Tasks in NMR data analysis for Nature Products

Hsiao-Ching Lin (林曉青)
Associate Research Fellow,
Institute of Biological Chemistry, Academia Sinica

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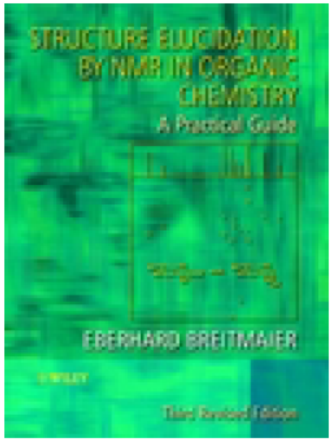
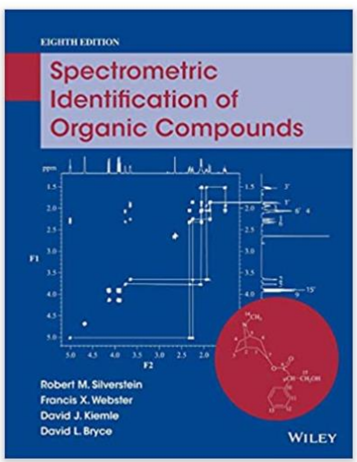
Textbook references:



Structure Elucidation by NMR in Organic Chemistry (Third revised edition)

Author(s): Eberhard Breitmaier

ISBN: 0470853069 (Electronic) 047085006X (Print)



Spectrometric Identification of Organic Compounds 8th

Robert M. Silverstein, Francis X. Webster, David J. Kiemle, David L. Bryce
ISBN: 978-0-470-61637-6 September 2014 464 Pages

1.1 Chemical shift

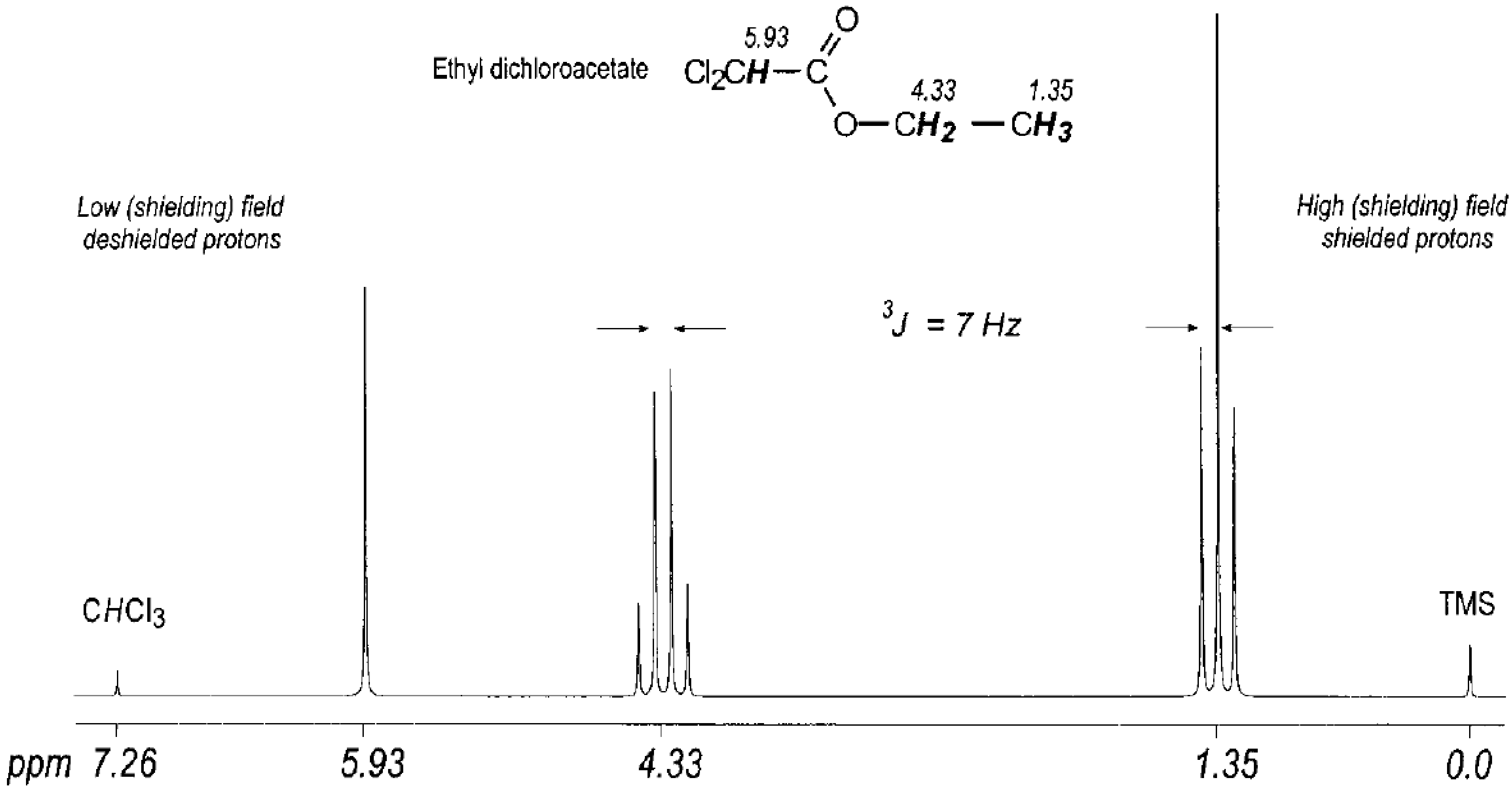
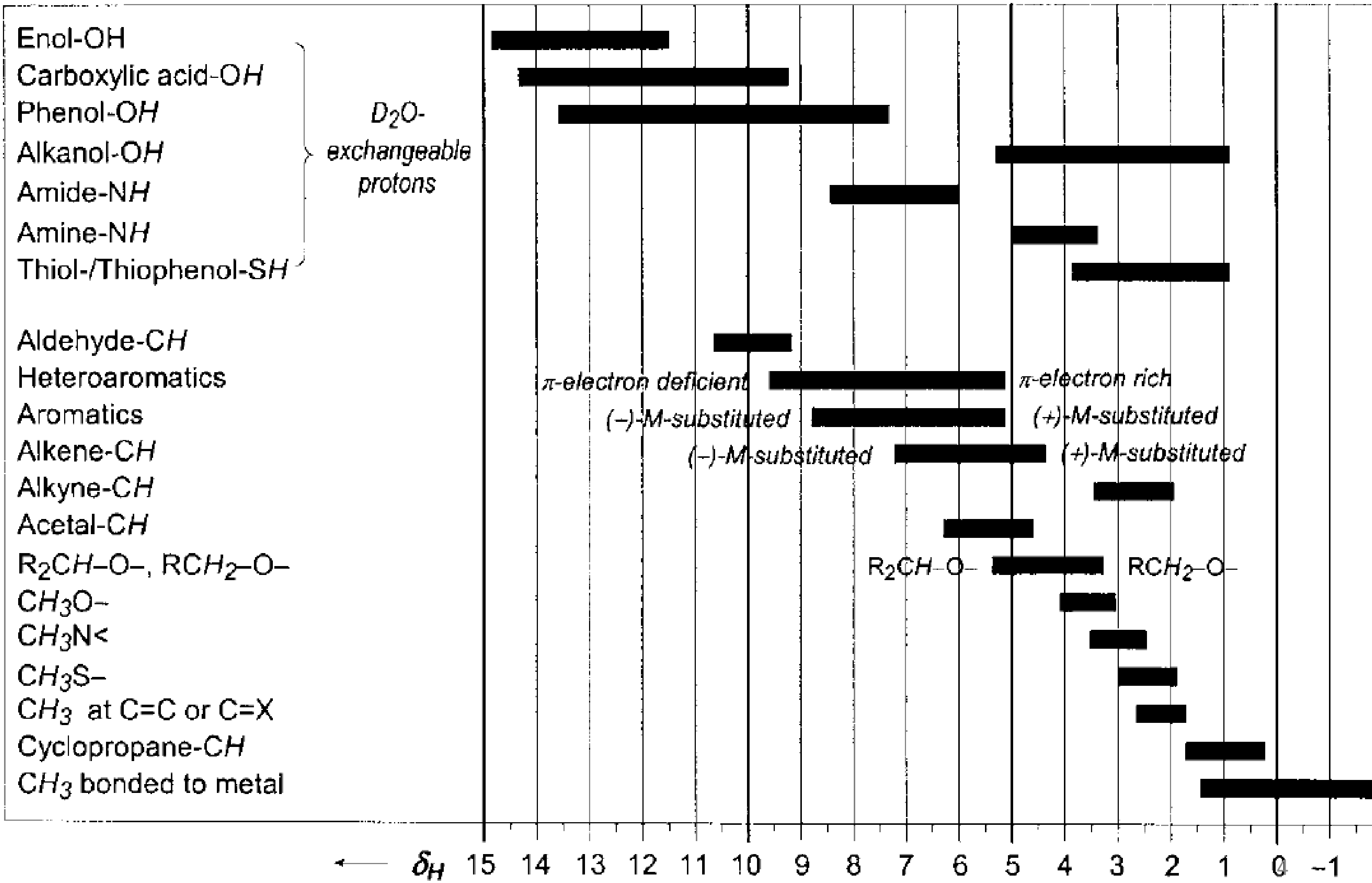


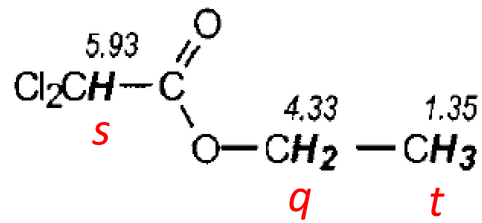
Figure 1.2. ^1H NMR spectrum of ethyl dichloroacetate ($CDCl_3$, 25 °C, 80 MHz). The proton of the $CHCl_2$ group is less shielded (more strongly deshielded) in comparison with the protons of the CH_2 and CH_3 residues

Table 2.1. ^{1}H chemical shift ranges for organic compounds



Signal multiplicity (multiplets)

Ethyl dichloroacetate



The ^{1}H NMR spectrum of ethyl dichloroacetate (Fig. 1.2), as an example, displays a triplet for the CH_3 group (*two vicinal H*), a quartet for the OCH_2 group (three *vicinal H*) and a singlet for the $CHCl_2$ fragment (no *vicinal H* for coupling).

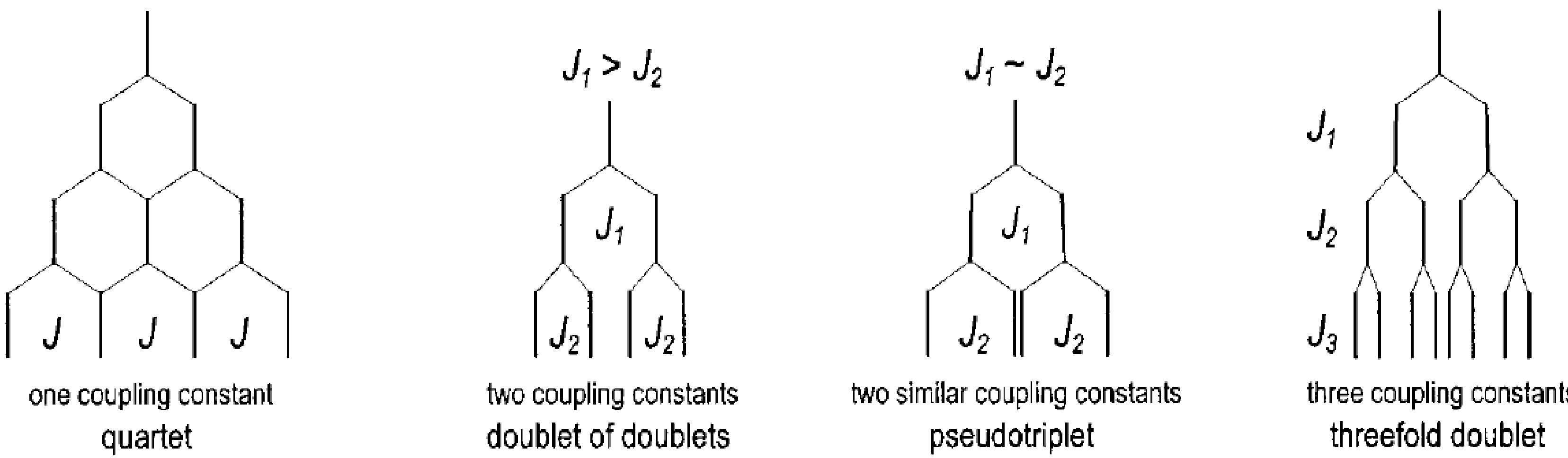


Figure 1.3. Quartet, doublet of doublets, pseudotriplet and threefold doublet (doublet of doublets of doublets)

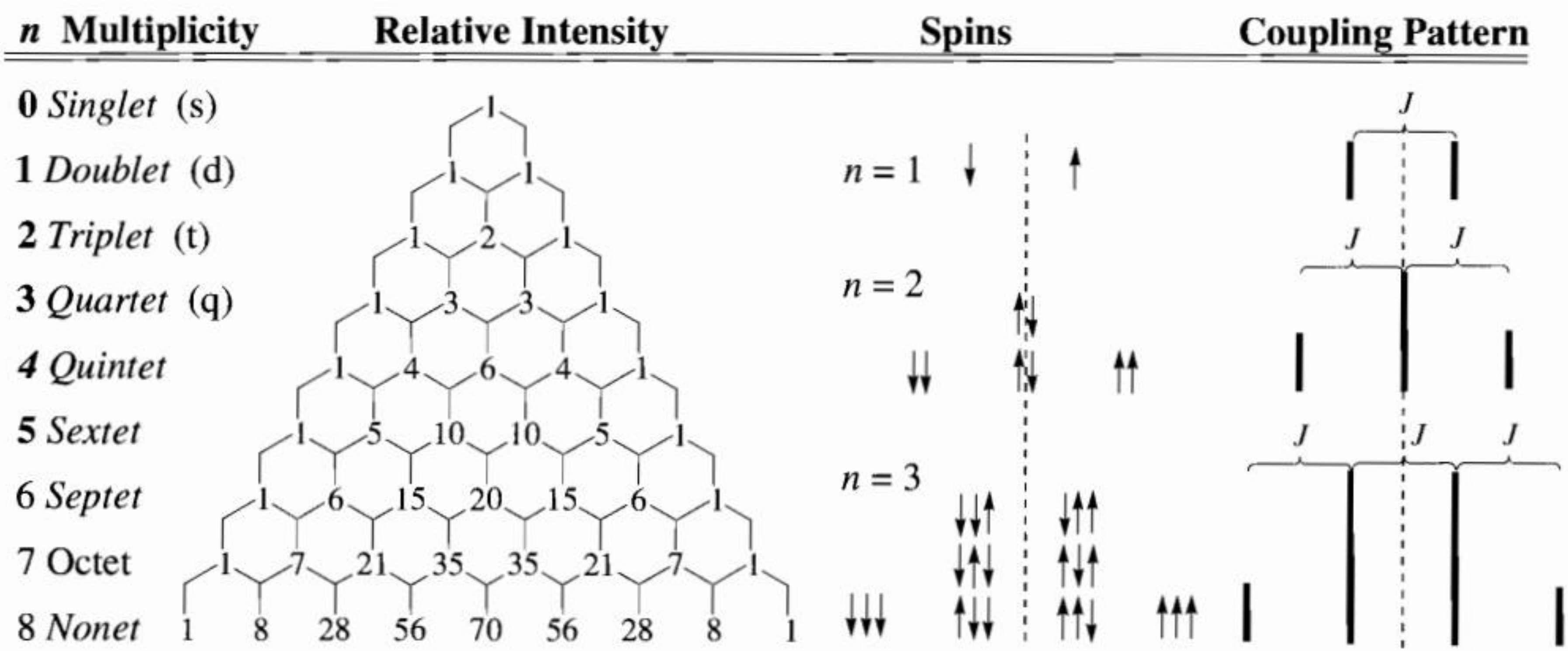
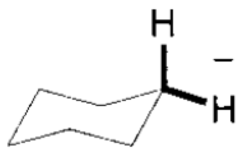
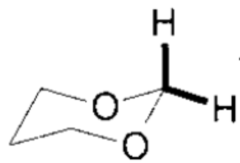
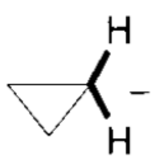
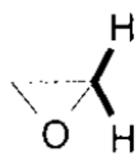
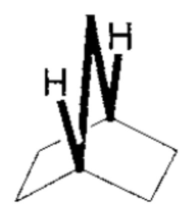
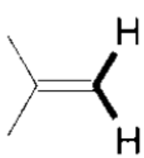
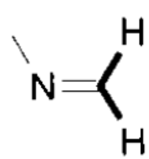
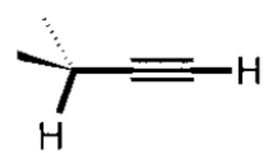
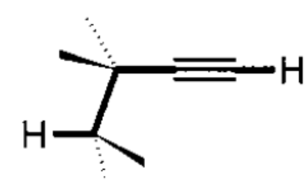


FIGURE 3.32 Pascal's triangle. Relative intensities of first-order multiplets; n = number of equivalent coupling nuclei of spin 1/2 (e.g., protons).

2.2.3 *HH* Coupling constants

Table 2.4. Typical *HH* coupling constants (*Hz*) of some units in alicycles, alkenes and alkynes [2,3]

$^2J_{HH}$ *geminal protons*		$^4J_{HH}$ *protons with w-relationships*	$^5J_{HH}$
 – 12.5	 – 6.0	 – 2.5	
 – 4.5	 5.5	 – 2.0	
 2.5	 16.5	 – 3.0	 1.0

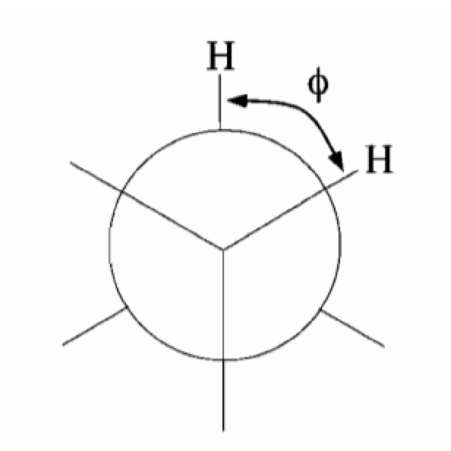
2.3 Relative configuration and conformation

2.3.1 *HH* Coupling constants

Vicinal coupling constants $^3J_{HH}$ indicate very clearly the relative configuration of the coupling protons. Their contribution depends, according to the Karplus-Conroy equation [2,3],

$$^3J_{HH} = a\cos^2\varphi - 0.28 \quad (\text{up to } \varphi = 90°\text{: } a \approx 10\text{; above } \varphi = 90°\text{: } a \approx 15) \qquad (2)$$

on the dihedral angle φ, enclosed by the *CH* bonds as shown in Fig. 2.18, which sketches the Karplus-Conroy curves for dihedral angles from 0 to 180°. Experimental values correspond to those given by the curve shown; deviations are up to *3 Hz*; electronegative substituents on the coupling path, for example, reduce the magnitude of $^3J_{HH}$.



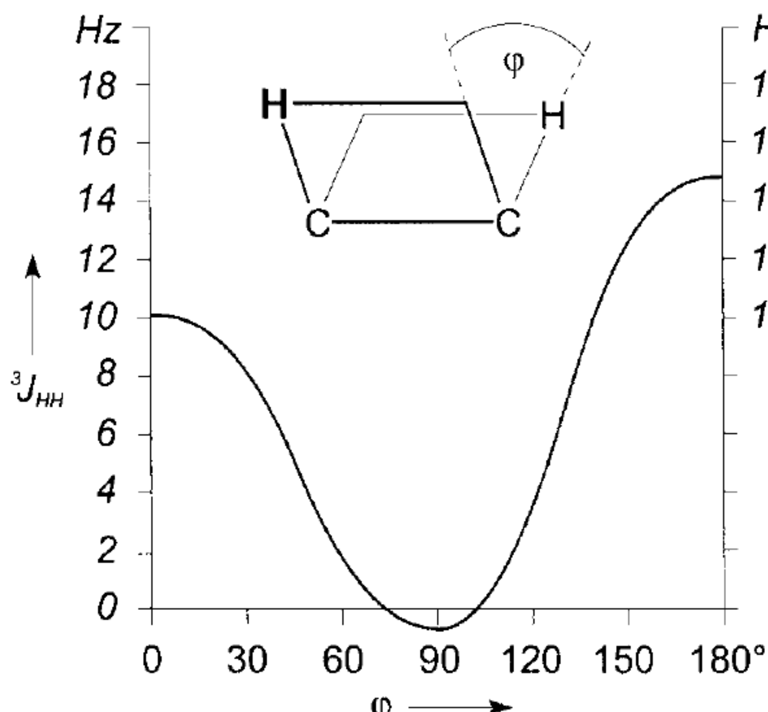


TABLE 3.6. Calculated and observed coupling constants, J, in cyclohexanes based on bond angle.

	Dihedral Angle	Calculated J(Hz)	Observed J(Hz)
Axial-axial	180°	9	8–14 (usually 8–10)
Axial-equatorial	60°	1.8	1–7 (usually 2–3)
Equatorial-equatorial	60°	1.8	1–7 (usually 2–3)

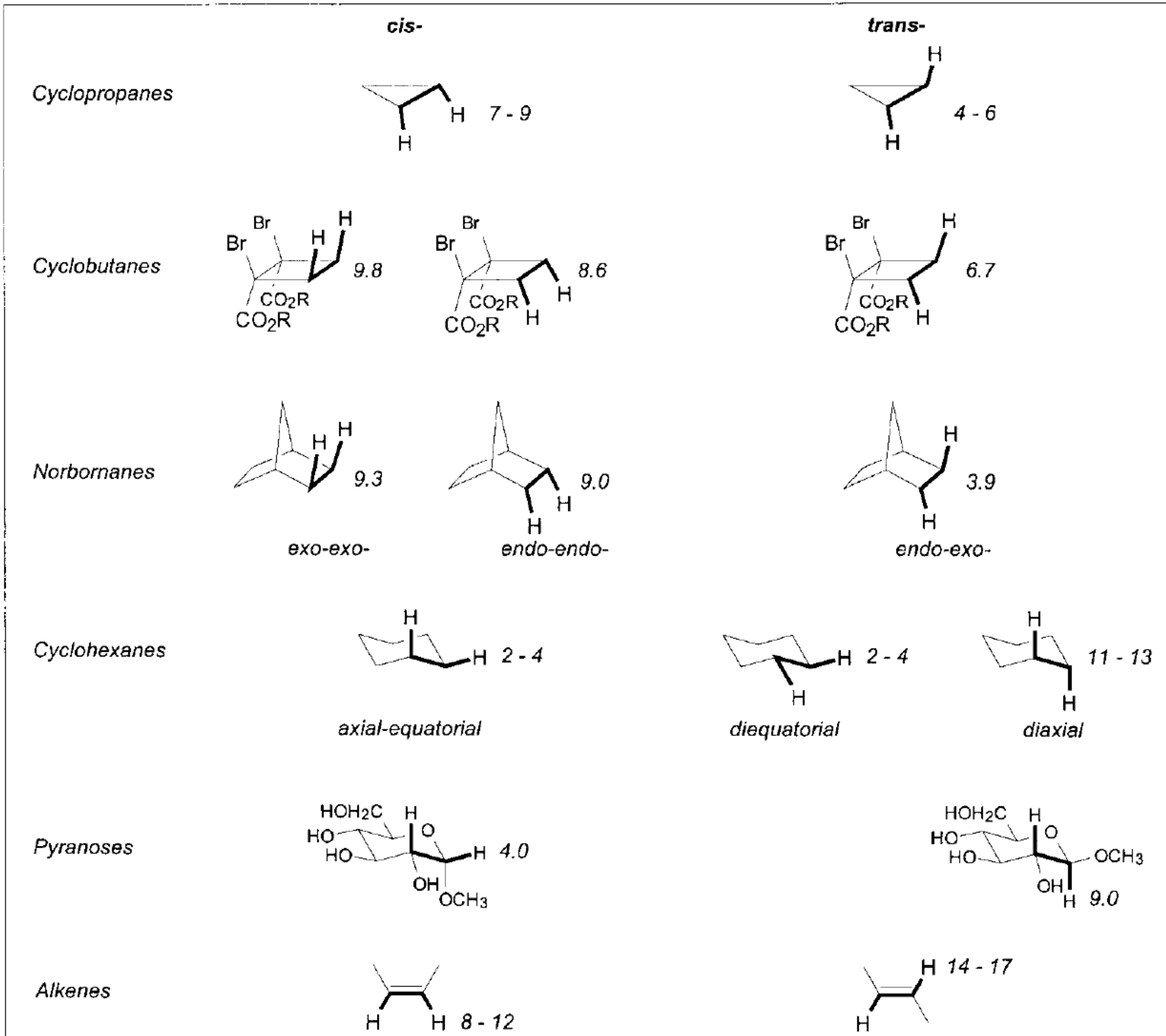
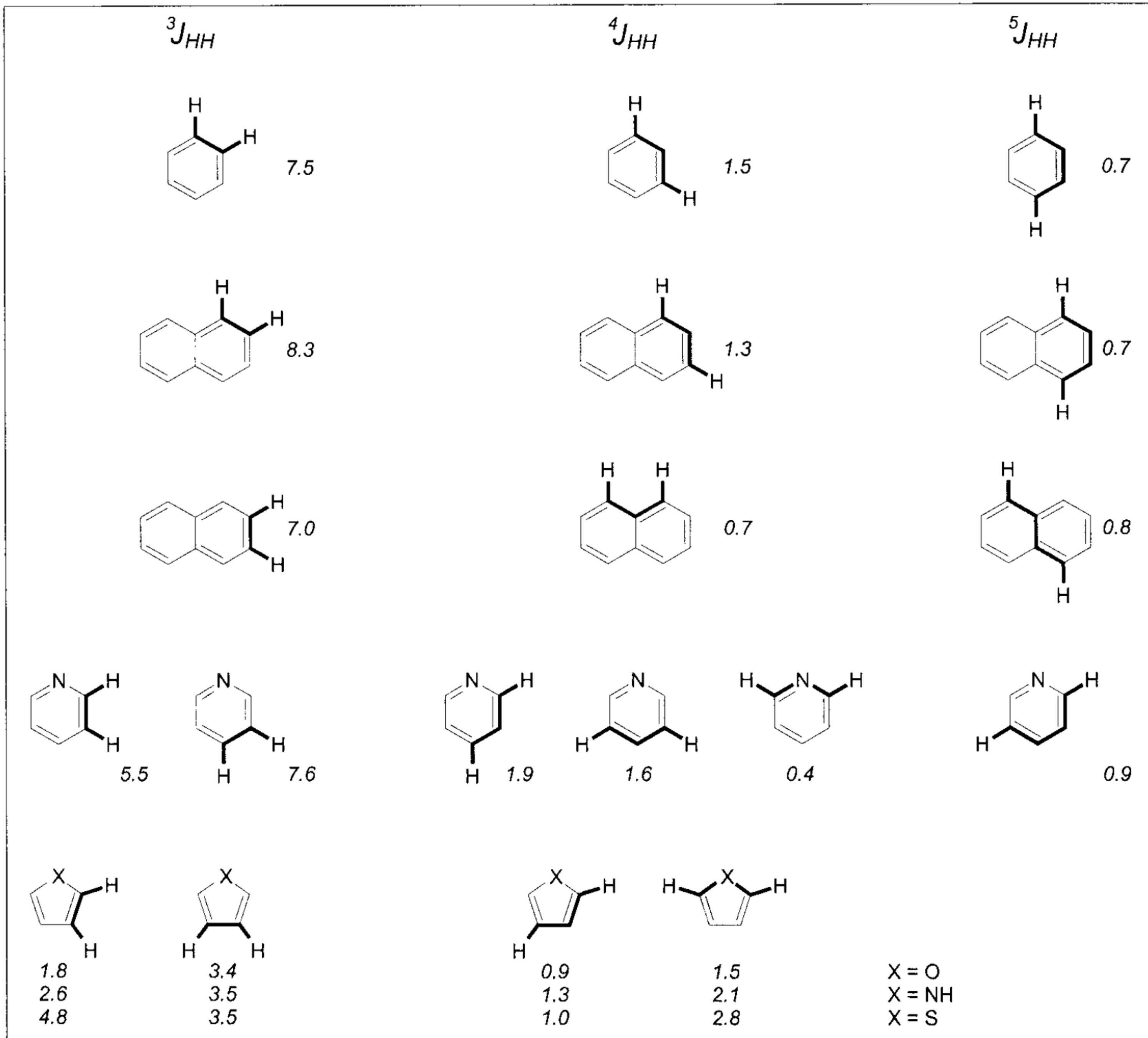
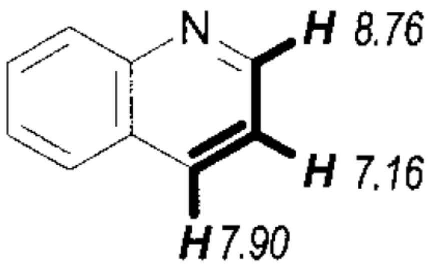

cis-
trans-
Cyclopropanes
7 - 9
4 - 6
Cyclobutanes
Br
CO2R
9.8
8.6
6.7
Norbornanes
9.3
9.0
3.9
exo-exo-
endo-endo-
endo-exo-
Cyclohexanes
2 - 4
2 - 4
11 - 13
axial-equatorial
diequatorial
diaxial
Pyranoses
HOH2C
HO
OH
OCH3
4.0
9.0
Alkenes
8 - 12
14 - 17

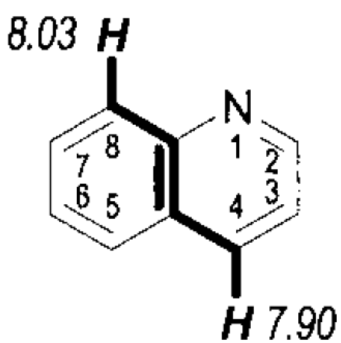
Table 2.5. Typical *HH* coupling constants (*Hz*) of aromatic and heteroaromatic compounds [2,3]



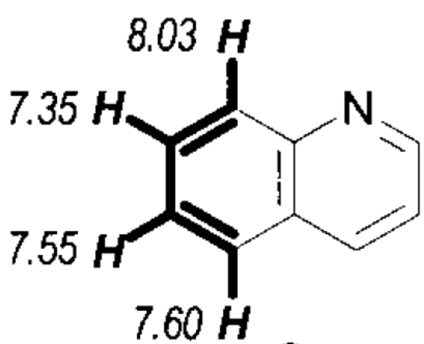
HH COSY (*geminal*, *vicinal*, *w*-relationships of protons)



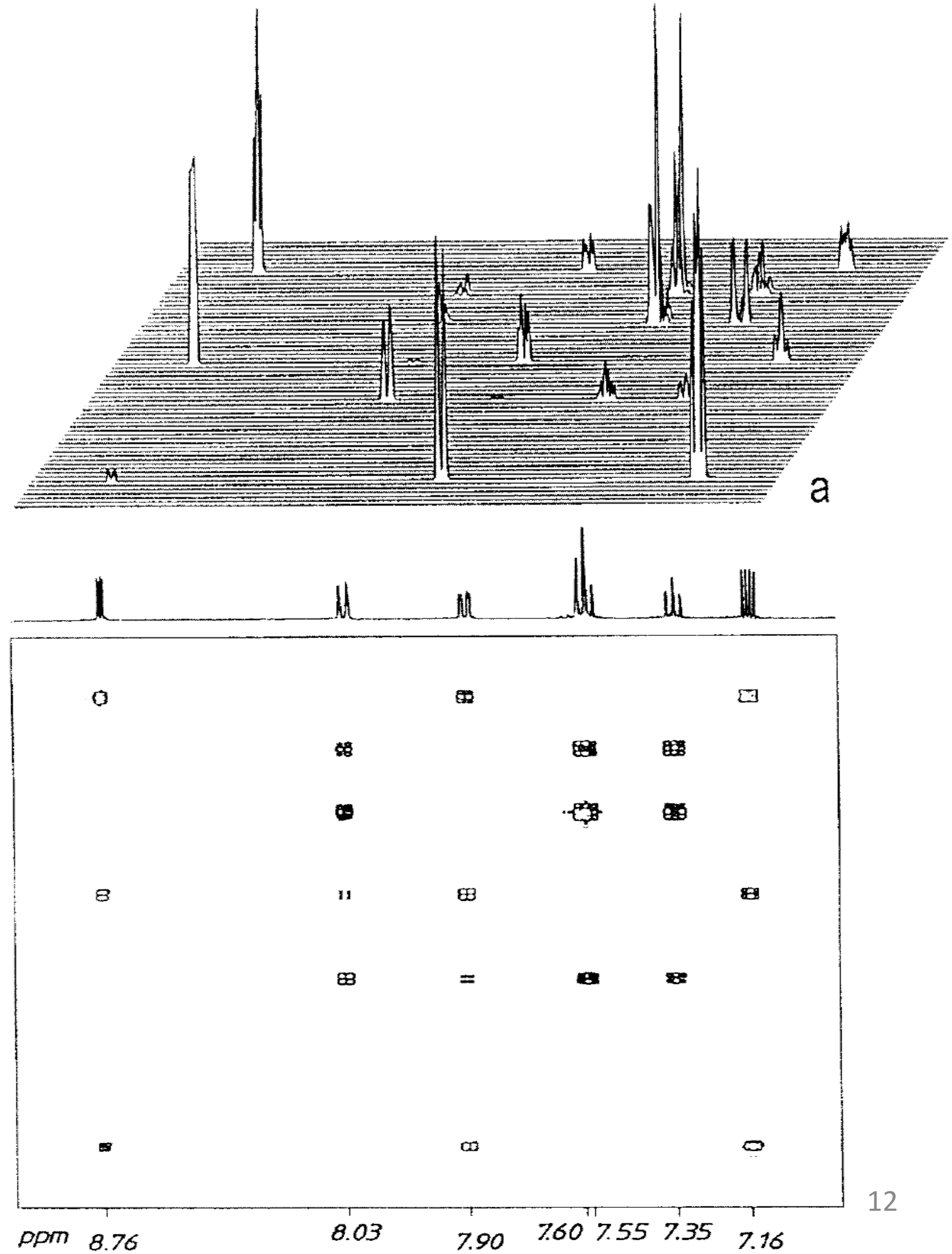
a

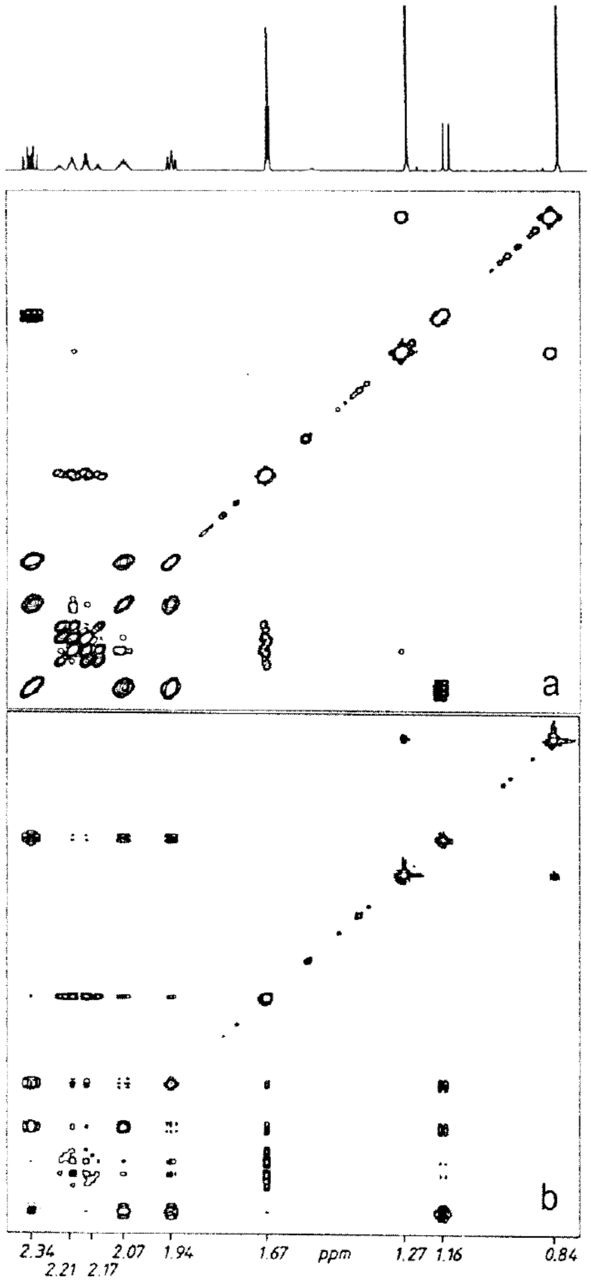


b

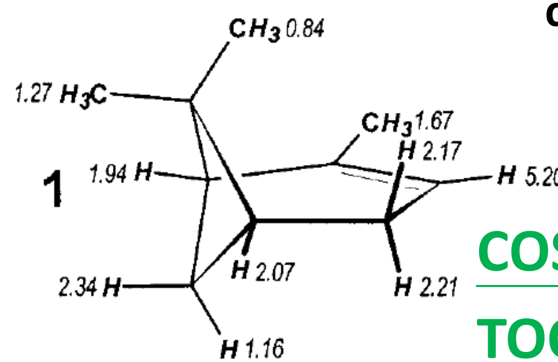


c



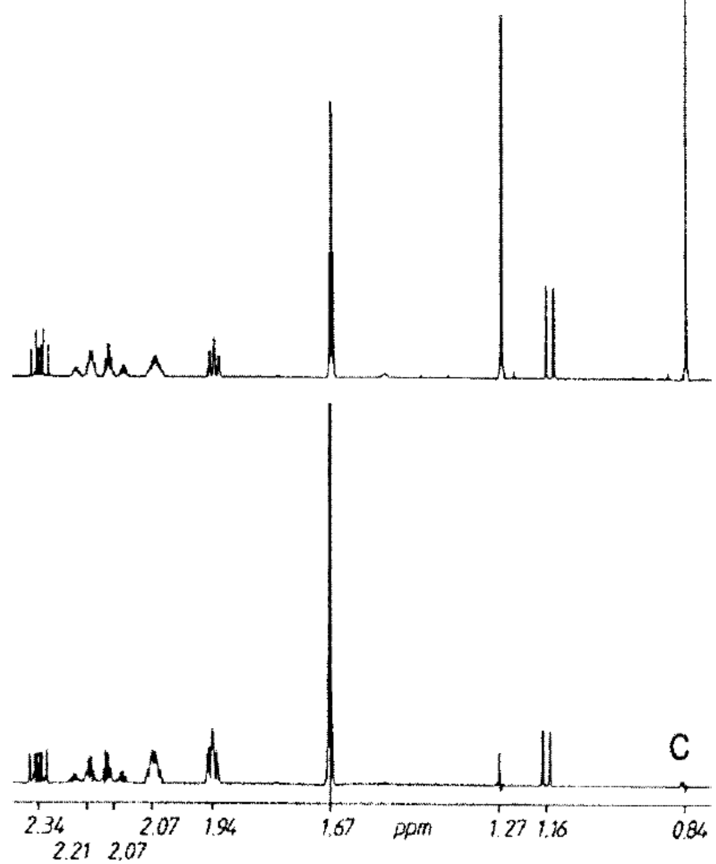


α-pinene



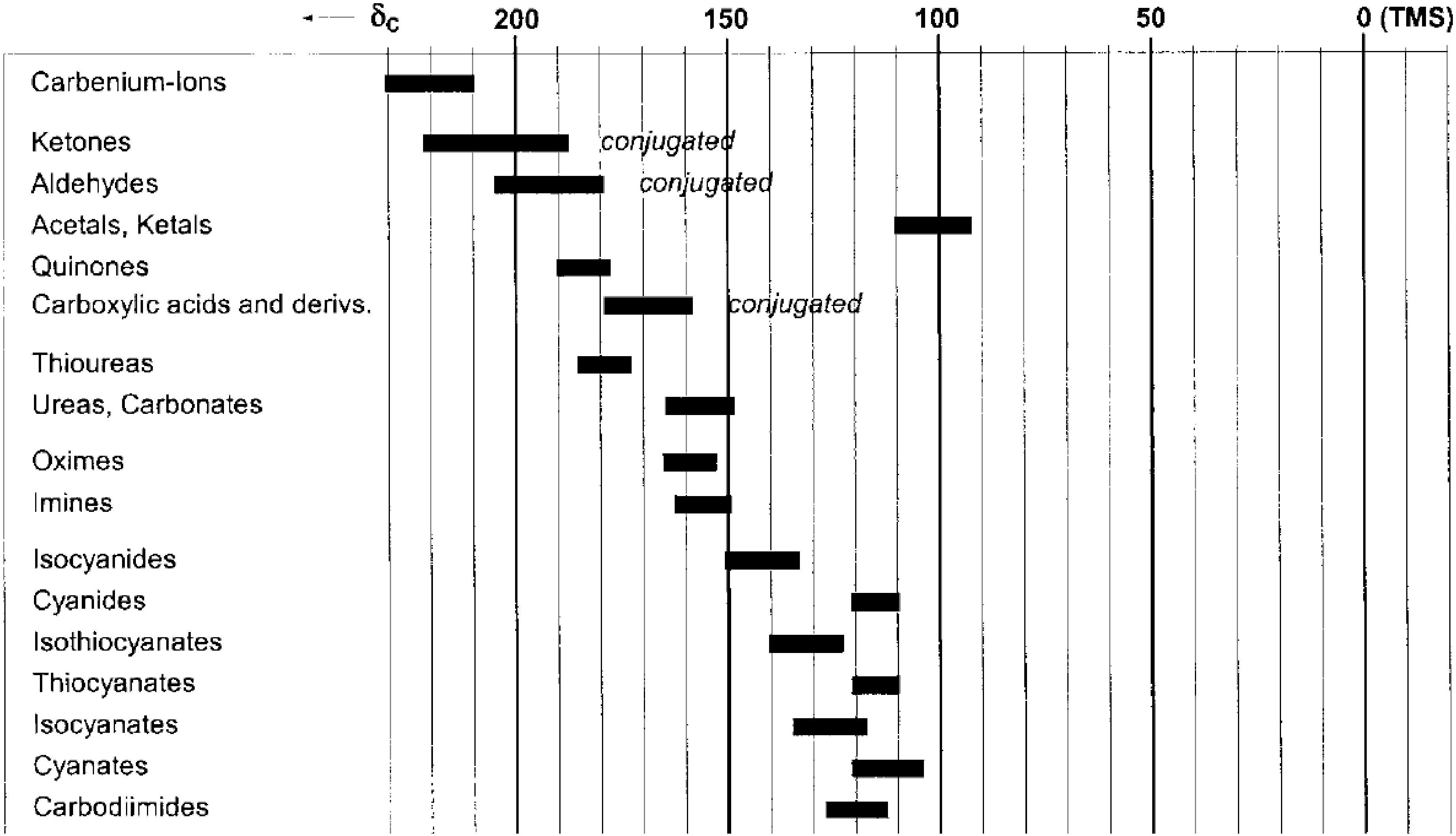
COSY: H to H (^2J or ^3J)

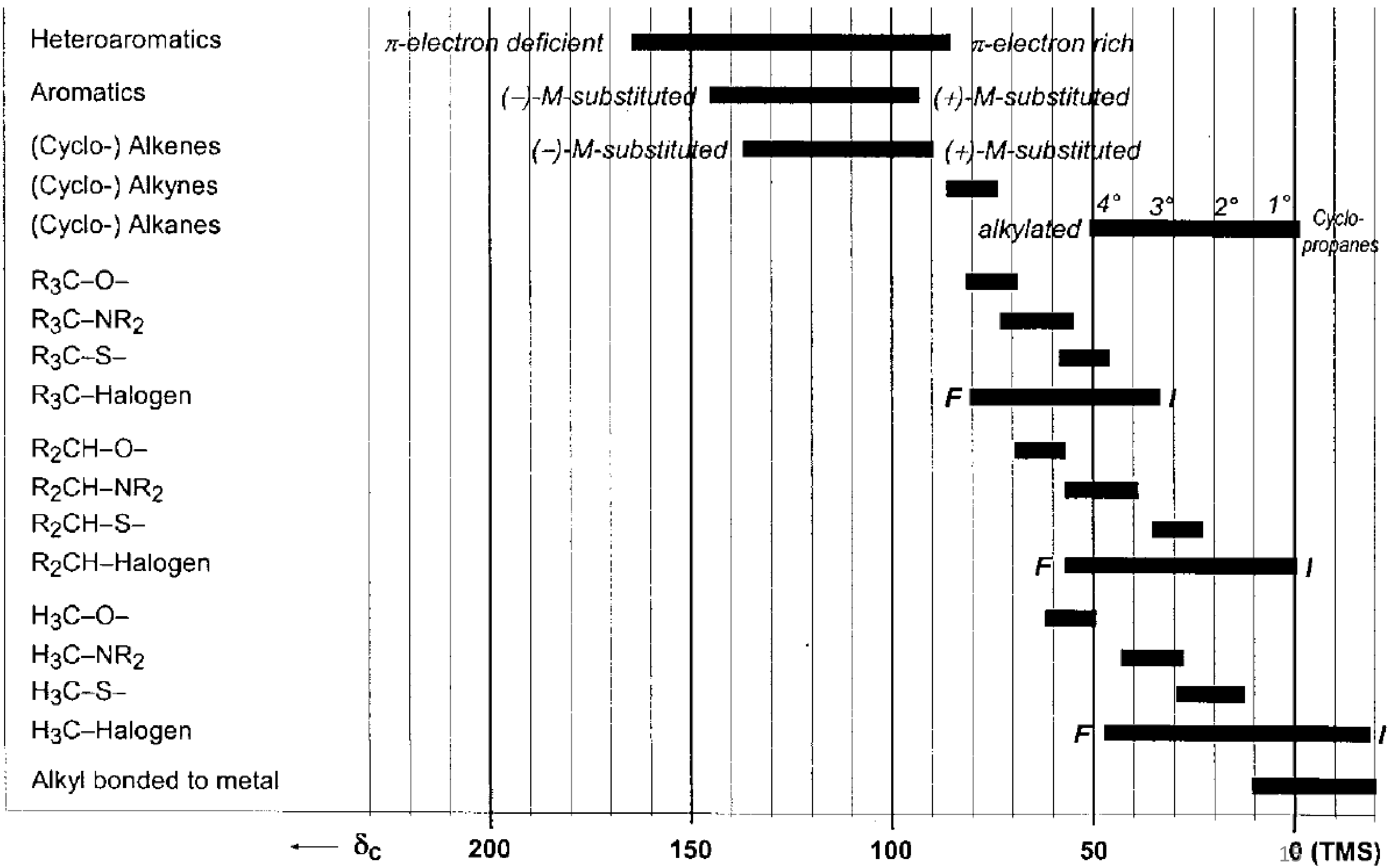
TOCSY: H to H (^2J to ^5J)

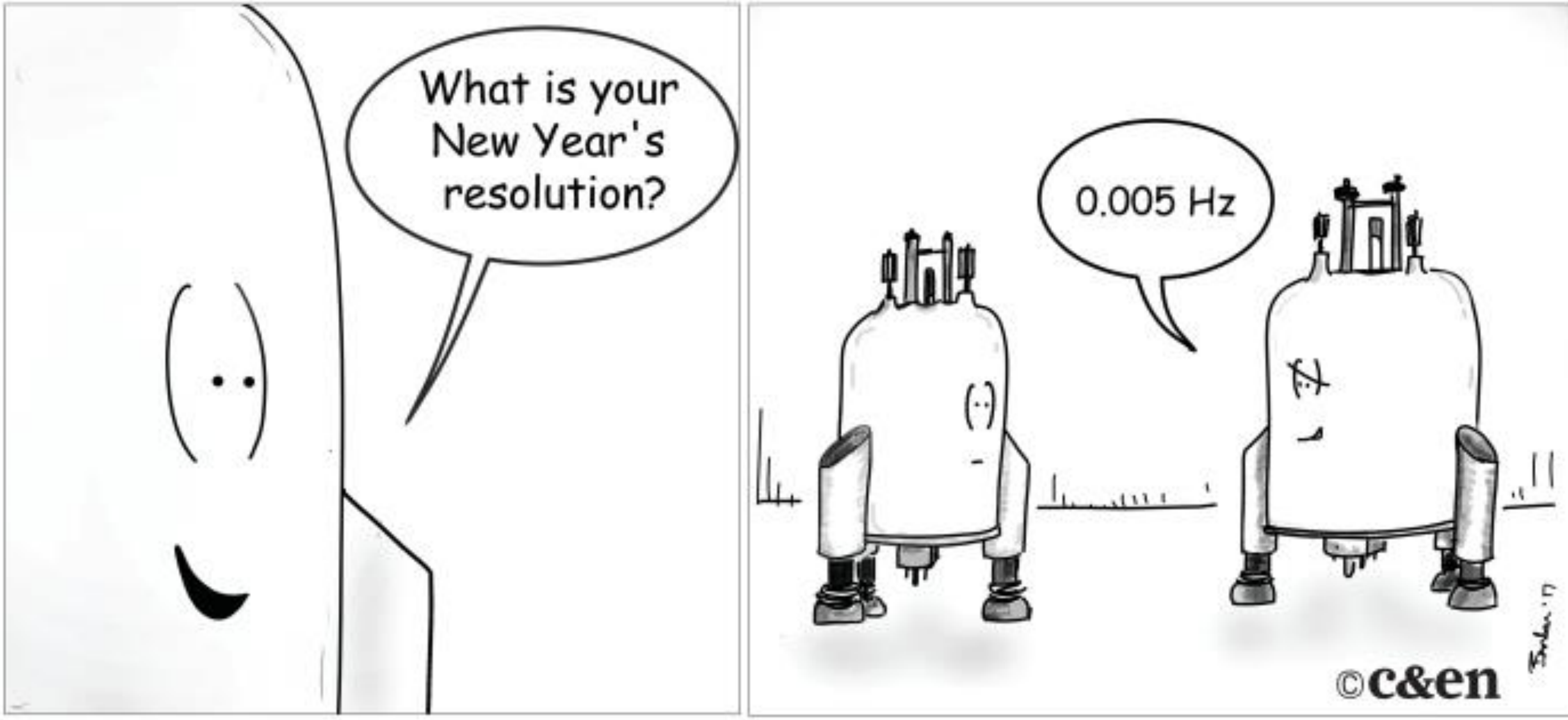


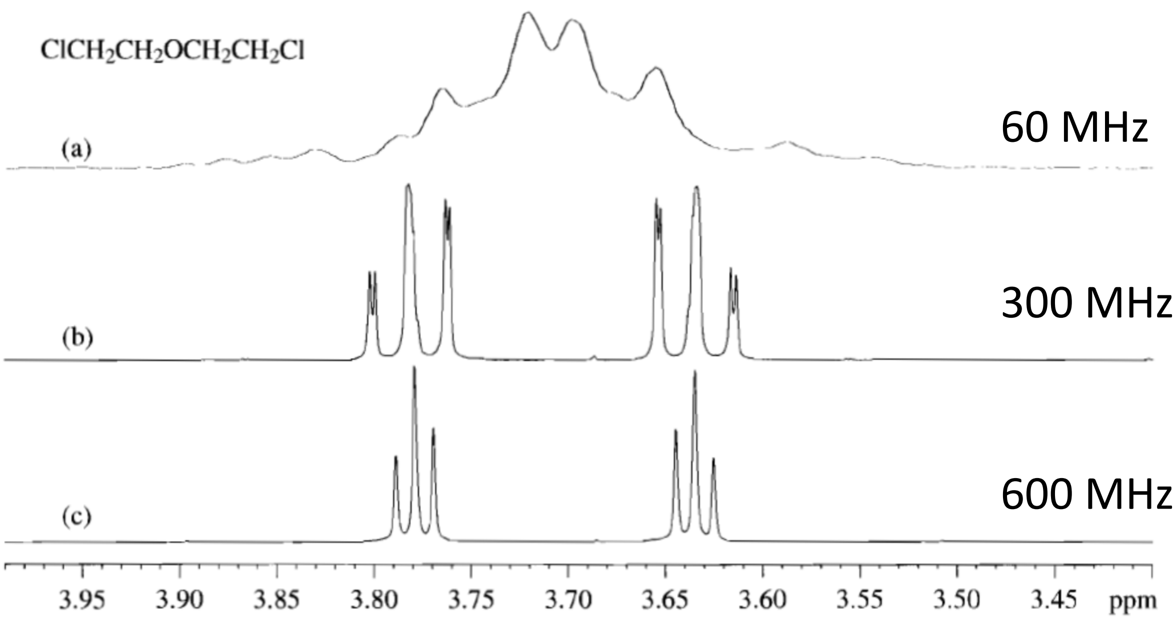
scans, 256 experiments]. (**a**) *HH* COSY; (**b**) *HH* TOCSY; (**c**) selective one-dimensional *HH* TOCSY, soft pulse irradiation at $\delta_H = 5.20$ (signal not shown), compared with the ^1H NMR spectrum on top; deviations of chemical

Table 2.2. ^{13}C chemical shift ranges for organic compounds




Heteroaromatics
π-electron deficient
π-electron rich
Aromatics
(–)-M-substituted
(+)-M-substituted
(Cyclo-) Alkenes
(–)-M-substituted
(+)-M-substituted
(Cyclo-) Alkynes
(Cyclo-) Alkanes
alkylated
4°
3°
2°
1°
Cyclo-propanes
R_3C–O–
R_3C–NR_2
R_3C–S–
R_3C–Halogen
F
I
R_2CH–O–
R_2CH–NR_2
R_2CH–S–
R_2CH–Halogen
F
I
H_3C–O–
H_3C–NR_2
H_3C–S–
H_3C–Halogen
F
I
Alkyl bonded to metal
← δ_C
200
150
100
50
0 (TMS)


What is your New Year's resolution?
0.005 Hz
©c&en


ClCH2CH2OCH2CH2Cl
(a)
60 MHz
(b)
300 MHz
(c)
600 MHz
3.95 3.90 3.85 3.80 3.75 3.70 3.65 3.60 3.55 3.50 3.45 ppm

Tips on Structure Elucidation by NMR Spectroscopy

1. Molecular Formula

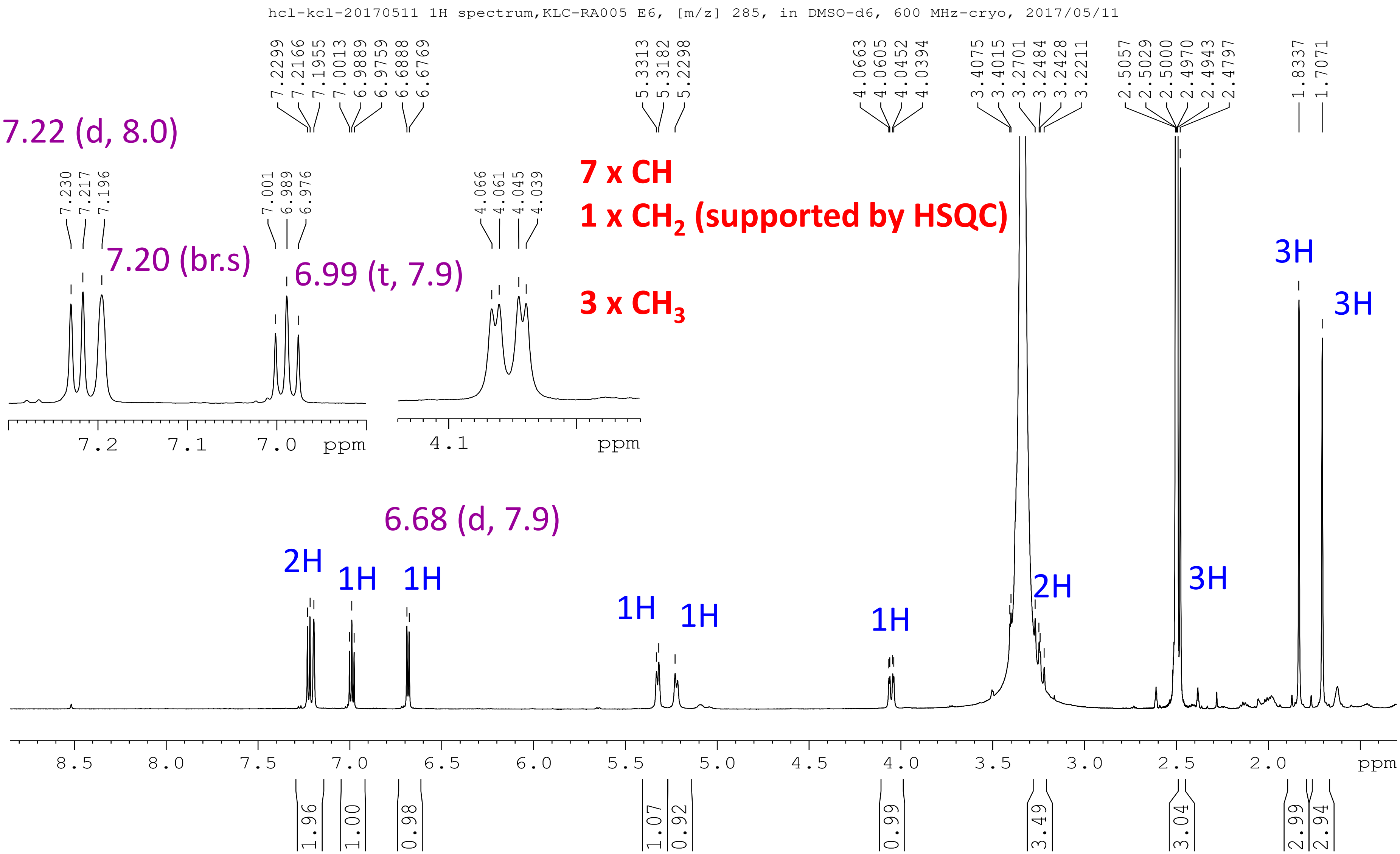
- +m/z = 285 (Mw = 284)
- Numbers of protons in ^{1}H spectrum
- Numbers of carbons and multiplicity in ^{13}C spectrum
- Propose number of heteroatoms

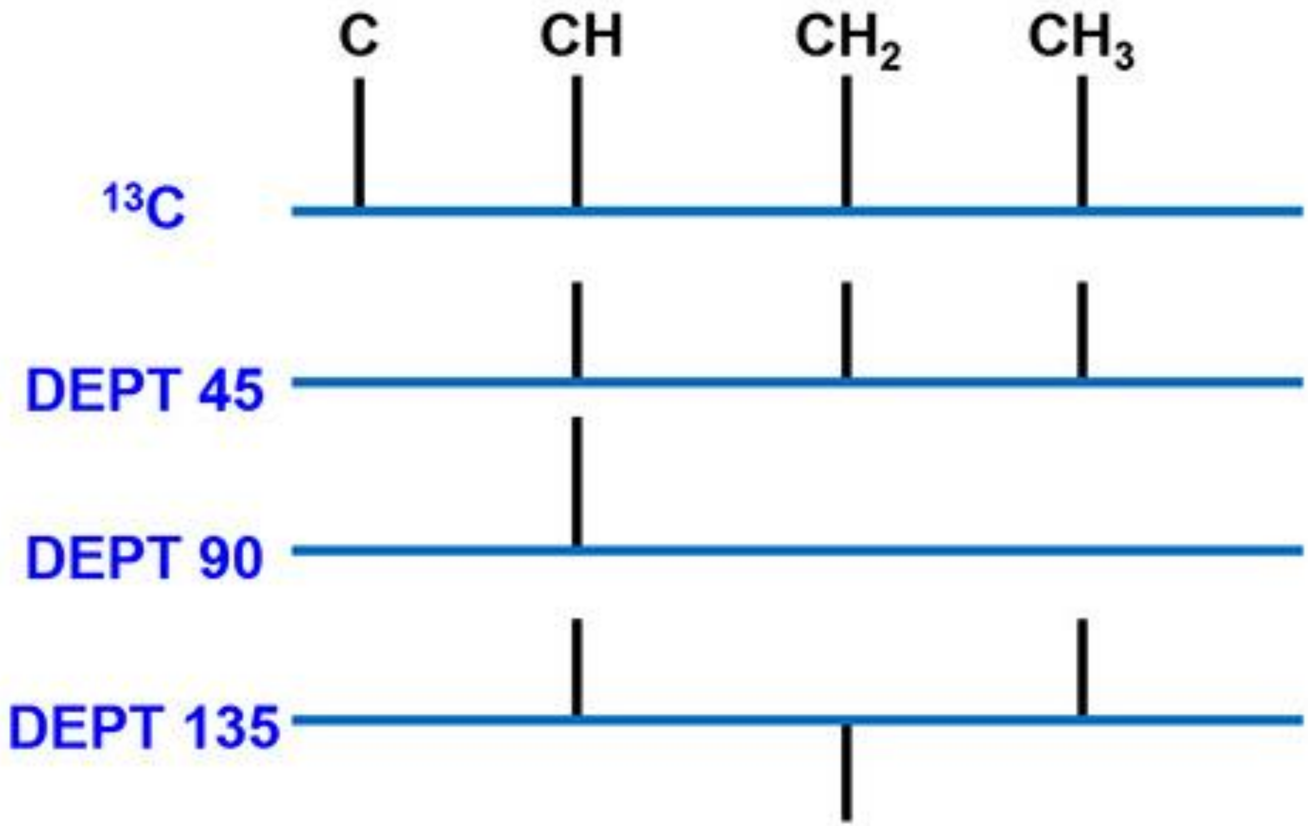
2. **Table of 1D (including coupling patterns) and 2D signals**

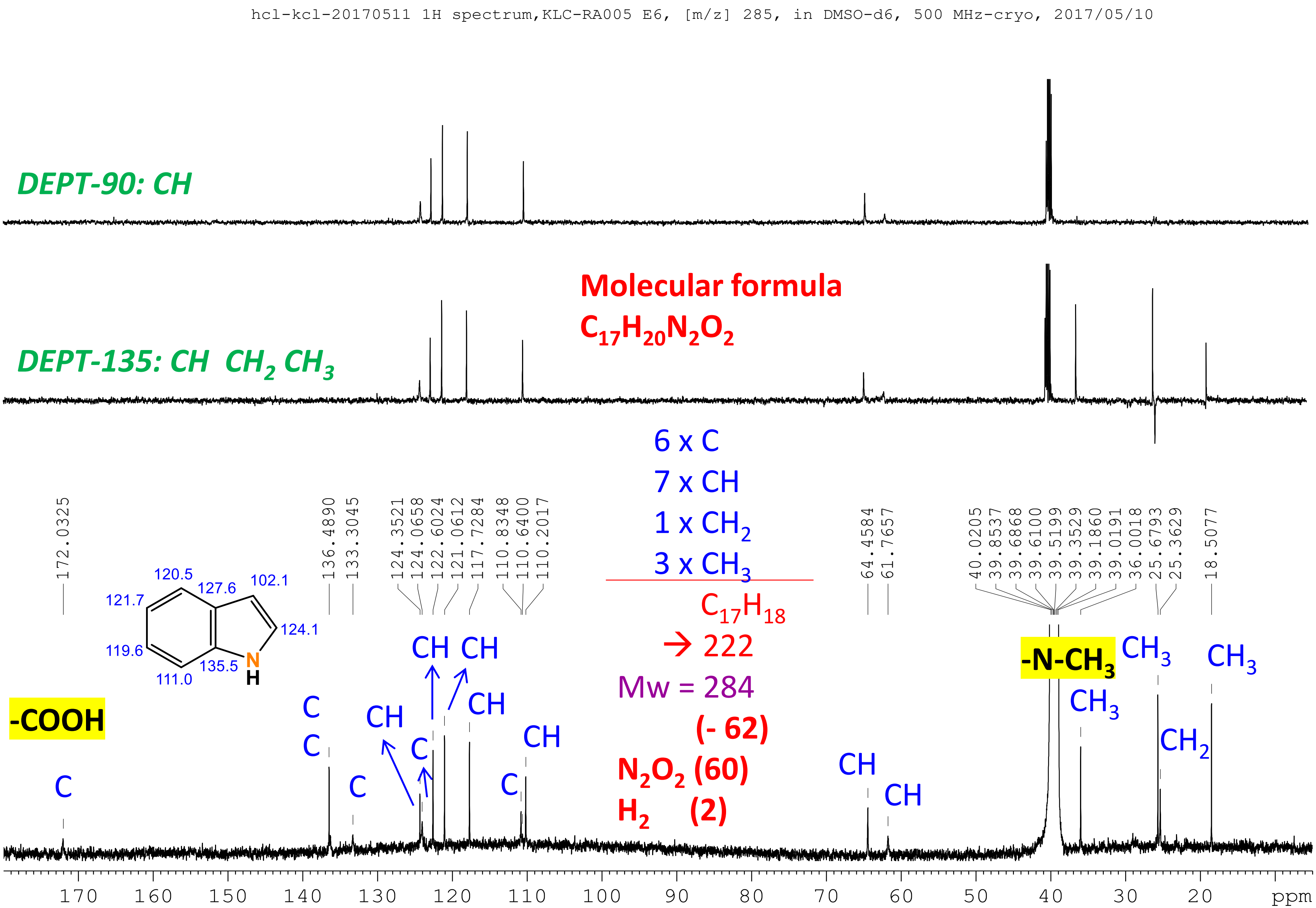
3. Characterize "**moieties**" based on **coupling patterns** and **2D spectra**

4. **Link moieties** together based on **HMBC**, **COSY** and **TOCSY**..etc.

5. Identify **Relative Stereochemistry** based on **NOESY**

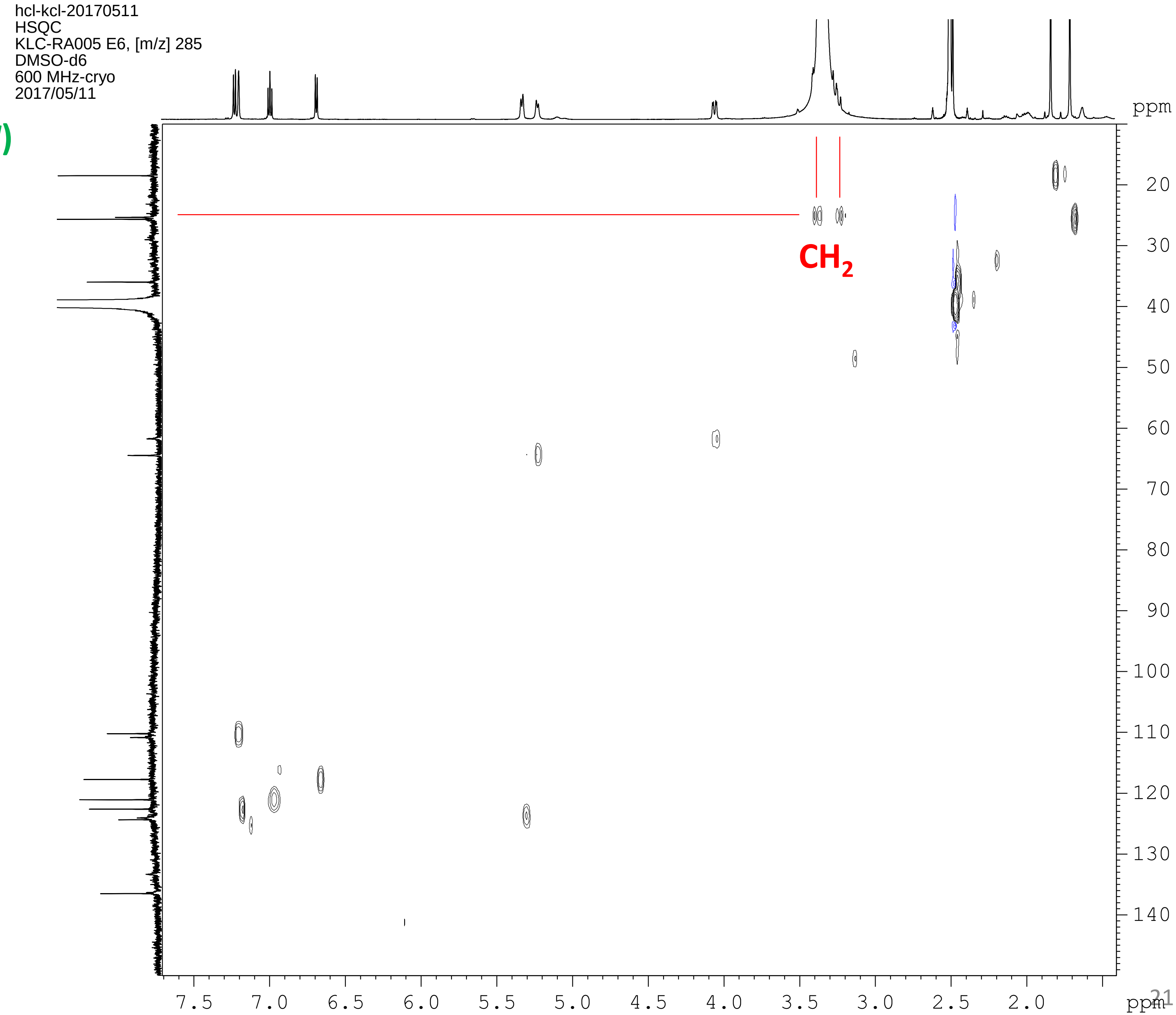

hcl-kcl-20170511 1H spectrum,KLC-RA005 E6, [m/z] 285, in DMSO-d6, 600 MHz-cryo, 2017/05/11
7.2299 7.2166 7.1955 7.0013 6.9889 6.9759 6.6888 6.6769
5.3313 5.3182 5.2298
4.0663 4.0605 4.0452 4.0394
3.4075 3.4015 3.2701 3.2484 3.2428 3.2211
2.5057 2.5029 2.5000 2.4970 2.4943 2.4797
1.8337 1.7071
7.22 (d, 8.0)
7.230 7.217 7.196
7.001 6.989 6.976
7.20 (br.s)
6.99 (t, 7.9)
4.066 4.061 4.045 4.039
7 x CH
1 x CH_2 (supported by HSQC)
3 x CH_3
7.2 7.1 7.0 ppm
4.1 ppm
6.68 (d, 7.9)
2H
1H
1H
1H
1H
1H
2H
3H
3H
3H
8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 ppm
1.96 1.00 0.98 1.07 0.92 0.99 3.49 3.04 2.99 2.94


C
CH
CH_2
CH_3
^{13}C
DEPT 45
DEPT 90
DEPT 135


hcl-kcl-20170511 1H spectrum,KLC-RA005 E6, [m/z] 285, in DMSO-d6, 500 MHz-cryo, 2017/05/10
DEPT-90: CH
DEPT-135: CH CH2 CH3
Molecular formula
C17H20N2O2
6 x C
7 x CH
1 x CH2
3 x CH3
C17H18
→ 222
Mw = 284
(- 62)
N2O2 (60)
H2 (2)
-COOH
-N-CH3
172.0325
136.4890
133.3045
124.3521
124.0658
122.6024
121.0612
117.7284
110.8348
110.6400
110.2017
64.4584
61.7657
40.0205
39.8537
39.6868
39.6100
39.5199
39.3529
39.1860
39.0191
36.0018
25.6793
25.3629
18.5077
120.5
127.6
102.1
121.7
124.1
119.6
135.5
111.0
N
H
C
CH
CH2
CH3
ppm

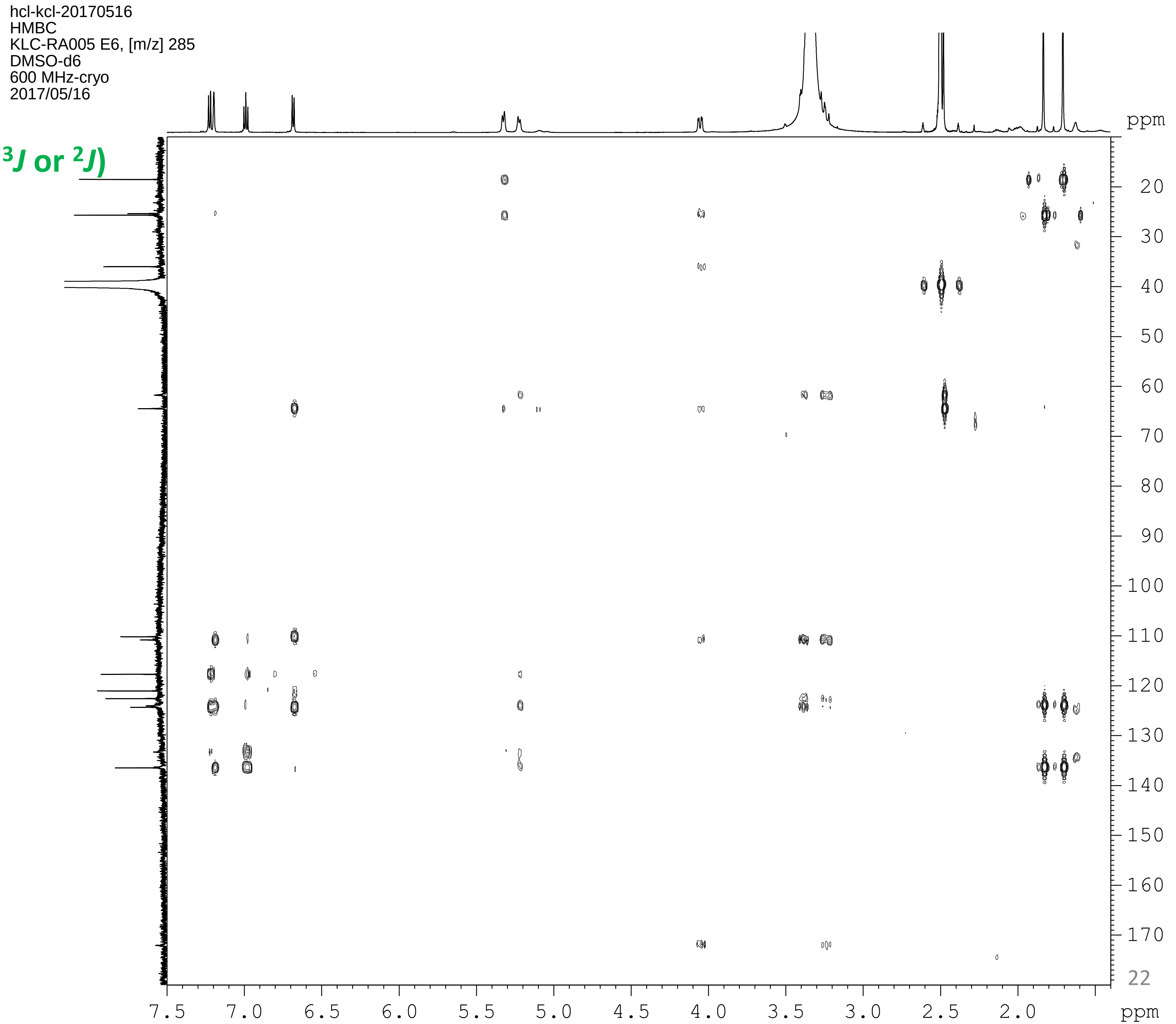
HSQC

C to H (^1J)

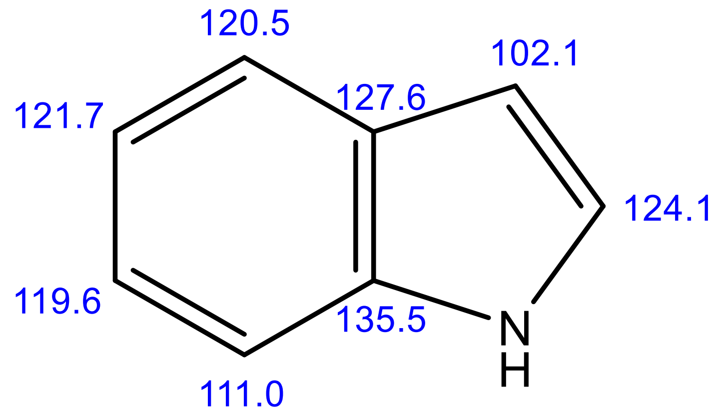


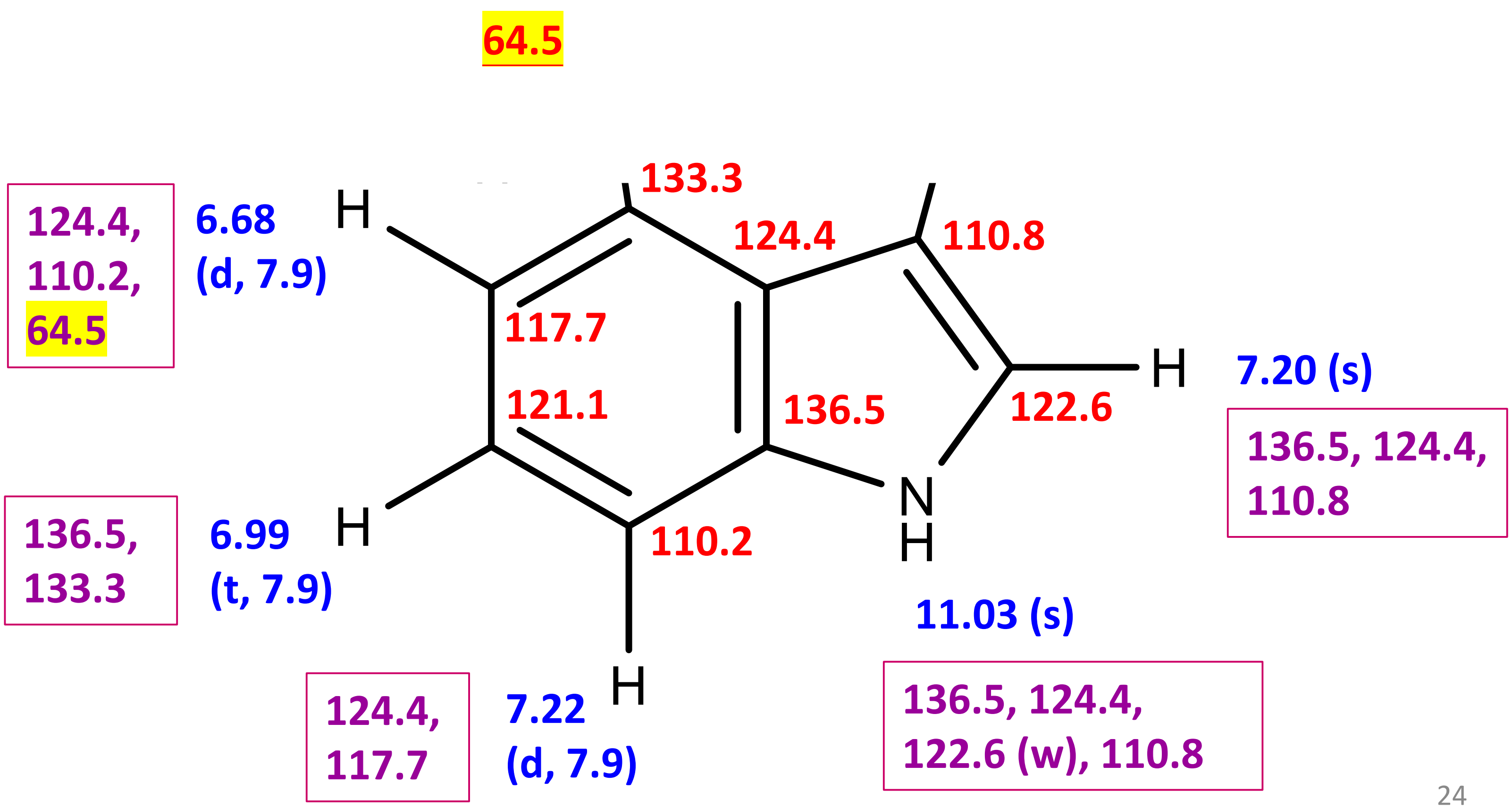
hcl-kcl-20170511
HSQC
KLC-RA005 E6, [m/z] 285
DMSO-d6
600 MHz-cryo
2017/05/11
CH2
ppm
20
30
40
50
60
70
80
90
100
110
120
130
140
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
ppm

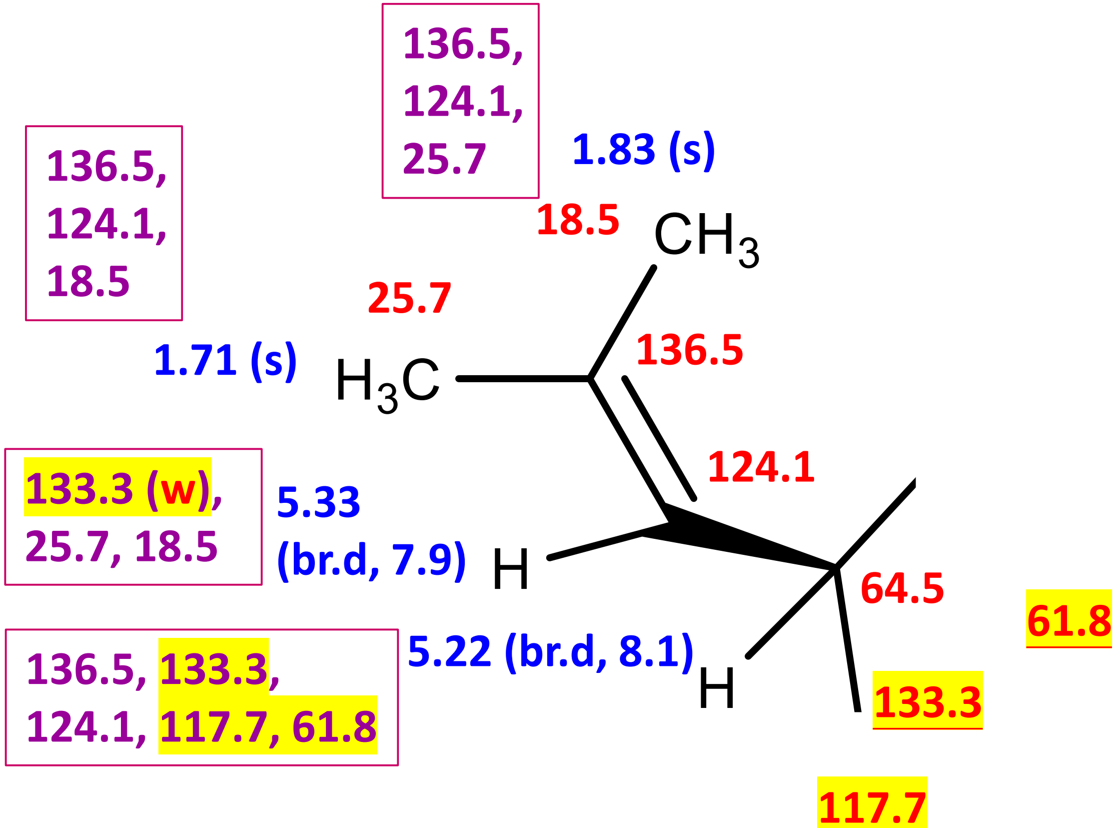
HMBC
C to H (^3J or ^2J)



δ_H (mult)	δ_C (mult)	HMBC	NOESY
11.03 (s)		136.5, 124.4, 122.6 (w), 110.8	6.48, 3.75
7.22 (d, 8.0)	110.2 d	124.4, 117.7	7.27. 7.00
7.20 (br.s)	122.6 d	136.5, 124.4, 110.8	7.48, 6.72
6.99 (t, 7.9)	121.1 d	136.5, 133.3	7.00, 5.18, 1.65
6.68 (d, 7.9)	117.7 d	124.4, 110.2, 64.5	
5.33 (br.d, 7.9)	124.4 d	133.3 (w), 25.7, 18.5	2.48, 2.38
5.22 (br.d, 8.0)	64.5 d	136.5, 133.3, 124.1, 117.7, 61.8	
4.05 (dd, 12.7, 3.6)	61.8 d	172.0, 110.8, 64.5, 36.0, 25.4	
3.39 (m)	25.4 t	124.4, 122.6, 110.8, 61.8	
3.25 (m)		172.0, 124.1, 122.6, 110.8, 61.8	
2.48 (s)	36.0 q	64.5, 61.8	3.91, 2.48, 1.82
1.83 (s)	18.5 q	136.5, 124.1, 25.7	6.72, 6.03, 1.65
1.71 (s)	25.7 q	136.5, 124.1, 18.5	6.88, 5.39, 5.18. 1.85, 1.65
	172.0 s		
	136.5 s		
	136.5 s		
	133.3 s		
	124.1 s		
	110.8 s		

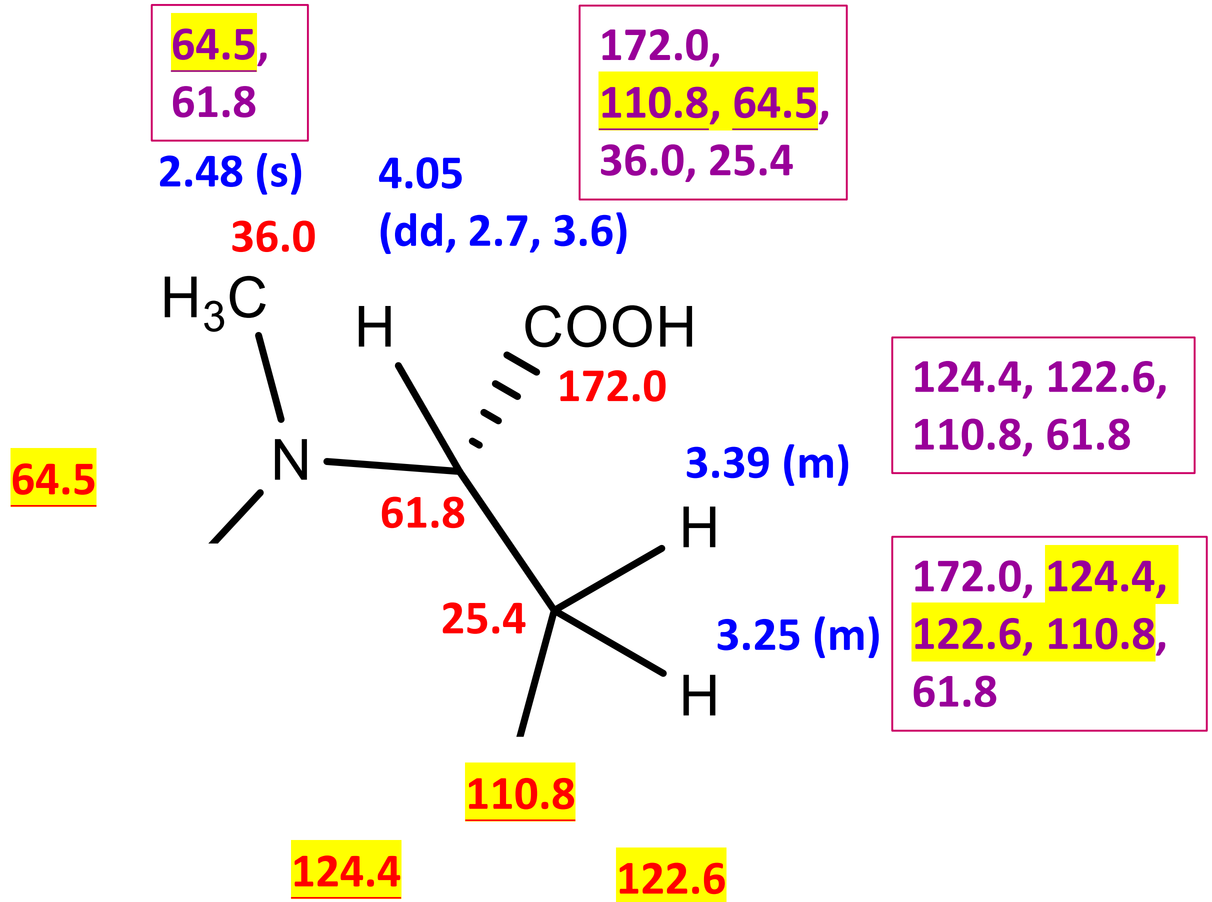

120.5
102.1
121.7
127.6
124.1
119.6
135.5
N
H
111.0

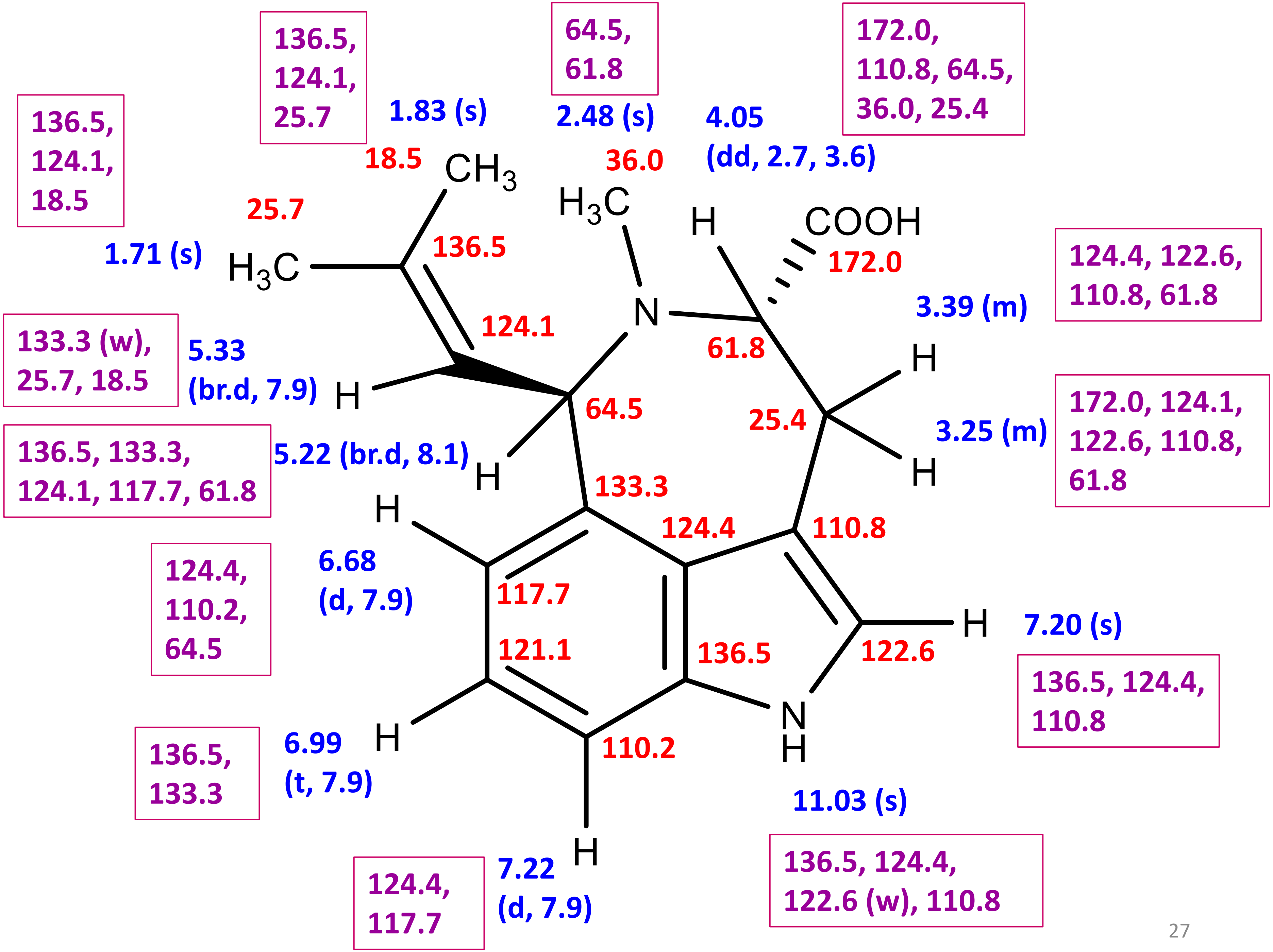

64.5
133.3
124.4, 110.2, 64.5
6.68 (d, 7.9)
H
124.4
110.8
117.7
121.1
136.5
122.6
H
7.20 (s)
136.5, 124.4, 110.8
N
H
136.5, 133.3
6.99 (t, 7.9)
H
110.2
11.03 (s)
H
124.4, 117.7
7.22 (d, 7.9)
136.5, 124.4, 122.6 (w), 110.8



- **HMBC**: C to H (^3J or ^2J)
- **COSY**: H to H (^2J or ^3J)

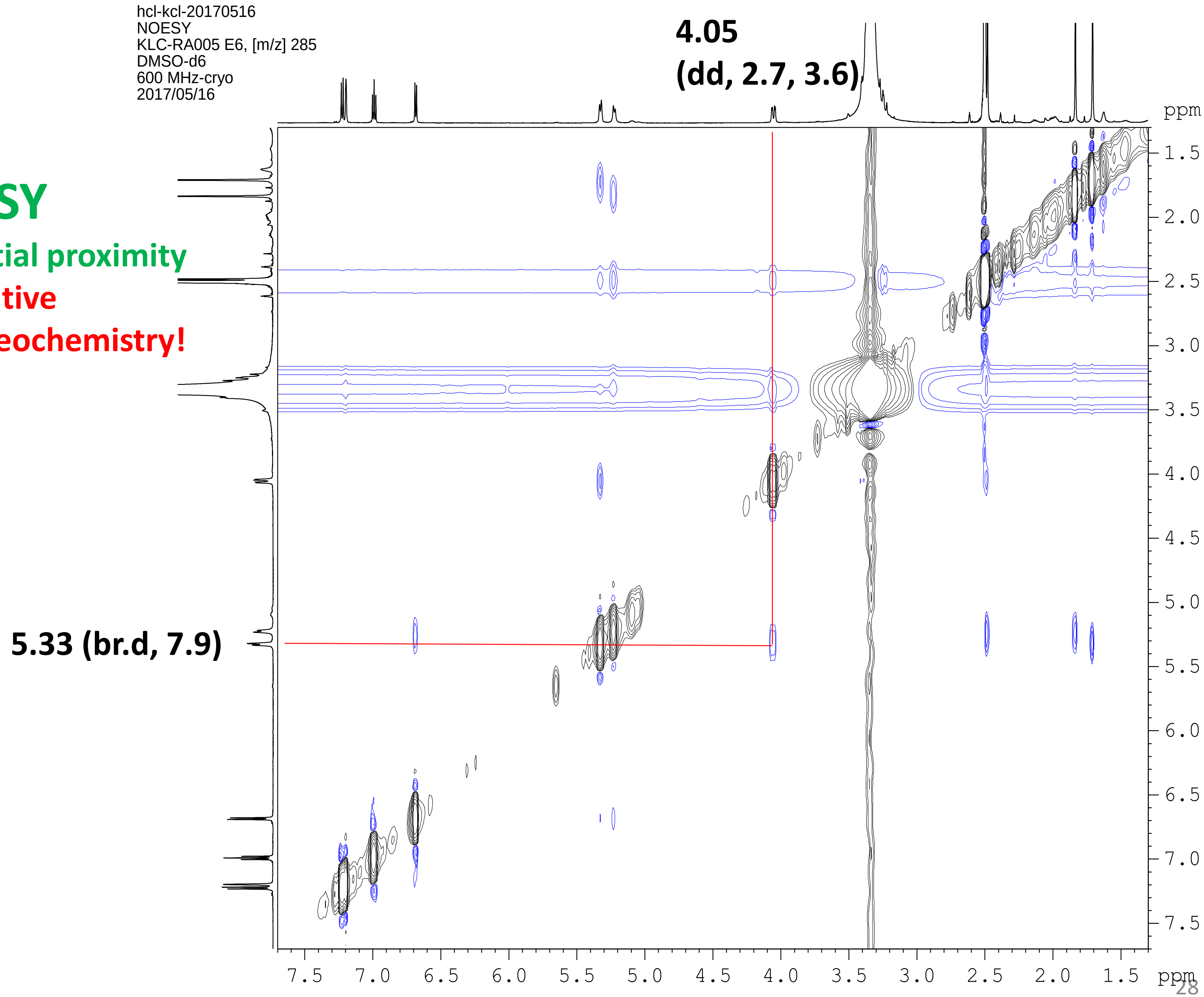
is also very informative that correlates those protons coupling to each other.

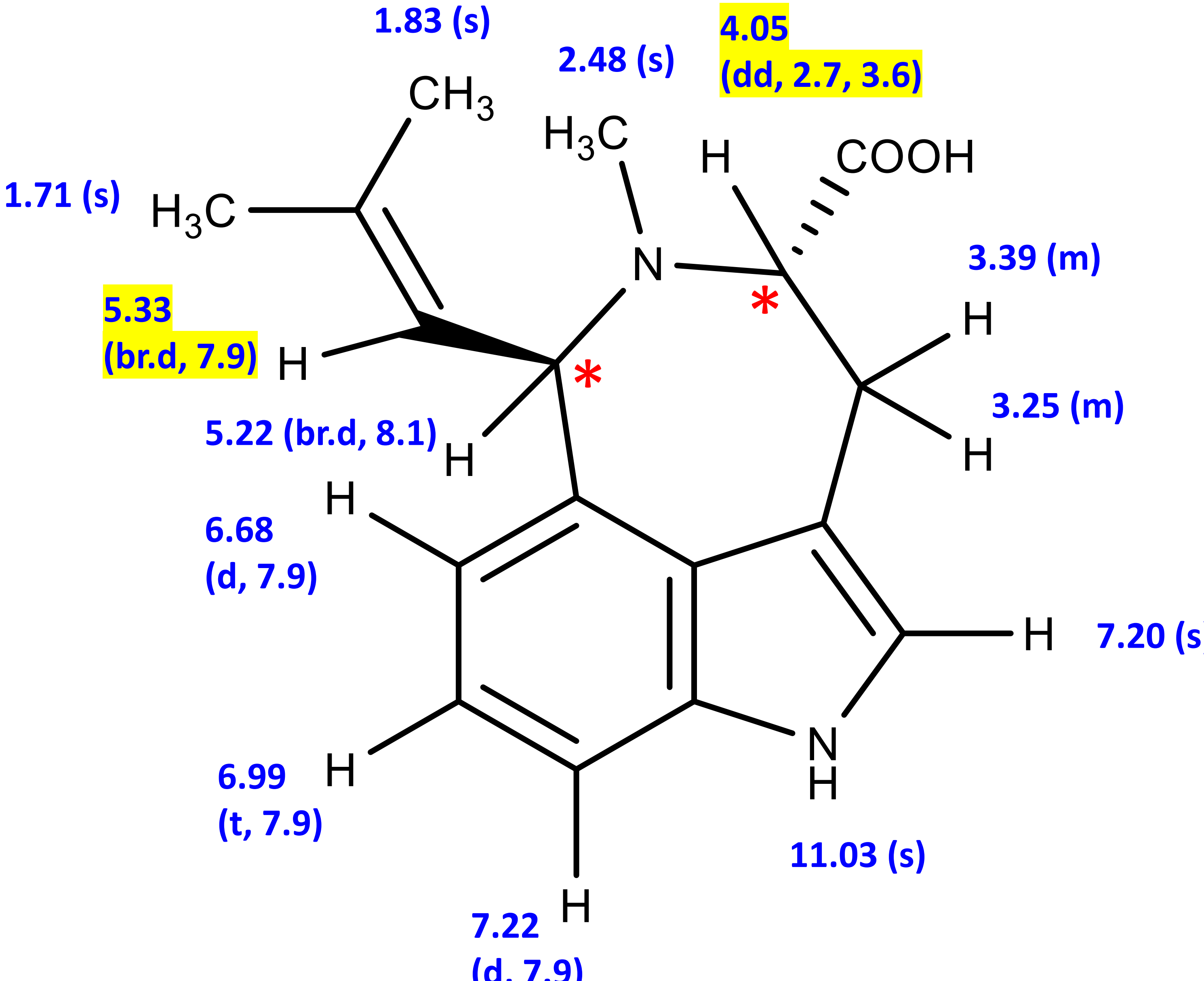

64.5, 61.8
172.0, 110.8, 64.5, 36.0, 25.4
2.48 (s)
4.05 (dd, 2.7, 3.6)
36.0
H3C
H
COOH
172.0
N
64.5
61.8
124.4, 122.6, 110.8, 61.8
3.39 (m)
H
172.0, 124.4, 122.6, 110.8, 61.8
25.4
3.25 (m)
H
110.8
124.4
122.6


136.5,
124.1,
18.5
136.5,
124.1,
25.7
1.83 (s)
18.5
CH3
64.5,
61.8
2.48 (s)
36.0
4.05
(dd, 2.7, 3.6)
172.0,
110.8, 64.5,
36.0, 25.4
25.7
1.71 (s)
H3C
136.5
H3C
H
COOH
172.0
124.4, 122.6,
110.8, 61.8
N
3.39 (m)
124.1
133.3 (w),
25.7, 18.5
5.33
(br.d, 7.9)
H
61.8
H
64.5
25.4
172.0, 124.1,
122.6, 110.8,
61.8
3.25 (m)
136.5, 133.3,
124.1, 117.7, 61.8
5.22 (br.d, 8.1)
H
133.3
H
H
124.4
110.8
124.4,
110.2,
64.5
6.68
(d, 7.9)
117.7
H
7.20 (s)
121.1
136.5
122.6
136.5, 124.4,
110.8
136.5,
133.3
6.99
(t, 7.9)
H
110.2
N
H
11.03 (s)
H
124.4,
117.7
7.22
(d, 7.9)
136.5, 124.4,
122.6 (w), 110.8

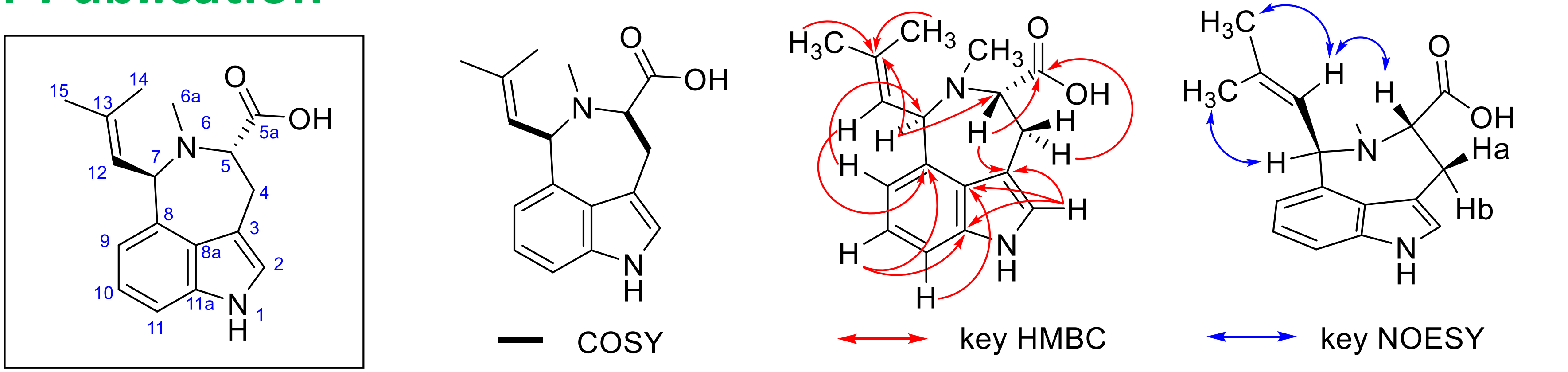
NOESY

- → Spatial proximity
- → Relative stereochemistry!



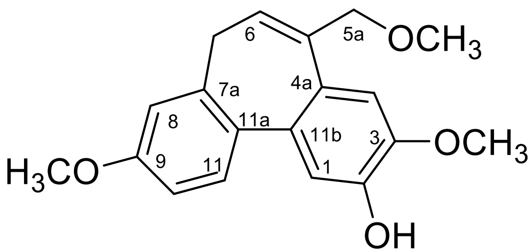

1.83 (s)
2.48 (s)
4.05
(dd, 2.7, 3.6)
CH3
H3C
H
COOH
1.71 (s)
H3C
N
3.39 (m)
5.33
(br.d, 7.9)
H
H
3.25 (m)
5.22 (br.d, 8.1)
H
H
H
6.68
(d, 7.9)
H
7.20 (s)
N
H
6.99
(t, 7.9)
H
11.03 (s)
7.22
(d, 7.9)
H

For Publication

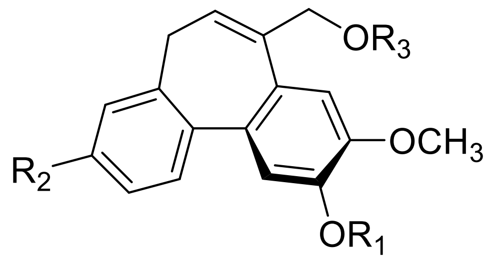


No.	δ_H (mult, *J* in Hz)	δ_C (type)	HMBC	COSY	NOESY
1	11.04 (s)		110.8, 122.6, 124.4, 136.5		
2	7.20 (s)	122.6 (CH)	25.4, 110.8, 124.4, 136.5		
3		110.8 (C)			
4a	3.42 (m)	25.4 (CH_2)	61.8, 110.8, 122.6		
4b	3.27 (m)		61.8, 110.8, 122.6, 172.0	4.05	
5	4.05 (dd, 12.0, 3.6)	61.8 (CH)	25.4, 36.0, 64.5, 110.8, 172.0	3.27	5.32
5a		172.0 (C)			
6a	2.48 (s)	36.0 (CH_3)	64.5		5.22
7	5.22 (d, 7.8)	64.5 (CH)	61.8, 124.1, 136.5	5.32	1.83, 2.48, 6.68
8		133.3 (C)			
8a		124.4 (C)			
9	6.68 (d, 7.8)	117.7 (CH)	64.5, 110.2, 124.4	6.99	5.22
10	6.99 (t, 7.8)	121.1 (CH)	133.3, 136.5	6.68, 6.99	
11	7.22 (d, 7.8)	110.2 (CH)	117.7, 124.4	6.99	
11a		136.5 (C)			
12	5.32 (d, 7.8)	124.1 (CH)	18.5, 25.7, 64.5, 133.3	5.22	1.70, 4.05
13		136.5 (C)			
14	1.83 (s)	18.5 (CH_3)	25.7, 124.1, 136.5		5.22
15	1.70 (s)	25.7 (CH_3)	18.5, 124.1, 136.5		5.32

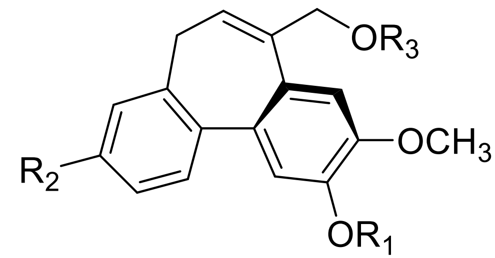
Dibenzocycloheptenes



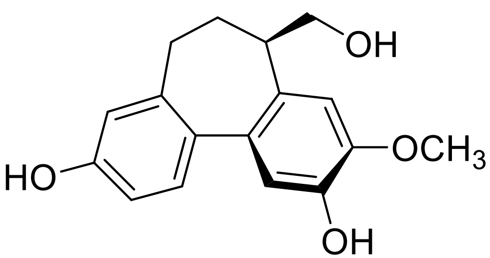
27



	R_1	R_2	R_3
28a	H	H	Glc
29a	H	OCH_3	Glc
30a	H	OH	Glc
31a	Glc	H	H
32a	Glc	OCH_3	CH_3
33a	Rha	OH	H



	R_1	R_2	R_3
28a	H	H	Glc
29b	H	OCH_3	Glc
30b	H	OH	Glc
31b	Glc	H	H
32b	Glc	OCH_3	CH_3
33b	Rha	OH	H



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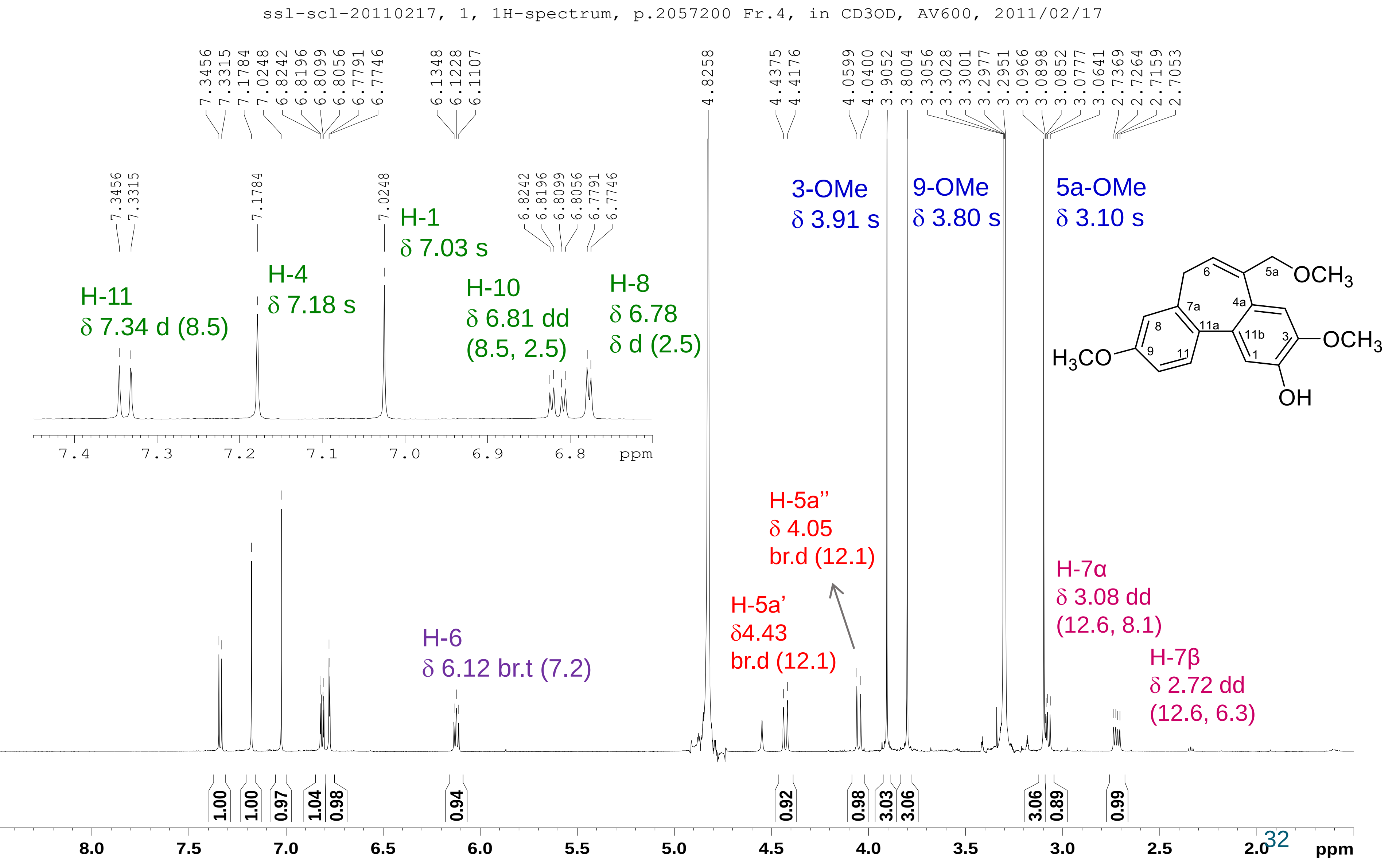
Lin, H. C., Lee, S. S.*. Dibenzocycloheptenes from the Leaves of *Cinnamomum subavenium.*
***J. Nat. Prod.* 2012**, *75*, 1735−1743

Cpd 27: cinnasubavene A

HR-ESI-MS: [M-H]$^{-}$= 335.1246 *m/z*

Calcd for $C_{19}H_{20}O_4$−H

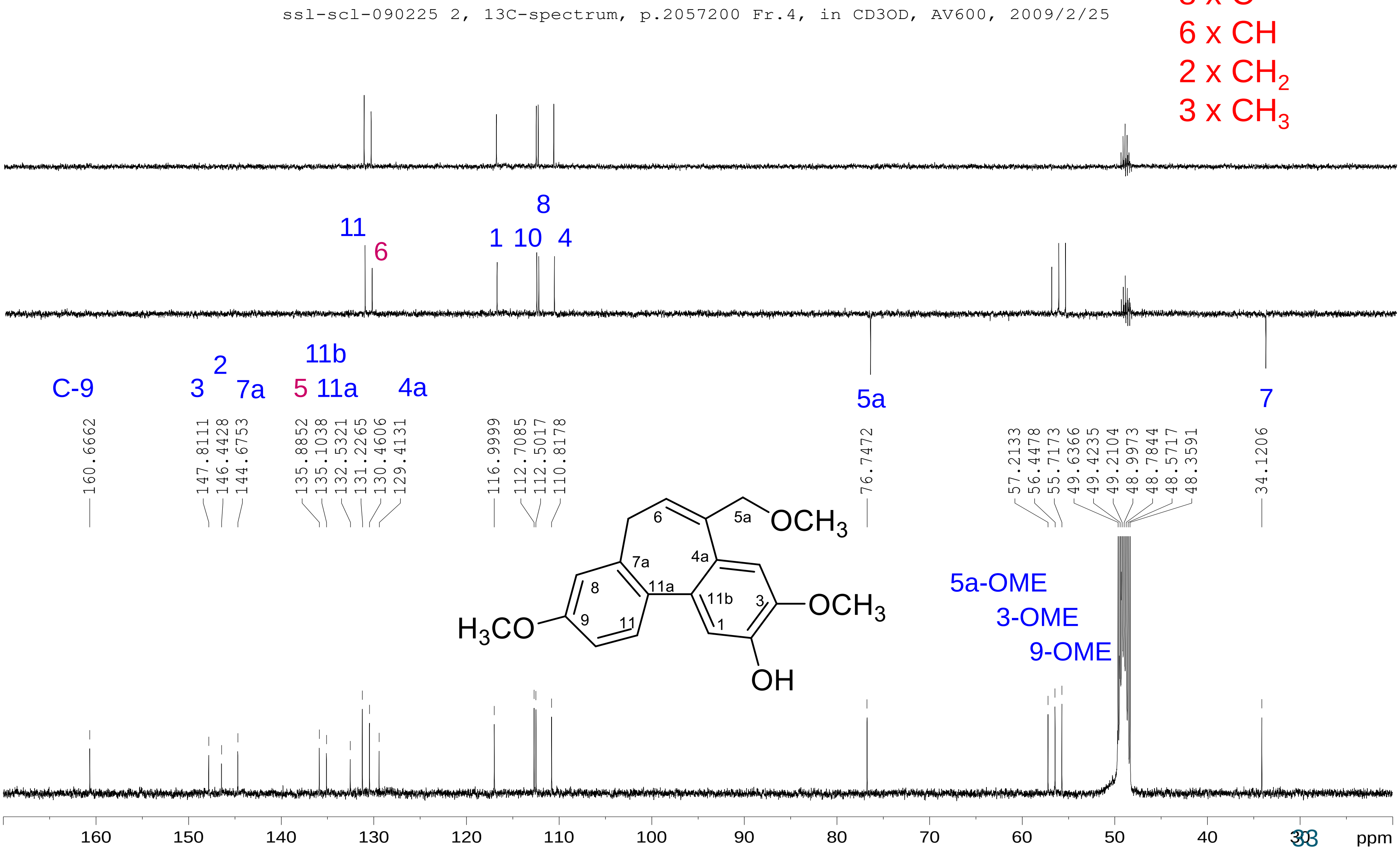


Cpd 27: cinnasubavene A

HR-ESI-MS: $[M-H]^-$= 335.1246 *m/z*

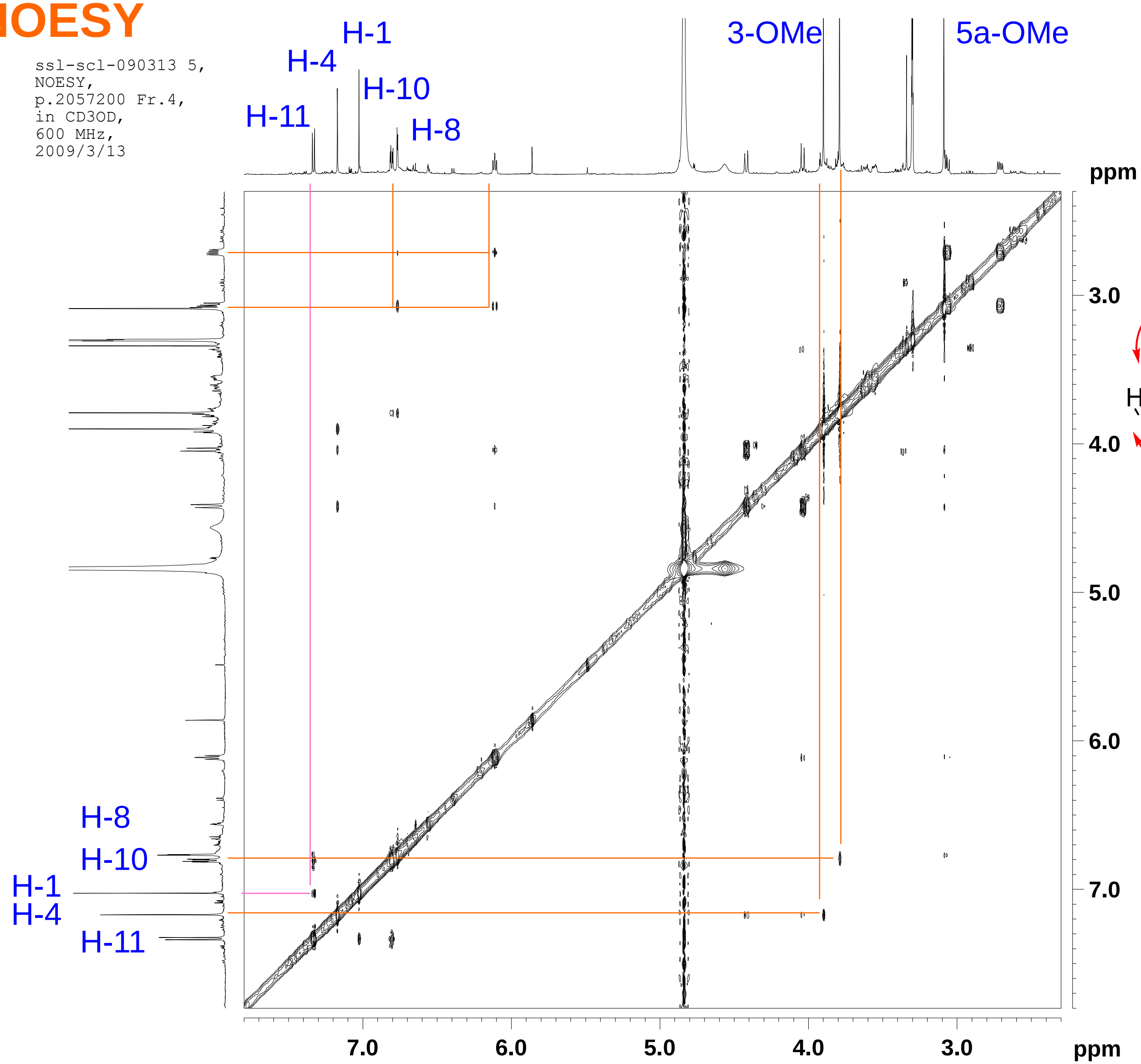
Calcd for $C_{19}H_{20}O_4$−H

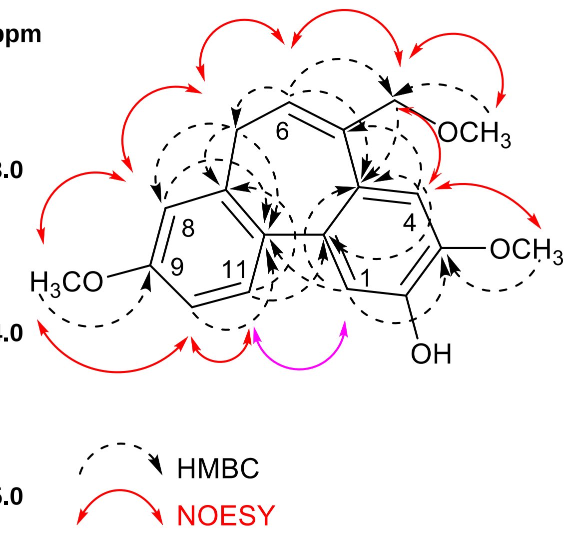
8 x C
6 x CH
$2 \times CH_2$
$3 \times CH_3$



Cpd 27: cinnasubavene A

NOESY

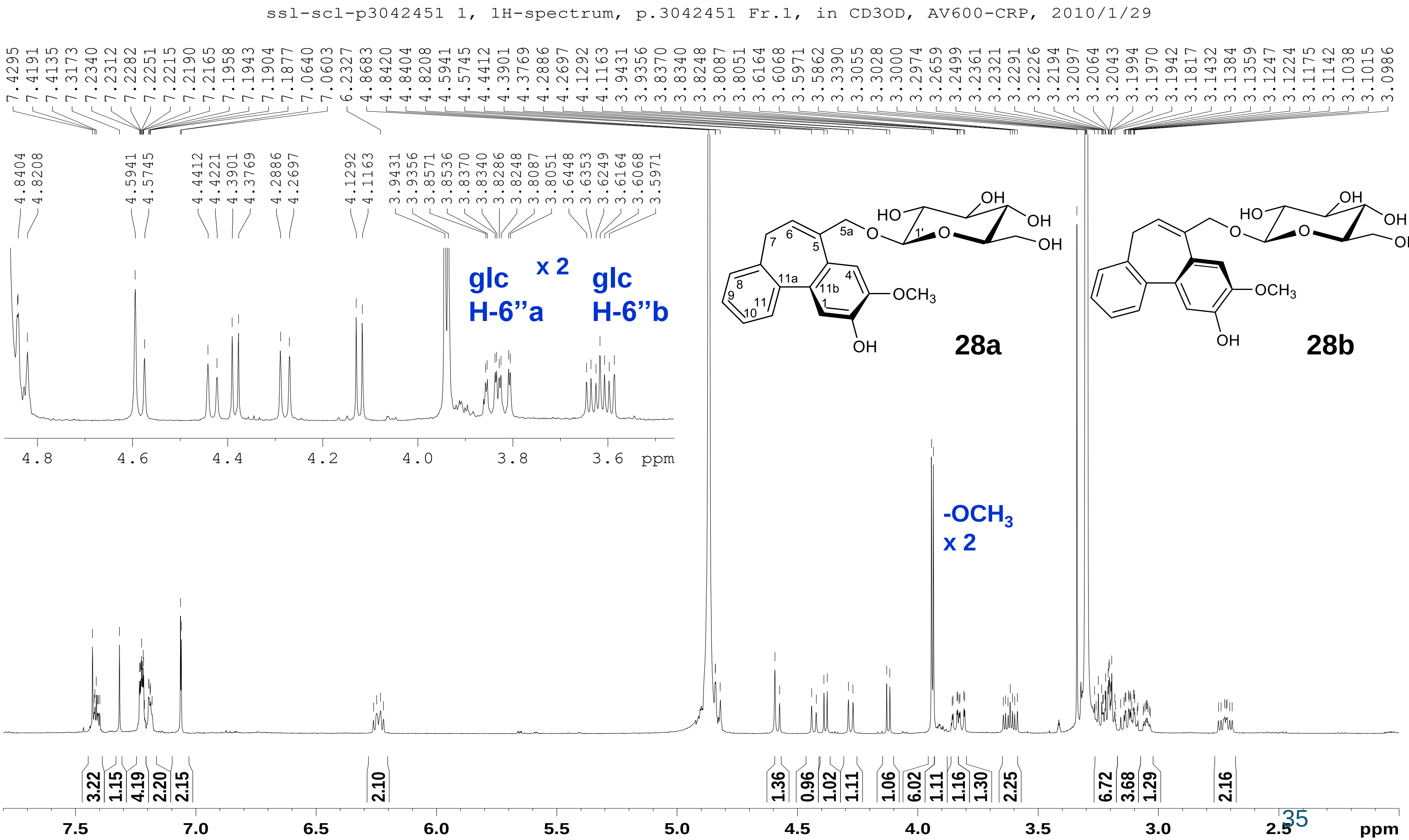




Cinnasubavene B (**28a**: *aR*, **28b**: *aS*)

HR-ESI-MS: $[M-H]^-$= 429.1555 *m/z*

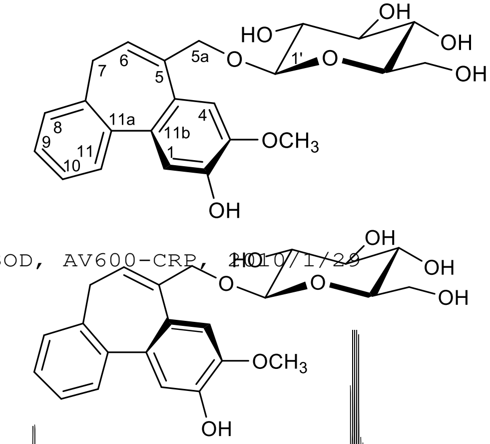
Calcd for $C_{23}H_{25}O_8-H$



Cinnasubavene B (**28a**: *aR*, **28b**: *aS*)

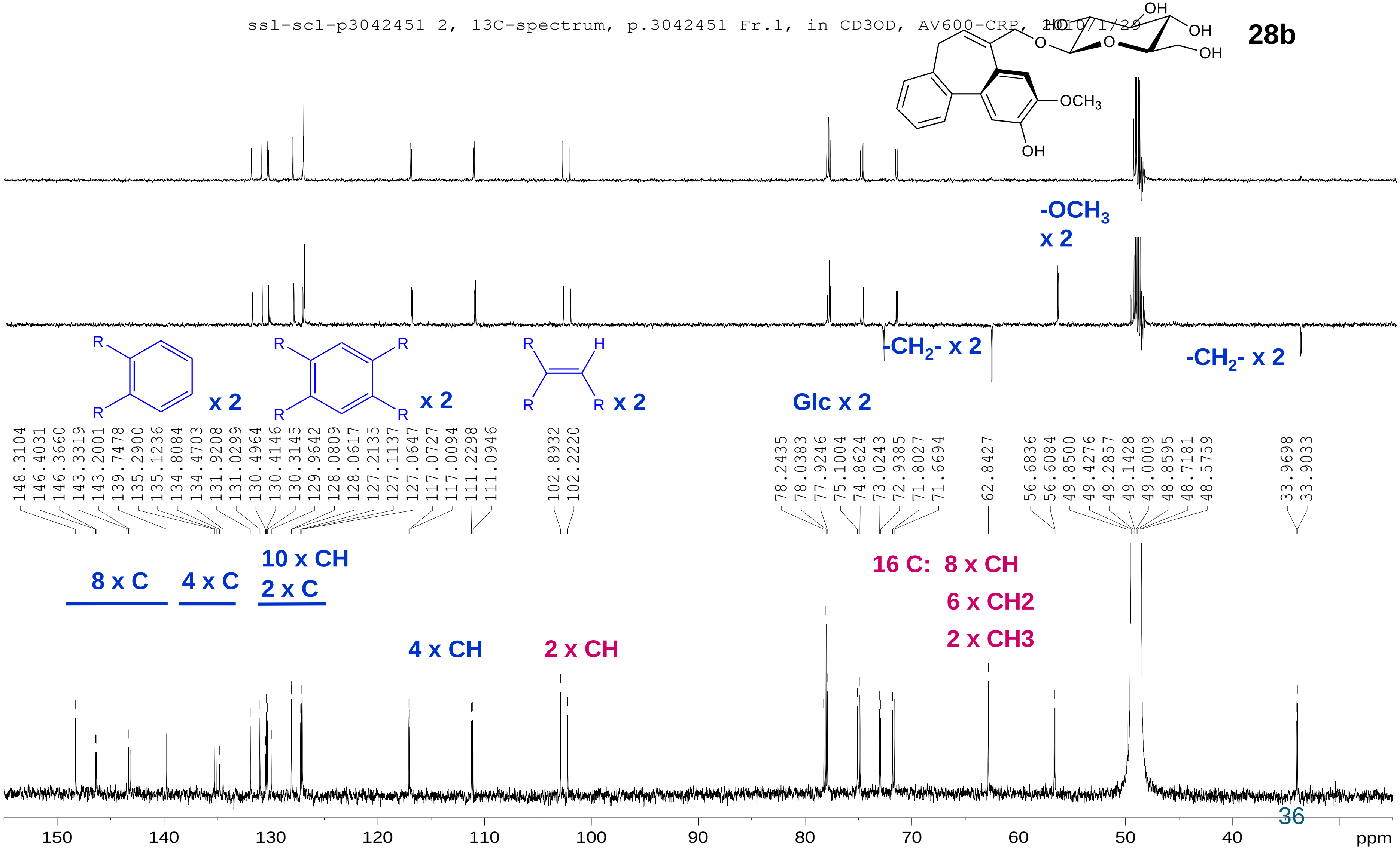
HR-ESI-MS: $[M-H]^-$= 429.1555 *m/z*

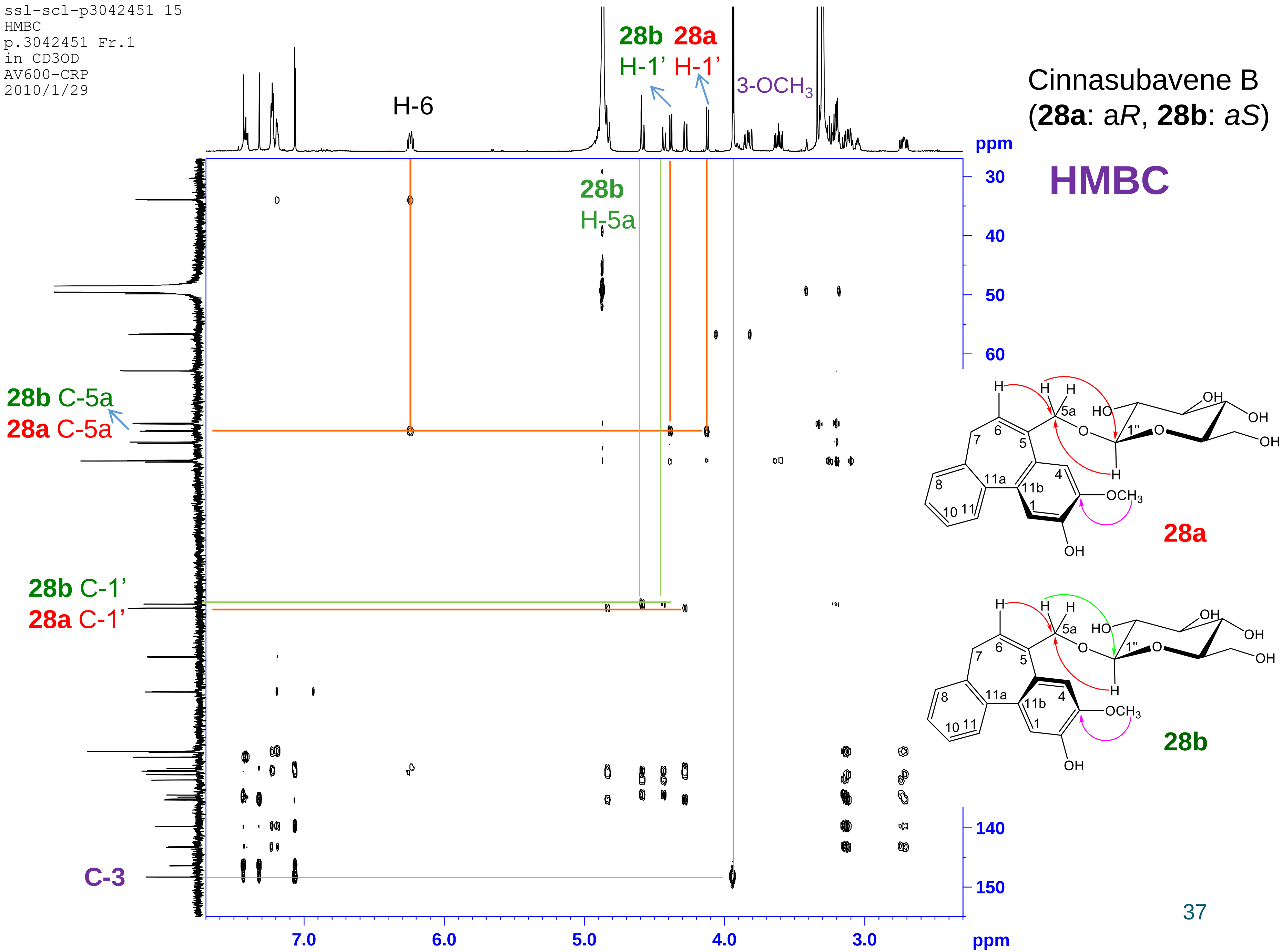
Calcd for $C_{23}H_{25}O_8$−H



28a

28b





ssl-scl-p3042451 15
HMBC
p.3042451 Fr.1
in CD3OD
AV600-CRP
2010/1/29
H-6
28b 28a
H-1' H-1'
3-OCH3
Cinnasubavene B
(28a: aR, 28b: aS)
HMBC
28b
H-5a
28b C-5a
28a C-5a
28b C-1'
28a C-1'
C-3
28a
28b
ppm
30
40
50
60
140
150
7.0
6.0
5.0
4.0
3.0

Verification of the presence of atropisomers with 1D-NOESY

1D-NOESY: selective excitation at the glc H-1 of 2a (δ 4.12) caused spontaneous excitation of the glc H-1 of 2b (δ 4.38) → This evidence confirms that the two atropisomers exist in equilibrium, rendering them inseparable.

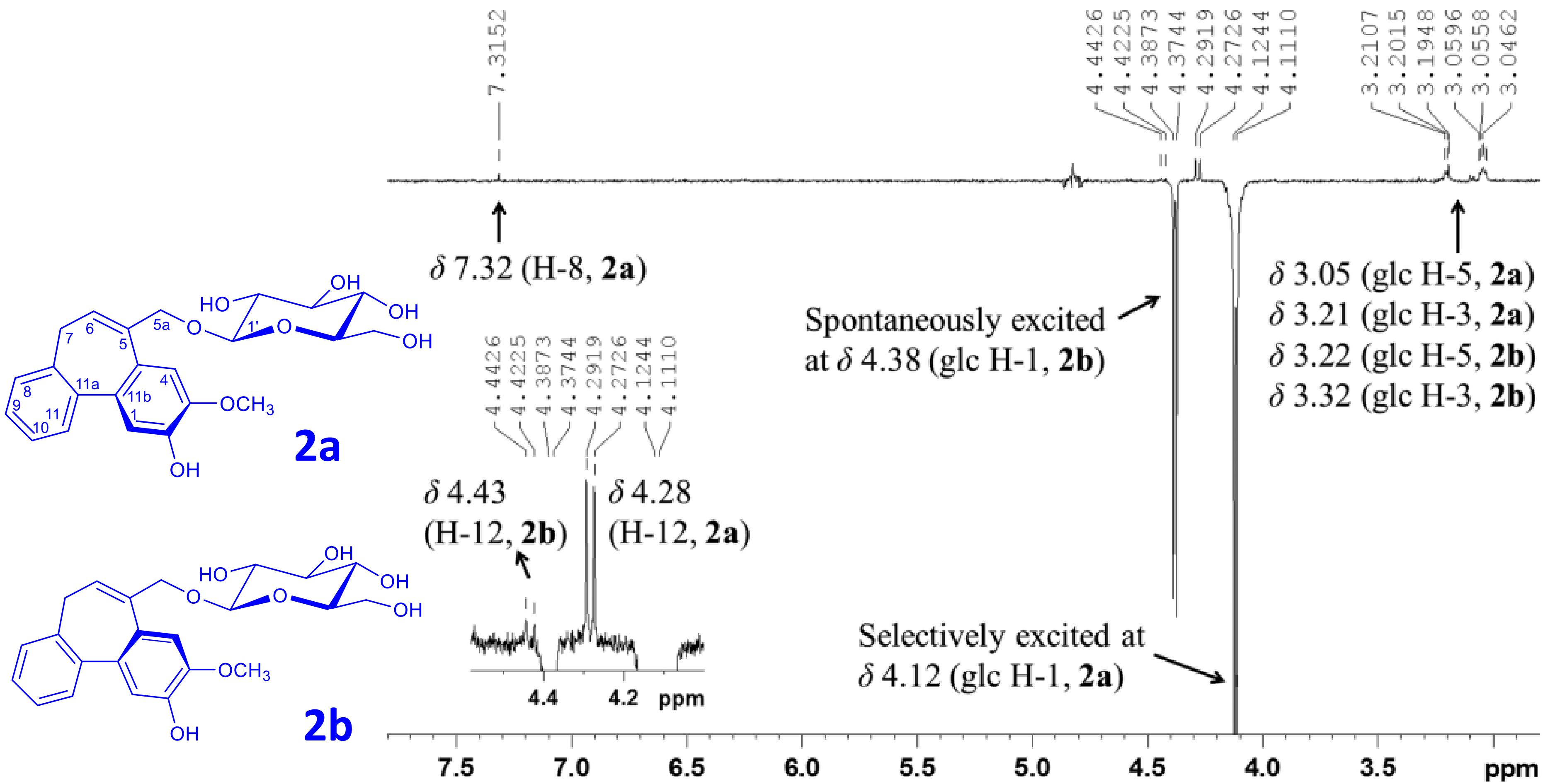
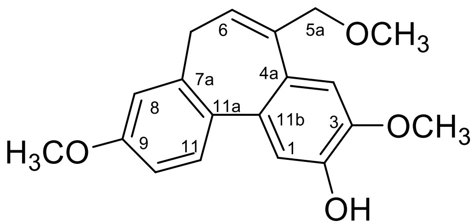
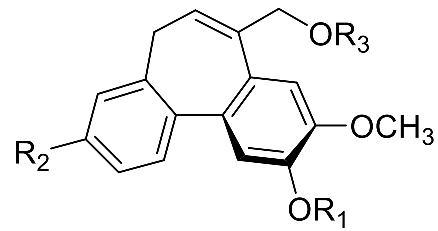
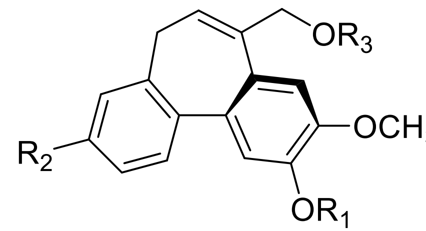


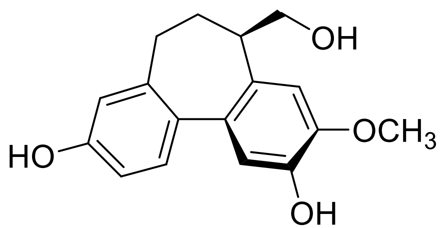
Figure 3. 1D NOESY of **2a** by selective excitation at δ 4.12 (glc H-1) (methanol-d_4, 600 MHz).



27







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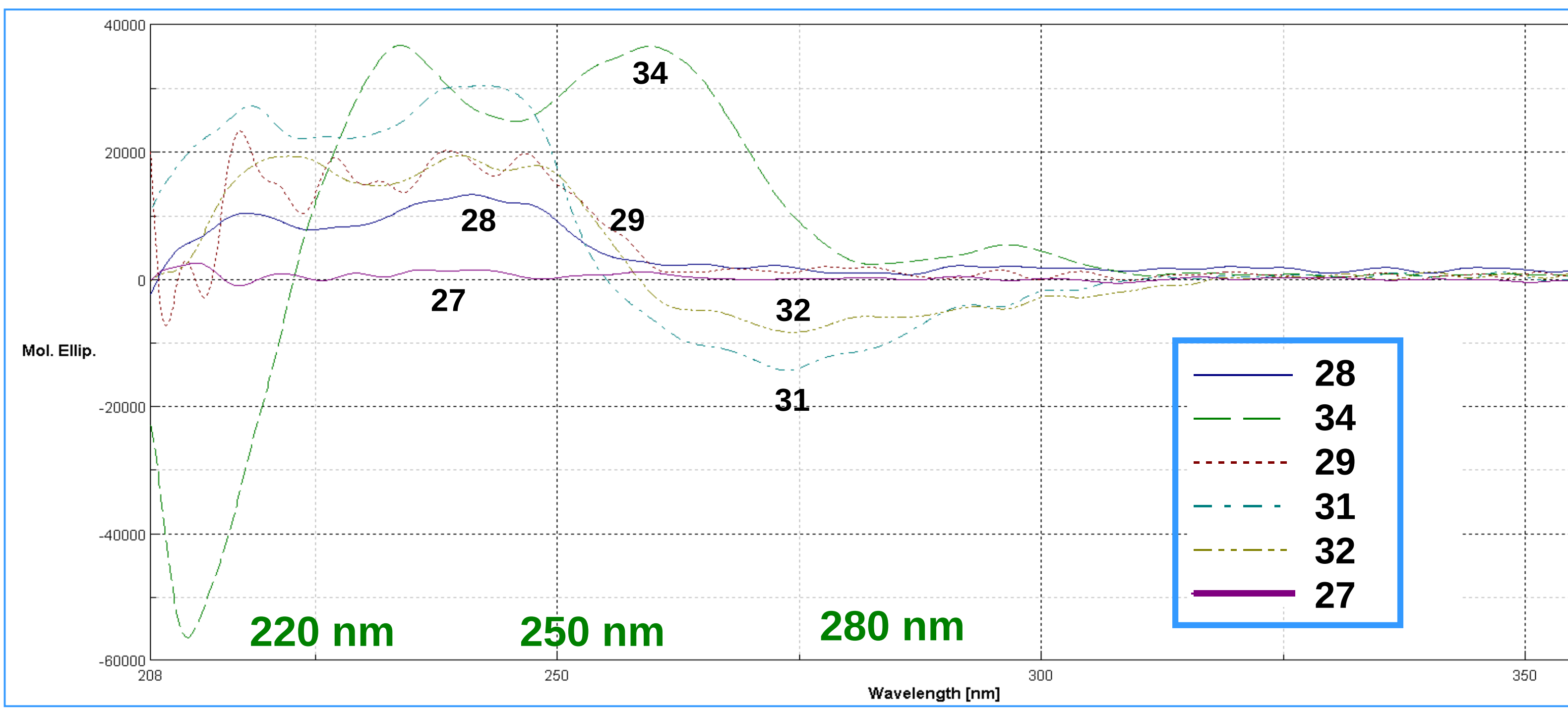
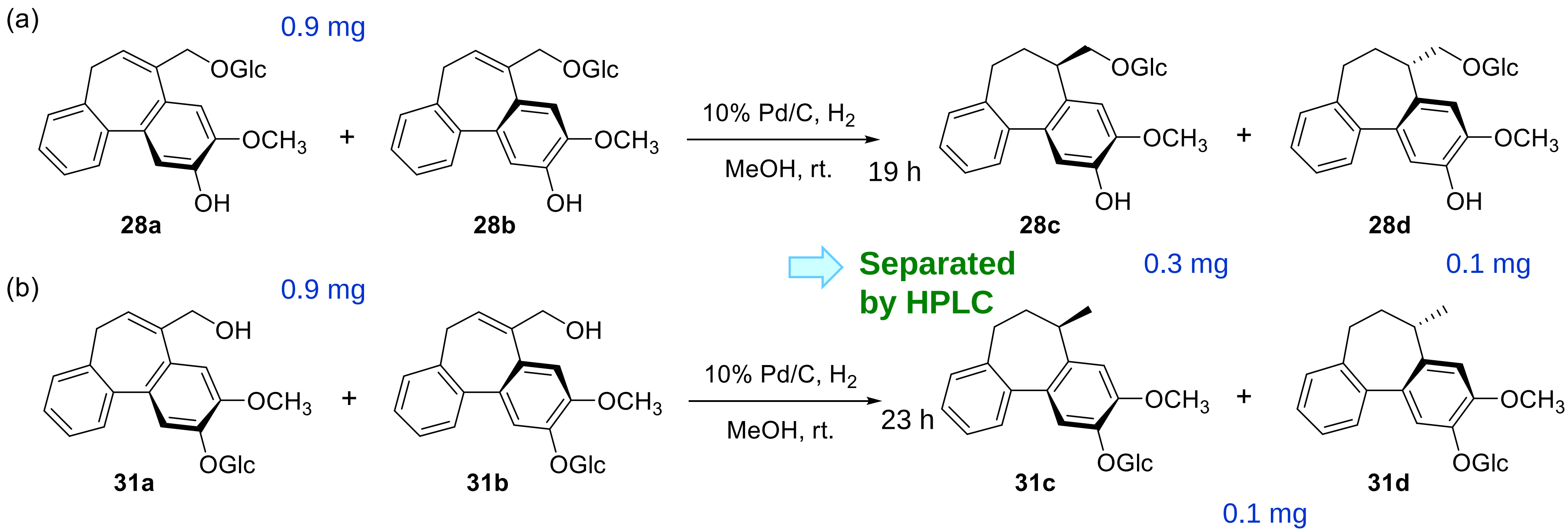
	R_1	R_2	R_3	ratio		R_1	R_2	R_3
28a	H	H	Glc	1.1:1	**28a**	H	H	Glc
29a	H	OCH_3	Glc	1:0.9	**29b**	H	OCH_3	Glc
30a	H	OH	Glc	1.1:1	**30b**	H	OH	Glc
31a	Glc	H	H	1:0.7	**31b**	Glc	H	H
32a	Glc	OCH_3	CH_3	2:1	**32b**	Glc	OCH_3	CH_3
33a	Rha	OH	H	1.1:1	**33b**	Rha	OH	H

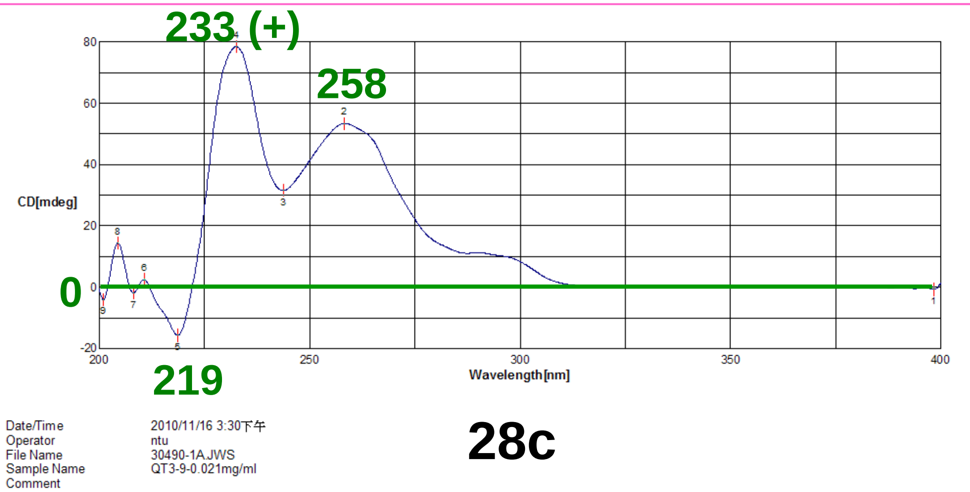


To confirm 1. diastereomeric property
2. absolute configuration

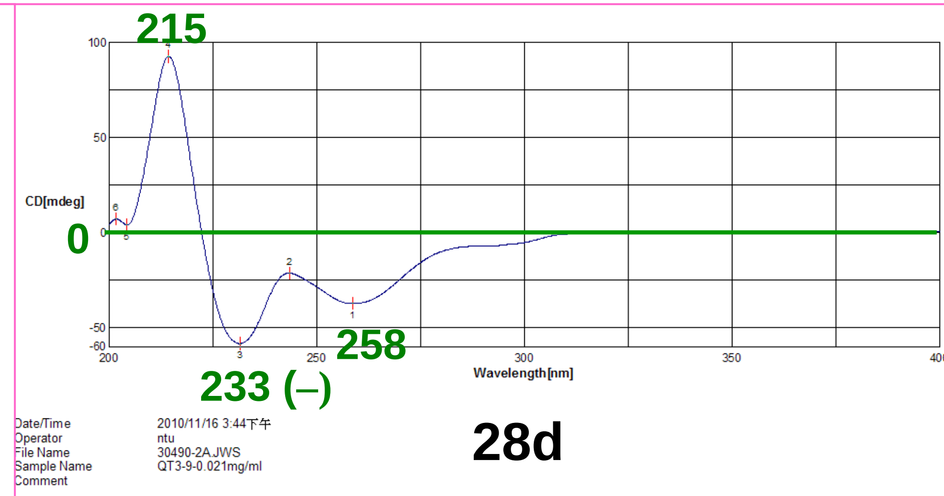
Hydrogenation of Mixture of 28a and 28b, and 31a and 31b



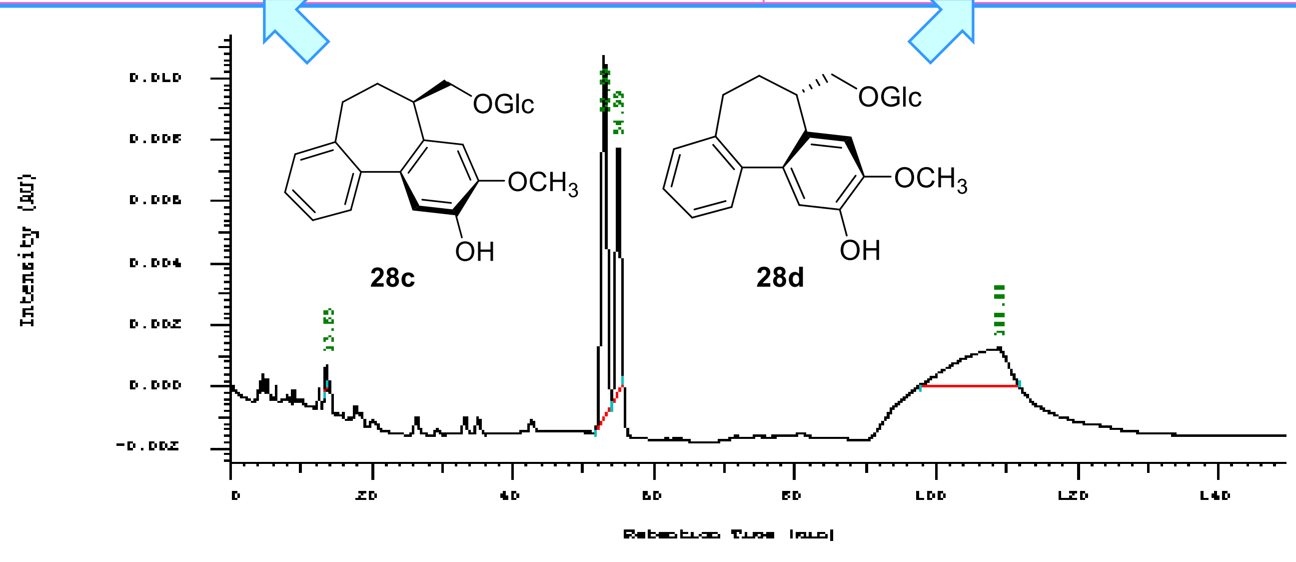
Axial chirality → cotton effect



28c



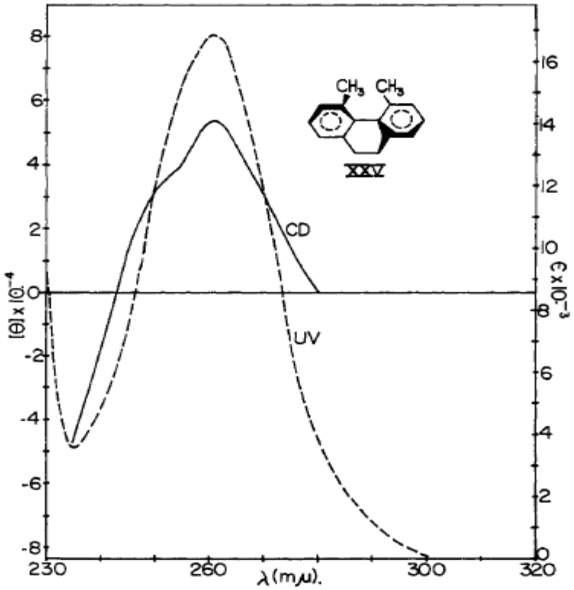
28d



Column Type: Phenomen-Prodigy-5um-ODS3-100A
Method Description: p.3042451 Fr.1->hydrogenation 1 mg/mL 1 uL, MeCN-H2O, 19%, 0.5 mL/min, UV 230 nm Phenomenex@Prodigy, 5u, , 250*4.6 mm

Mislow *et al.*

J. Am. Chem. Soc. **1963**, *85*, 1342−1349



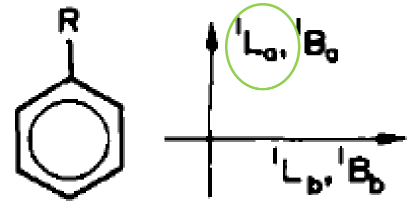
Without polar group substituted

→ *M* helicity

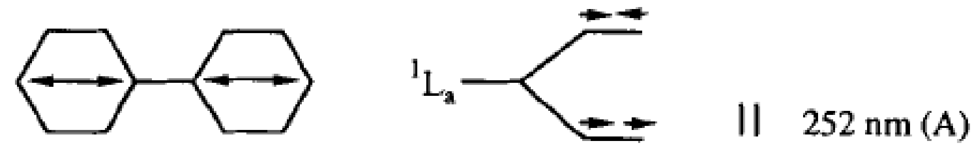
→ CE (+) (235—250 nm, A band)

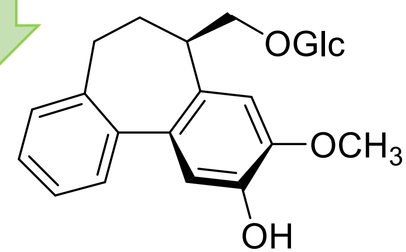
Tetrahedron. **1977**, *99*, 2320-2313

J. Am. Chem. Soc. **1977**, *99*, 6861−6869



^1L_a transitions: Polarization along the axis

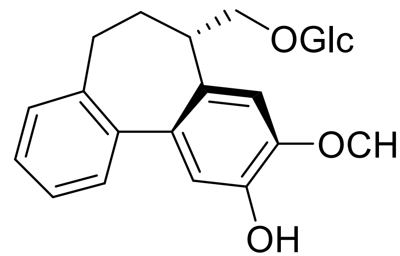




28c

CE (+)

→ *M* helicity → a*R*



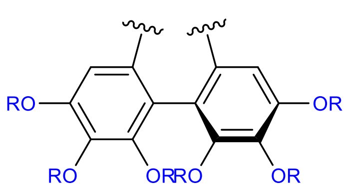
28d

CE (–)

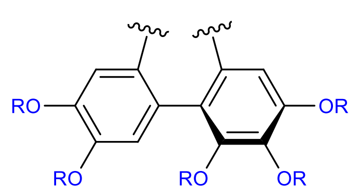
→ *P* helicity → a*S*

X-ray crystallography and **CD**

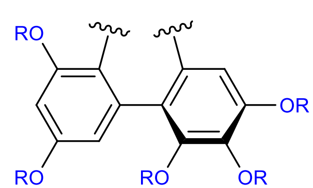
M helicity → CE (+)



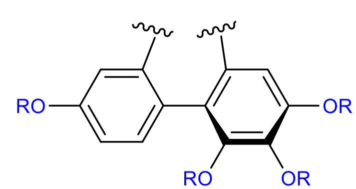
2,3,4- and 2',3',4'-



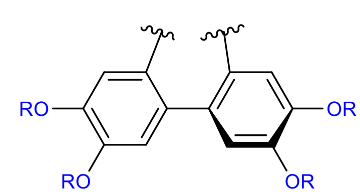
2,3,4- and 3',4'-



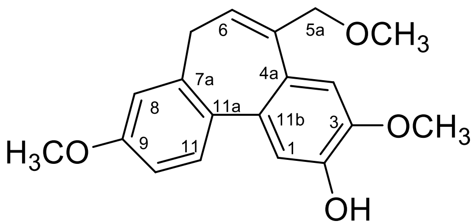
2,3,4- and 2',5'-



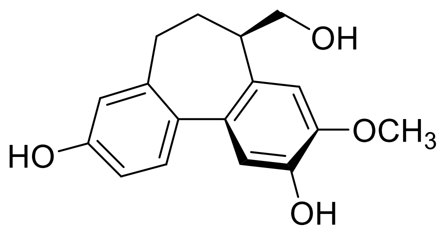
2,3,4- and 4'-



3,4- and 3',4'-



27



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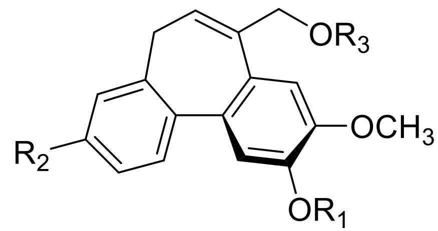
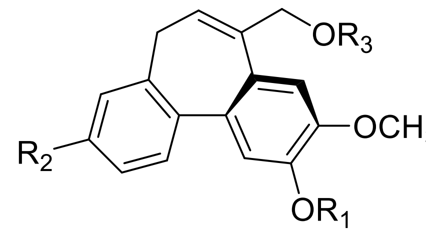
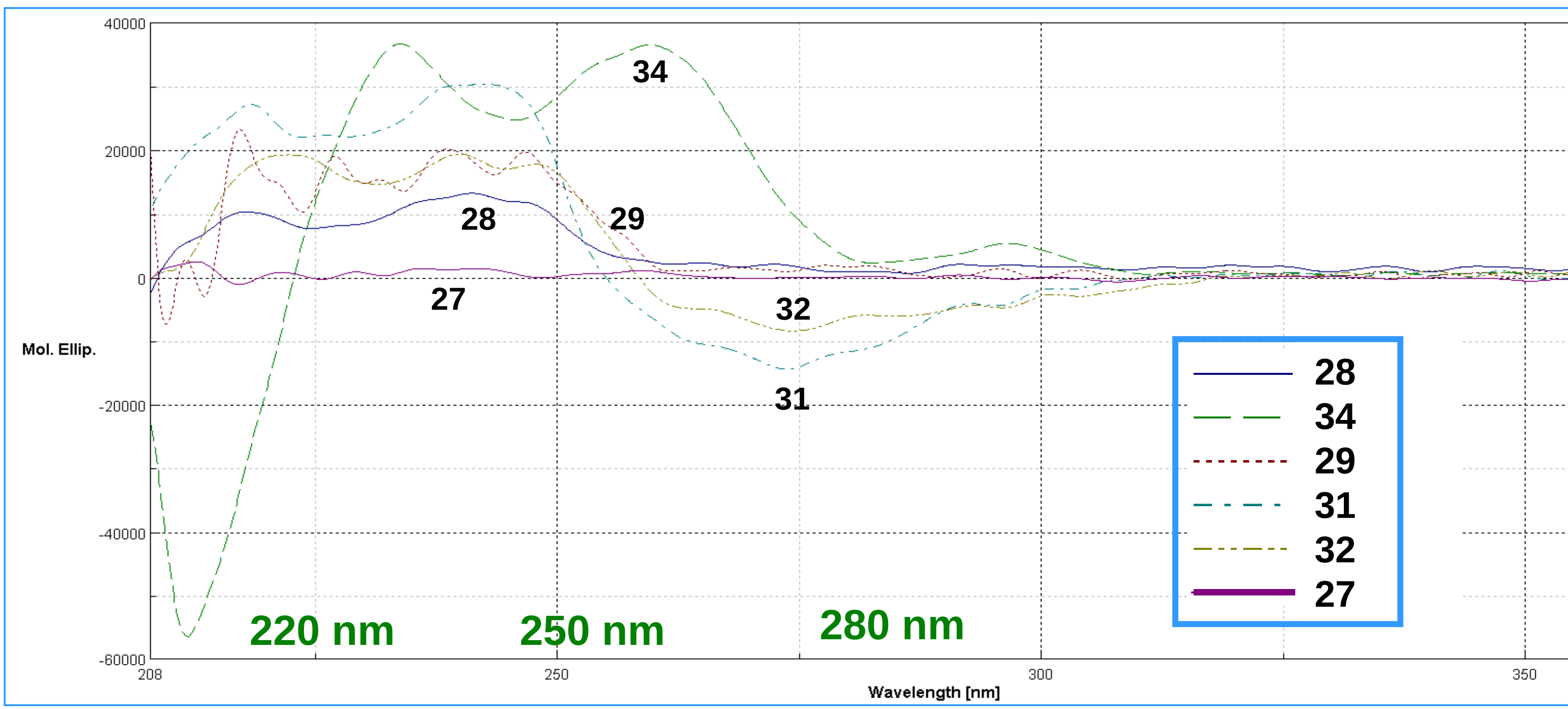




	R_1	R_2	R_3	ratio		R_1	R_2	R_3
28a	H	H	Glc	1.1:1	**28a**	H	H	Glc
29a	H	OCH_3	Glc	1:0.9	**29b**	H	OCH_3	Glc
30a	H	OH	Glc	1.1:1	**30b**	H	OH	Glc
31a	Glc	H	H	1:0.7	**31b**	Glc	H	H
32a	Glc	OCH_3	CH_3	2:1	**32b**	Glc	OCH_3	CH_3
33a	Rha	OH	H	1.1:1	**33b**	Rha	OH	H



Tips on Structure Elucidation by NMR Spectroscopy

1. Molecular Formula

- +m/z = 285 (Mw = 284)
- Numbers of protons in ^{1}H spectrum
- Numbers of carbons and multiplicity in ^{13}C spectrum
- Propose number of heteroatoms

2. **Table of 1D (including coupling patterns) and 2D signals**

3. Characterize "**moieties**" based on **coupling patterns** and **2D spectra**

4. **Link moieties** together based on **HMBC**, **COSY** and **TOCSY**..etc.

5. Identify **Relative Stereochemistry** based on **NOESY**
(**Absolute stereochemistry** can be further verified through other methods such as circular dichroism, chemical synthesis, Mosher's method, X-ray crystallography…etc.)


Thank You


Practice!