

Chasing molecules– a structure elucidation journey

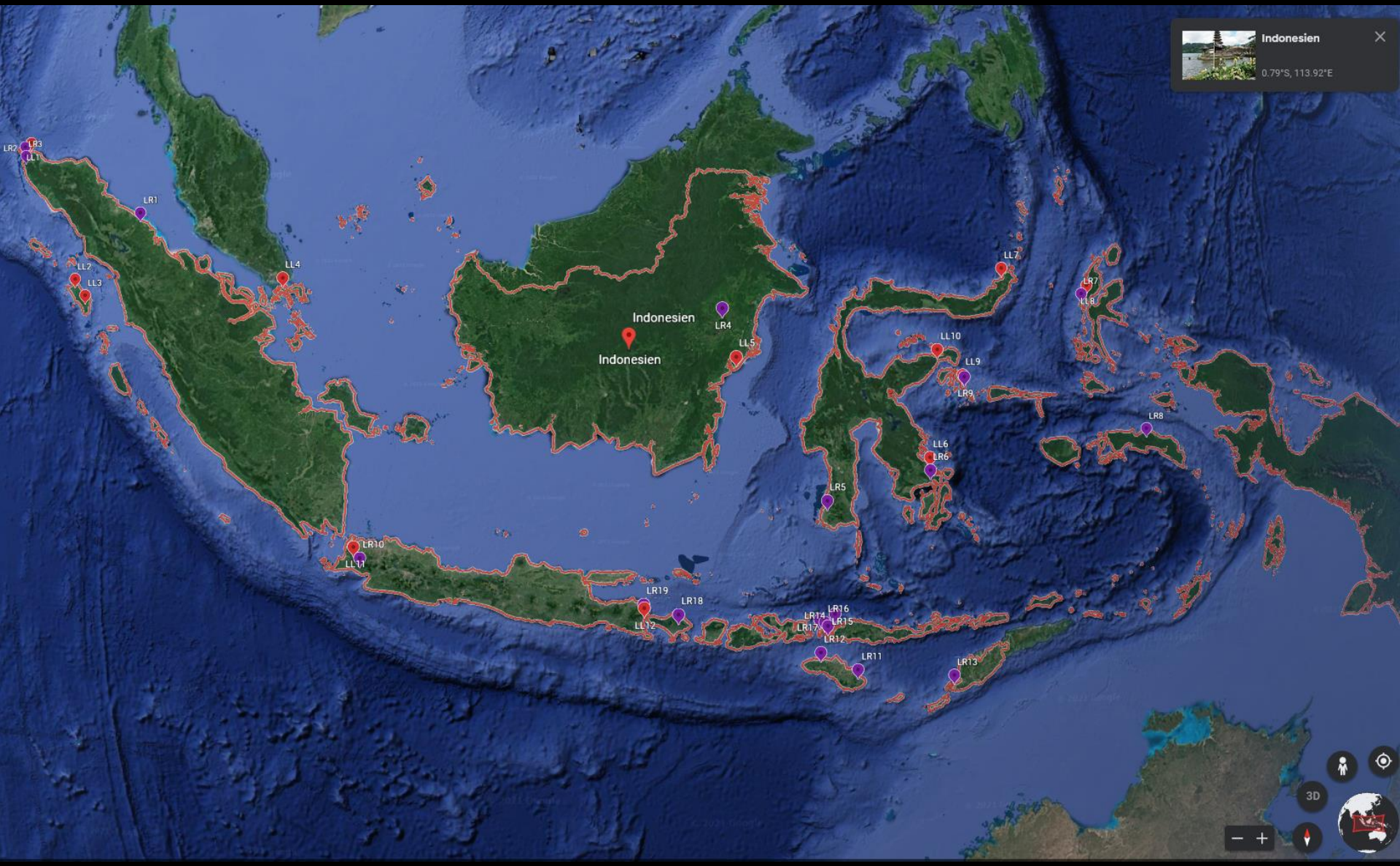
Jonas Kappen

Leibniz Institute of Plant Biochemistry 03.07.2023

Targetplant: mangrove






Indonesien
0.79°S, 113.92°E
Indonesien
Indonesien
LR1
LR2
LR3
LL1
LL2
LL3
LL4
LR4
LL5
LL7
LR7
LL8
LL10
LL9
LR9
LR8
LL6
LR6
LR5
LR10
LL11
LR19
LR18
LL12
LR16
LR14
LR15
LR17
LR12
LR11
LR13
3D

Extraction





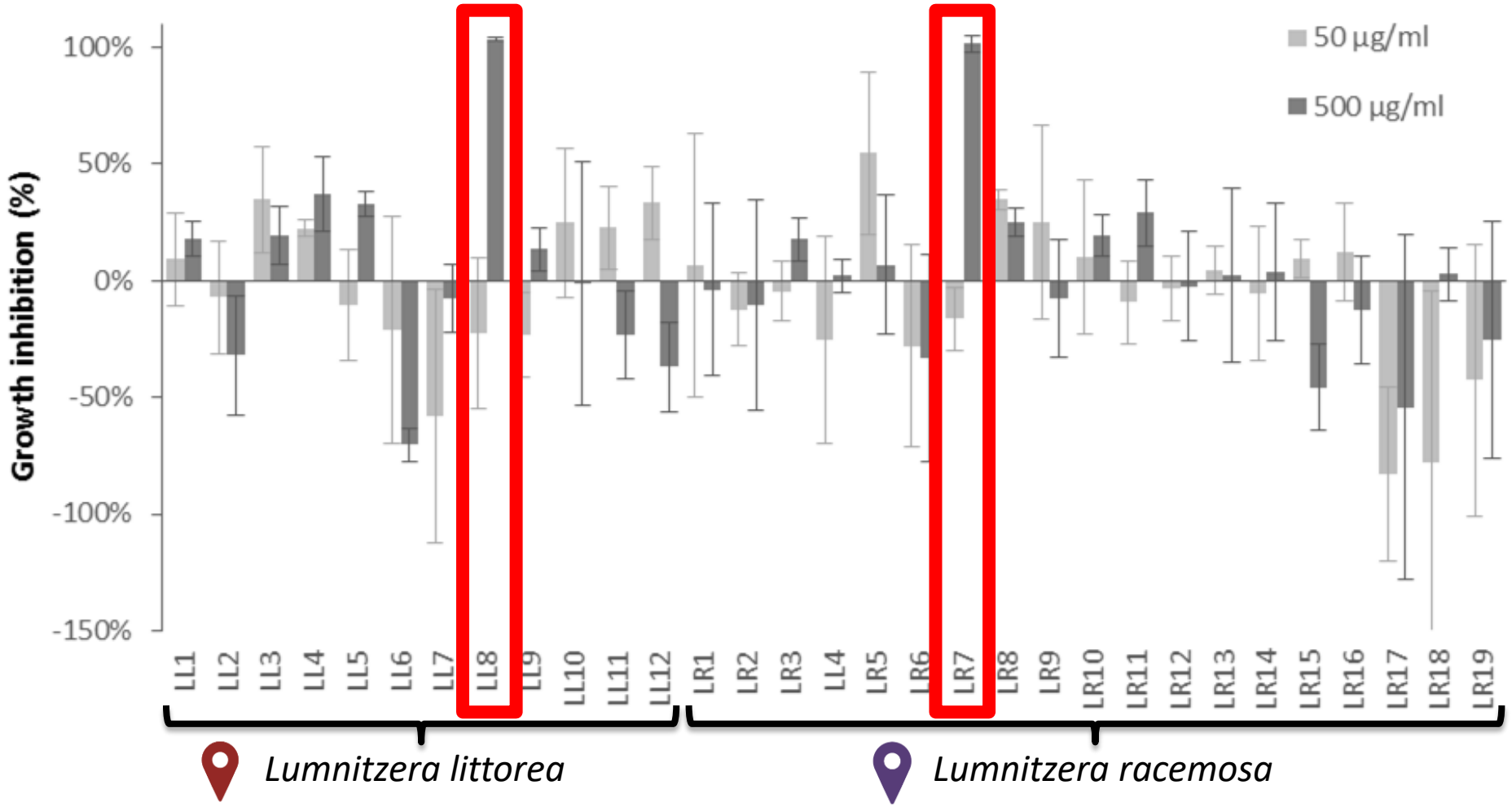
Antibacterial assay



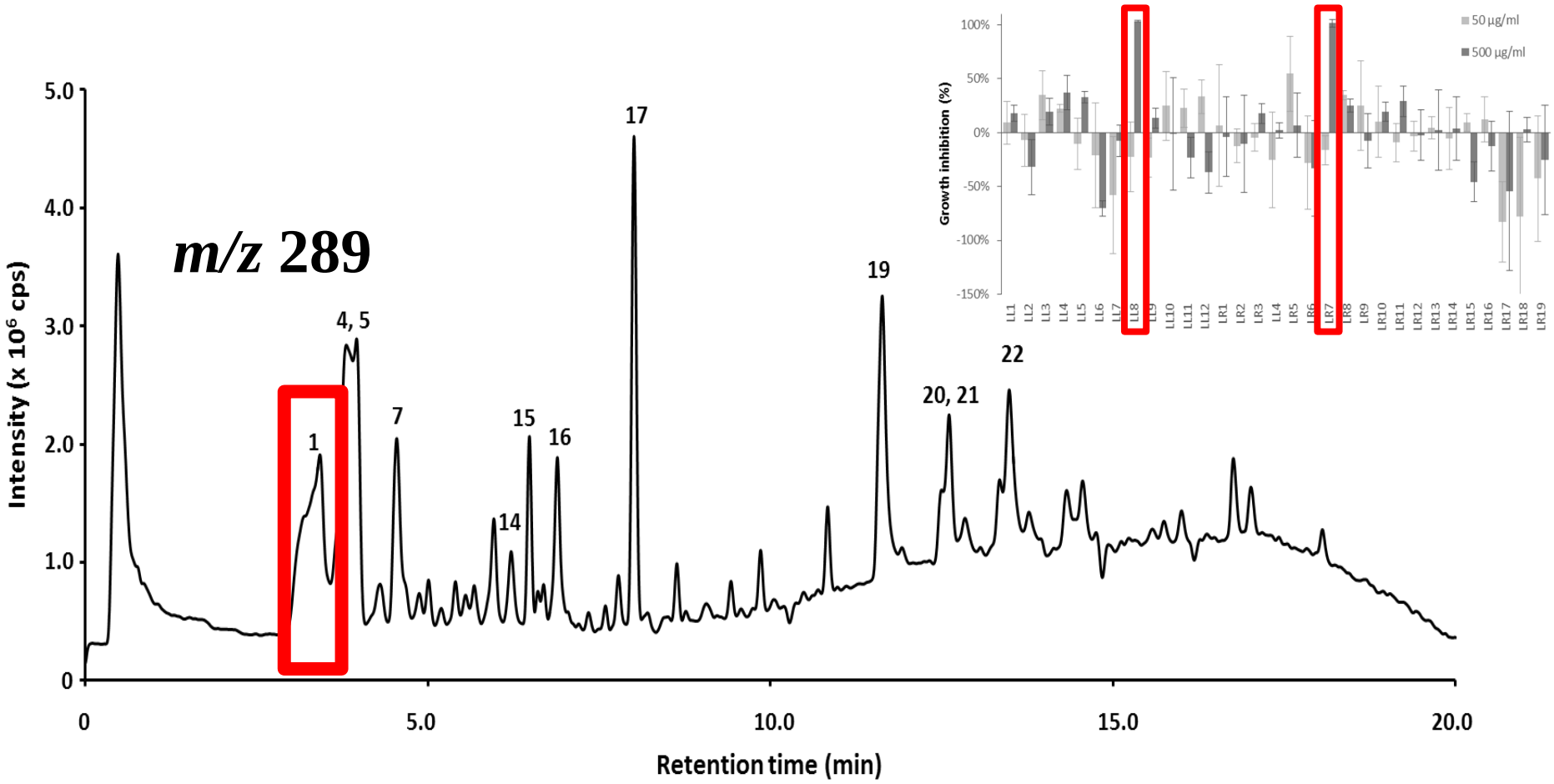


High Resolution MS

Antibacterial assay – *Bacillus subtilis*

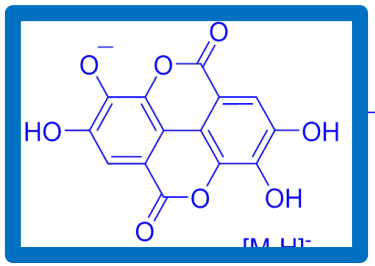


HRMS - Screening



Total ion chromatogram (TIC) acquired for the antibacterial sample LR7

Fragmentation



[M-H]⁻

-H → $C_{14}H_4O_8^{\bullet-}$ **7**: *m/z* 299.9906 → (-H, -CO) $C_{13}H_3O_7^-$ **7**: *m/z* 270.9868

$C_{14}H_5O_8^-$ **7**: *m/z* 300.9970 → (-OH) $C_{14}H_4O_7^{\bullet-}$ **7**: *m/z* 283.9956

[M-H]⁻ $C_{13}H_5O_8^-$ **1**: *m/z* 288.999

-CO_2 → $C_{12}H_5O_6^-$ **1**: *m/z* 245.0082, **7**: *m/z* 245.0077

$C_{12}H_5O_6^-$ → (-OH) $C_{12}H_4O_5^{\bullet-}$ **1**: *m/z* 228.0054, **7**: *m/z* 228.0054

-2CO → $C_{11}H_5O_5^-$ **1**: *m/z* 217.0133, **7**: *m/z* 217.0128

$C_{11}H_5O_5^-$ → (-CO_2) $C_{10}H_5O_3^-$ **1**: *m/z* 173.0236, **7**: *m/z* 173.0241

$C_{11}H_5O_5^-$ → (-OH) $C_{11}H_4O_4^{\bullet-}$ **1**: *m/z* 200.0104, **7**: *m/z* 200.0114

-CO → $C_{10}H_5O_4^-$ **1**: *m/z* 189.0183, **7**: *m/z* 189.0182

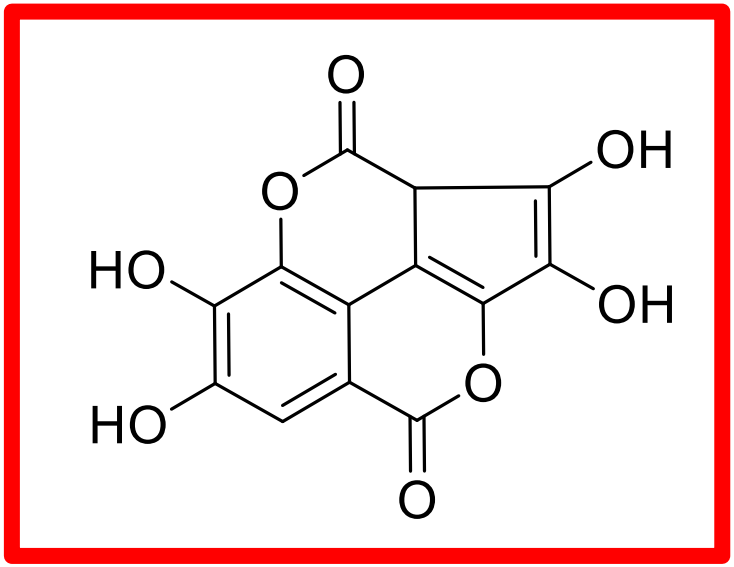
$C_{10}H_5O_4^-$ → (-CO_2) $C_9H_5O_2^-$ **1**: *m/z* 145.0286, **7**: *m/z* 145.0297

-CO → $C_9H_5O_3^-$ **1**: *m/z* 161.0235, **7**: *m/z* 161.0237

$C_9H_5O_3^-$ → (-CO_2) $C_8H_5O^-$ **1**: *m/z* 117.0337, **7**: *m/z* 117.0344

$C_9H_5O_3^-$ → (-CO) $C_8H_5O_2^-$ **1**: *m/z* 133.0285, **7**: *m/z*133.0287

$C_8H_5O_2^-$ → (-CO) $C_7H_5O^-$ **1**: *m/z* 105.0335, **7**: *m/z* 105.0341

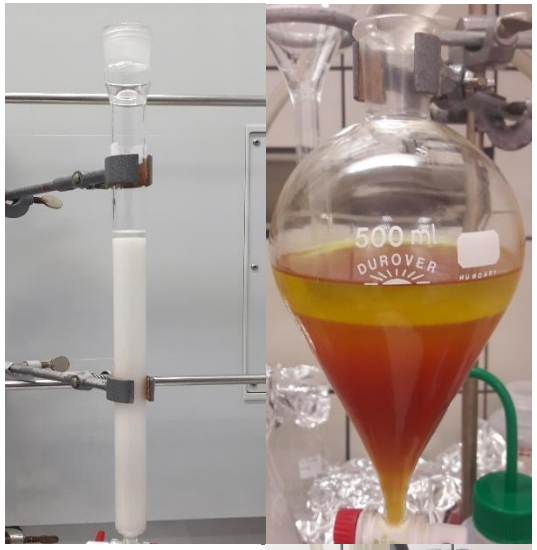


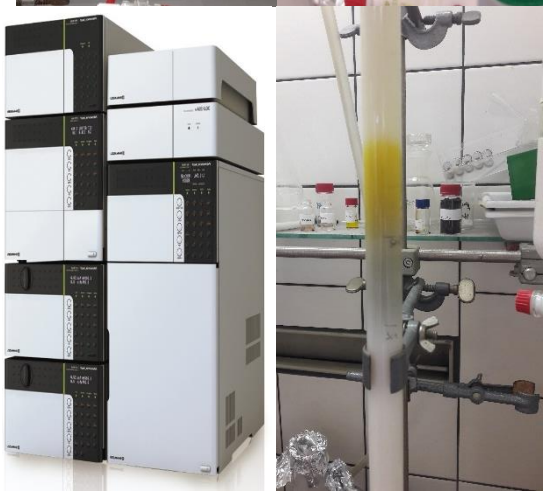
First proposal

Isolation







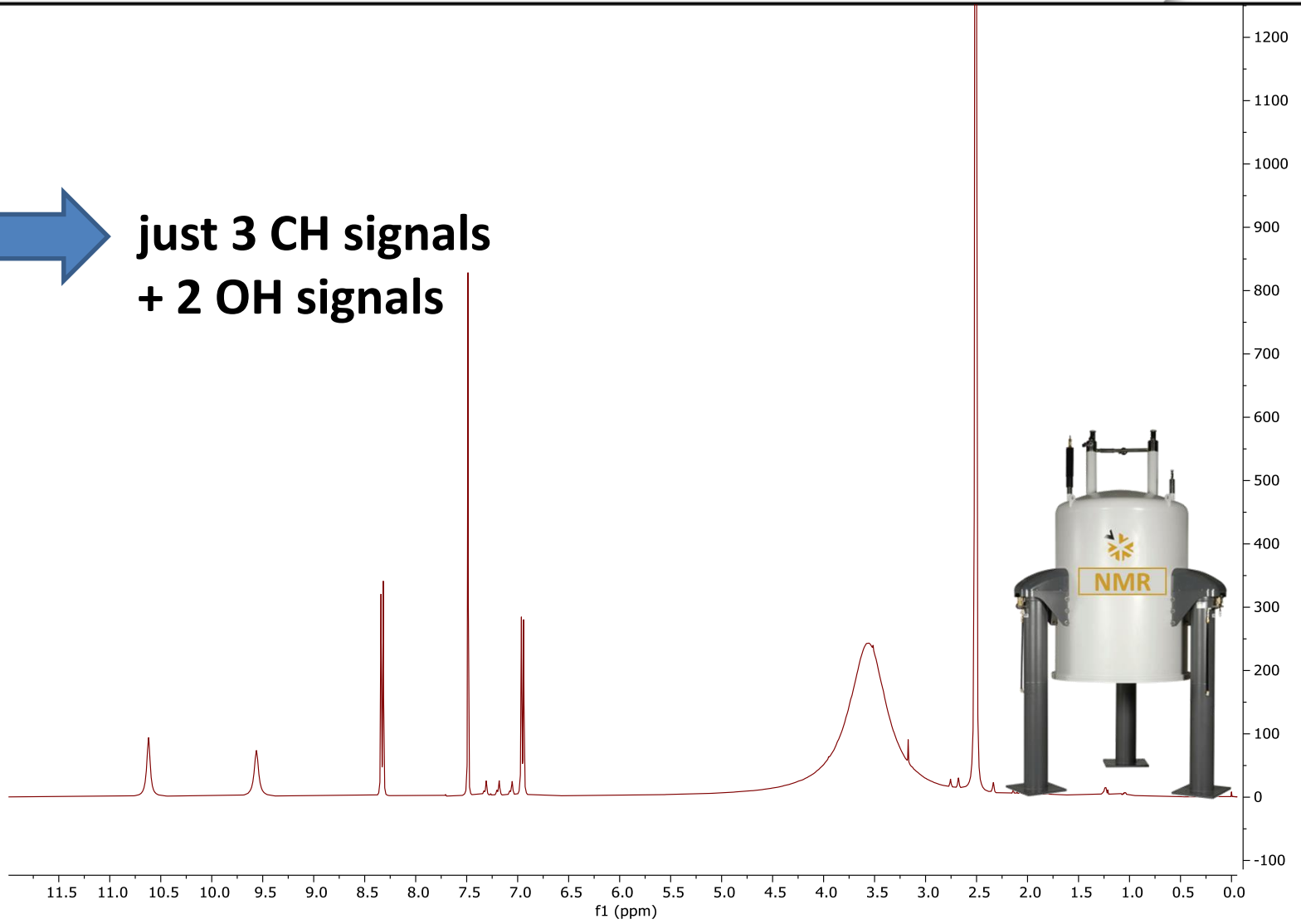






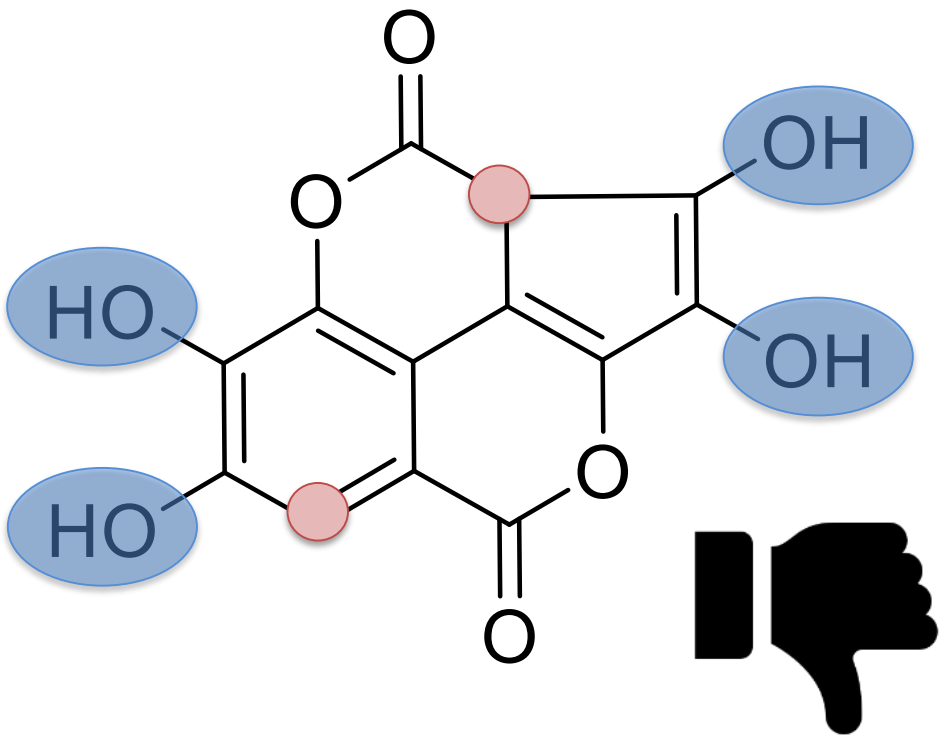
^{1}H in DMSO-d6, 400 MHz – $C_{13}H_6O_8$

just 3 CH signals
+ 2 OH signals



NMR evaluation

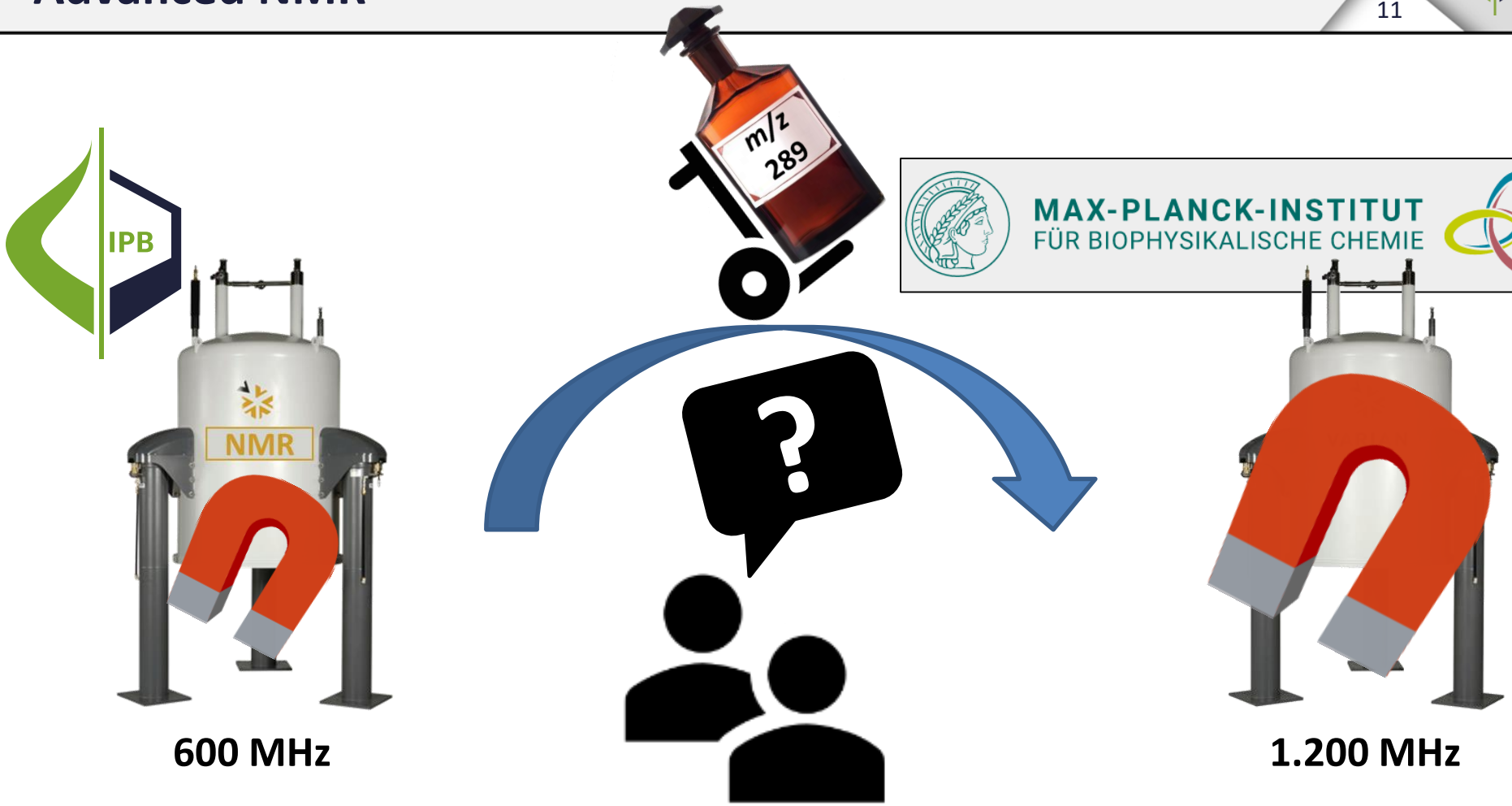
δ_C DMSO	δ_H (J in Hz) DMSO	HMBC DMSO
158.2, C		
145.8, C		
107.3, CH	7.49, s	145.8, 125.1, 158.3, 108.1, 127.9, 160.0
125.1, C		
158.3, C		
142.8, C		
132.7, C		
150.8, C		
113.2, CH	6.95, d (9.0)	142.8, 132.7, 150.8, 108.1
117.9, CH	8.33, d (9.0)	125.1, 142.8, 132.7, 150.8, 113.2, 127.9
108.1, C		
127.9, C		
160.0, C		
OH	9.56, s	
OH	10.62, s	



First proposal

Just 2 CH but 4 OH!

Advanced NMR



Advanced NMR





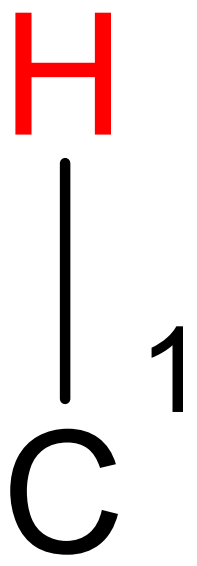
600 MHz



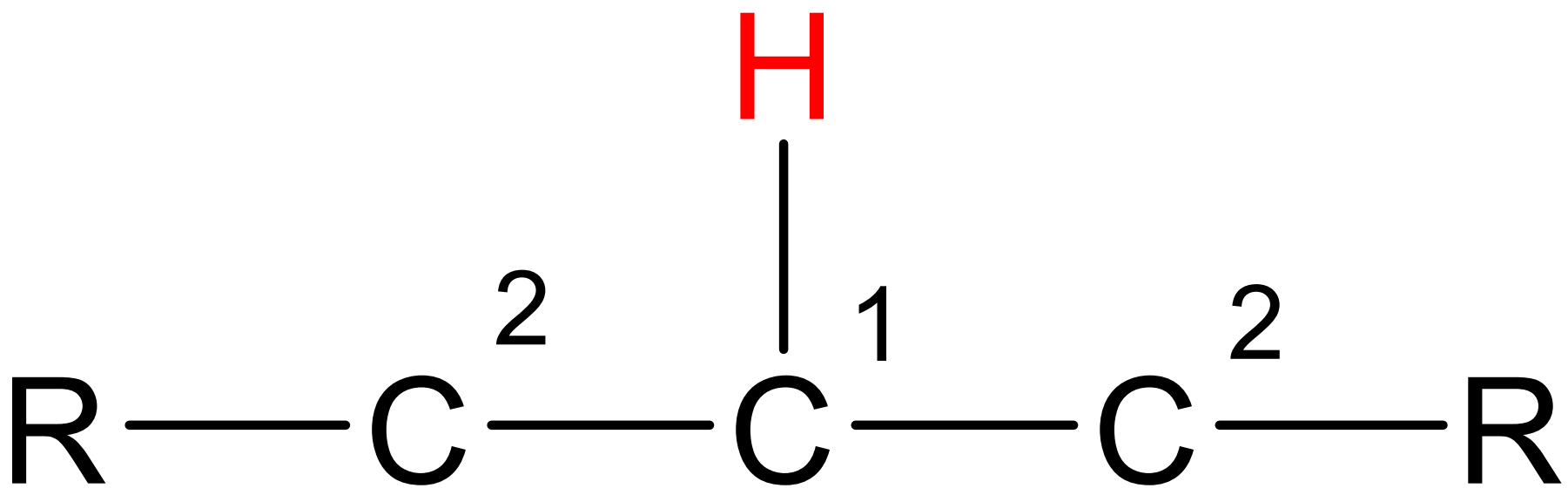


1.200 MHz

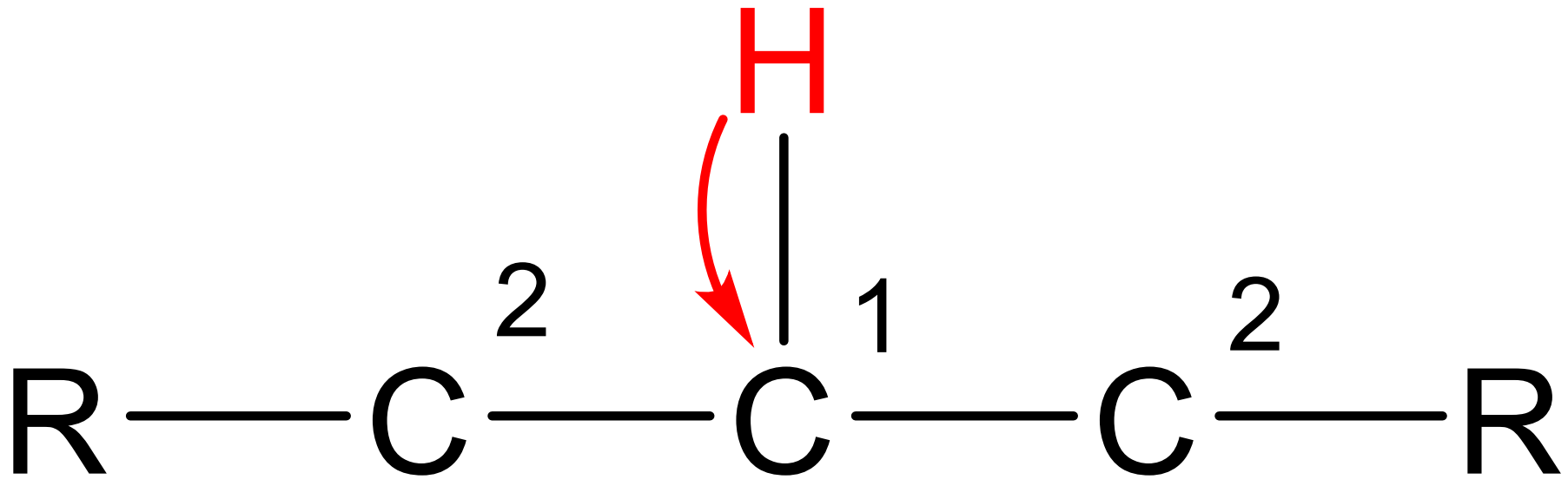
1,1 ADEQUATE in DMSO – $C_{13}H_6O_8$



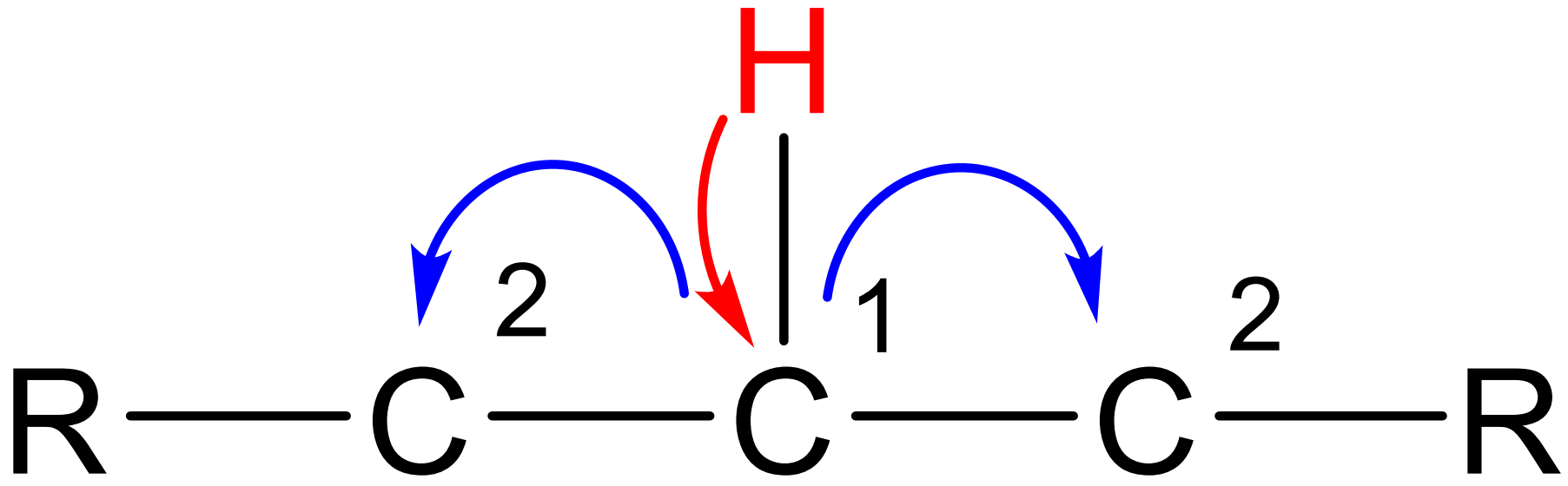
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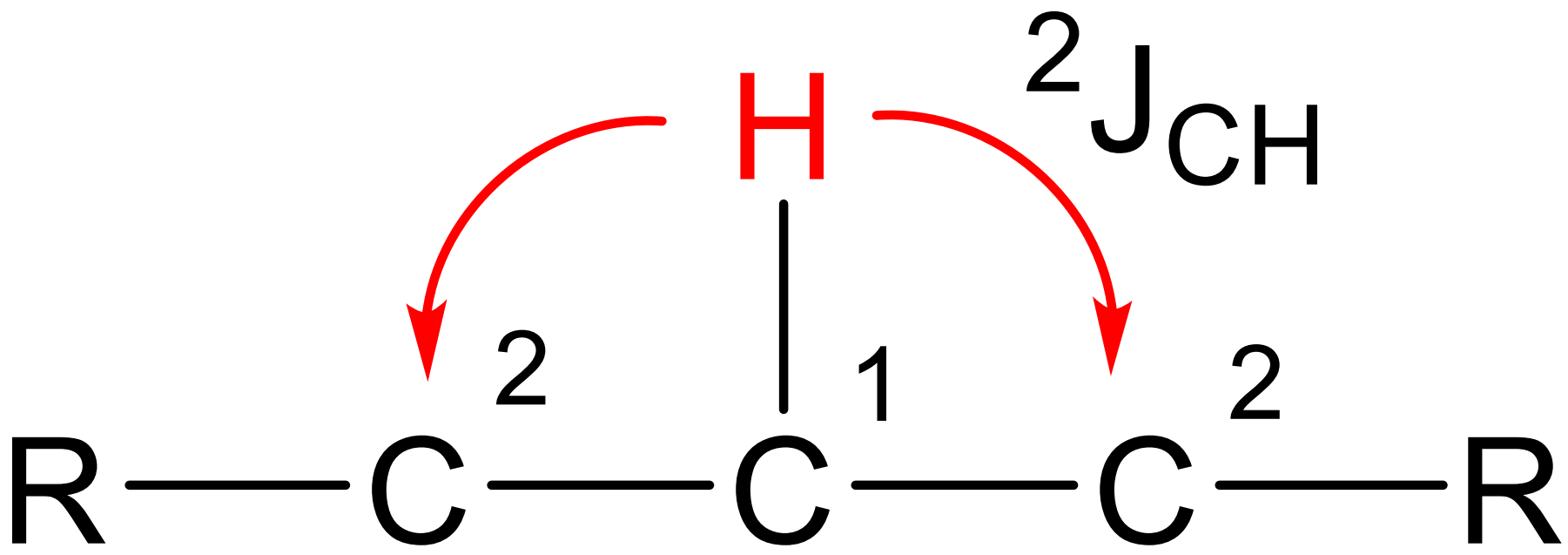
1,1 ADEQUATE in DMSO – $C_{13}H_6O_8$



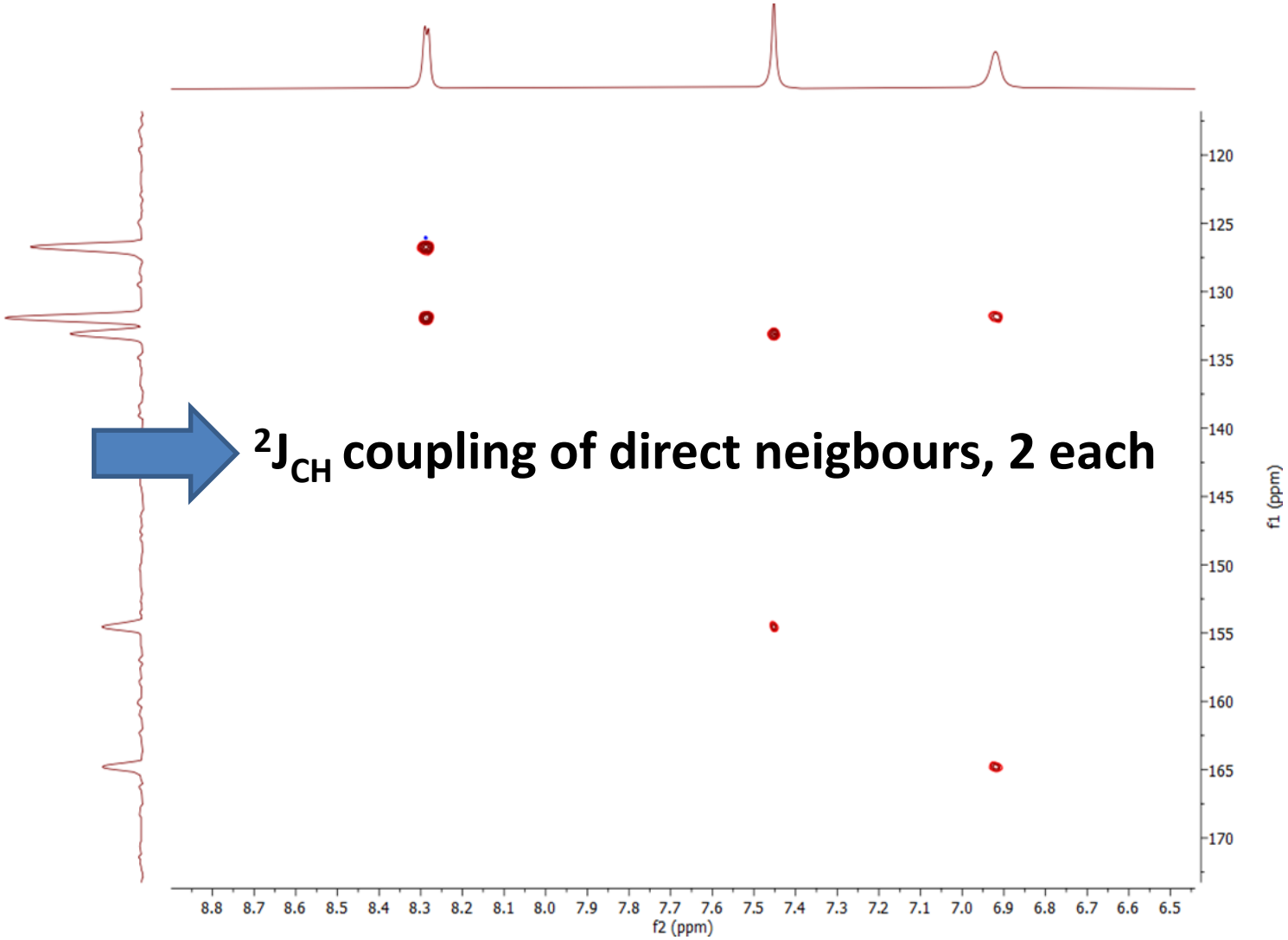
1,1 ADEQUATE in DMSO – $C_{13}H_6O_8$



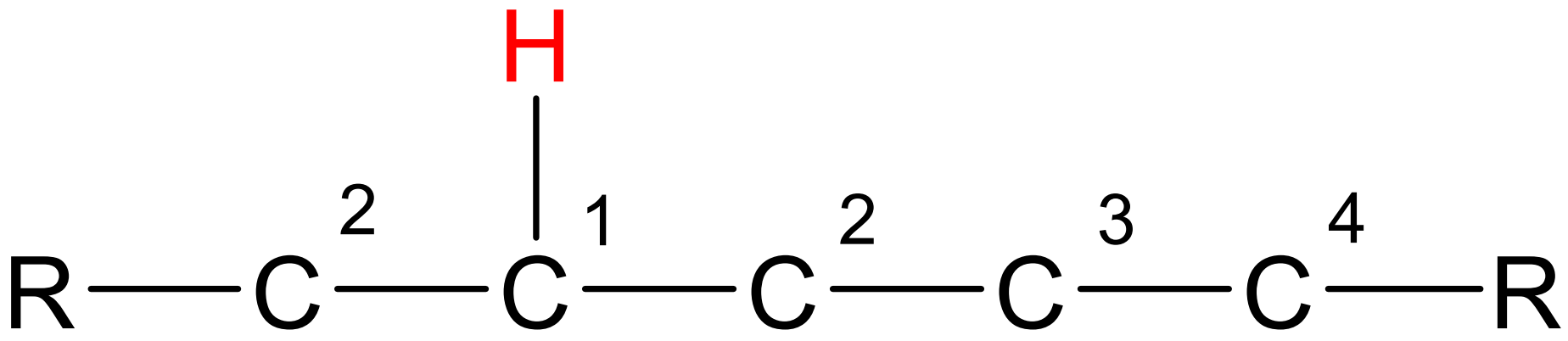
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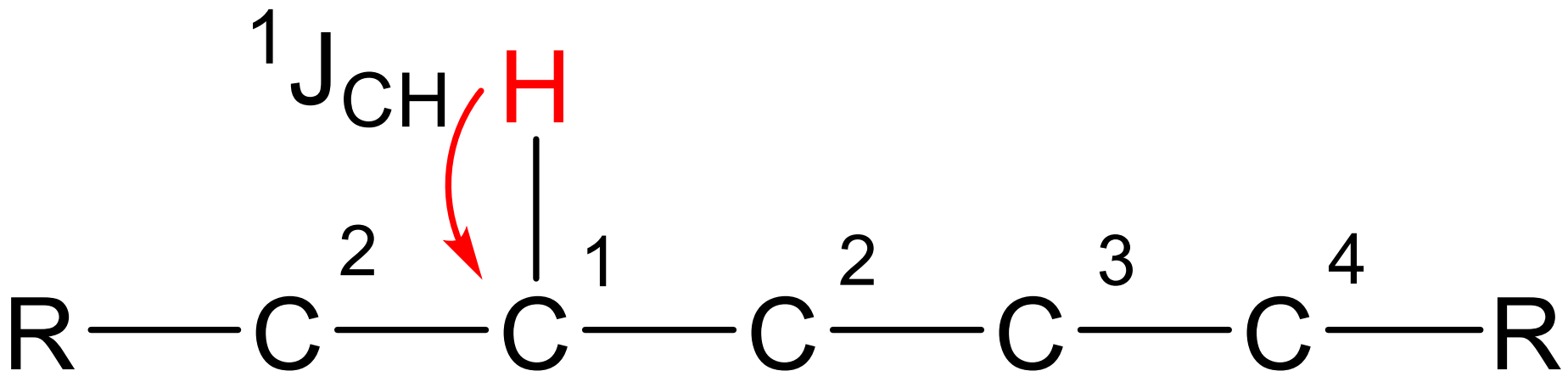
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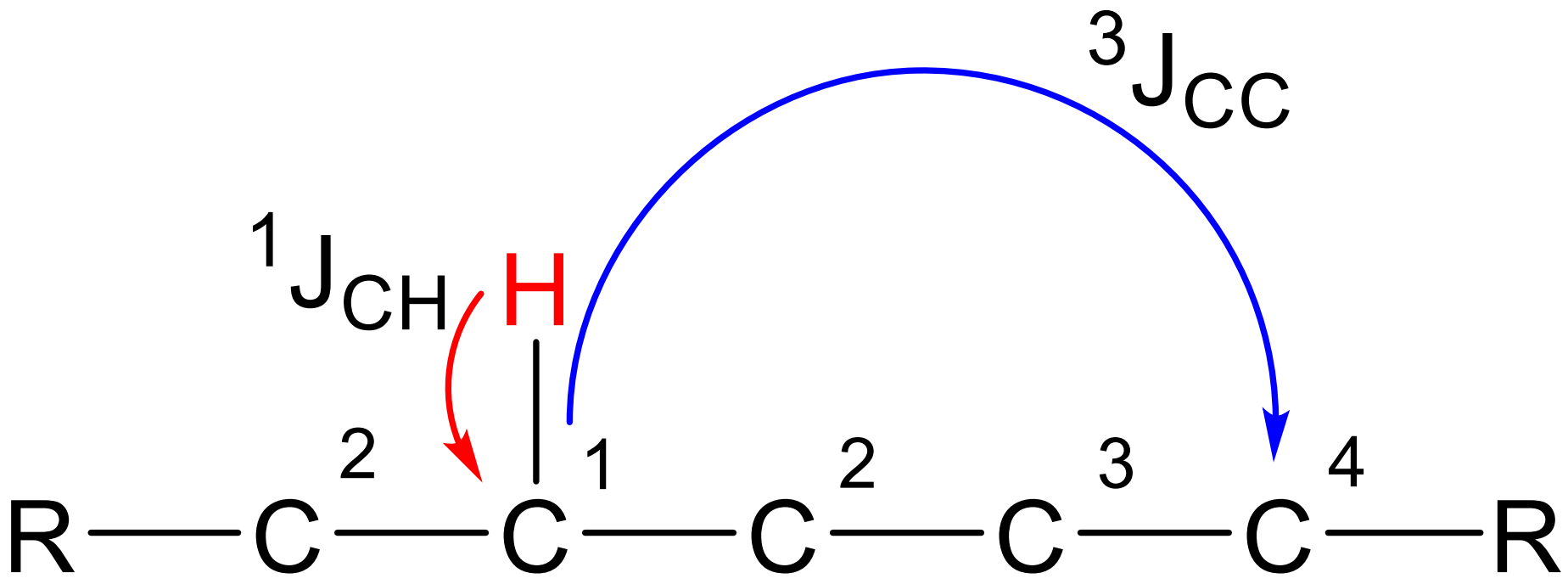
1,n ADEQUATE in DMSO – $C_{13}H_6O_8$



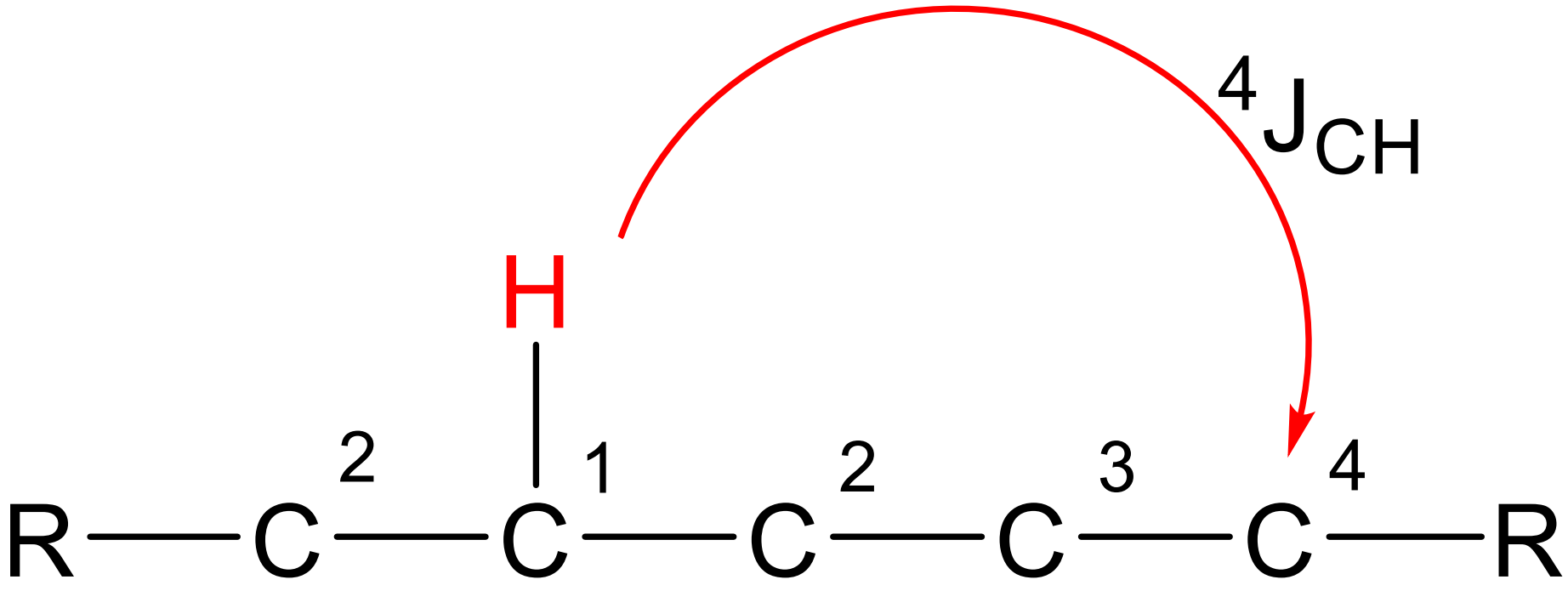
1,n ADEQUATE in DMSO – $C_{13}H_6O_8$



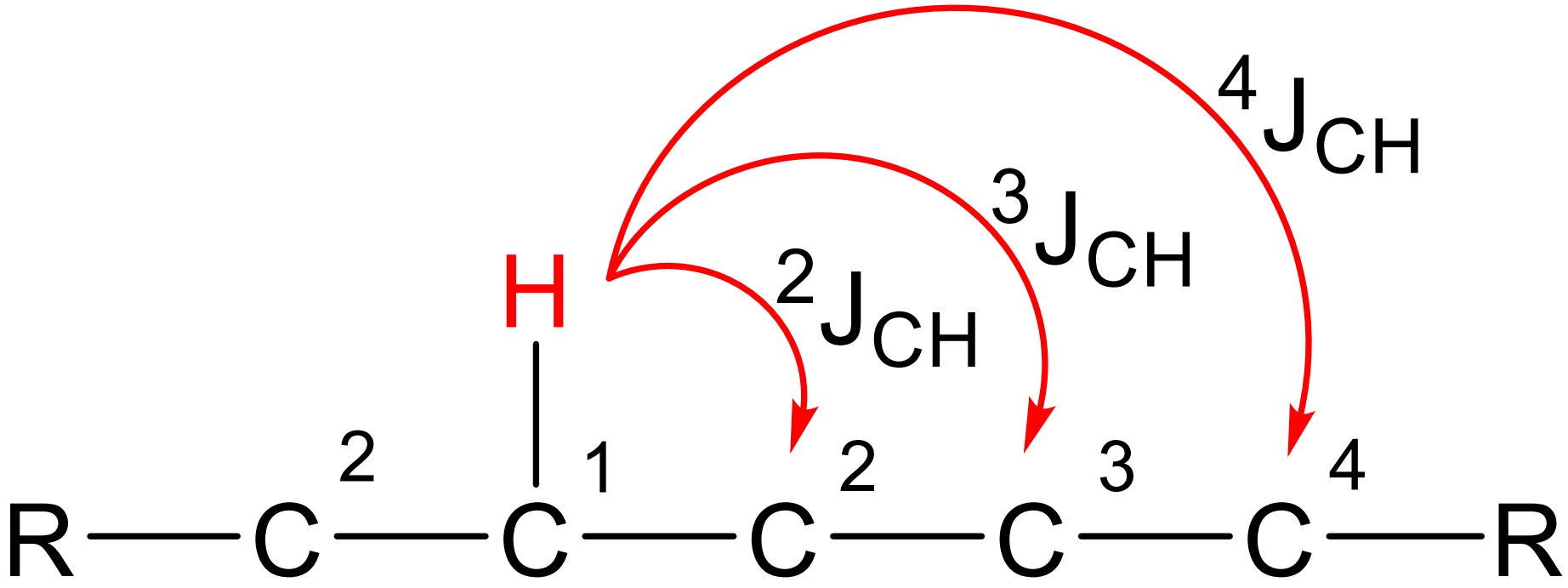
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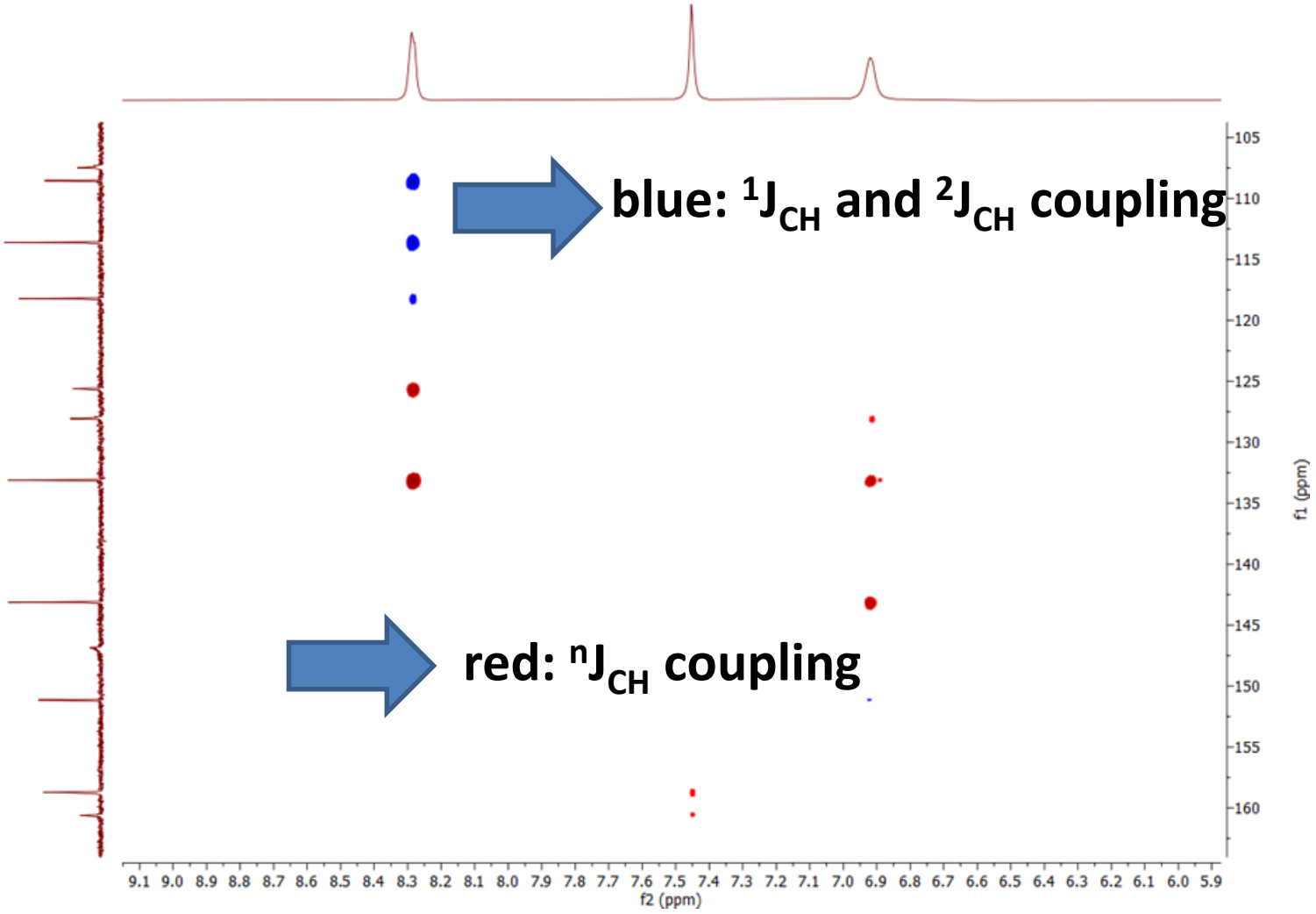
1,n ADEQUATE in DMSO – $C_{13}H_6O_8$



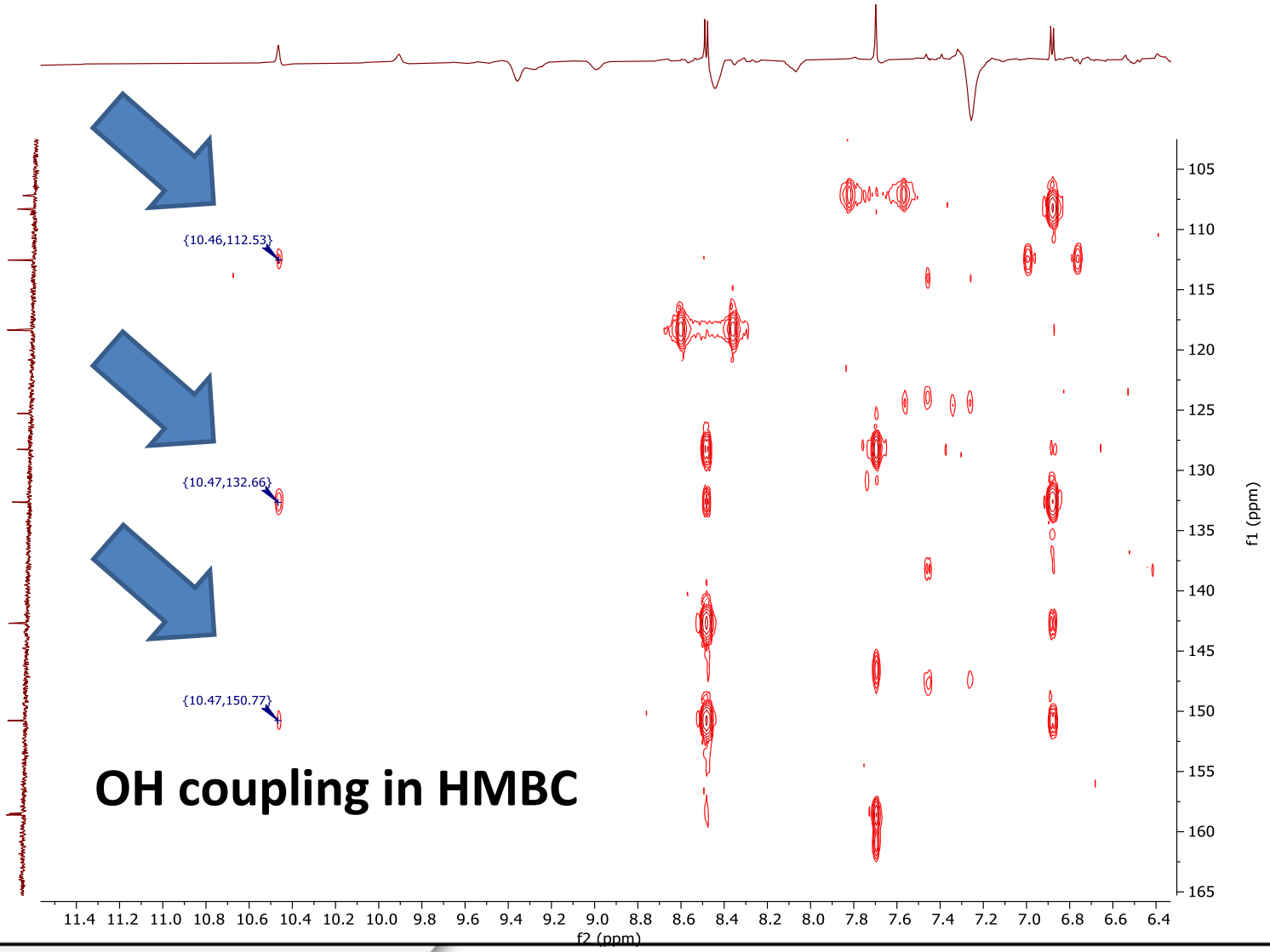
1,n ADEQUATE in DMSO – $C_{13}H_6O_8$



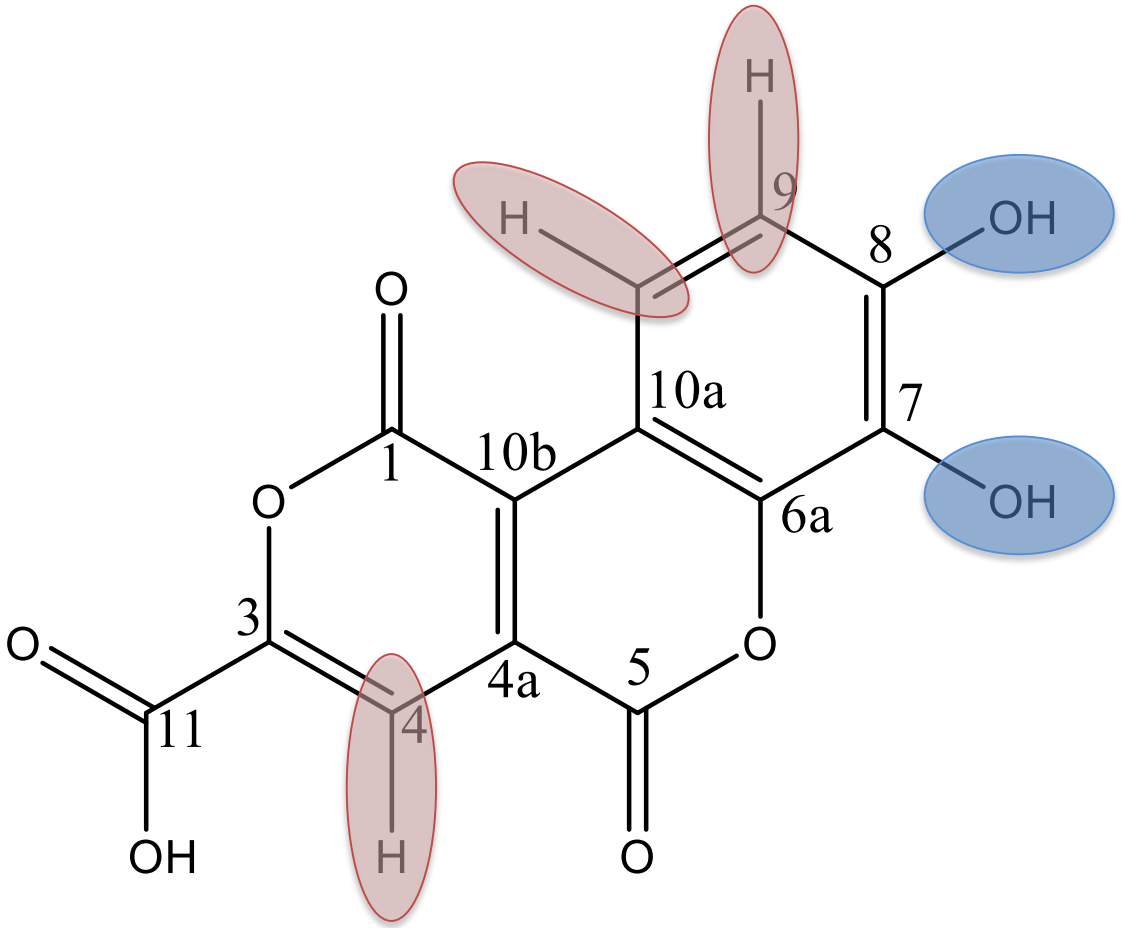
1,n ADEQUATE in DMSO – $C_{13}H_6O_8$



HMBC in CD_3OH – $C_{13}H_6O_8$

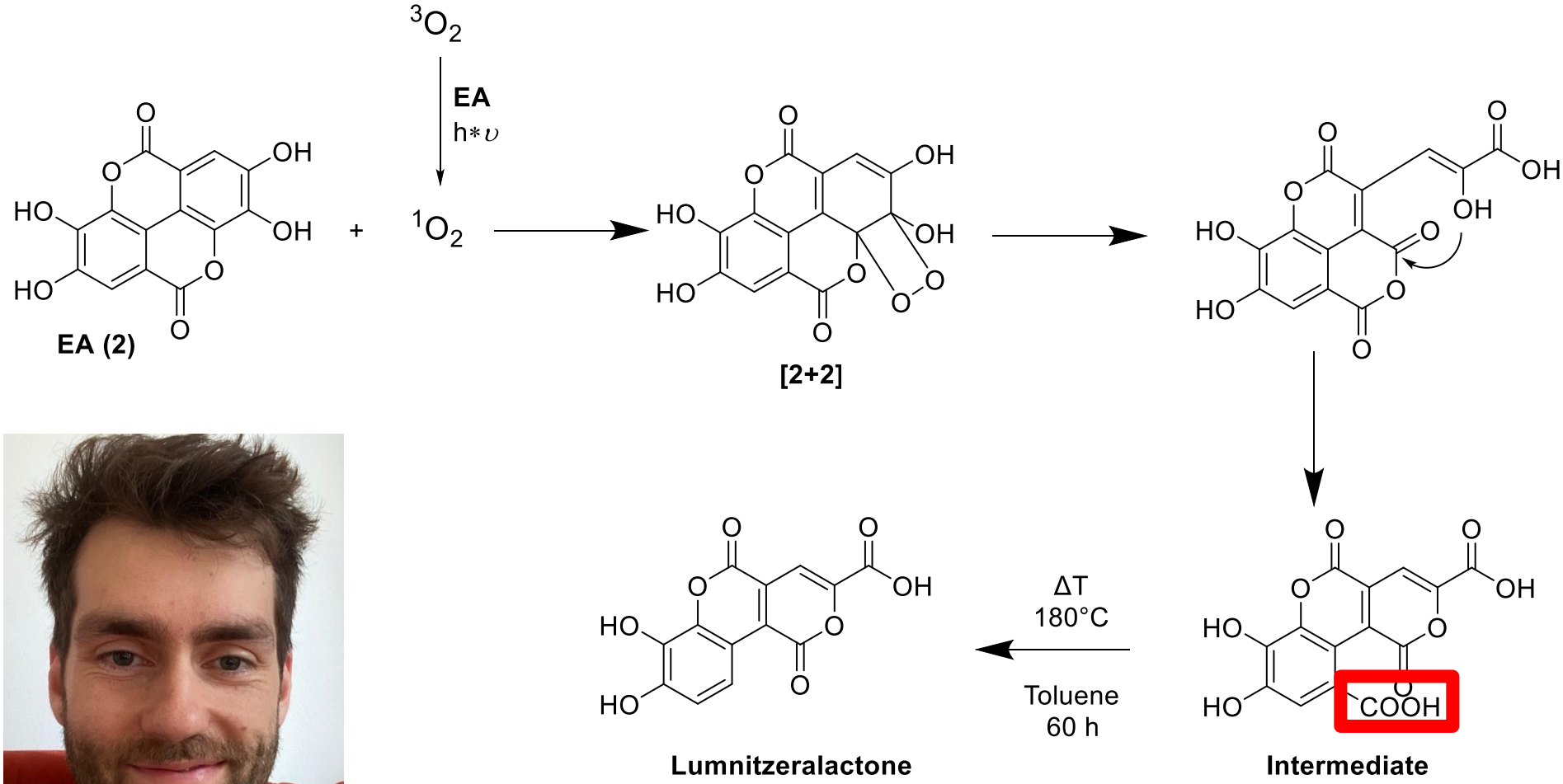


Structur proposal Lumnitzeralacton



3 CH (two as neighbours) and 2 OH!

Synthesis


$^3O_2$
EA
h∗υ
$^1O_2$
EA (2)
[2+2]
ΔT
180°C
Toluene
60 h
Lumnitzeralactone
Intermediate
COOH

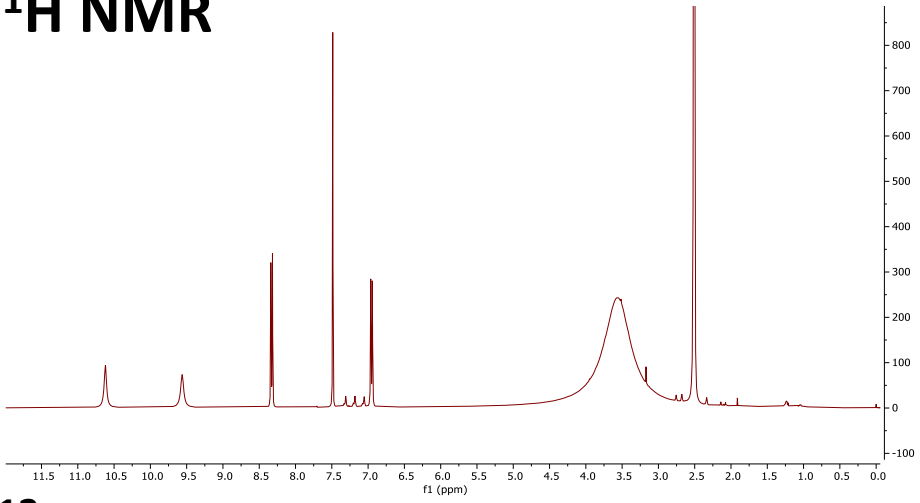
Tristan Fuchs

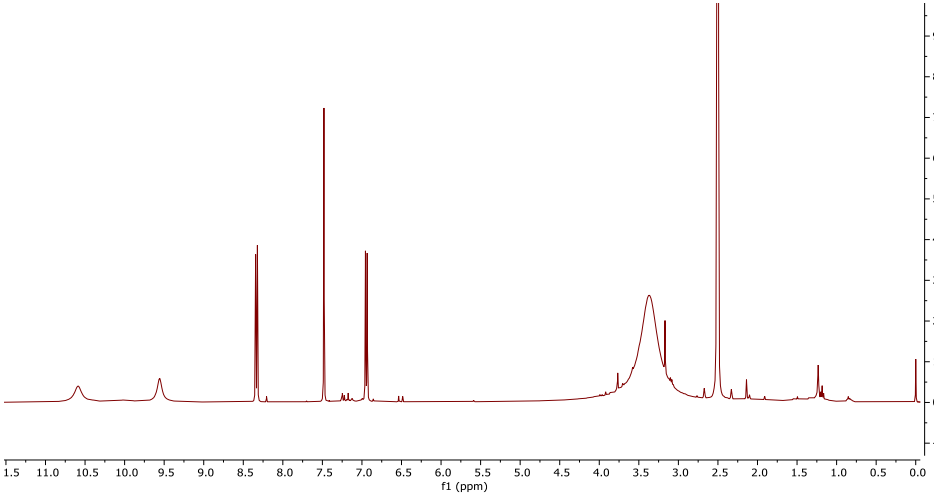
Comparison

Natural product

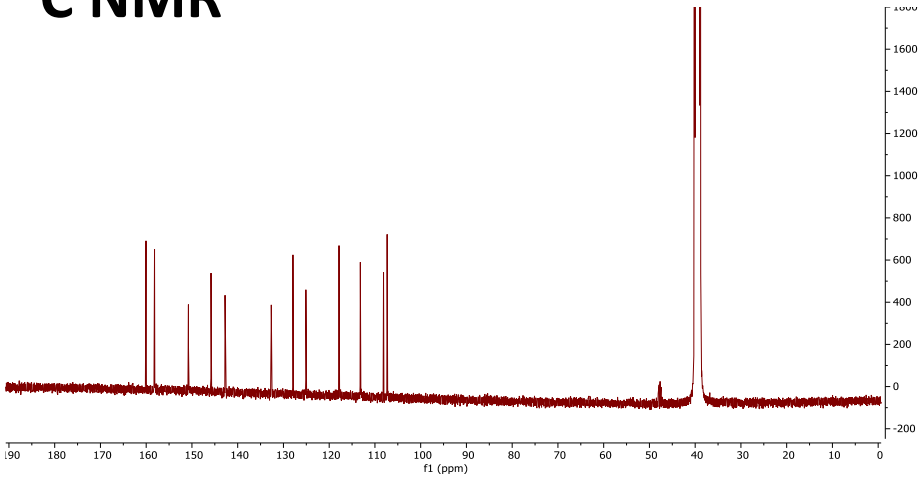
Synthesis product

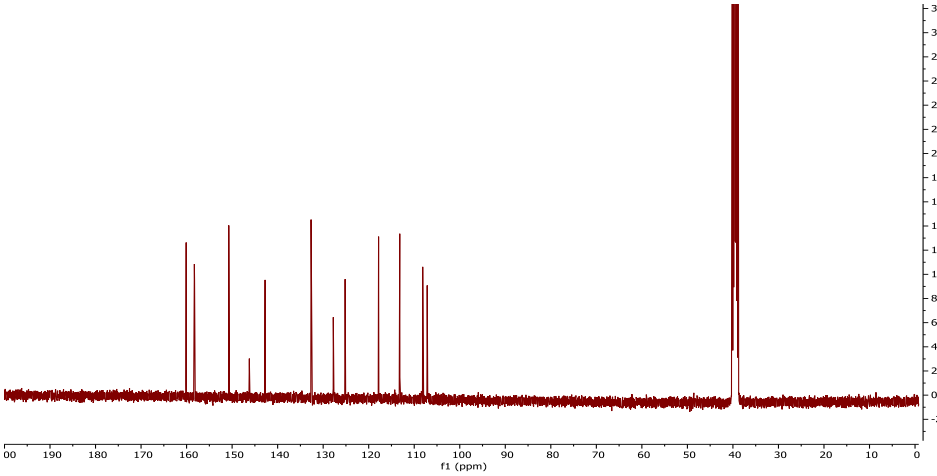
^{1}H NMR





^{13}C NMR





Publication 14.04.2023

marine drugs

MDPI

Article

Challenging Structure Elucidation of Lumnitzeralactone, an Ellagic Acid Derivative from the Mangrove *Lumnitzera racemosa*

Jonas Kappen [1], Jeprianto Manurung [1,2,3], Tristan Fuchs [1], Sahithya Phani Babu Vemulapalli [4,5], Lea M. Schmitz [1], Andrej Frolov [1], Andria Agusta [6], Alexandra N. Muellner-Riehl [2,3], Christian Griesinger [4,*], Katrin Franke [1,3,7,*] and Ludger A. Wessjohann [1,3,*]

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6 Research Center for Pharmaceutical Ingredients and Traditional Medicine, National Research and Innovation Agency (BRIN), Jl. M.H. Thamrin No. 8, Jakarta 10340, Indonesia; andr005@brin.go.id
7 Institute of Biology/Geobotany and Botanical Garden, Martin Luther University Halle-Wittenberg, 06108 Halle (Saale), Germany
* Correspondence: cigr@mpinat.mpg.de (C.G.); kfranke@ipb-halle.de (K.F.); wessjohann@ipb-halle.de (L.A.W.); Tel.: +49-551-201-2201 (C.G.); +49-345-5582-1313 (K.F.); +49-345-5582-1300 (L.A.W.)

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Tristan Fuchs
Prof. Luay Rashan
Martina Brode
Marvin Hempel
Prof. Alexandra Muellner-Rhiel
Prof. Andria Agusta
Jeprianto Manurung
R2 Lab Group
NWC-Department



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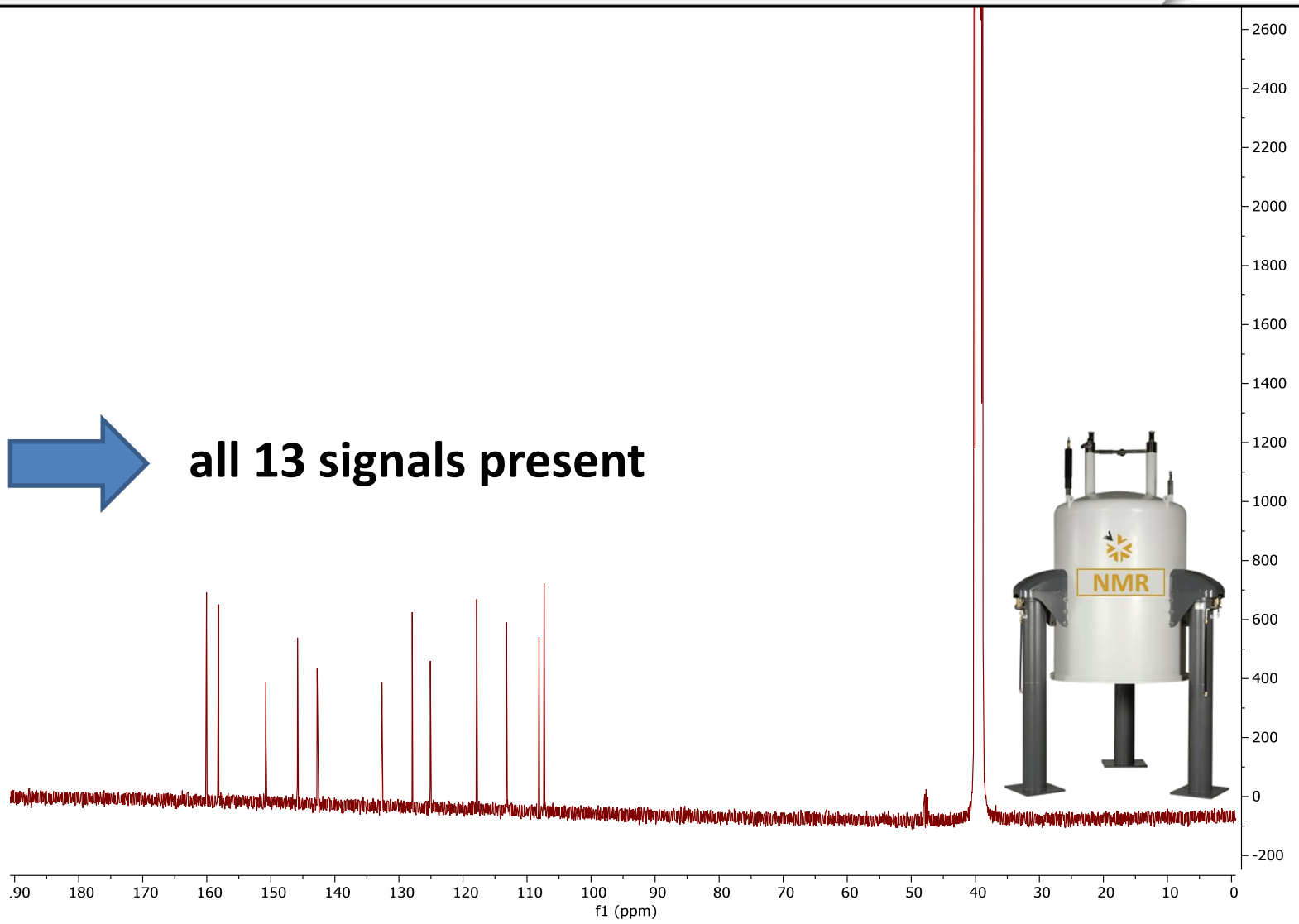
Antibacterial assay – *Bacillus subtilis*



Antibacterial assay – *Bacillus subtilis*



^{13}C in DMSO-d6, 100 MHz – $C_{13}H_6O_8$



DFT calculations

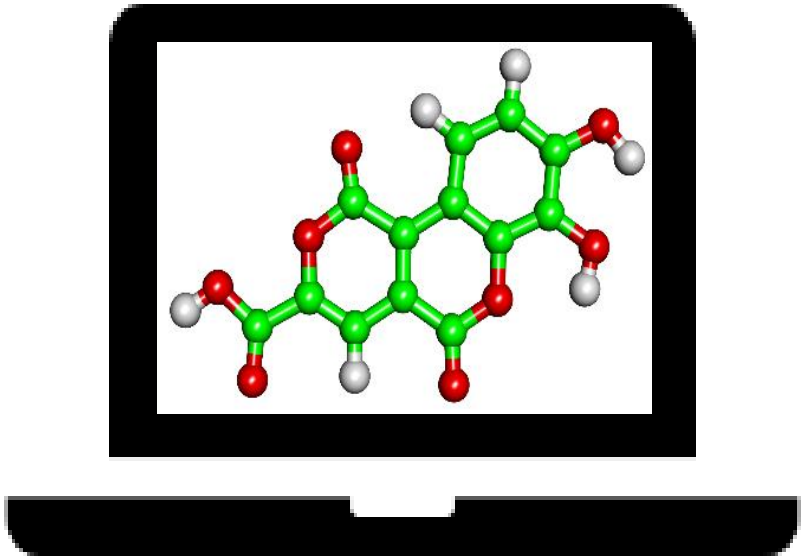
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Carl von Ossietzky Universität Oldenburg



Dr. S. Phani B. Vemulapalli





HMBC in DMSO-d6 – $C_{13}H_6O_8$

