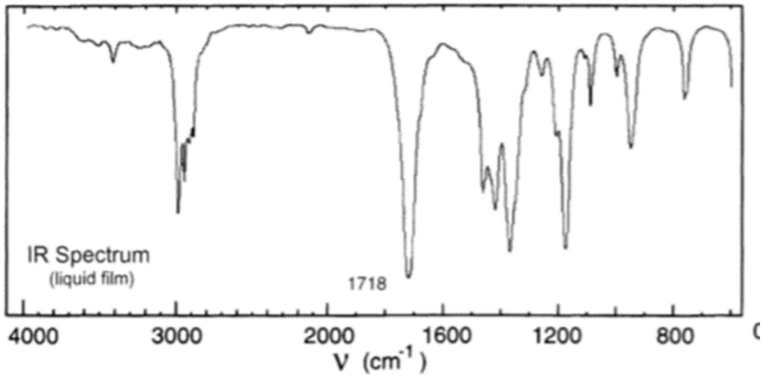


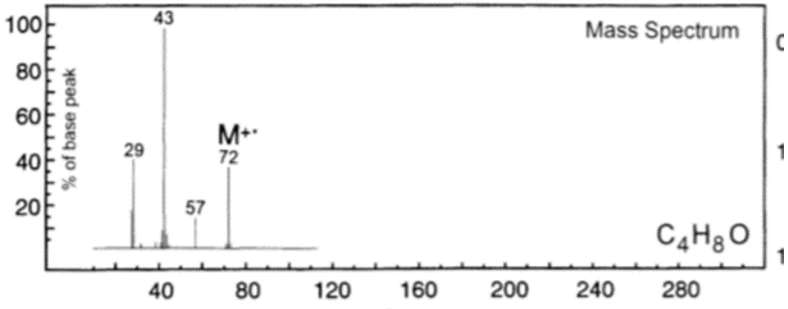
nitrobenzene

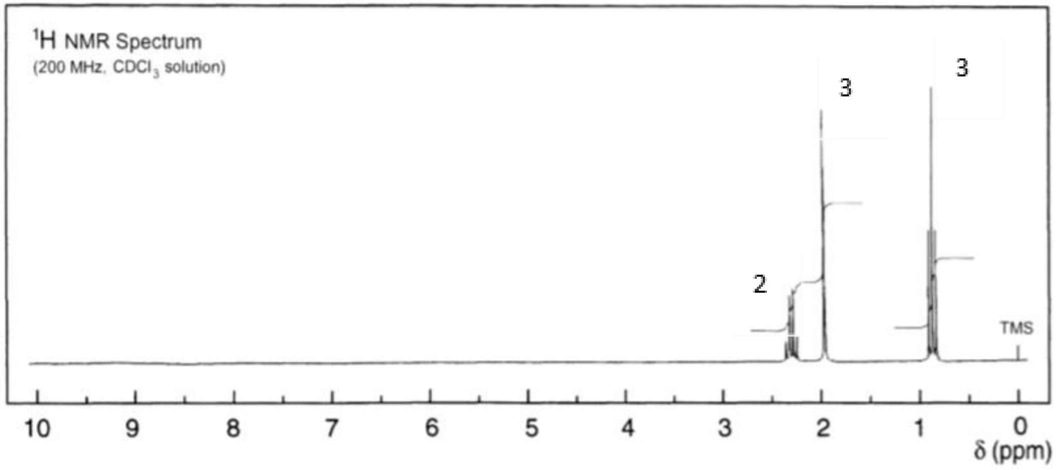
Atração de elétrons pelo efeito de ressonância (mesomérico)



Determine a estrutura do composto através dos dados de IV, EM e RMN de ^1H







RMN de ^{13}C:

1) Deslocamentos químicos
   (δ / 0-210ppm);

2) Acoplamentos ^{1}H e ^{13}C

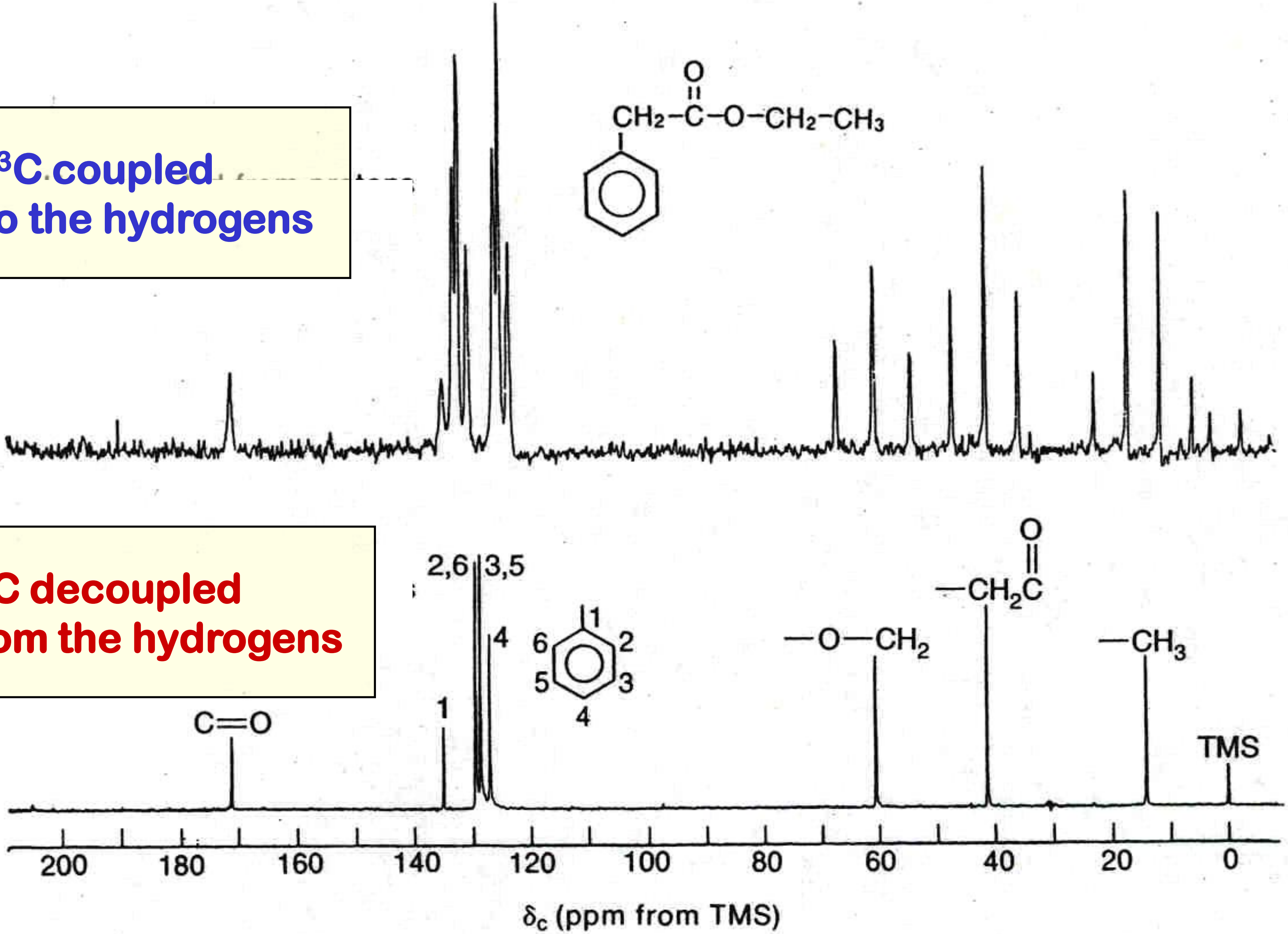
3) Integrações

Tipos de espectros de RMN de ^{13}C

PND: proton noise decoupled

(não há acoplamento entre entre ^{13}C e ^{1}H e cada carbono (C, CH, CH_3 e CH_3) se apresenta como um *singleto*)

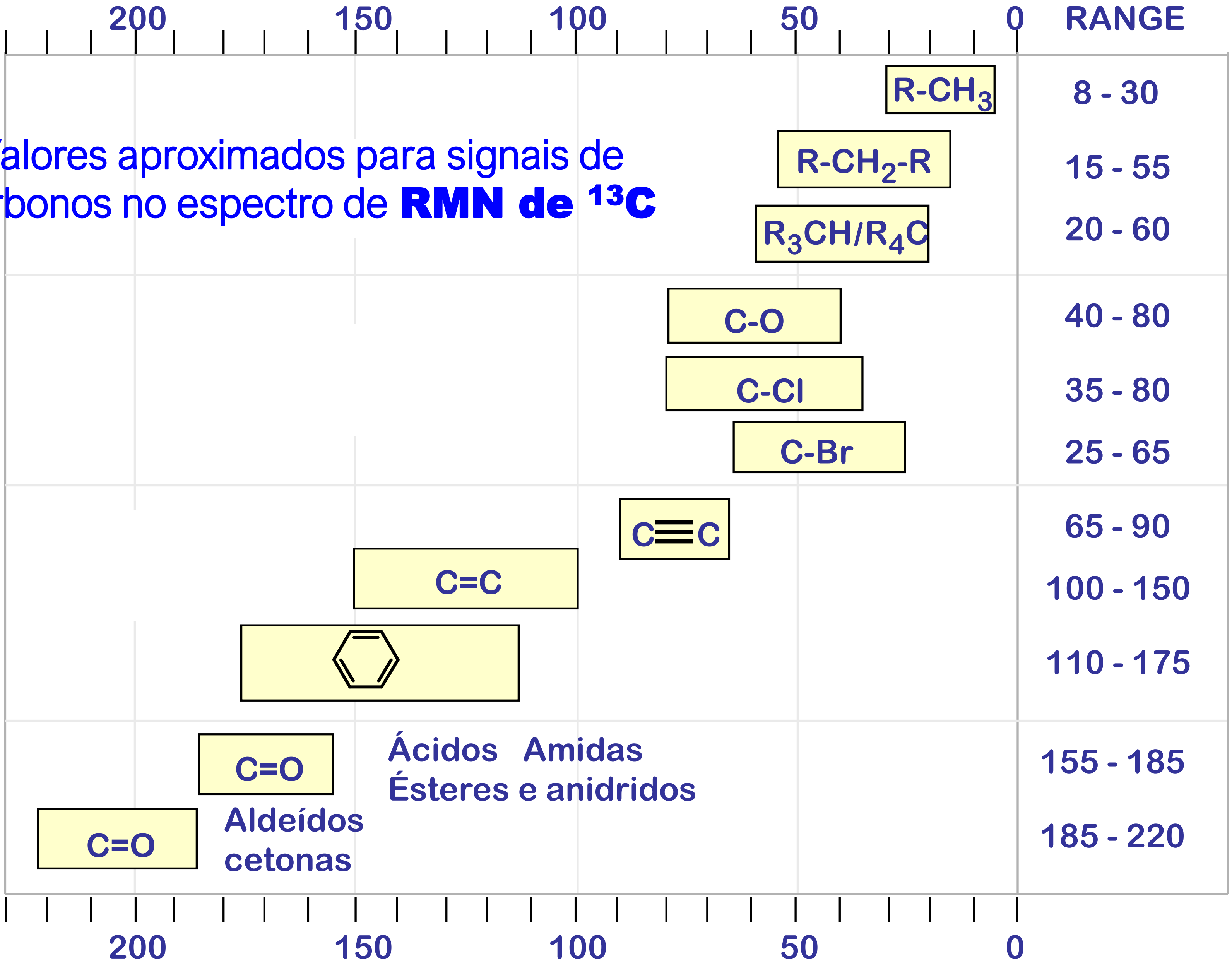
ETHYL PHENYLACETATE



^{13}C coupled
to the hydrogens

^{13}C decoupled
from the hydrogens

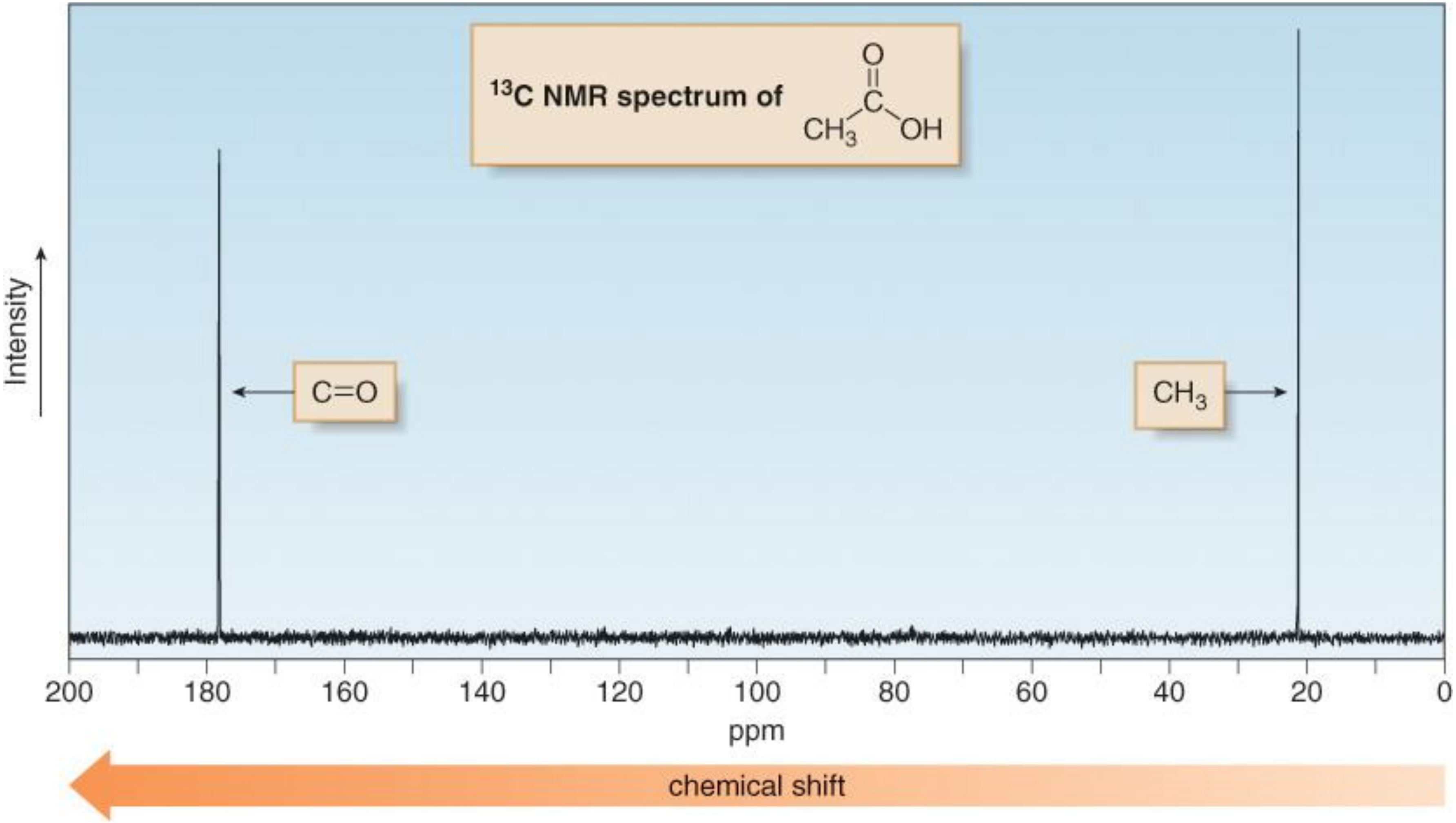
Valores aproximados para signais de carbonos no espectro de **RMN de ^{13}C**



	RANGE
$R-CH_3$	8 - 30
$R-CH_2-R$	15 - 55
R_3CH/R_4C	20 - 60
C-O	40 - 80
C-Cl	35 - 80
C-Br	25 - 65
C≡C	65 - 90
C=C	100 - 150
	110 - 175
C=O Ácidos Amidas Ésteres e anidridos	155 - 185
C=O Aldeídos cetonas	185 - 220

^{13}C Nuclear Magnetic Resonance Spectroscopy

^{13}C NMR spectra are easier to analyze than ^{1}H spectra because the signals are not split. Each type of carbon atom appears as a single peak.

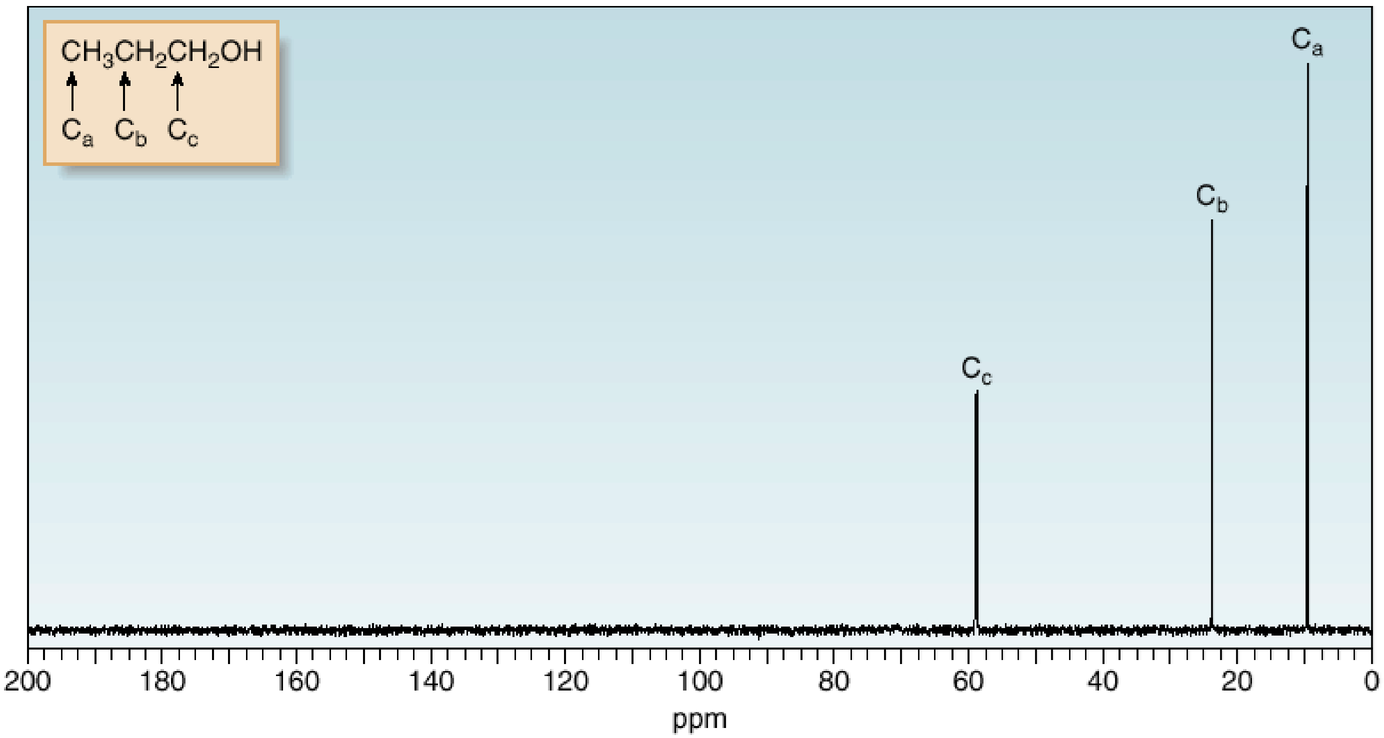


Nuclear Magnetic Resonance Spectroscopy

^{13}C NMR—Number of Signals

- The three types of C's in 1-propanol—identified as C_a, C_b, and C_c—give rise to three ^{13}C NMR signals.
- Deshielding increases with increasing proximity to the electronegative O atom, and the absorption shifts downfield; thus, in order of increasing chemical shift: $C_a < C_b < C_c$.

a. 1-Propanol

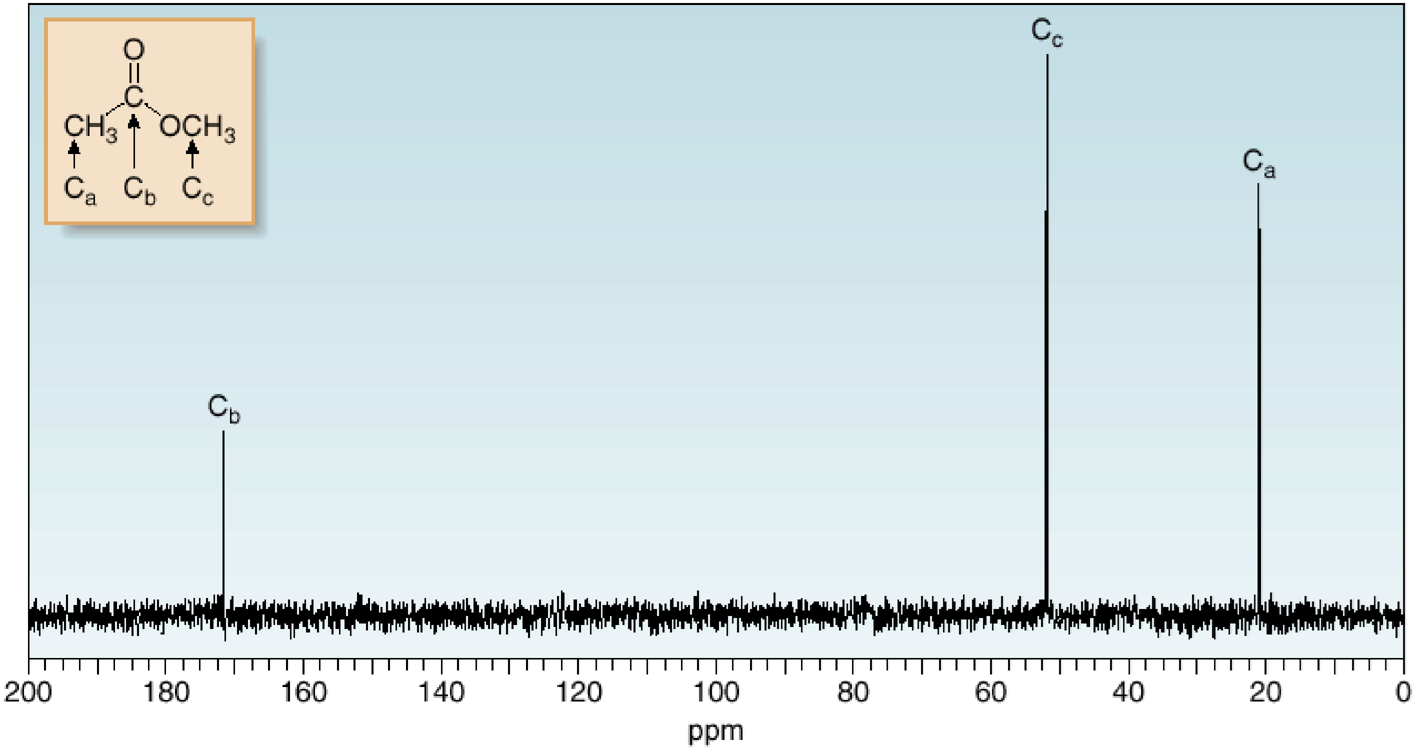


Nuclear Magnetic Resonance Spectroscopy

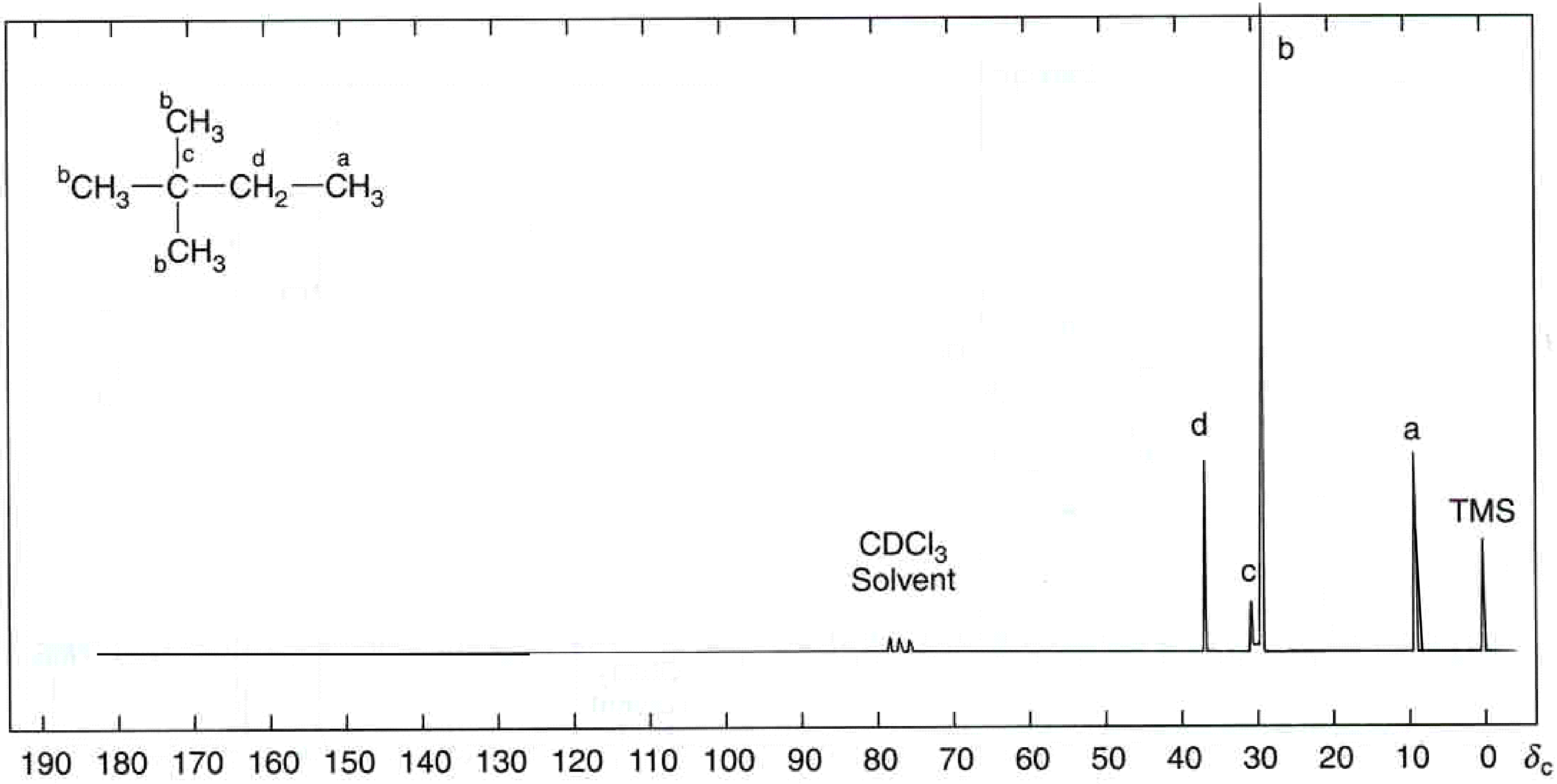
^{13}C NMR—Number of Signals

- The three types of C's in methyl acetate—identified as C_a, C_b, and C_c—give rise to three ^{13}C NMR signals.
- The carbonyl carbon (C_b) is highly deshielded, so it absorbs farthest downfield.
- C_a, an *sp*^{3} hybridized C that is not bonded to an O atom, is the most shielded, and so it absorbs farthest upfield.
- Thus, in order of increasing chemical shift: $C_a < C_c < C_b$.

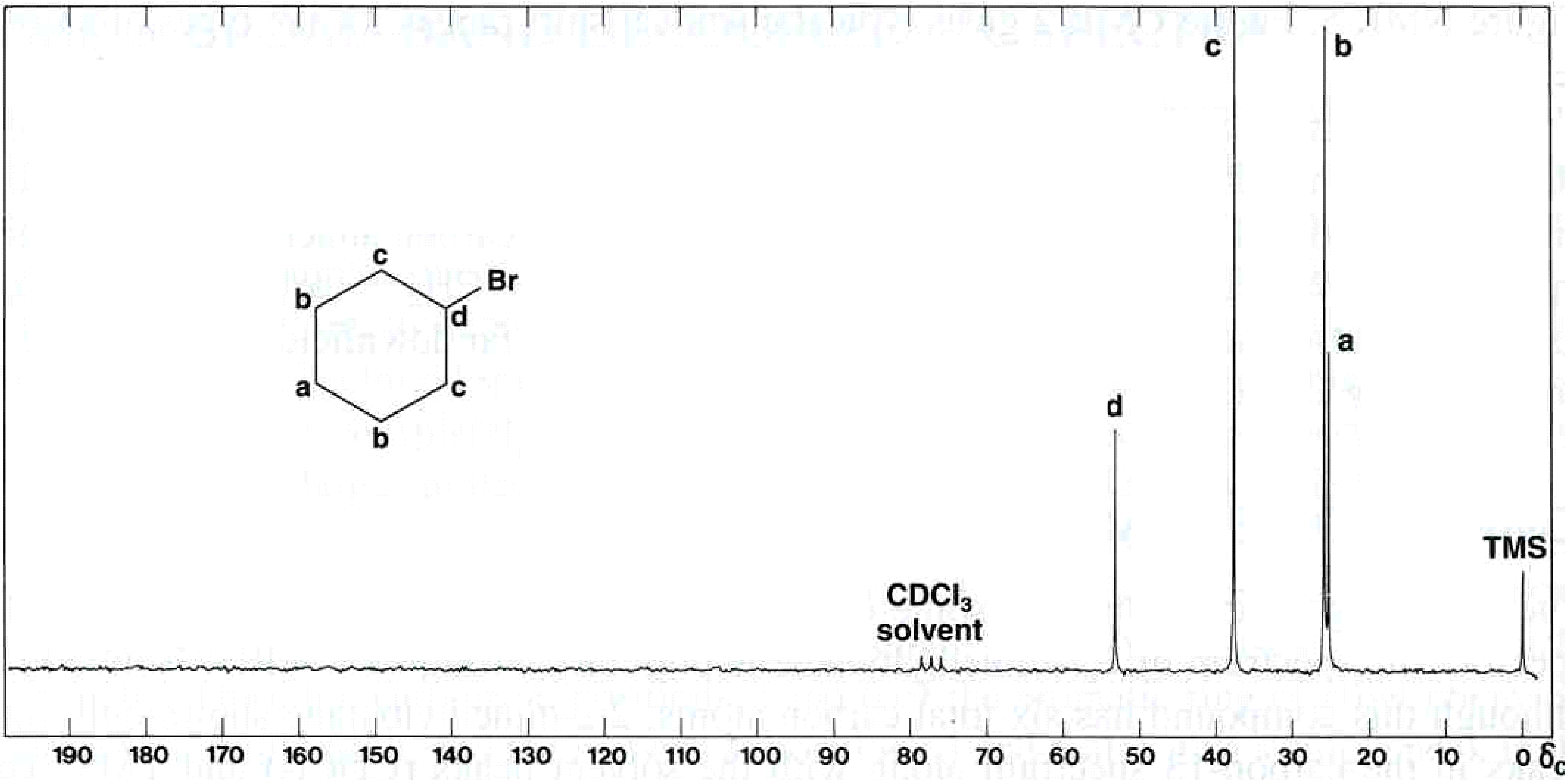
b. Methyl acetate



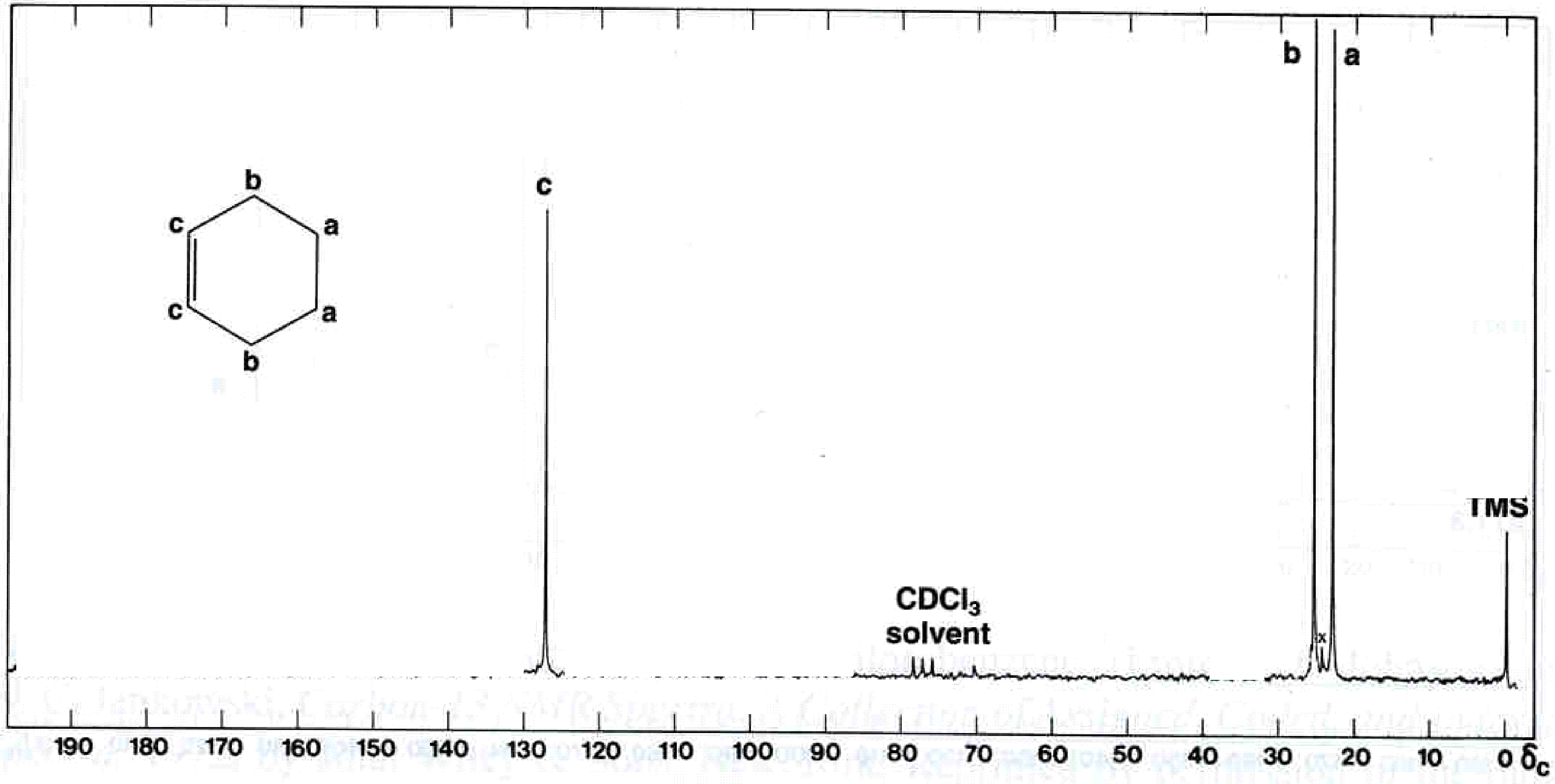
2,2-DIMETILBUTANO



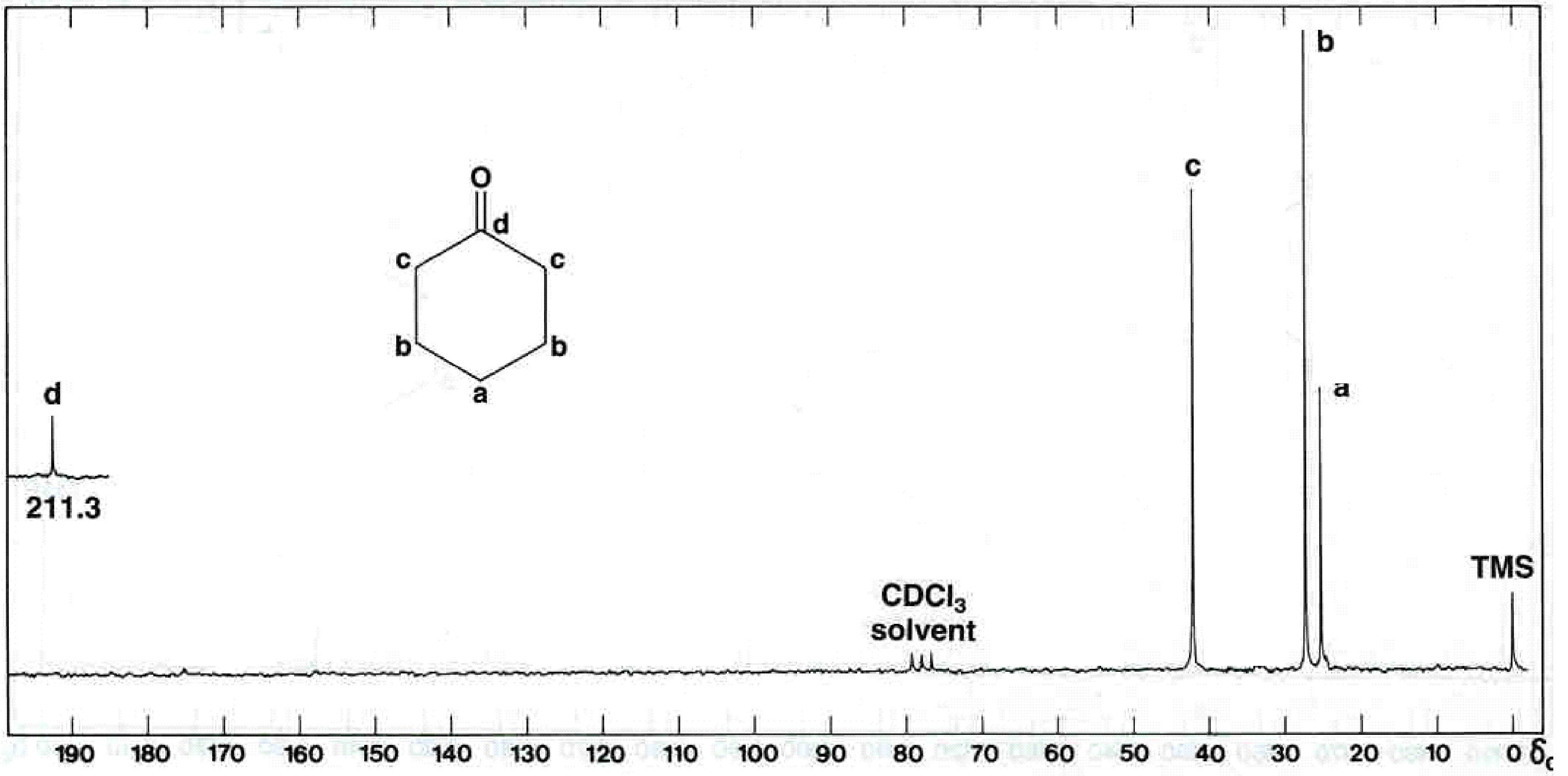
BROMOCICLOHEXANO

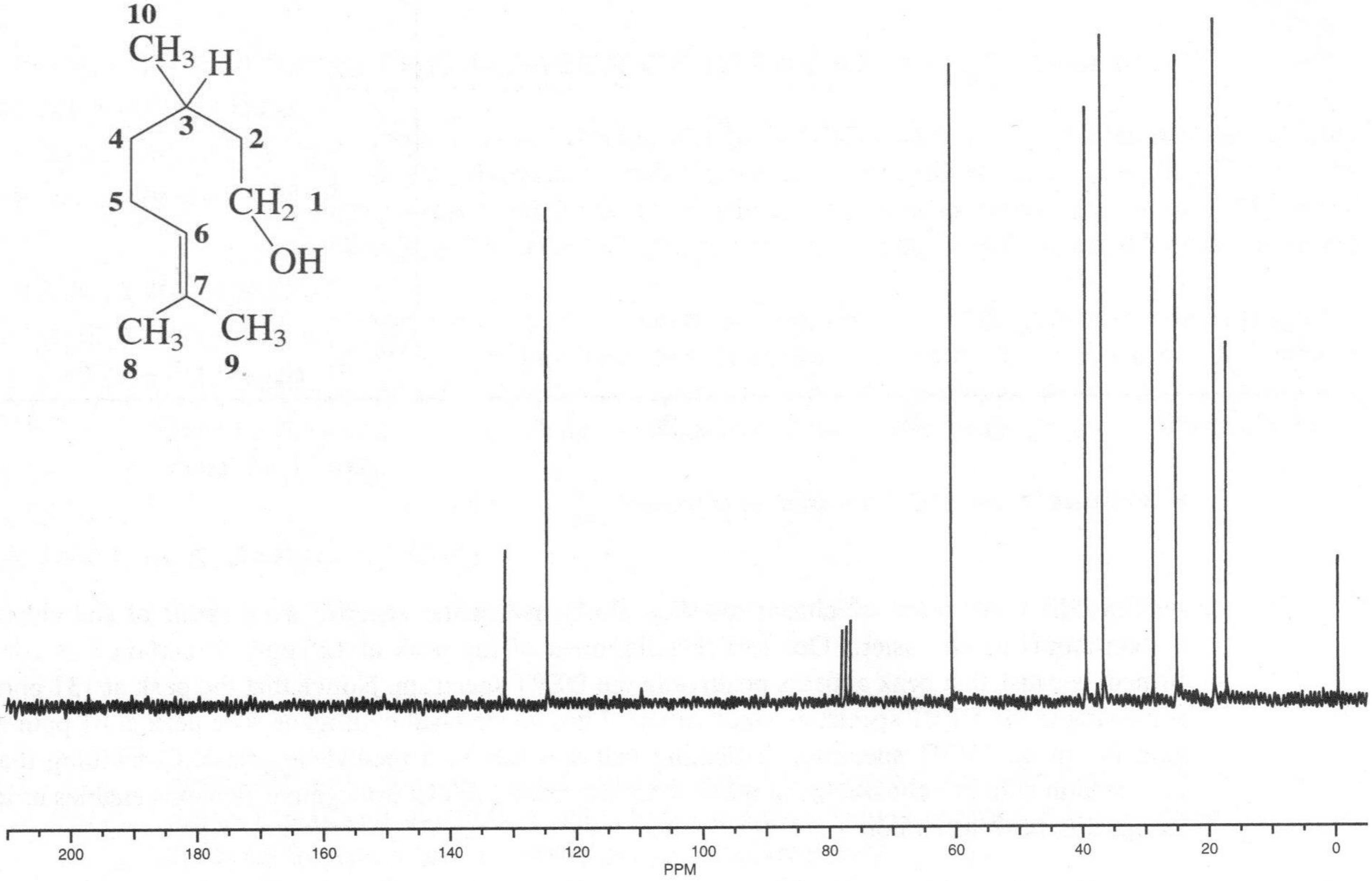


CICLOHEXENO

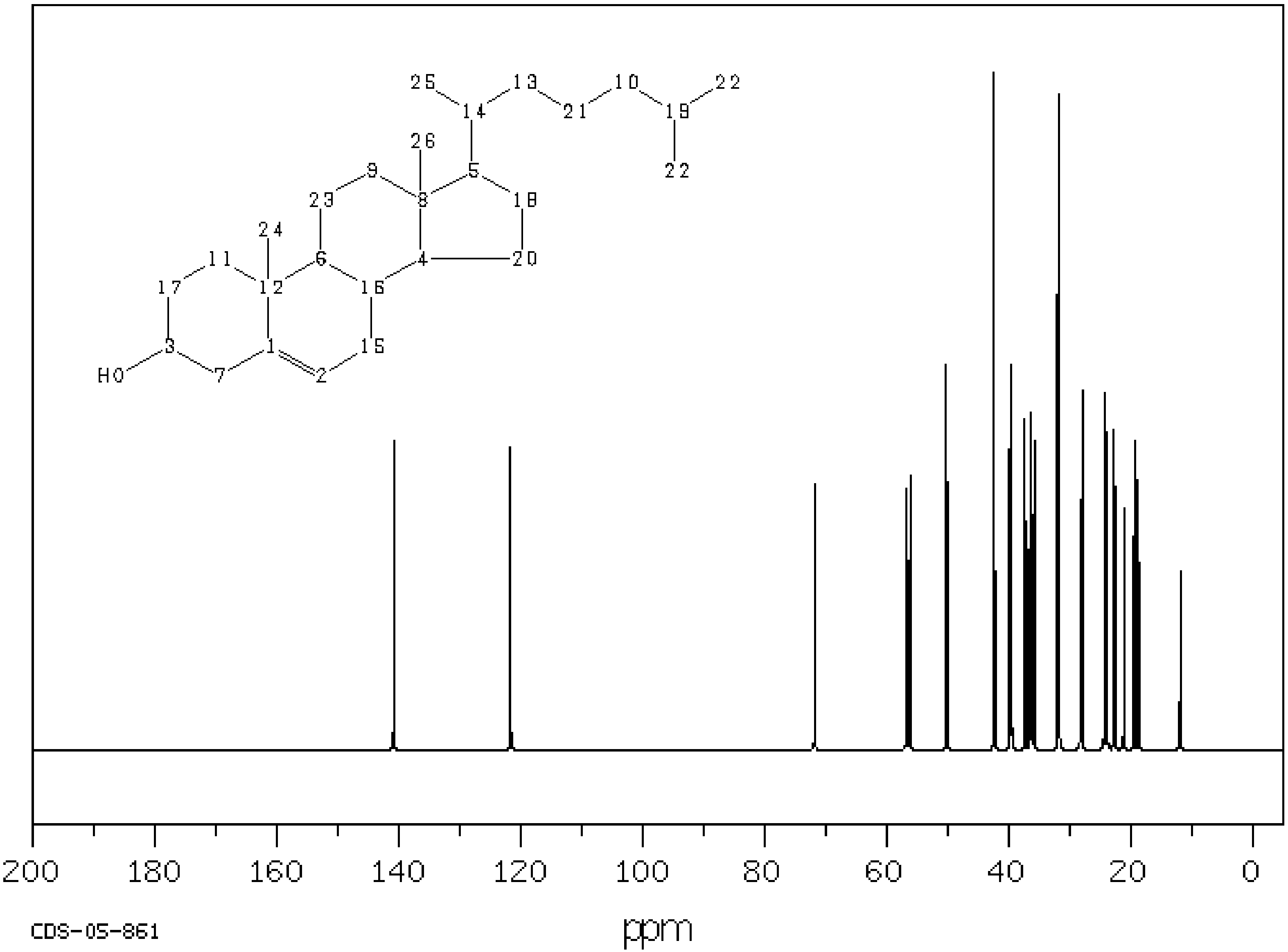


CICLOHEXANONA





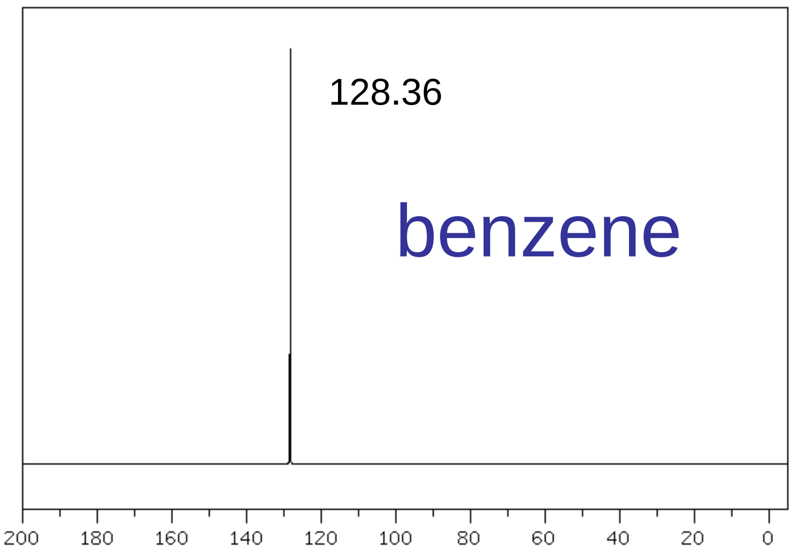
▶ **FIGURE 9.10** ^{13}C NMR spectrum of citronellol.

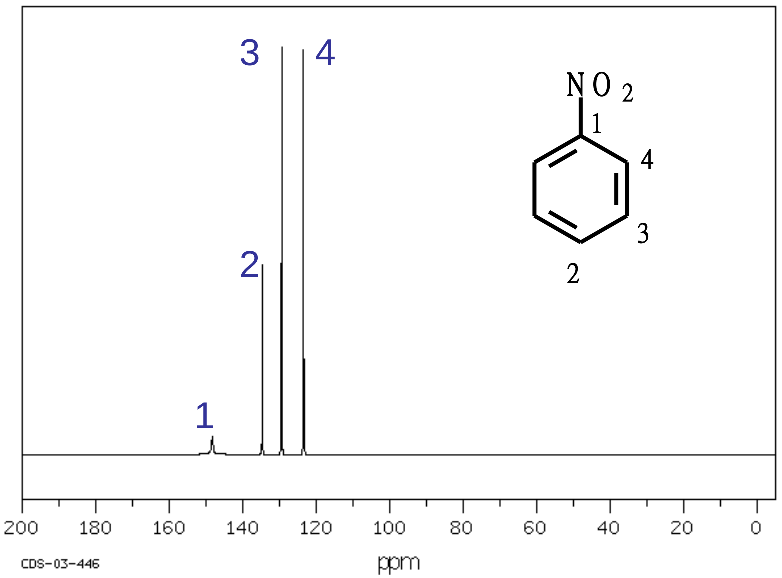


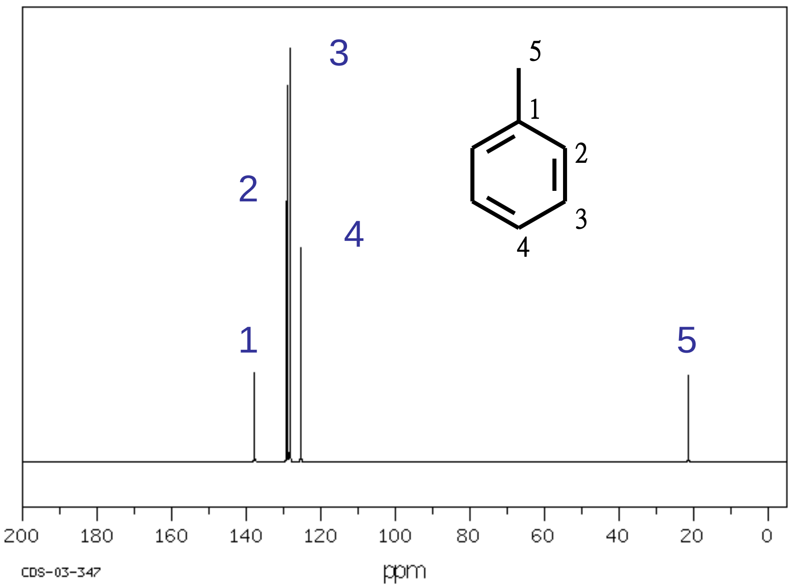
25
14
13
21
10
19
22
22
26
9
5
23
8
18
24
6
4
20
11
17
12
16
3
1
15
HO
7
2
200
180
160
140
120
100
80
60
40
20
0
ppm

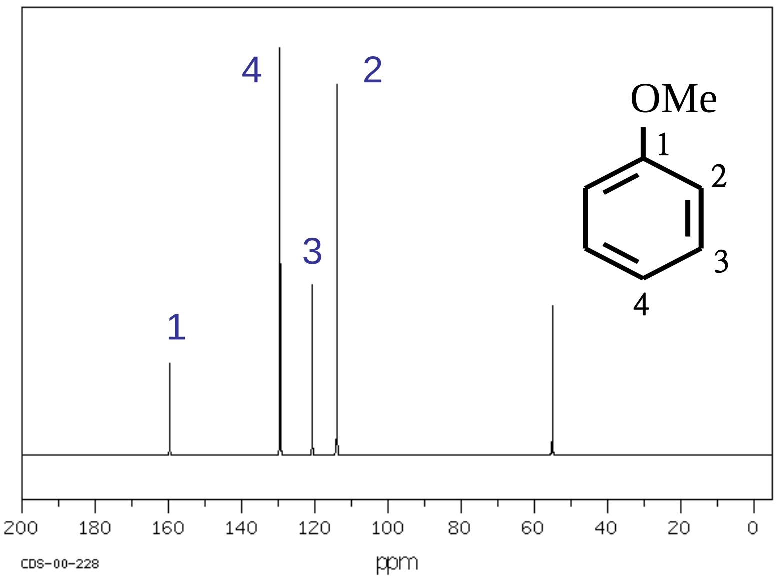
CDS-05-861

NMR ^{13}C of benzene derivatives

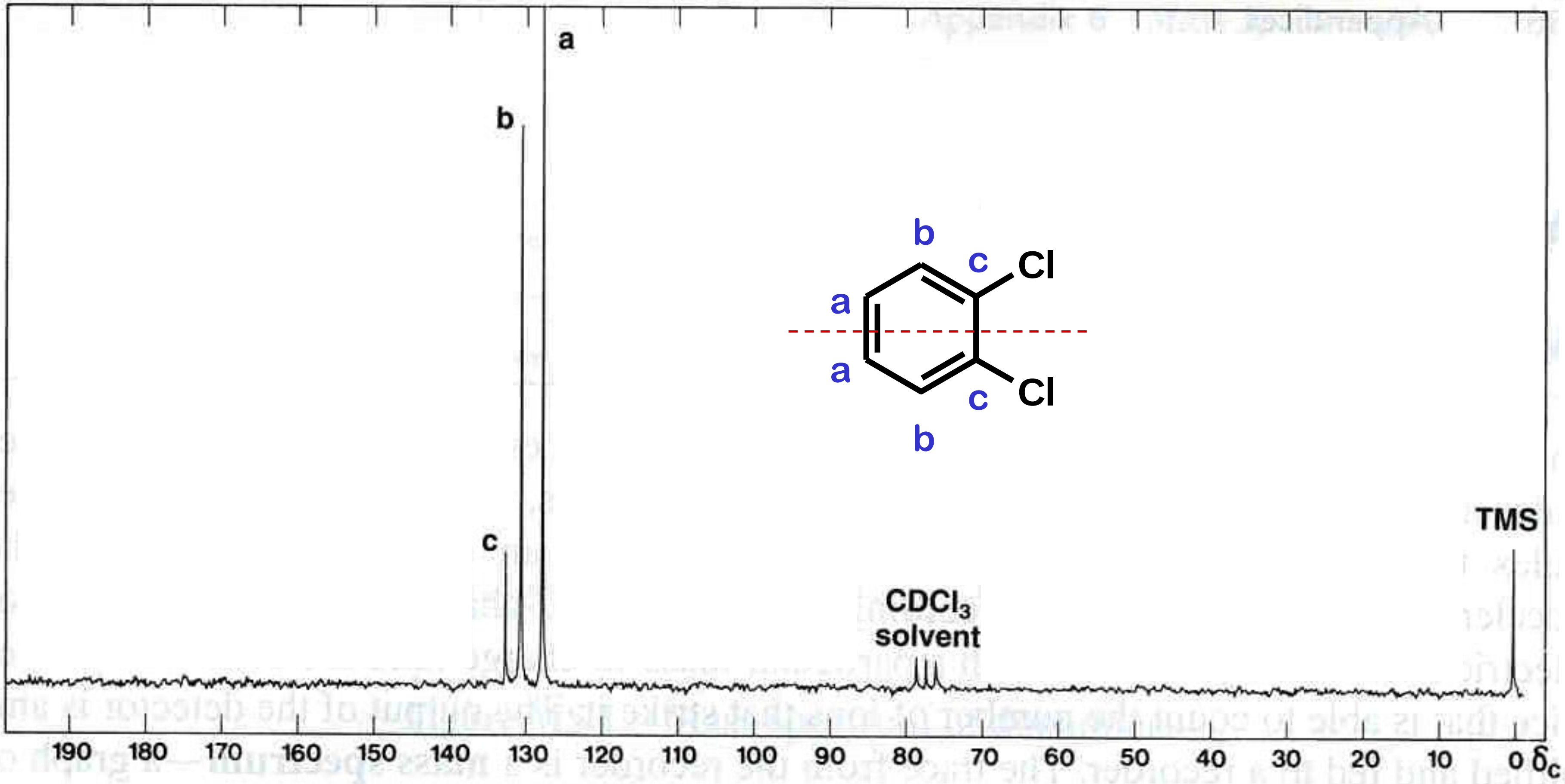




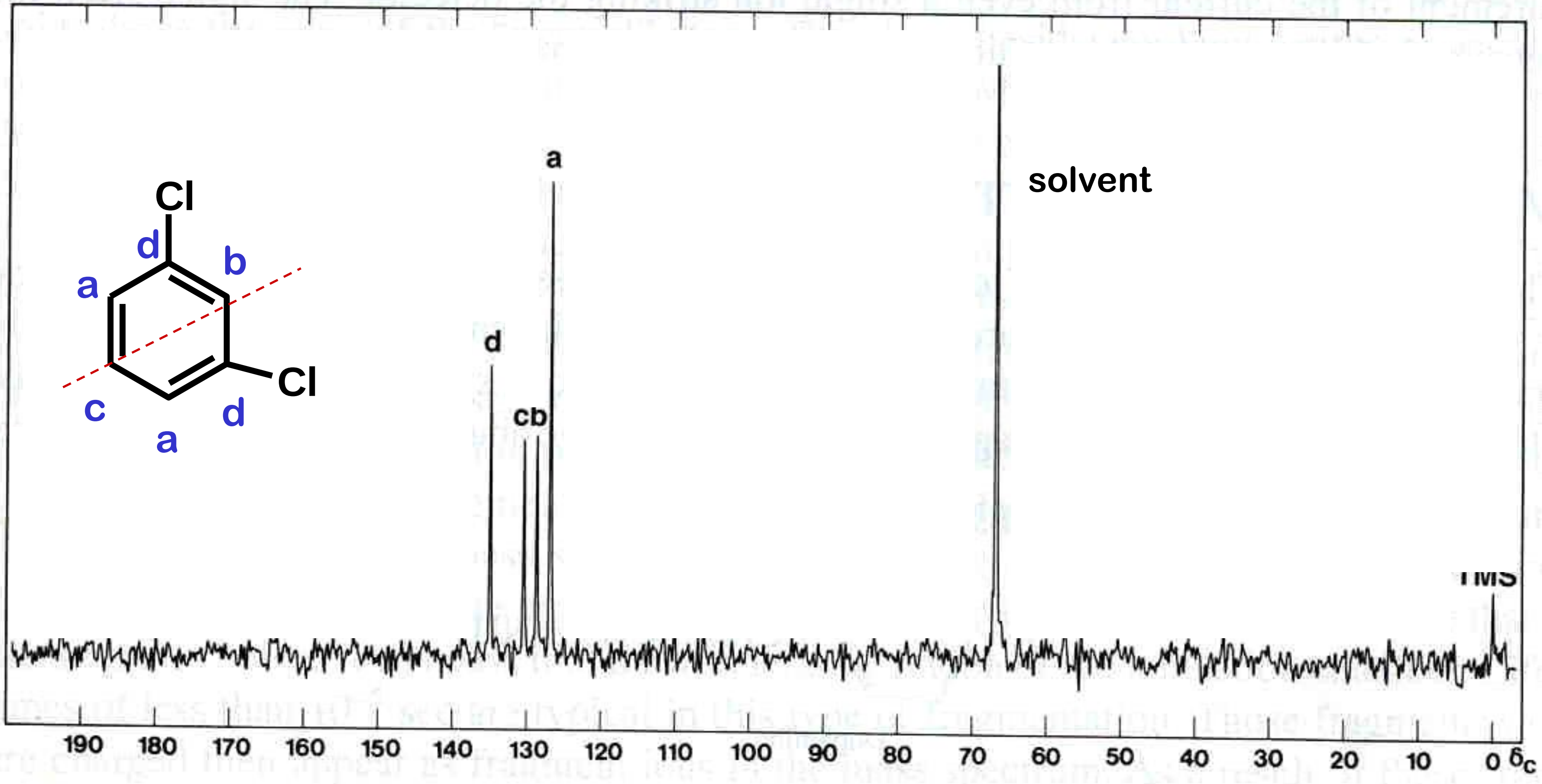




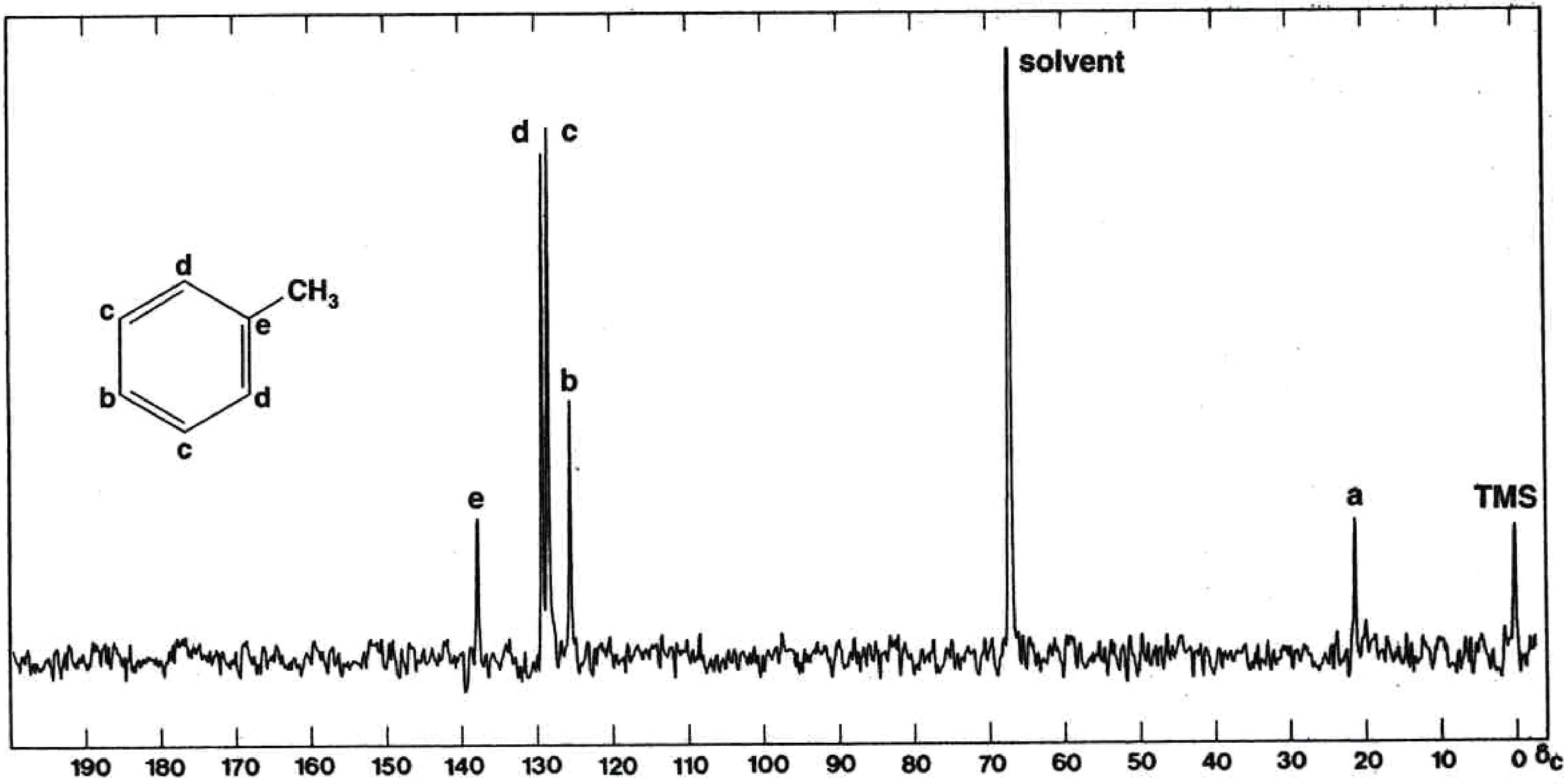
1,2-DICLOROBENZENO

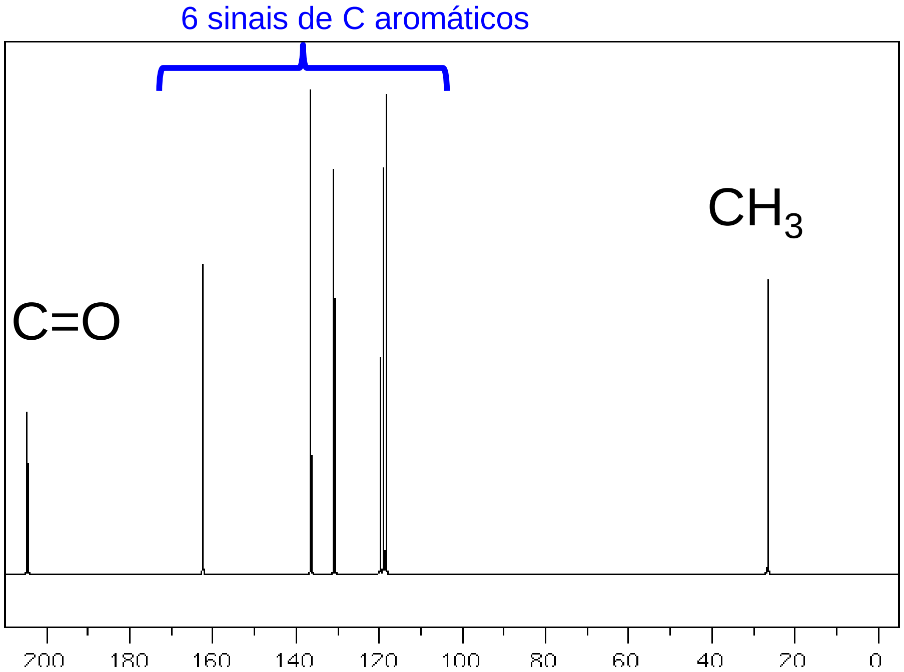


1,3-DICLOROBENZENO

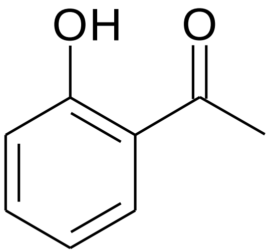


TOLUENO

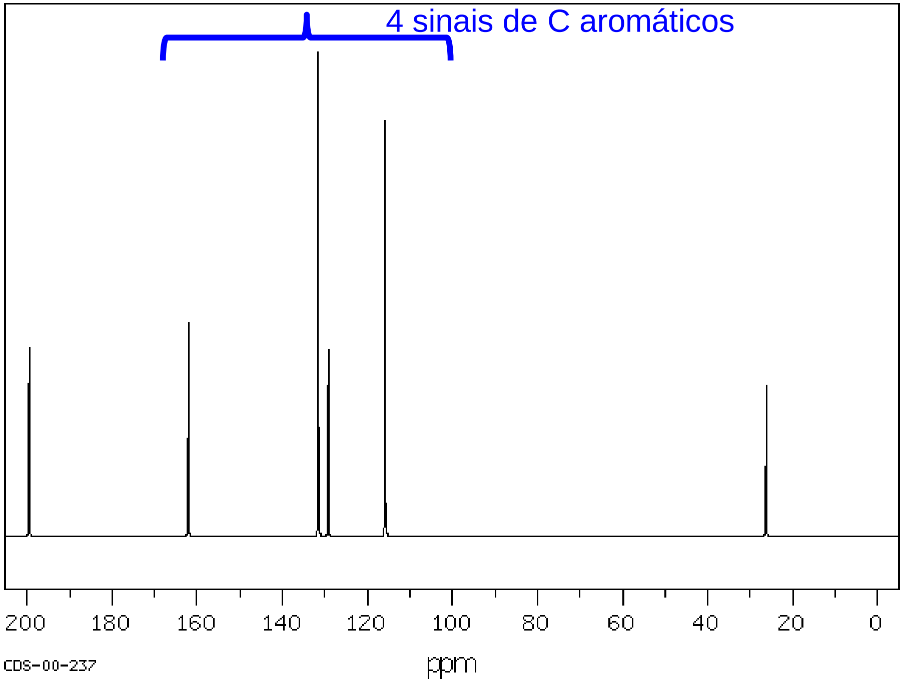

solvent
d
c
b
e
a
TMS
d
c
CH3
e
b
d
c
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
δC



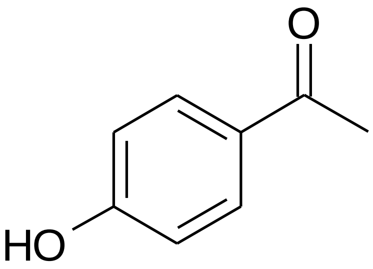
o-hidroxiacetofenona

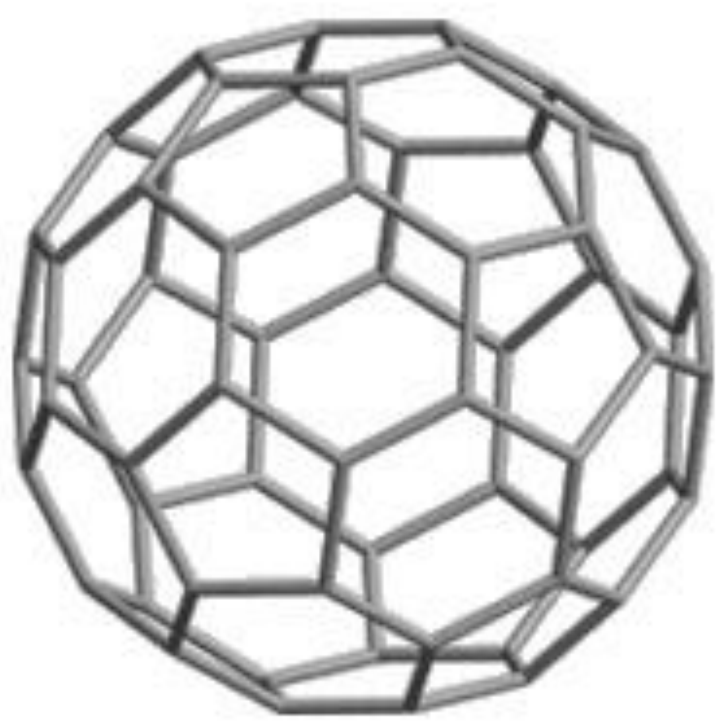


Espectros de RMN de ^{13}C

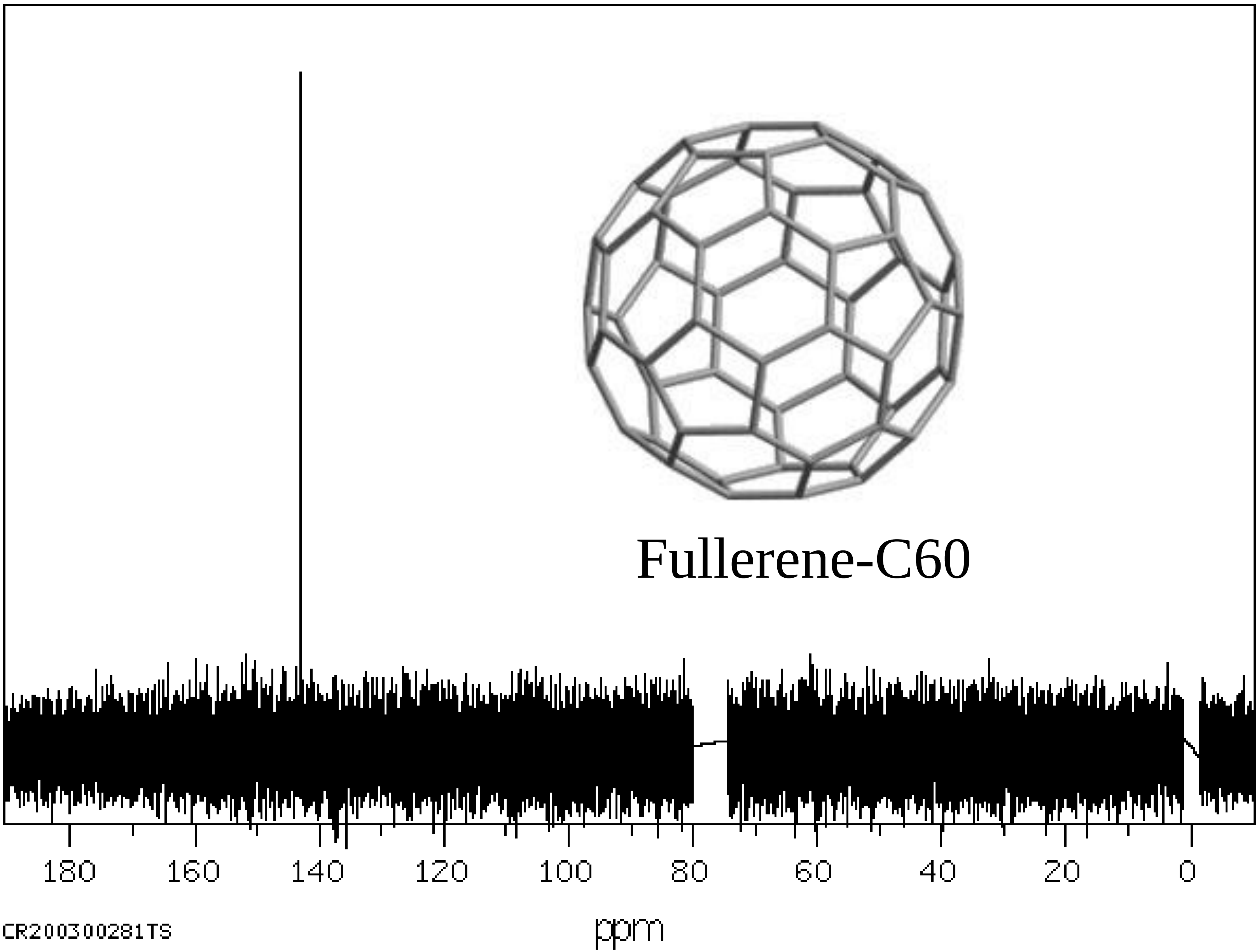


p-hidroxiacetofenona





Fullerene-C60 (all carbons are sp^2)


Fullerene-C60
180
160
140
120
100
80
60
40
20
0
ppm
CR200300281TS

ESPECTROS DE RMN FREQUENTEMENTE OBTIDOS PARA ELUCIDAÇÃO ESTRUTURAL

1D

^{1}H NMR; NOE

^{13}C NMR (PND; DEPT 135)

2D homo nuclear

^{1}H-^{1}H COSY

^{1}H-^{1}H NOESY

2D hetero nuclear

HMQC (Heteronuclear Multiple Quantum Correlation)

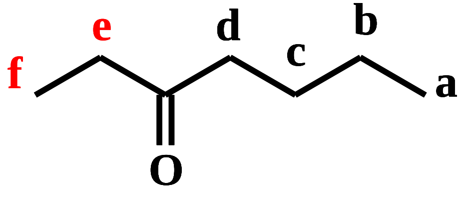
HSQC (Heteronuclear Single Quantum Coherence)

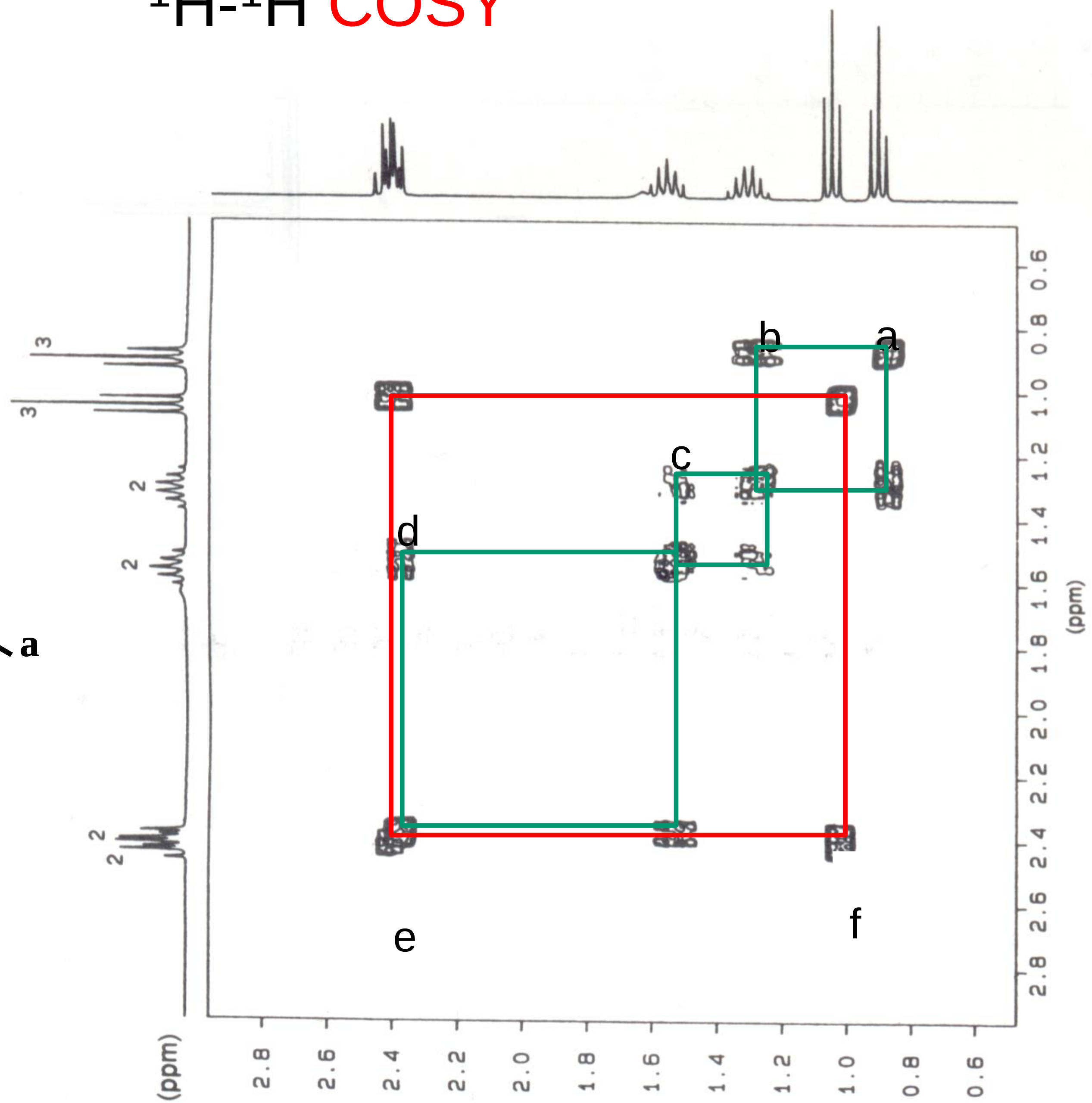
HMBC ^{1}H-^{13}C (J= 5-10 Hz)

(heteronuclear multiple Bond Correlation)

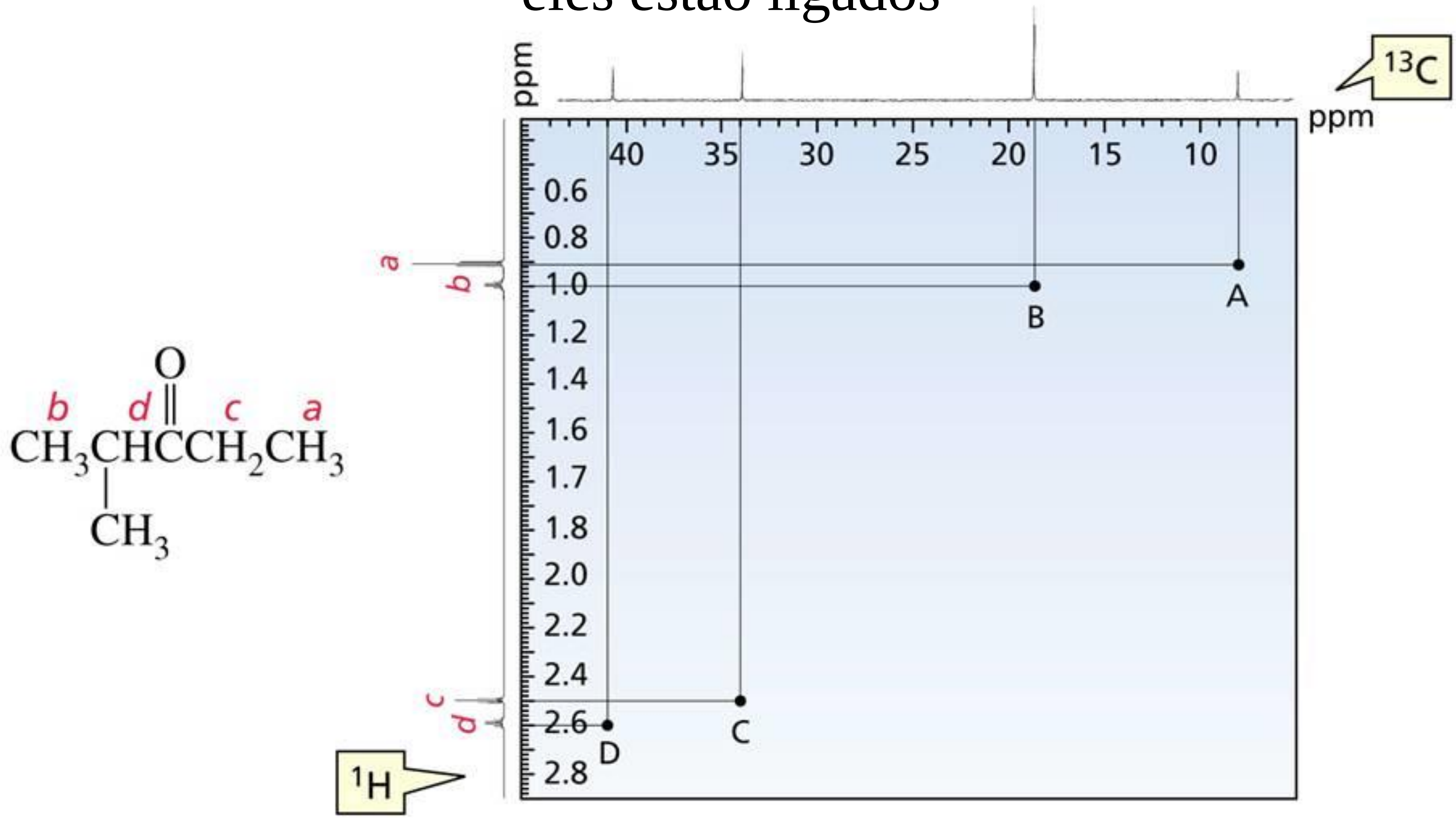
^{1}H-^{1}H COSY

$C_7H_{14}O$

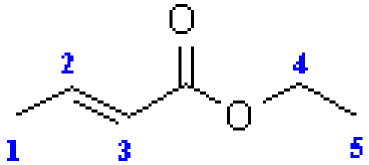




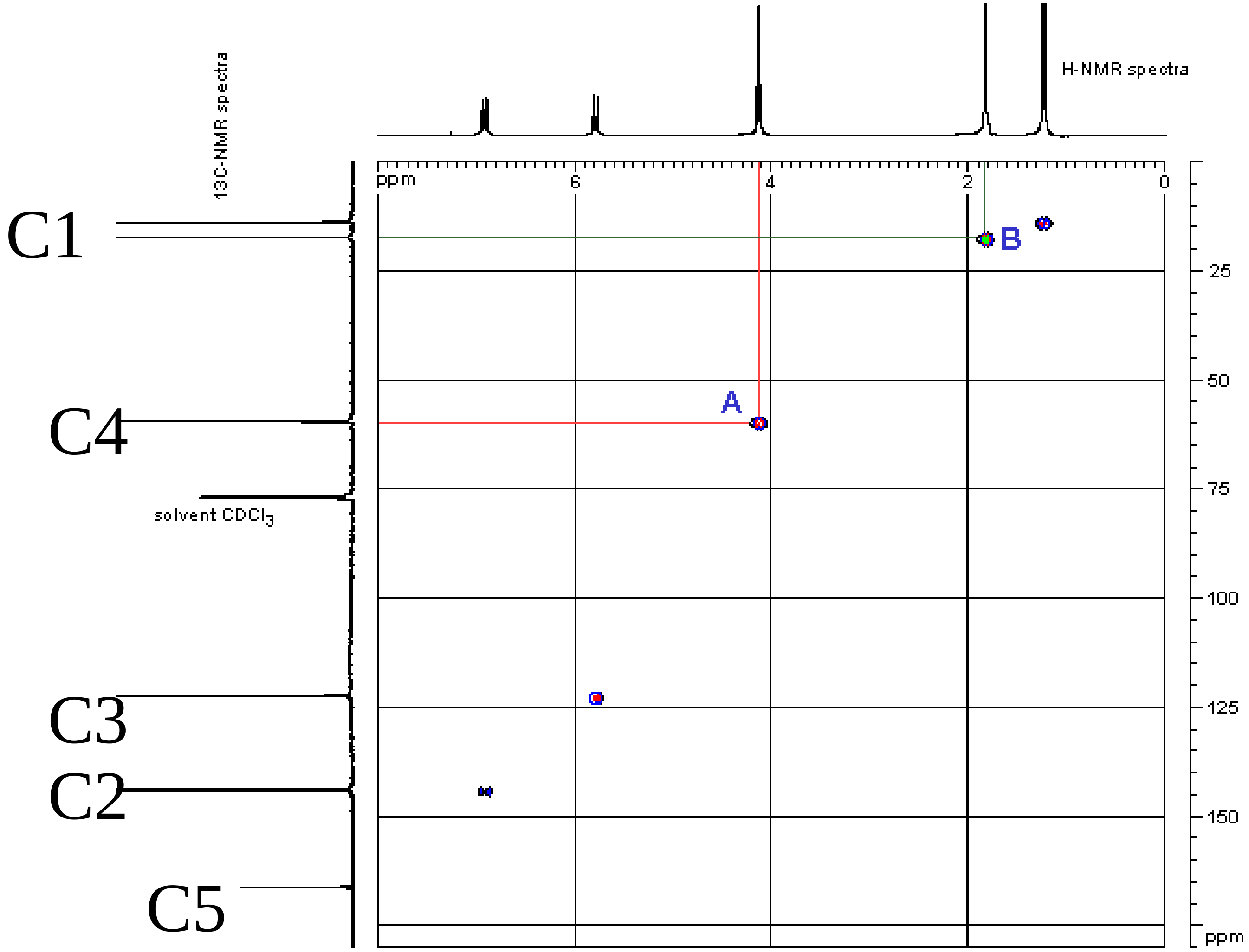
O espectro HETCOR da 2-metil-3-pentanona mostra os acoplamentos entre prótons e os carbonos aos quais eles estão ligados



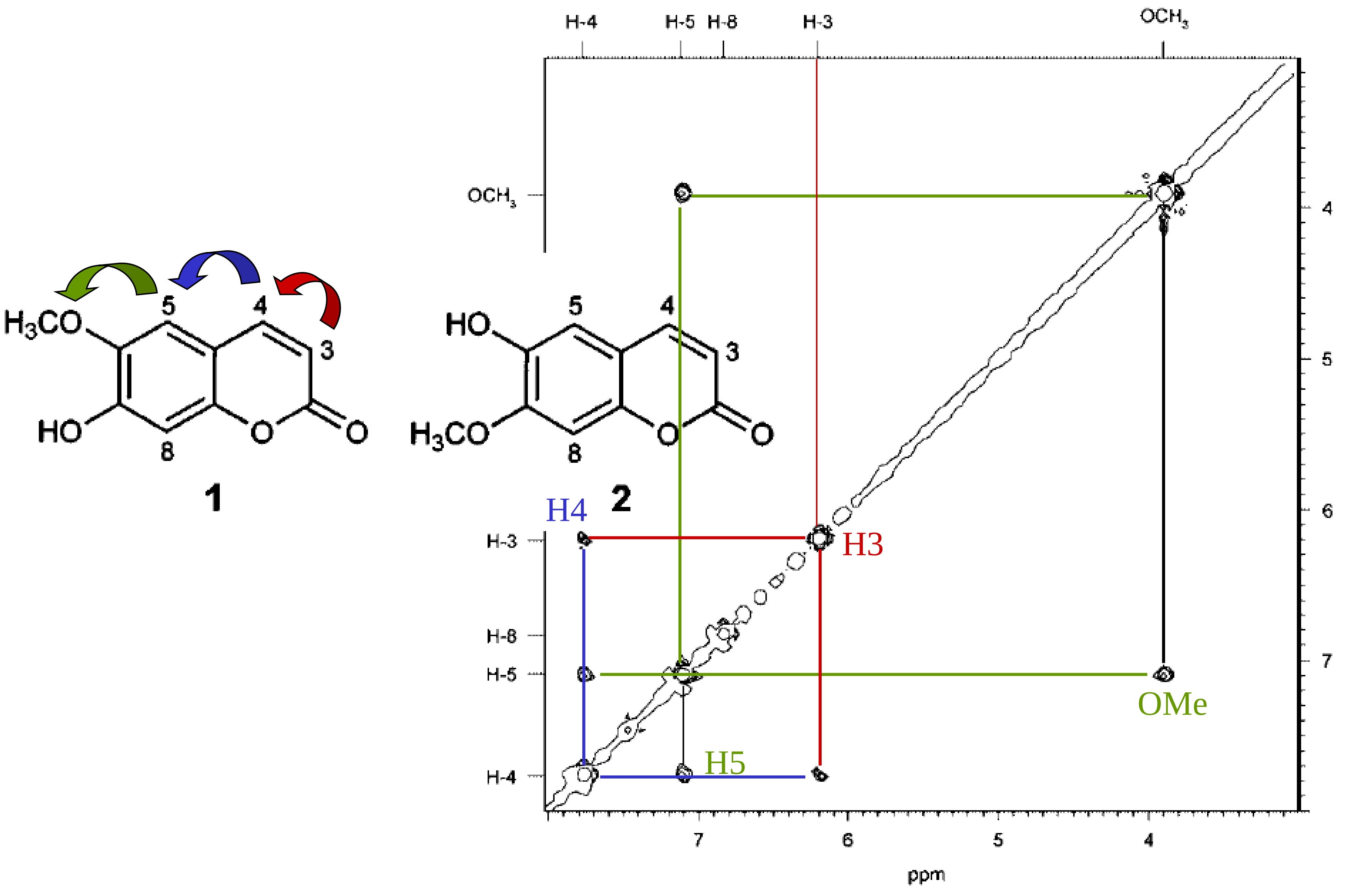
$CH_3CH(CH_3)C(=O)CH_2CH_3$



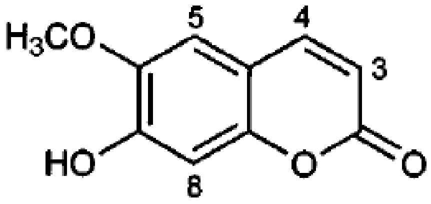
HMQC (Heteronuclear Multiple Quantum Coherence)



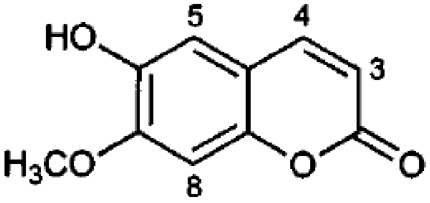
NOESY (Nuclear Overhauser Effect Spectroscopy)
(interações através do espaço e não através de ligações)



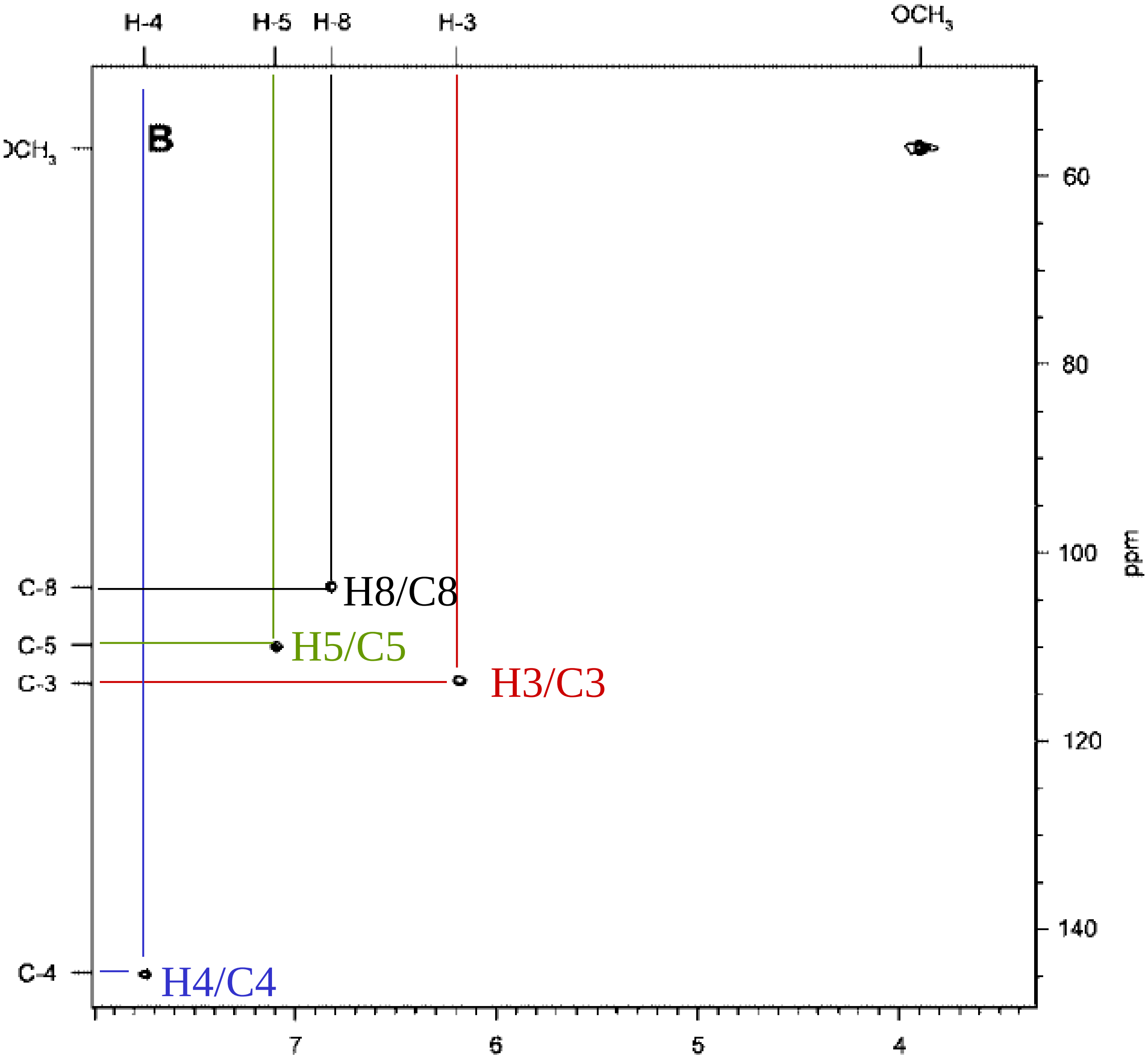
HSQC (Heteronuclear Single Quantum Coherence)
(quais hidrogênios estão ligados aos seus respectivos carbonos)



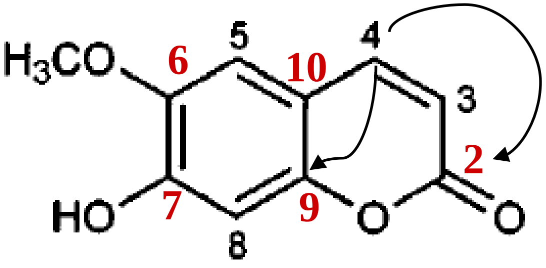
1



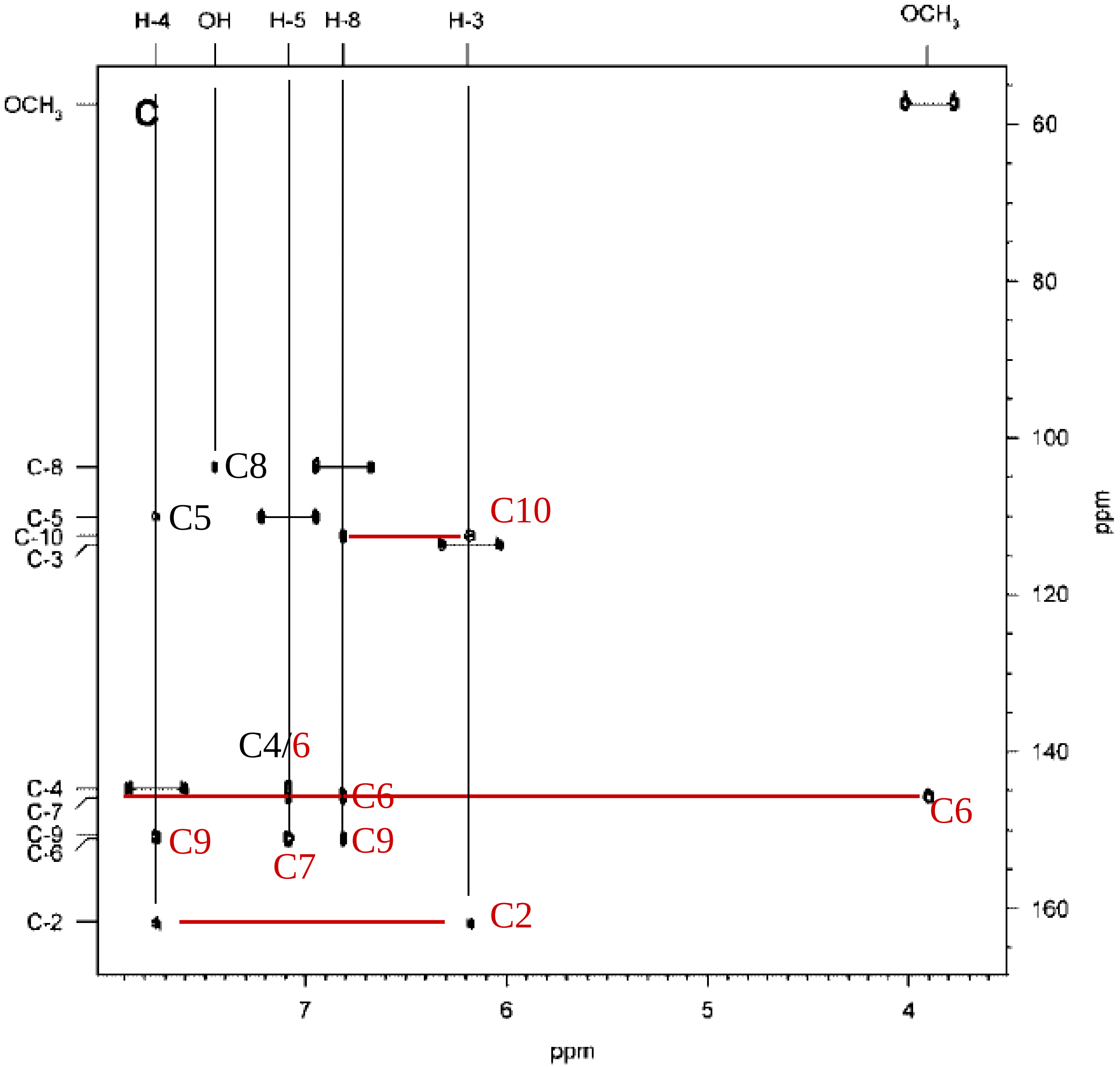
2



HMBC (Heteronuclear Multiple-Bond Correlation)
(correlação através de acoplamentos a longa distância entre H e C)



Por exemplo:
H4 está correlacionado
Como C9 e C2



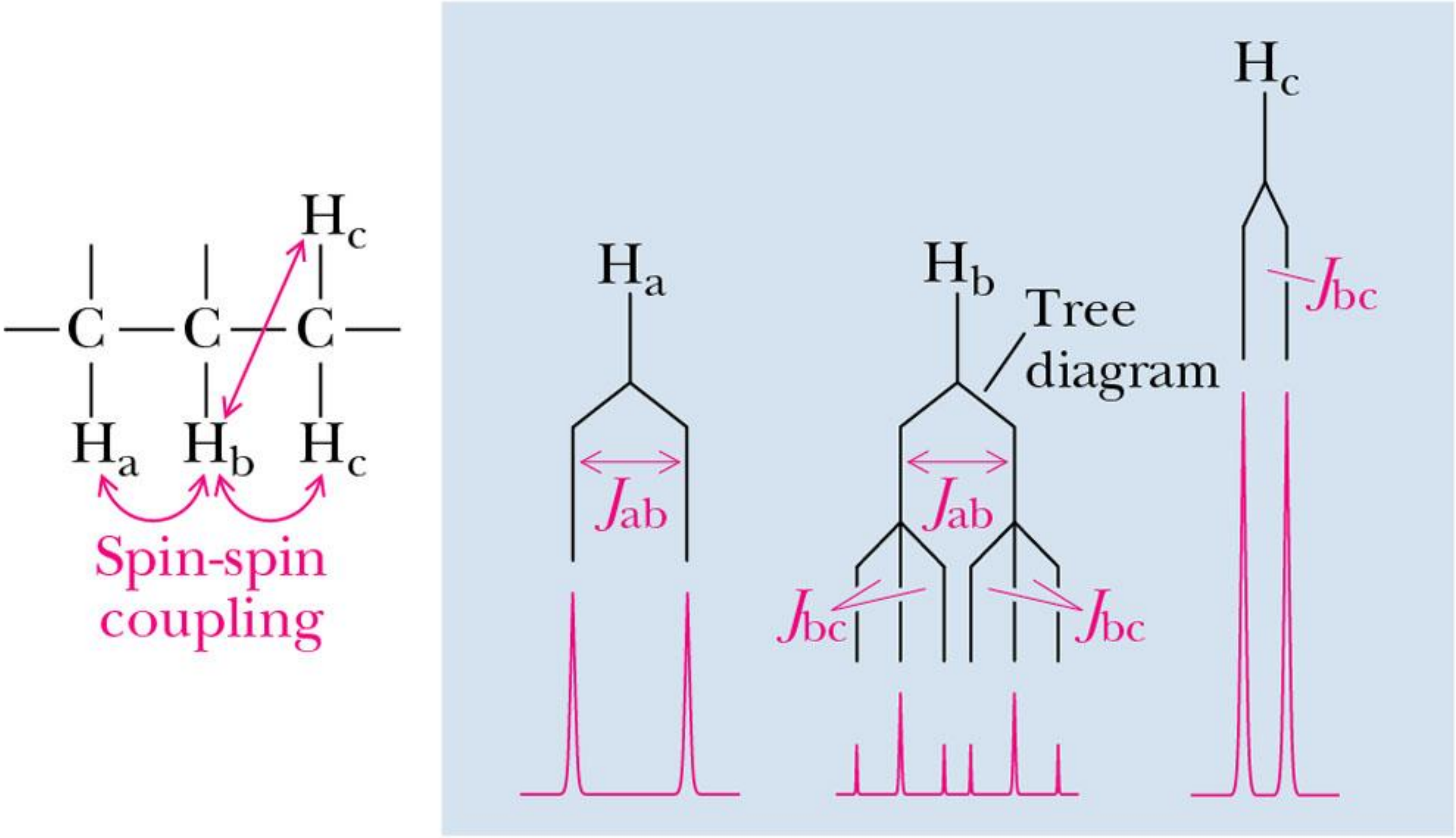
Nuclear Magnetic Resonance Spectroscopy

Use of more complex splitting patterns in structural assignment:

Non-equivalent coupling

Complex Splitting Patterns: Non-equivalent coupling

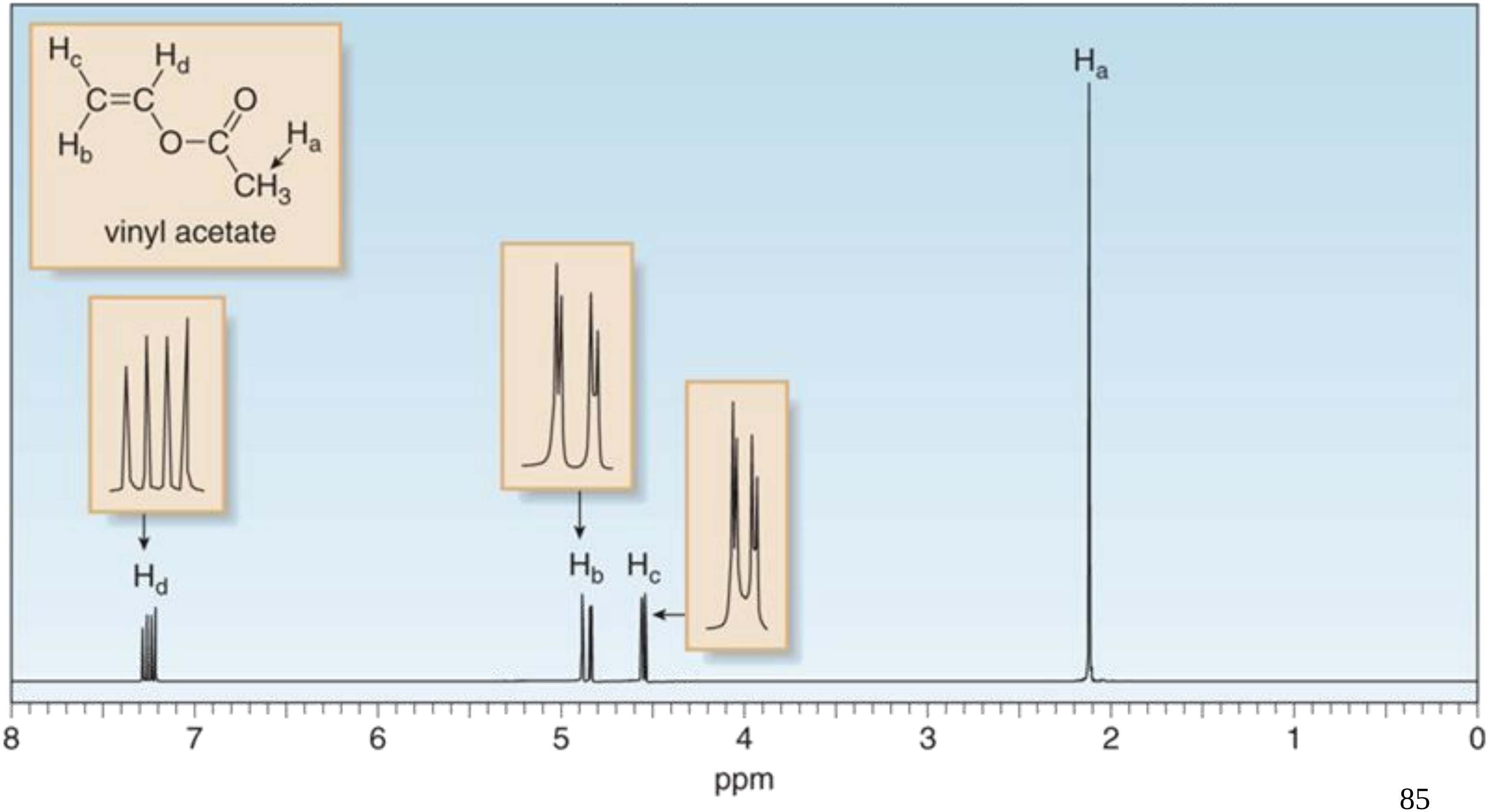
if H_c is a set of two equivalent H, then the observed splitting for Hb is a doublet of triplets (um dupleto desdobrado por um tripleto)



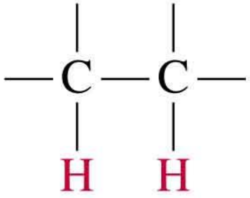
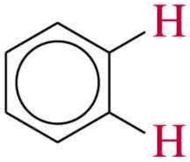
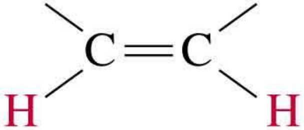
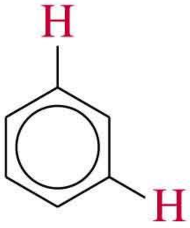
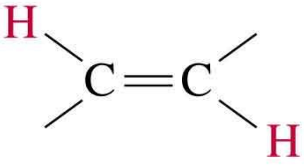
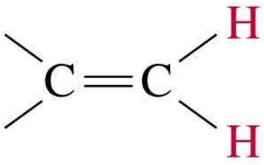
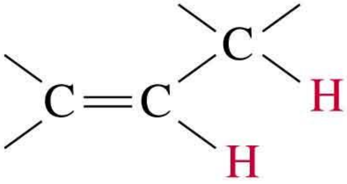
d dt d

^{1}H NMR—Spin-Spin Splitting: Non-equivalent coupling

H_a, H_b, H_c and H_d are not equivalent to each other



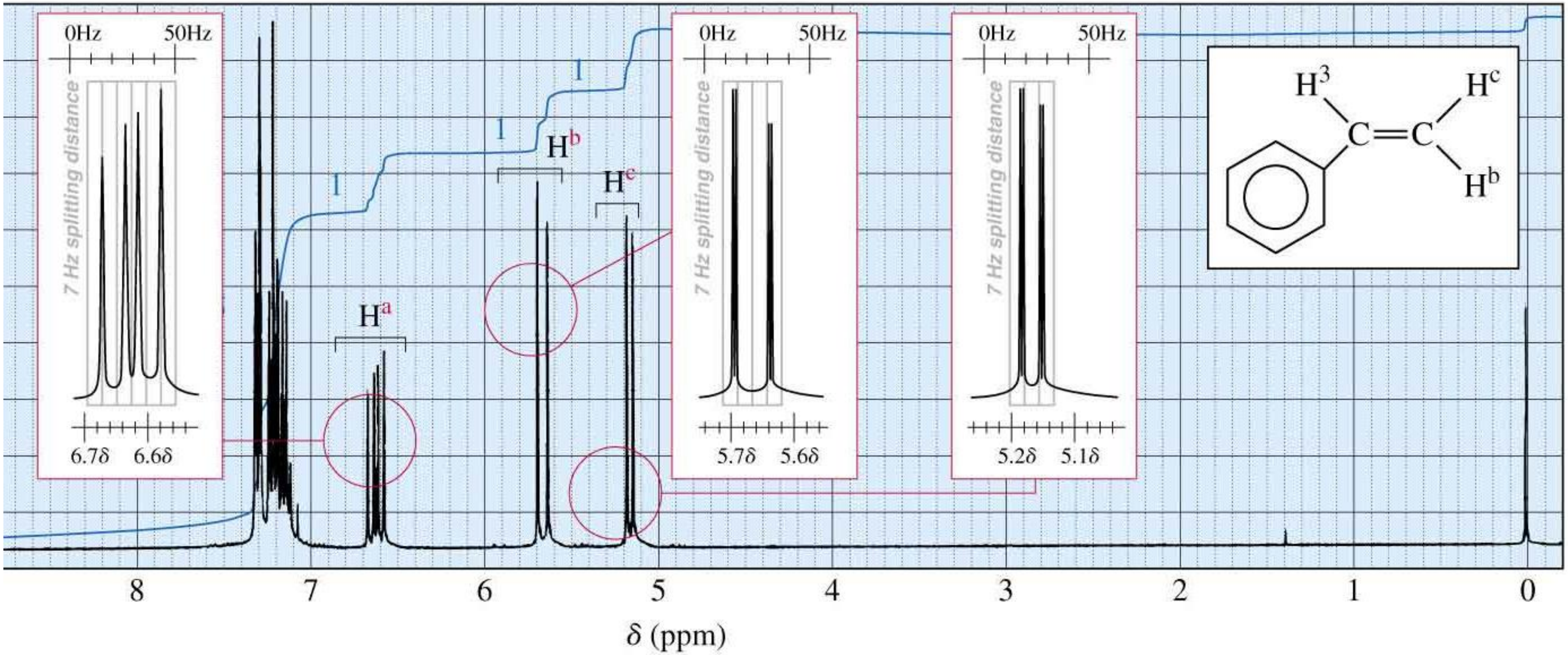
Typical Values for Coupling Constants

		Approx. *J*			Approx. *J*
	(free rotation)	7 Hz[a]		(ortho)	8 Hz
	(cis)	10 Hz		(meta)	2 Hz
	(trans)	15 Hz			
	(geminal)	2 Hz		(allylic)	6 Hz

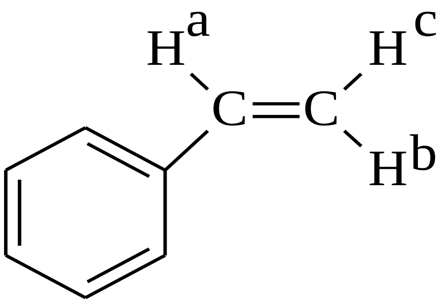
=>

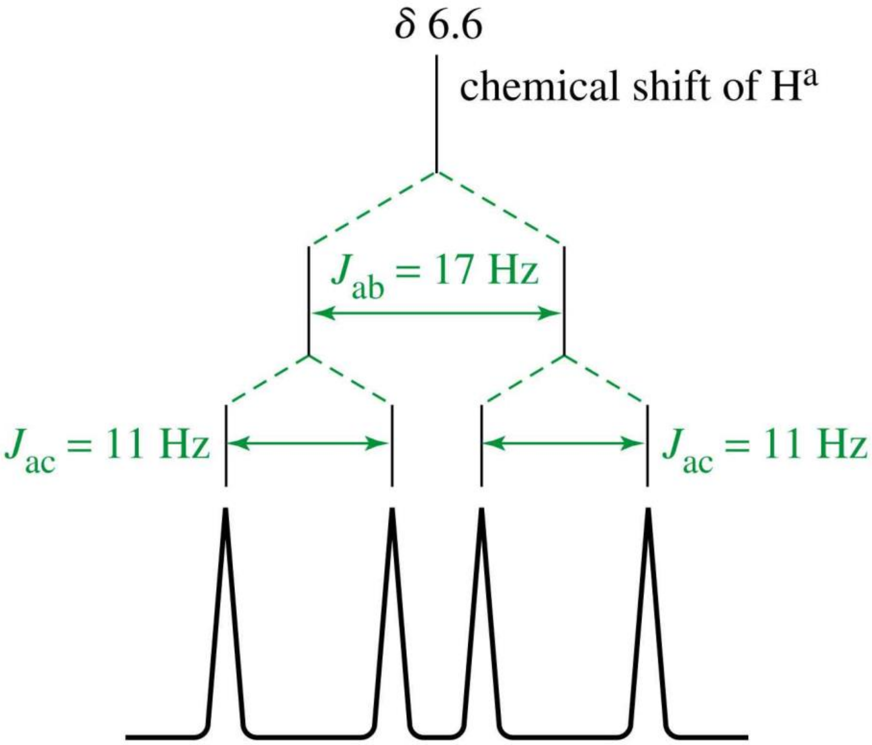
[a] The value of 7 Hz in an alkyl group is averaged for rapid rotation about the carbon–carbon bond. If rotation is hindered by a ring or bulky groups, other splitting constants may be observed.

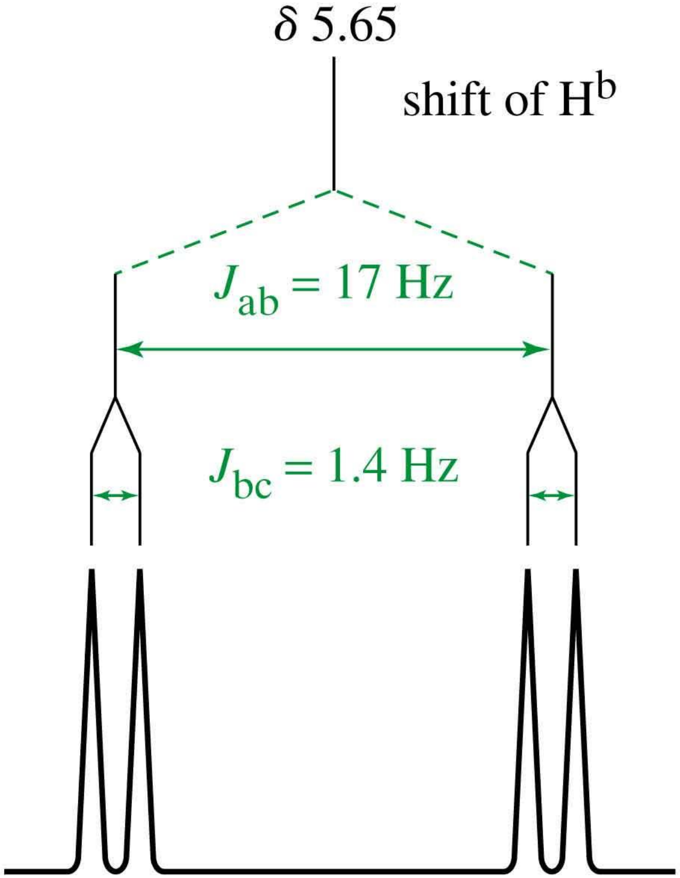
Spectrum for Styrene



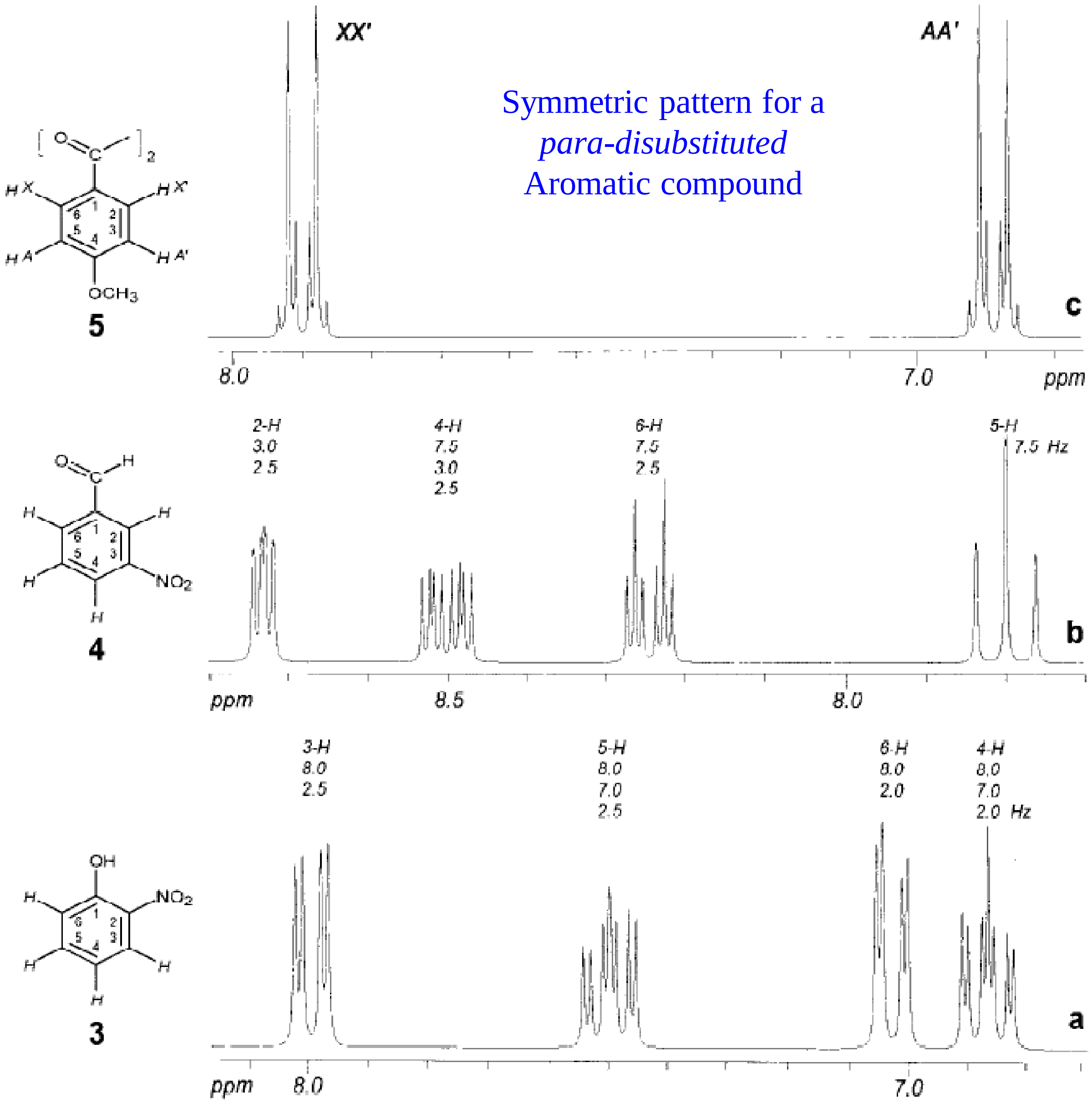
Splitting Tree







Splitting Pattern
For aromatic compounds



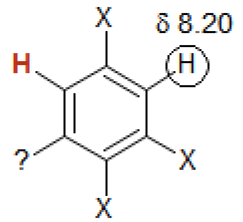
Symmetric pattern for a
para-disubstituted
Aromatic compound

Four different
hydrogens

Four different
hydrogens

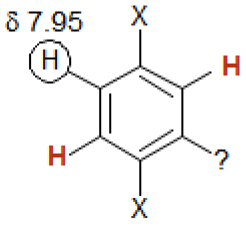
Figure 2.6. ^1H NMR spectra of disubstituted benzene rings [$CDCl_3$, 25 °C, 200 MHz]. (**a**) o-Nitrophenol (**3**); (**b**) *m*-nitrobenzaldehyde (**4**); (**c**) 4,4'-dimethoxybenzil (**5**)

Trisubstituted Aromatic ring

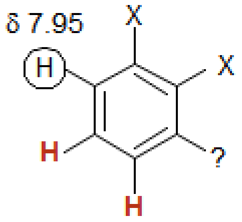


A

J ca 2 Hz
one meta H only

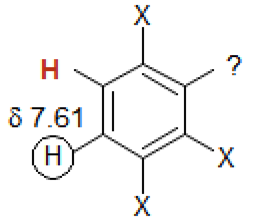


B



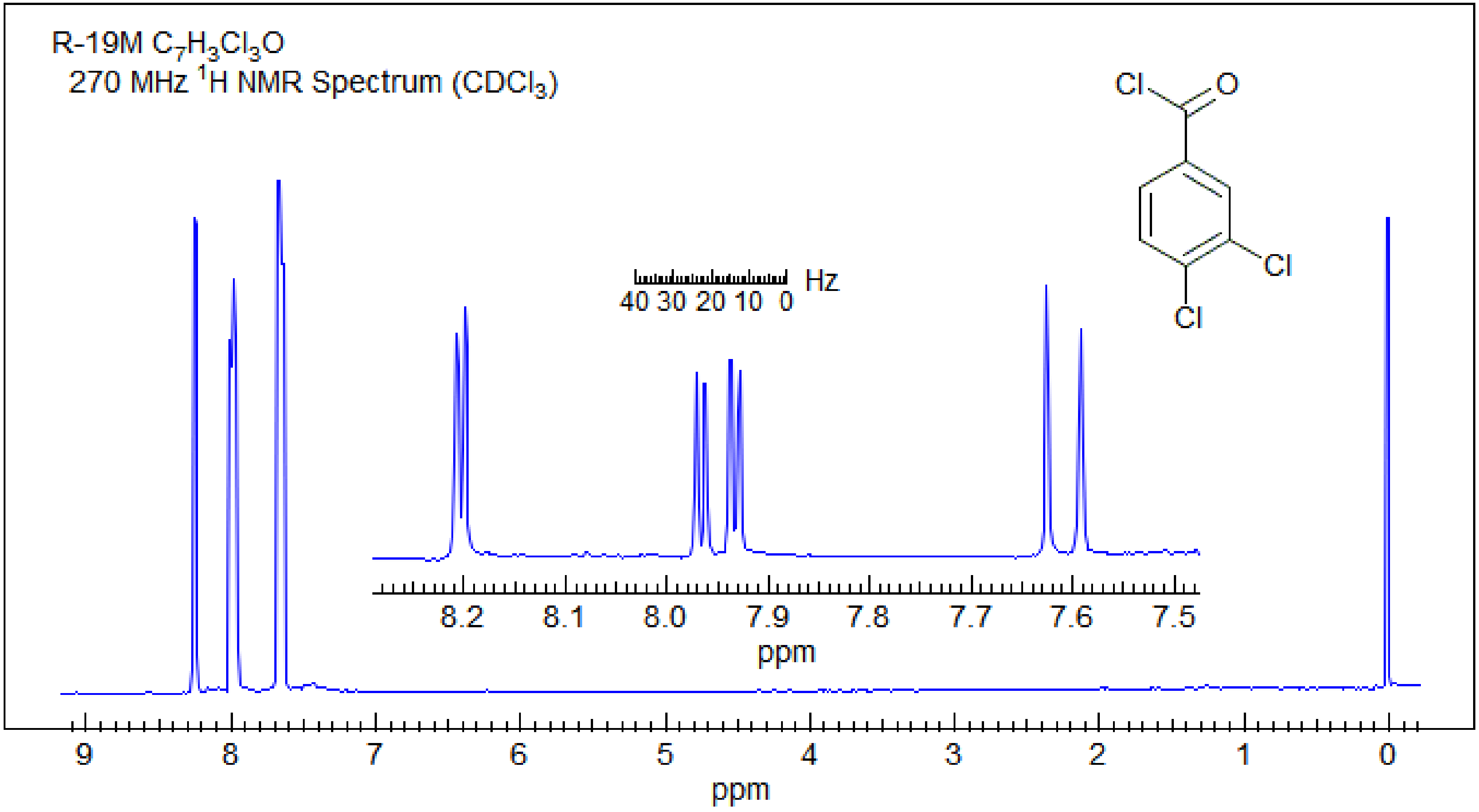
C

J ca 2 and 8 Hz
one ortho and one meta H



D

J = 8 Hz
one ortho H



R-19M $C_7H_3Cl_3O$
270 MHz ^{1}H NMR Spectrum ($CDCl_3$)