Problem of the Month:
February 2021

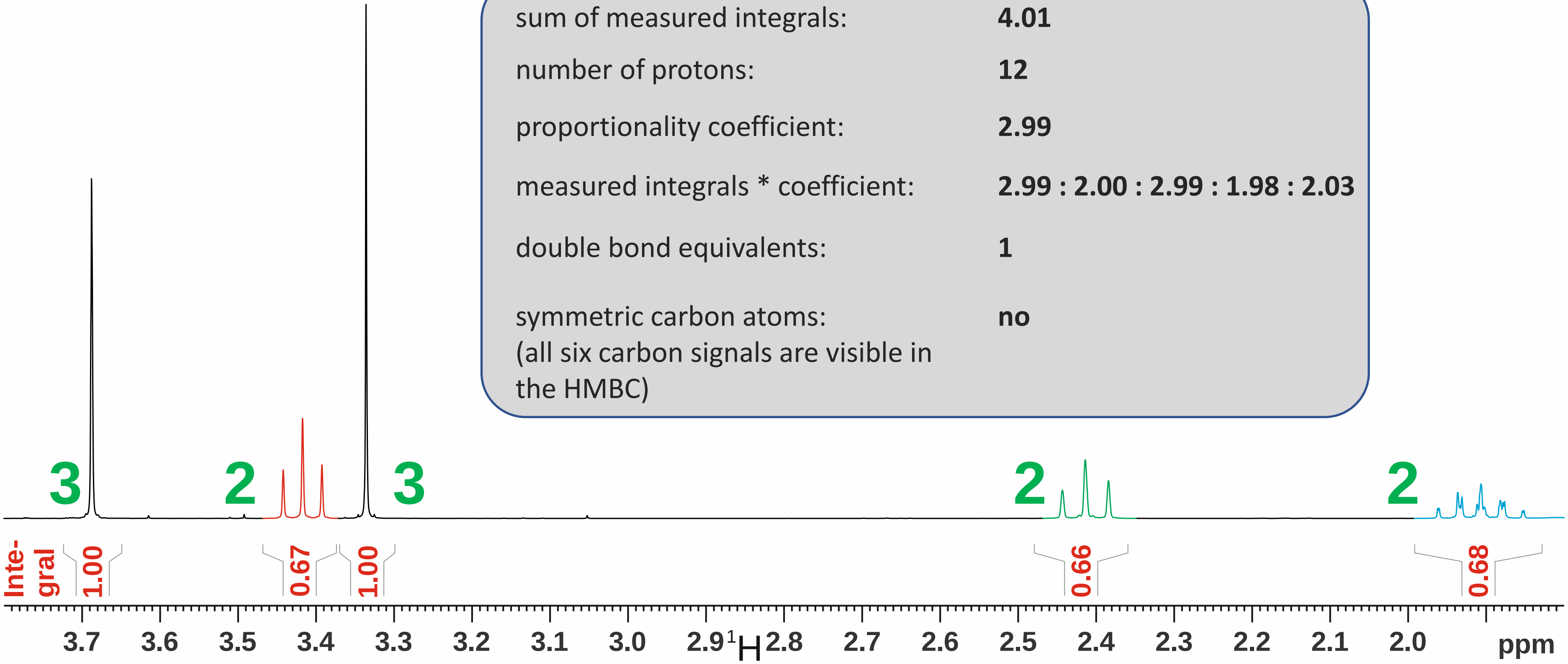
Solution

Basics

Integration,double bond equivalents, symmetry

$C_6H_{12}O_3$

sum of measured integrals:	**4.01**
number of protons:	**12**
proportionality coefficient:	**2.99**
measured integrals * coefficient:	**2.99 : 2.00 : 2.99 : 1.98 : 2.03**
double bond equivalents:	**1**
symmetric carbon atoms: (all six carbon signals are visible in the HMBC)	**no**



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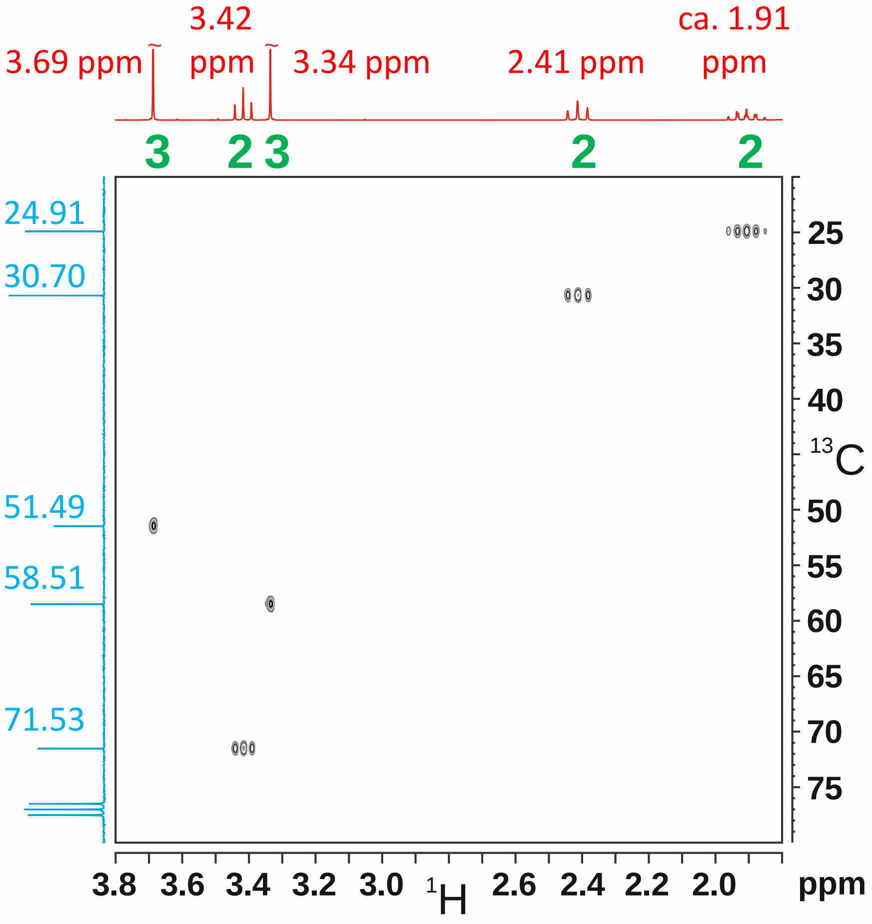
Building blocks

CH$_n$-fragments



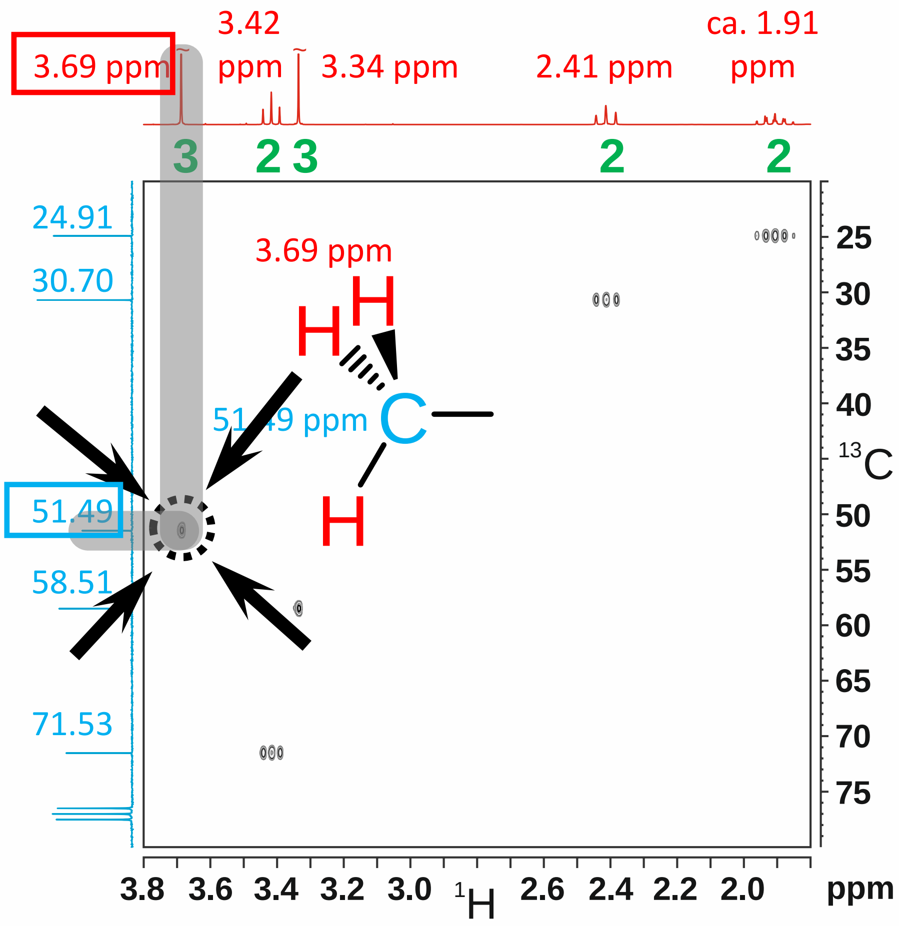
It is very easy to evaluate a HSQC. The sensitivity , of course, is below the sensitivity of a one dimensional proton spectrum but much higher in comparison to a one dimensional carbon spectrum. Therefore, the measurement of a HSQC is always recommended, if somehow feasible.

We need some data for the projections, chemical shifts and integrals from the one dimensional proton spectrum and the carbon chemical hifts from the one dimensional carbon spectrum.

The one dimensional carbon spectrum is not explicitely given here but used as a pseudo projection for the HMBC. The chemical shifts can be picked there.

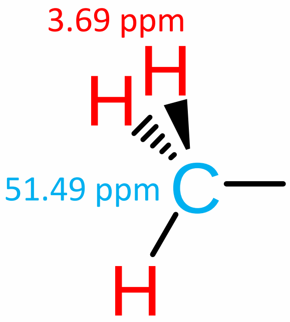
Building blocks

CH$_n$-fragments



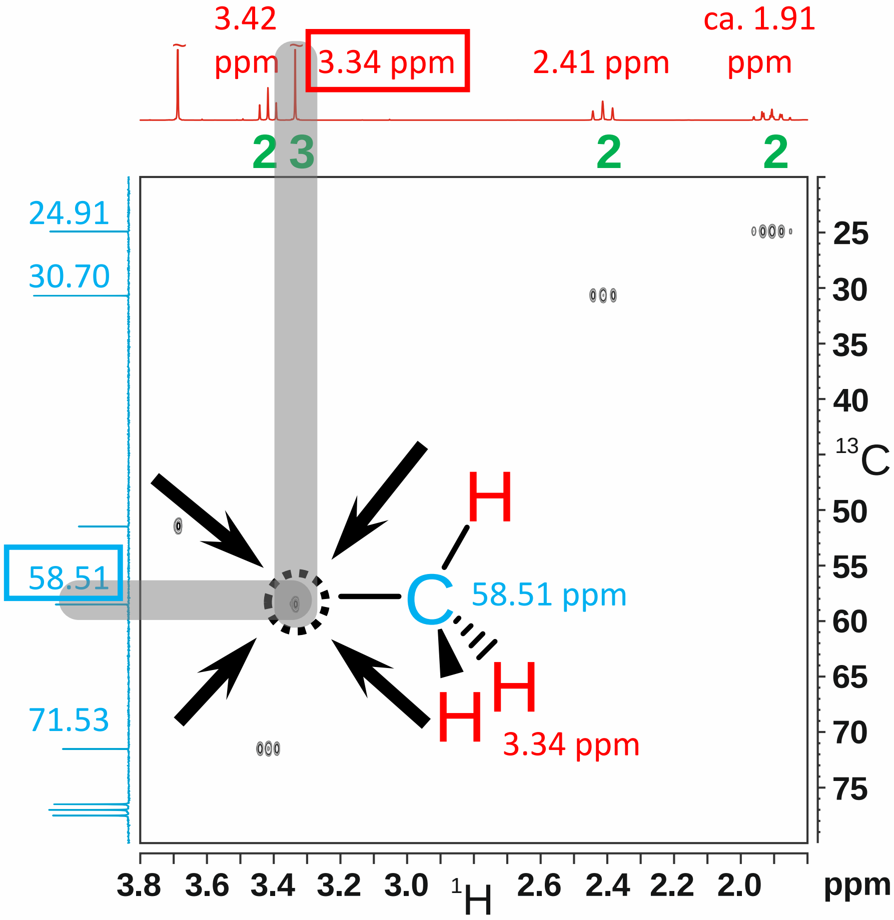
The proton signals at 3.69 ppm and 3.34 ppm could only belong to methyl groups according to their integral. Three symmetric CH-groups became already excluded at the very first beginning.

Please continue to the second methyl group …



Building blocks

CH$_n$-fragments



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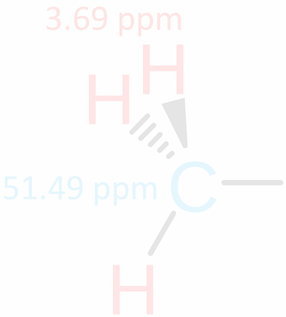
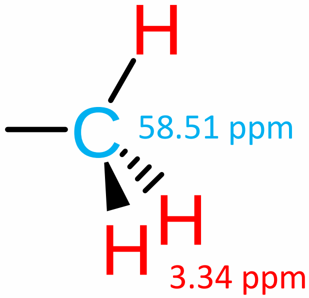
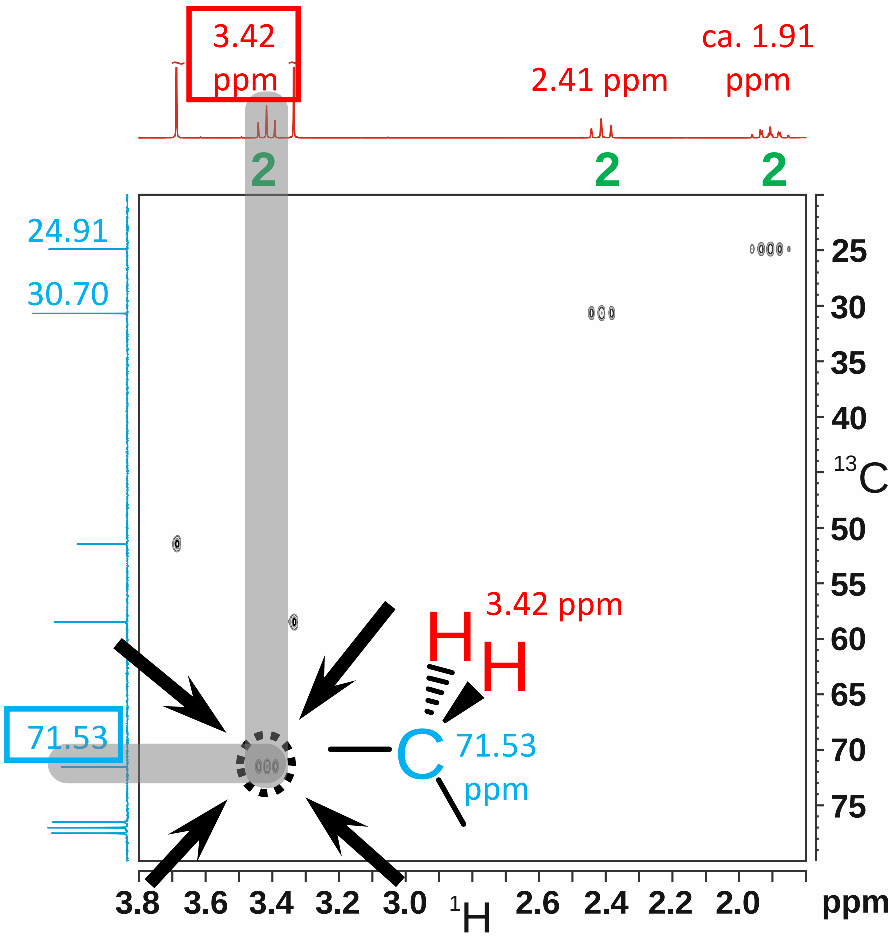


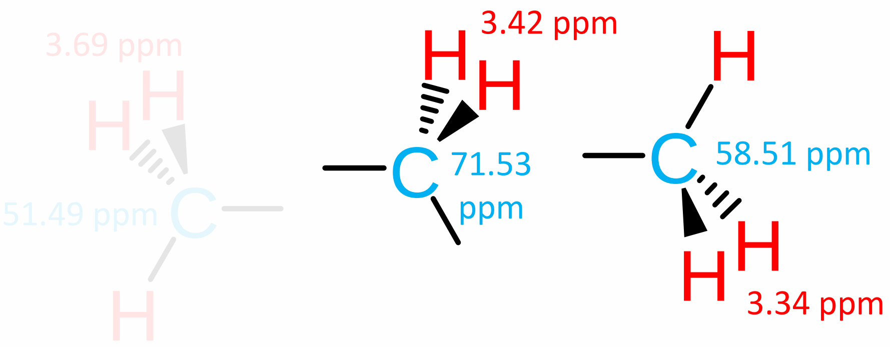


Building blocks

CH_n-fragments

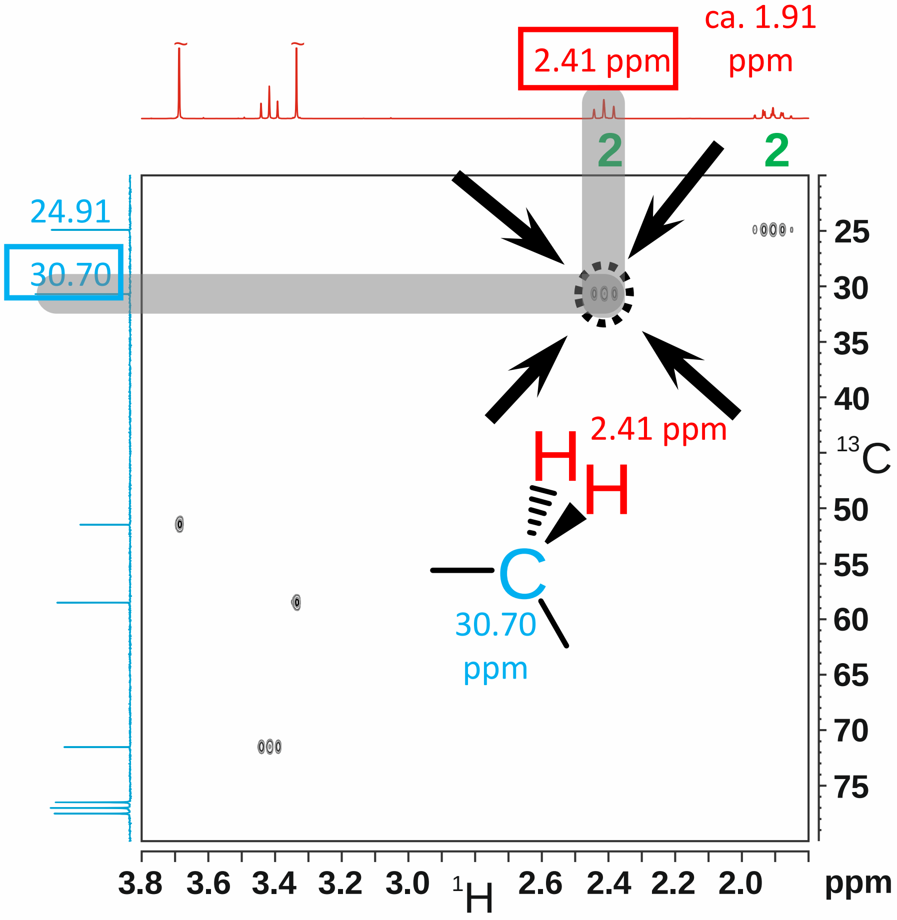


The remaining cross peaks belong to methylene groups. Let us extract them step by step.



Building blocks

CH_n-fragments



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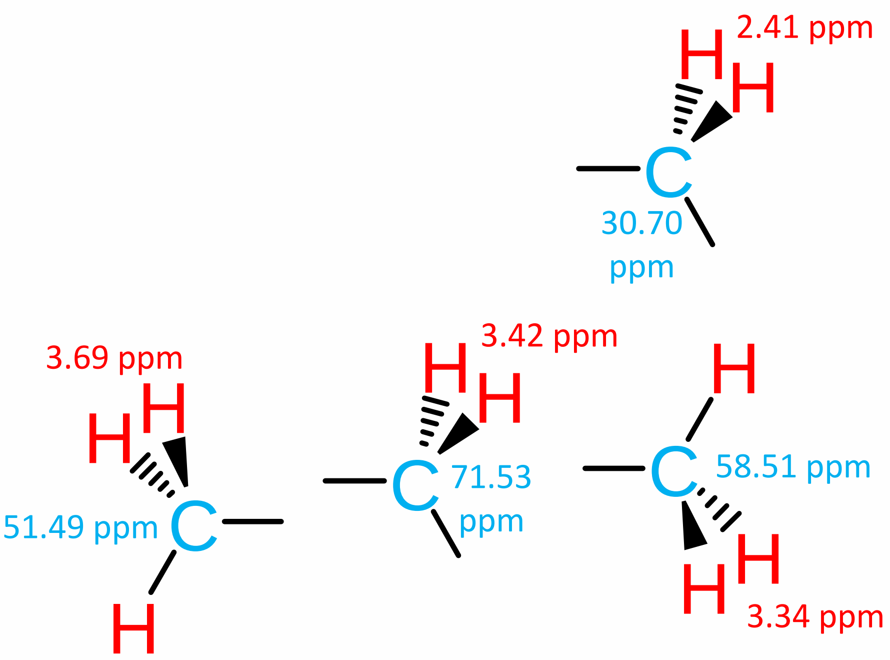

Building blocks

CH_n-fragments

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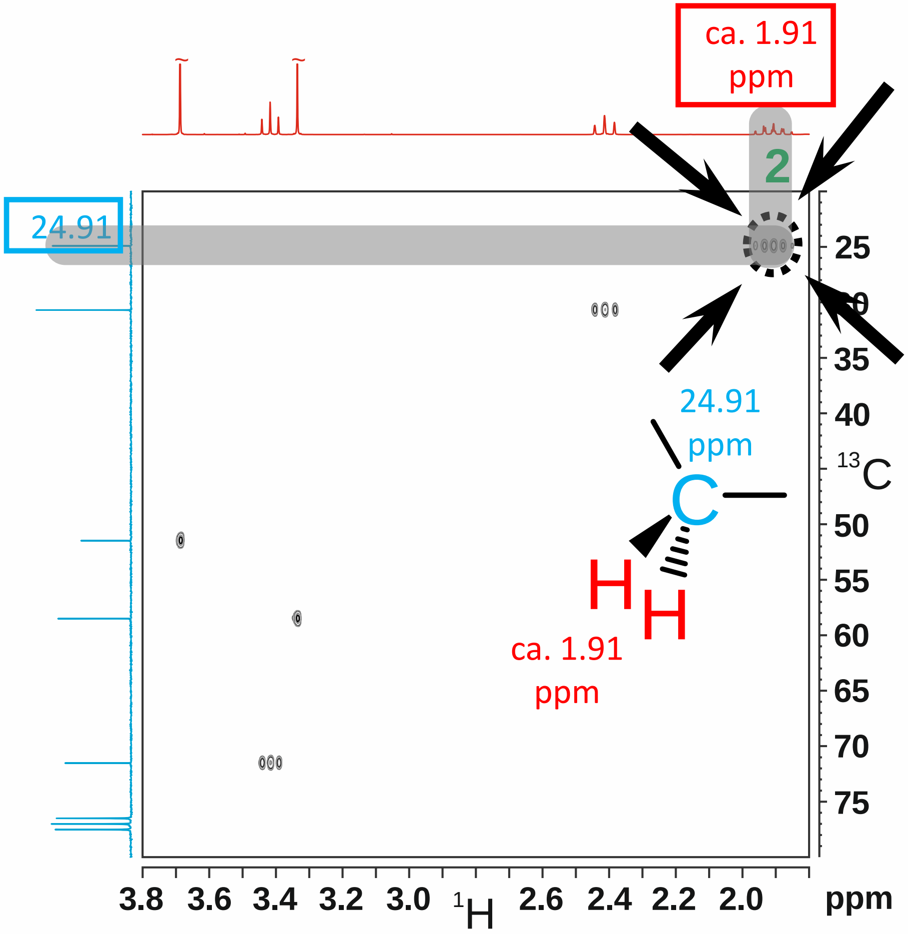
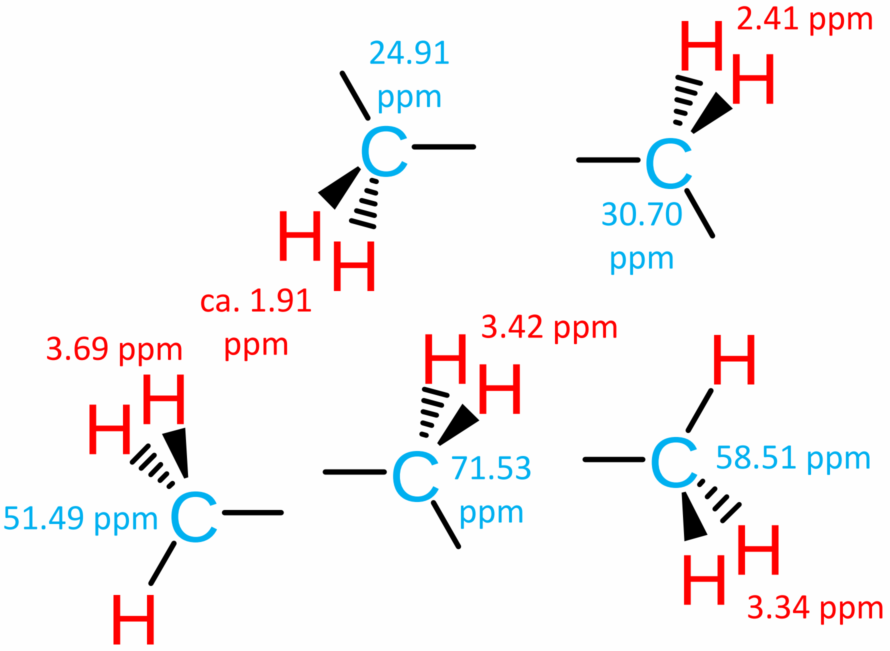
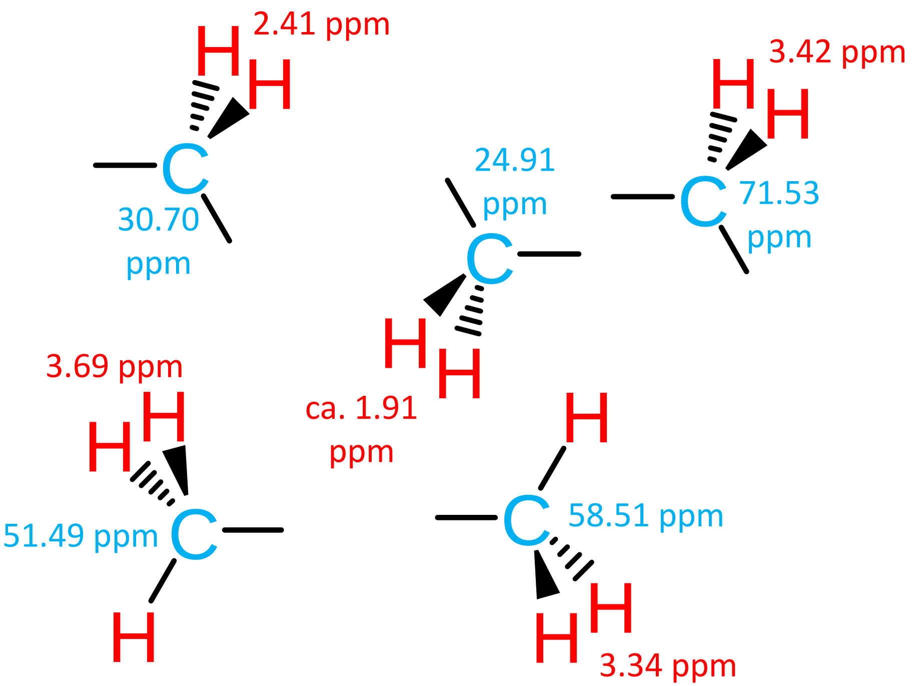




Linking the pieces

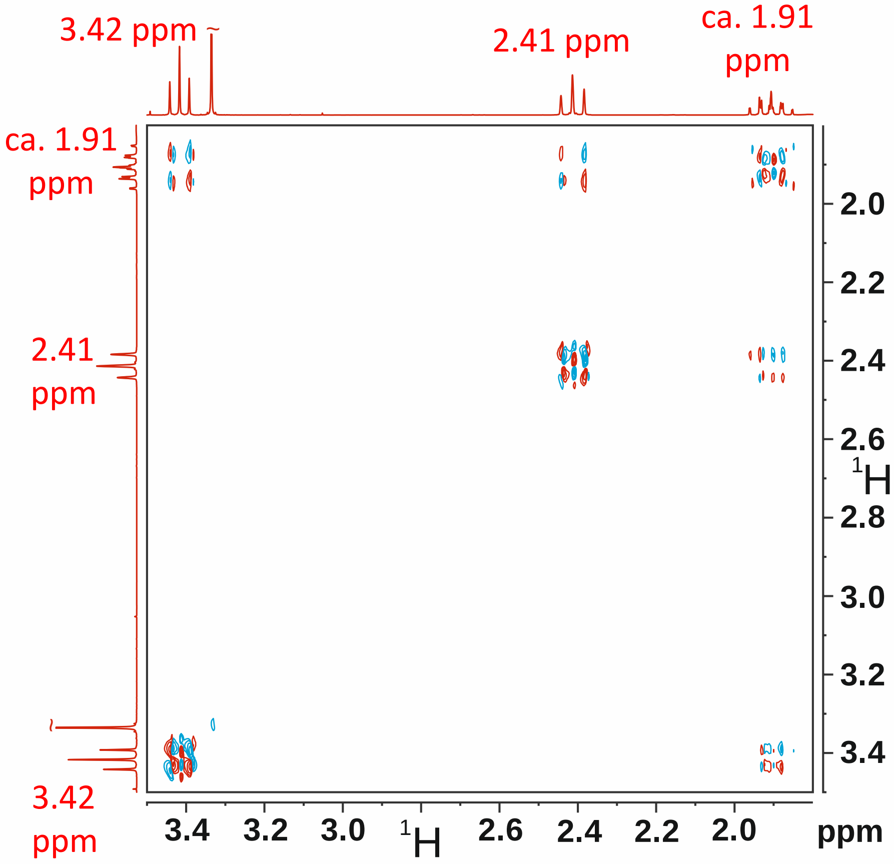
part 1 – alkyl chain

First let us reorder the fragments a little bit to make the next steps easier.

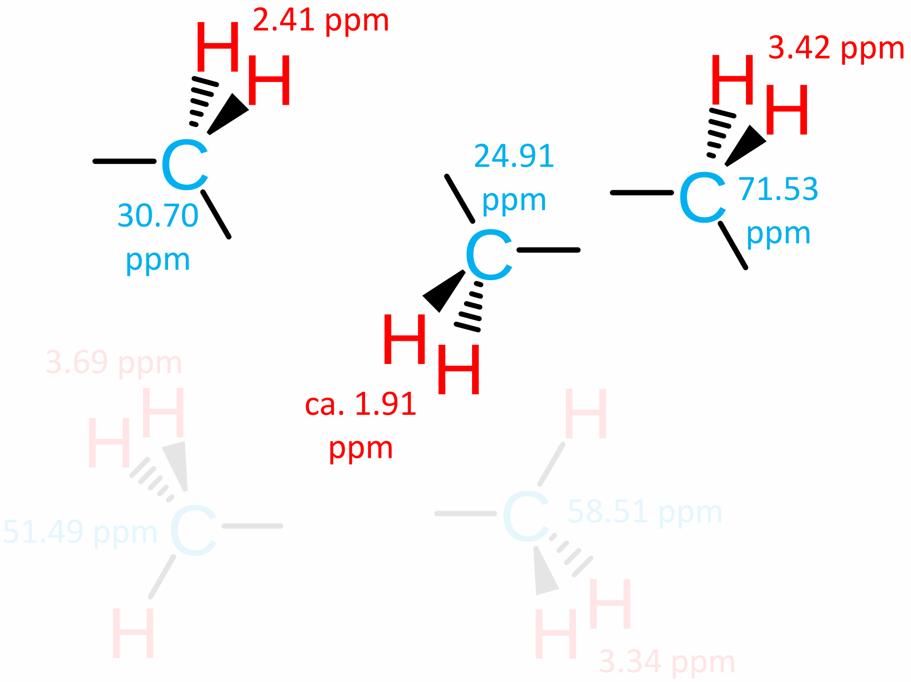


Linking the pieces

part 1 – alkyl chain

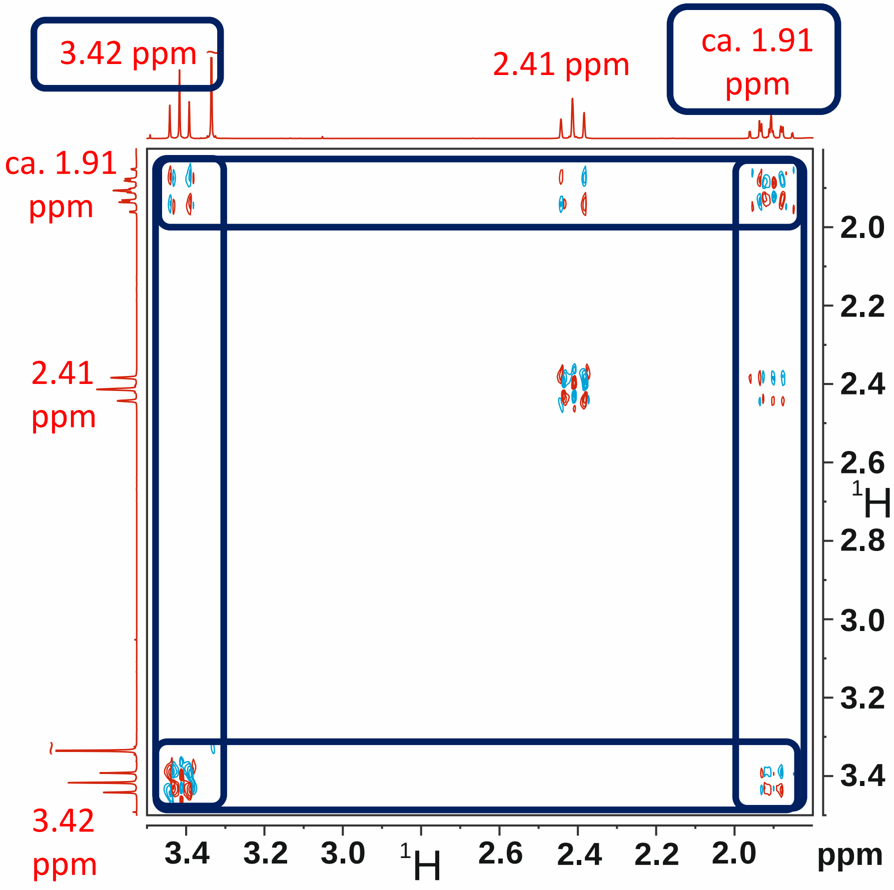


During our work with the COSY we have no use for the methyl groups..

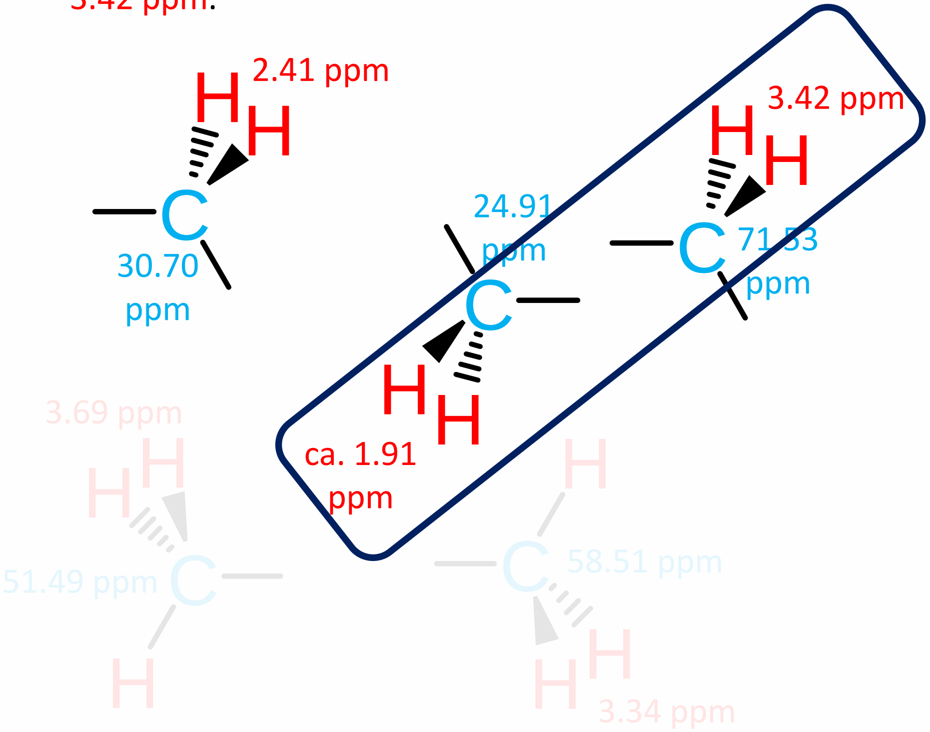


Linking the pieces

part 1 – alkyl chain

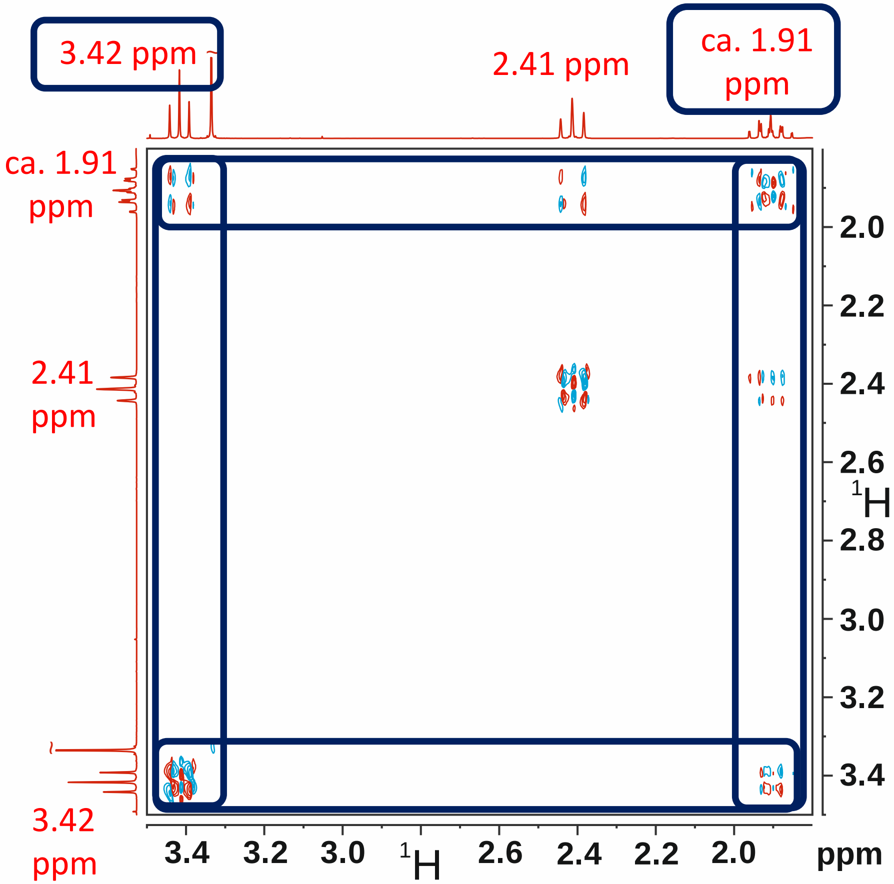


The first visible proximity in the COSY, is between the protons with the chemical shifts of ca. 1.91 ppm and 3.42 ppm.



Linking the pieces

part 1 – alkyl chain



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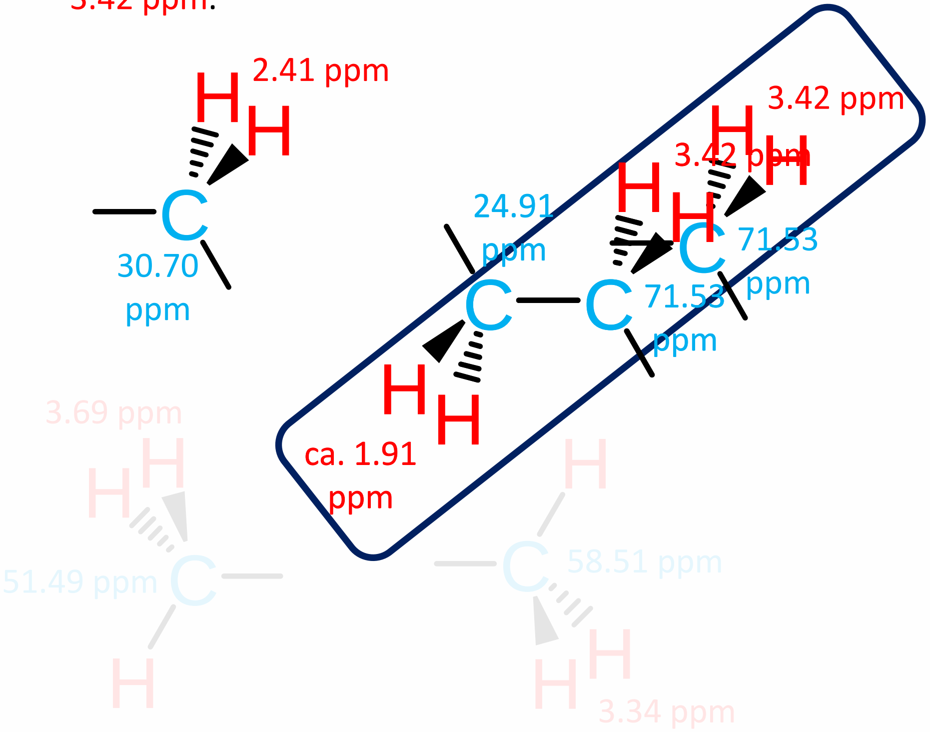
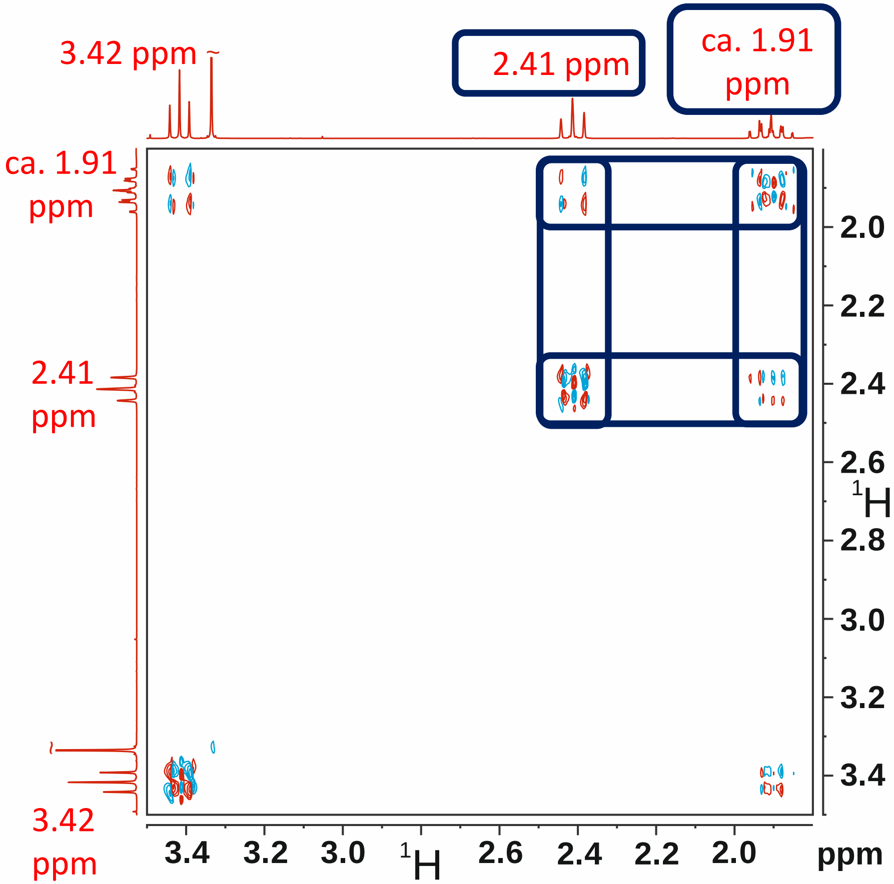
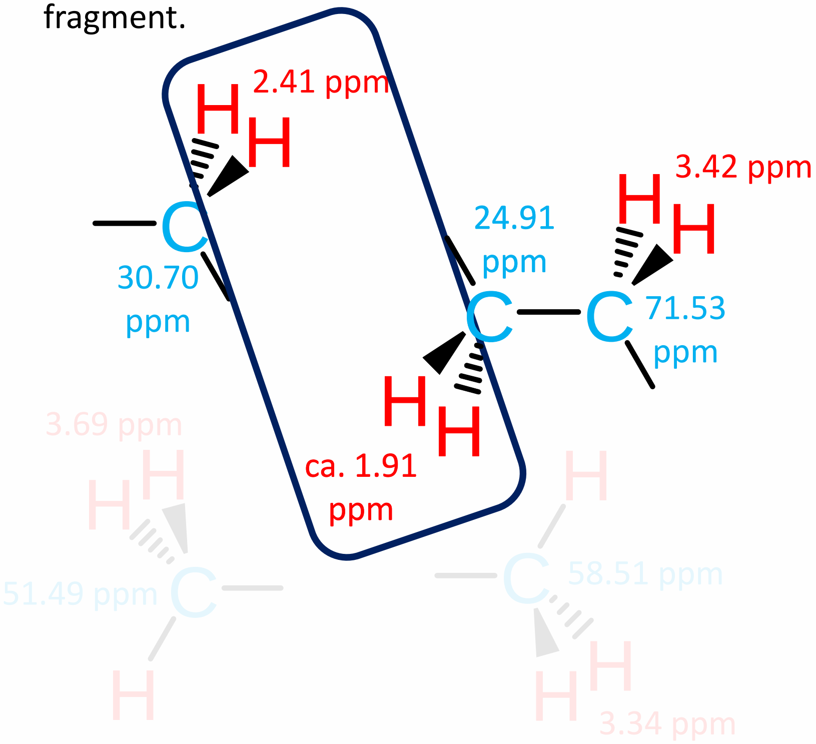


Linking the pieces

part 1 – alkyl chain

The second proximity, visible in the COSY, is between the protons with the chemical shifts of ca. 1.91 ppm and 2.41 ppm, which allows us to complete the already known ethyl fragment.



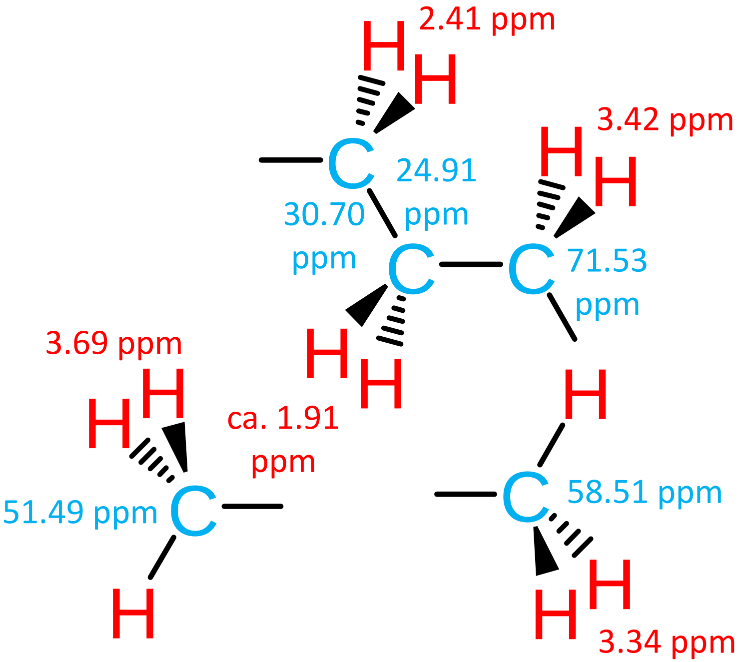


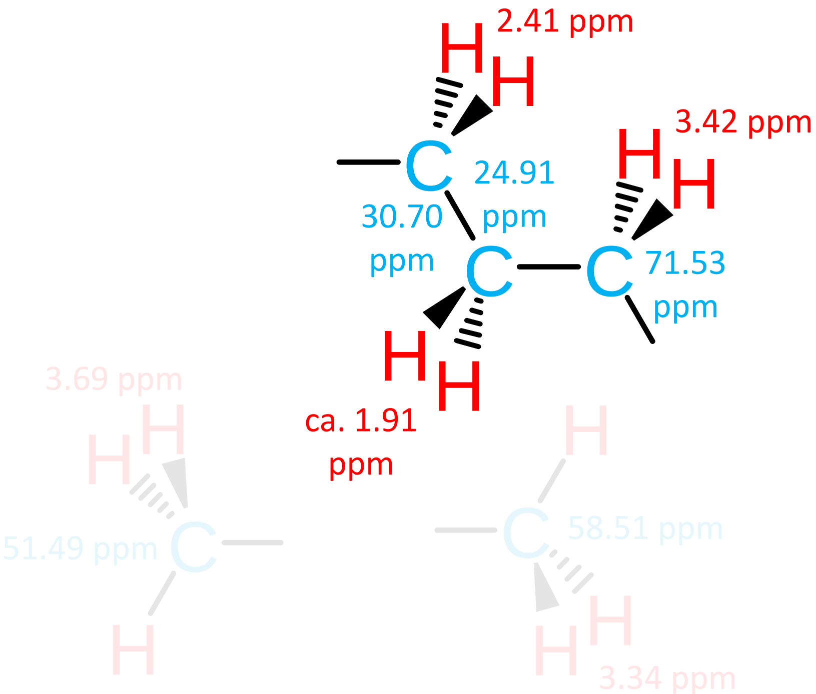
Something missing?

time for a short inventory

We no longer need the COSY.

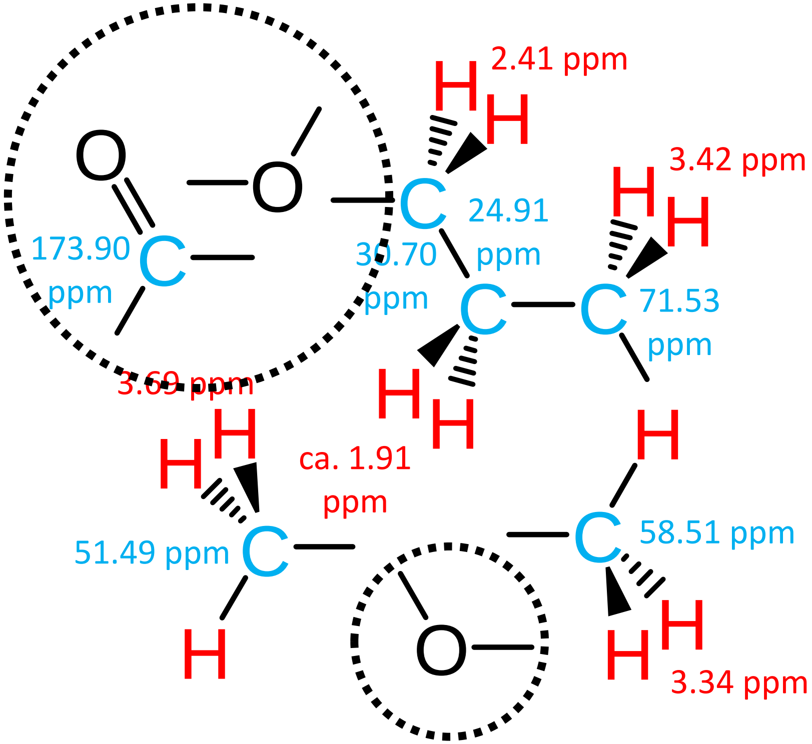
To continue let us unhide the methyl groups and rearrange the fragments a little bit for further use.





Something missing?

time for a short inventory



molecular formula	$C_6H_{12}O_3$
known fragments	C_5H_{12}
unassigned carbon atom without attached hydrogen	173.9 ppm
missing	CO_3 one double bond equivalent

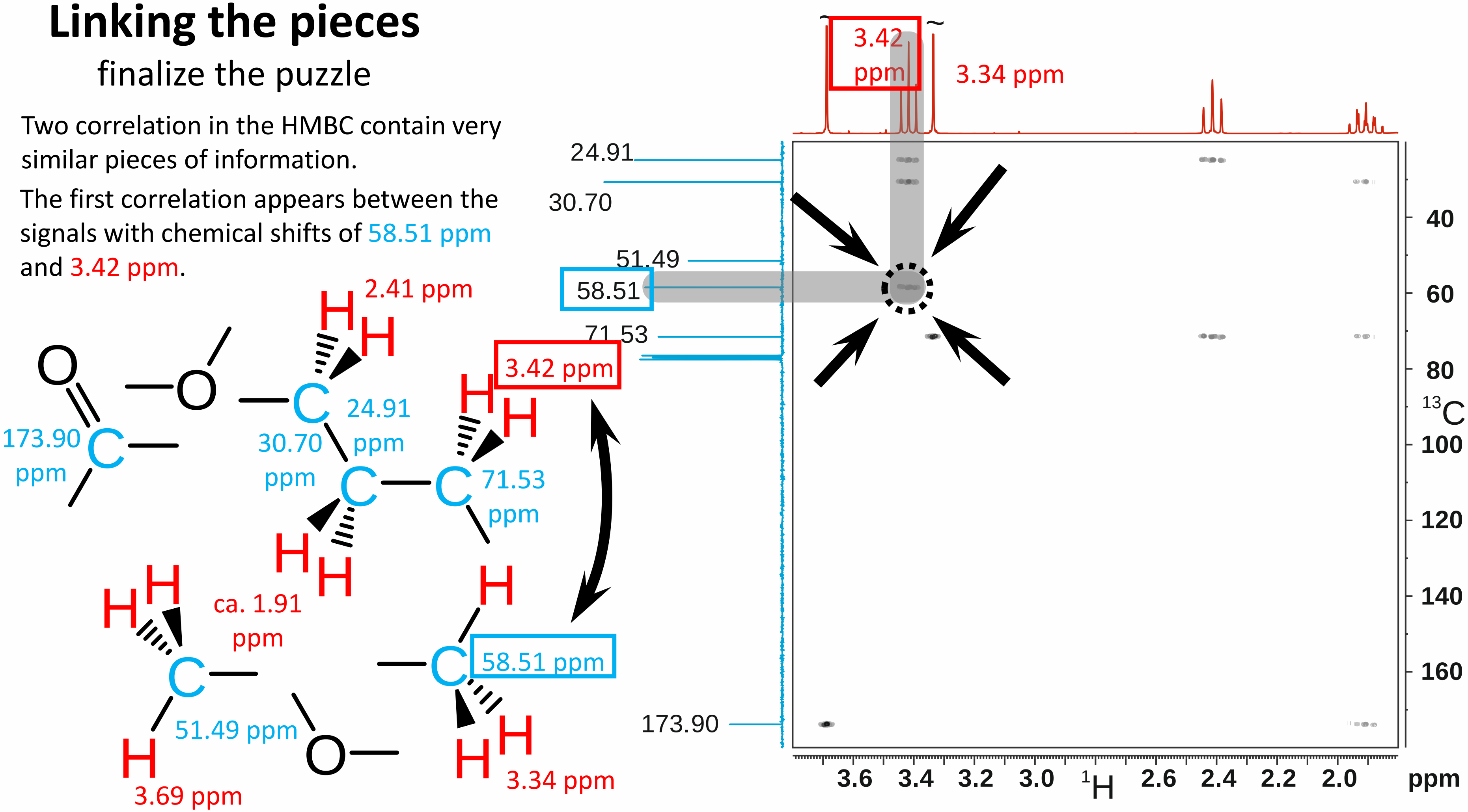
As a result let us increase our unordered pile of building blocks by three hydrogen free fragments.

Linking the pieces

finalize the puzzle

Two correlation in the HMBC contain very similar pieces of information.

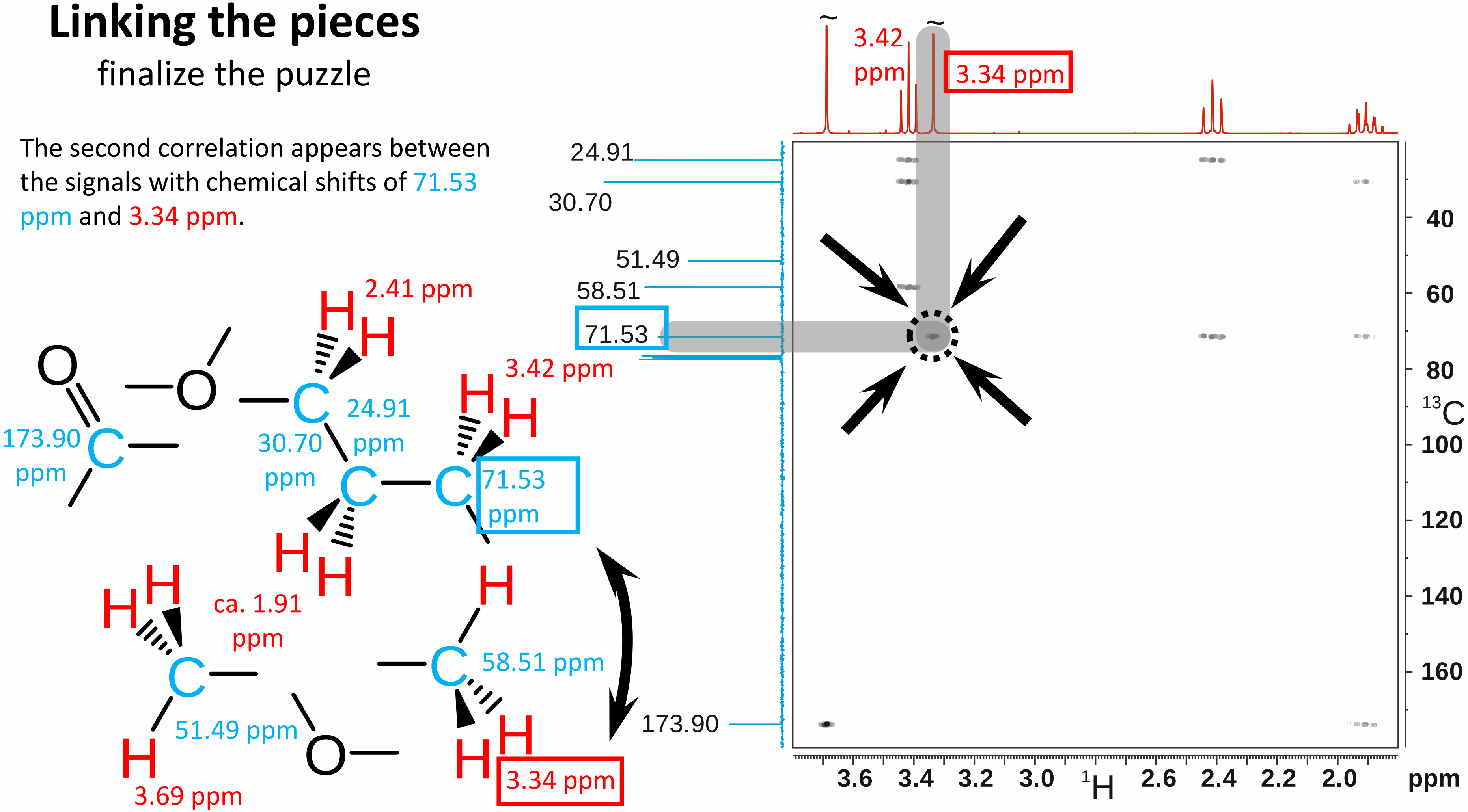
The first correlation appears between the signals with chemical shifts of 58.51 ppm and 3.42 ppm.



Linking the pieces

finalize the puzzle

The second correlation appears between the signals with chemical shifts of 71.53 ppm and 3.34 ppm.

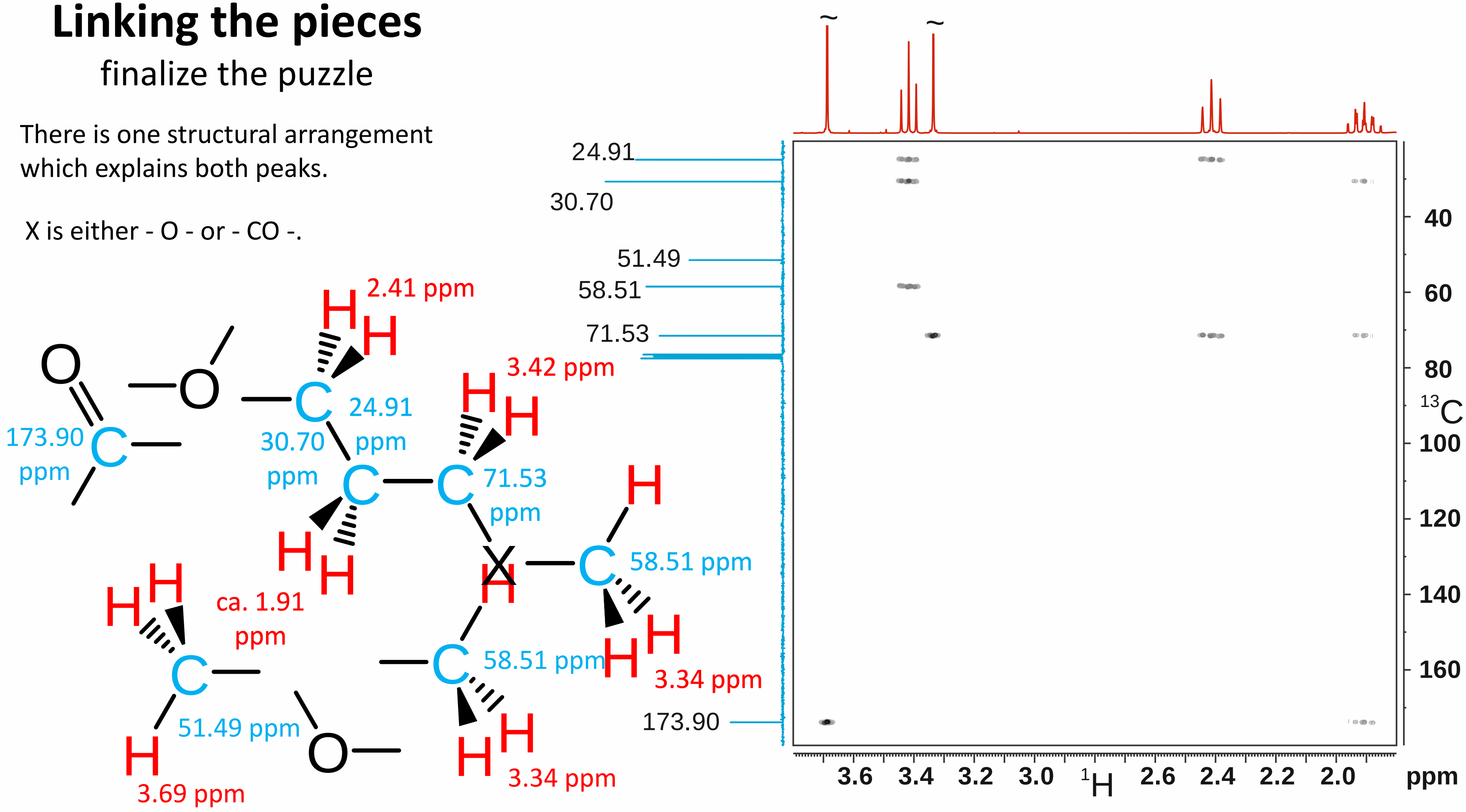


Linking the pieces

finalize the puzzle

There is one structural arrangement which explains both peaks.

X is either - O - or - CO -.

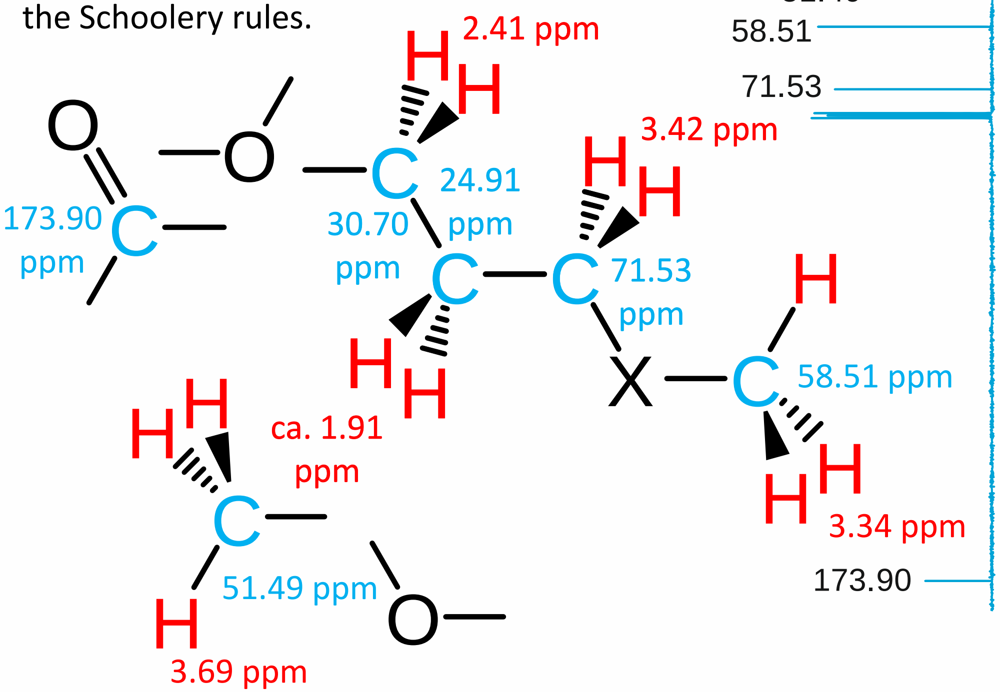


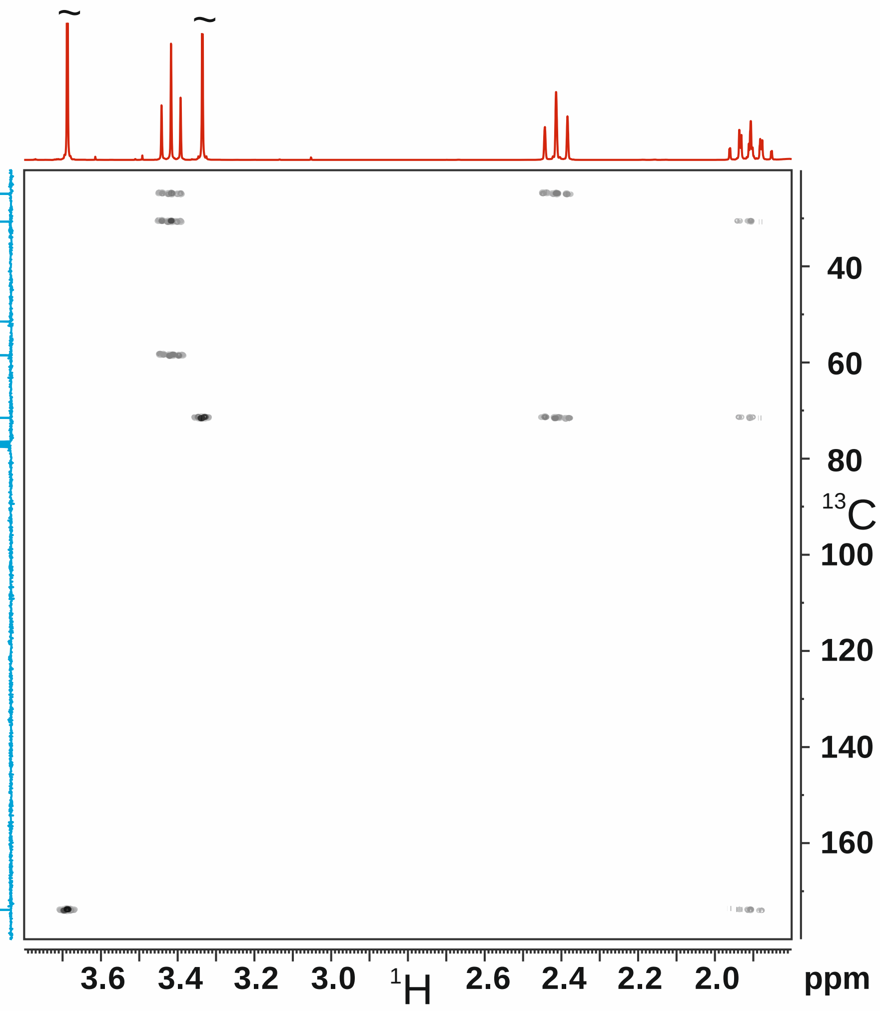
Linking the pieces

finalize the puzzle

Would it be possible to replace X by - CO - ?

Calculate by yourself the chemical shift of the methylene protons bond to the carbon with the chemical shift of 71.53 ppm using the Schoolery rules.



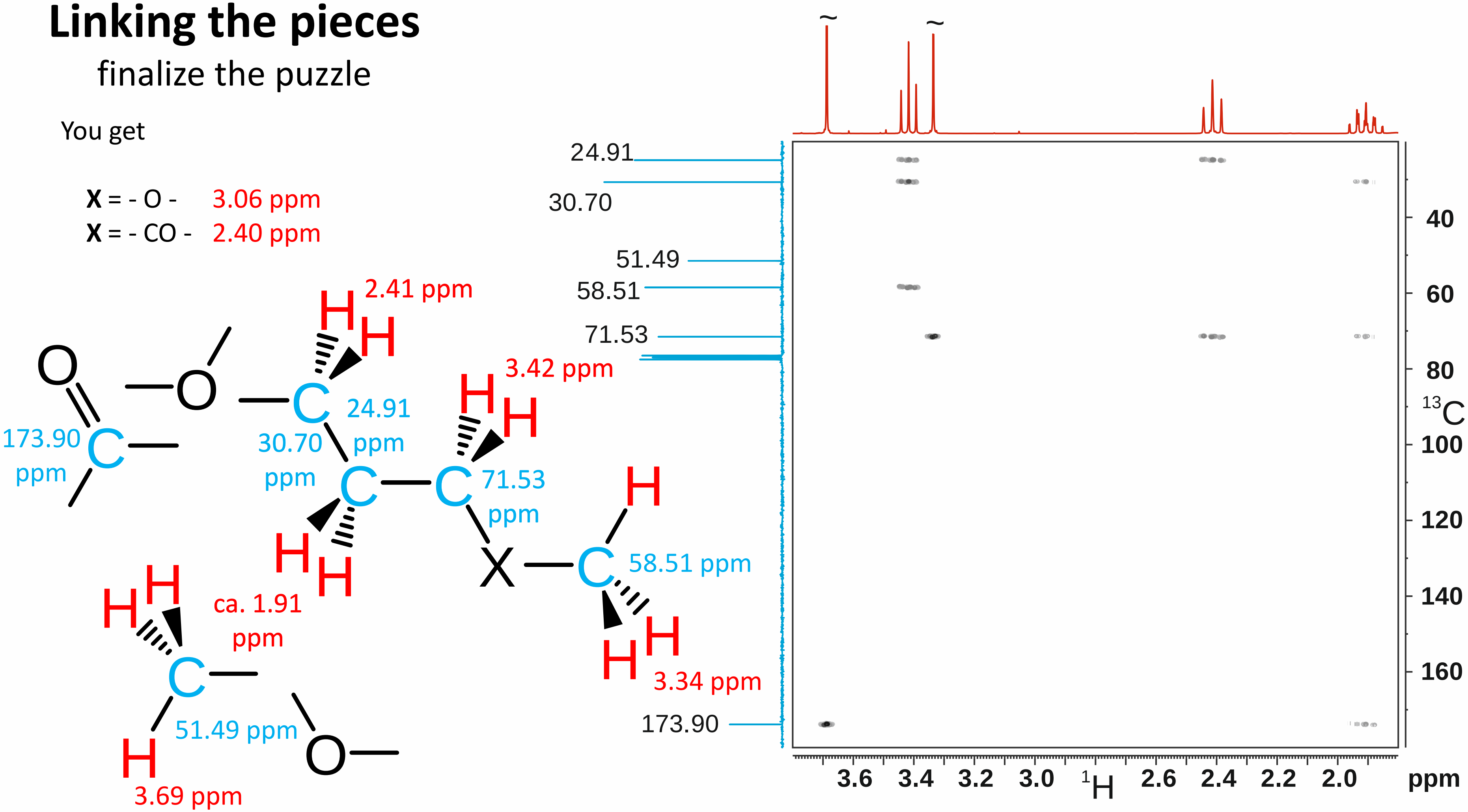


Linking the pieces

finalize the puzzle

You get

X = - O - 3.06 ppm
X = - CO - 2.40 ppm

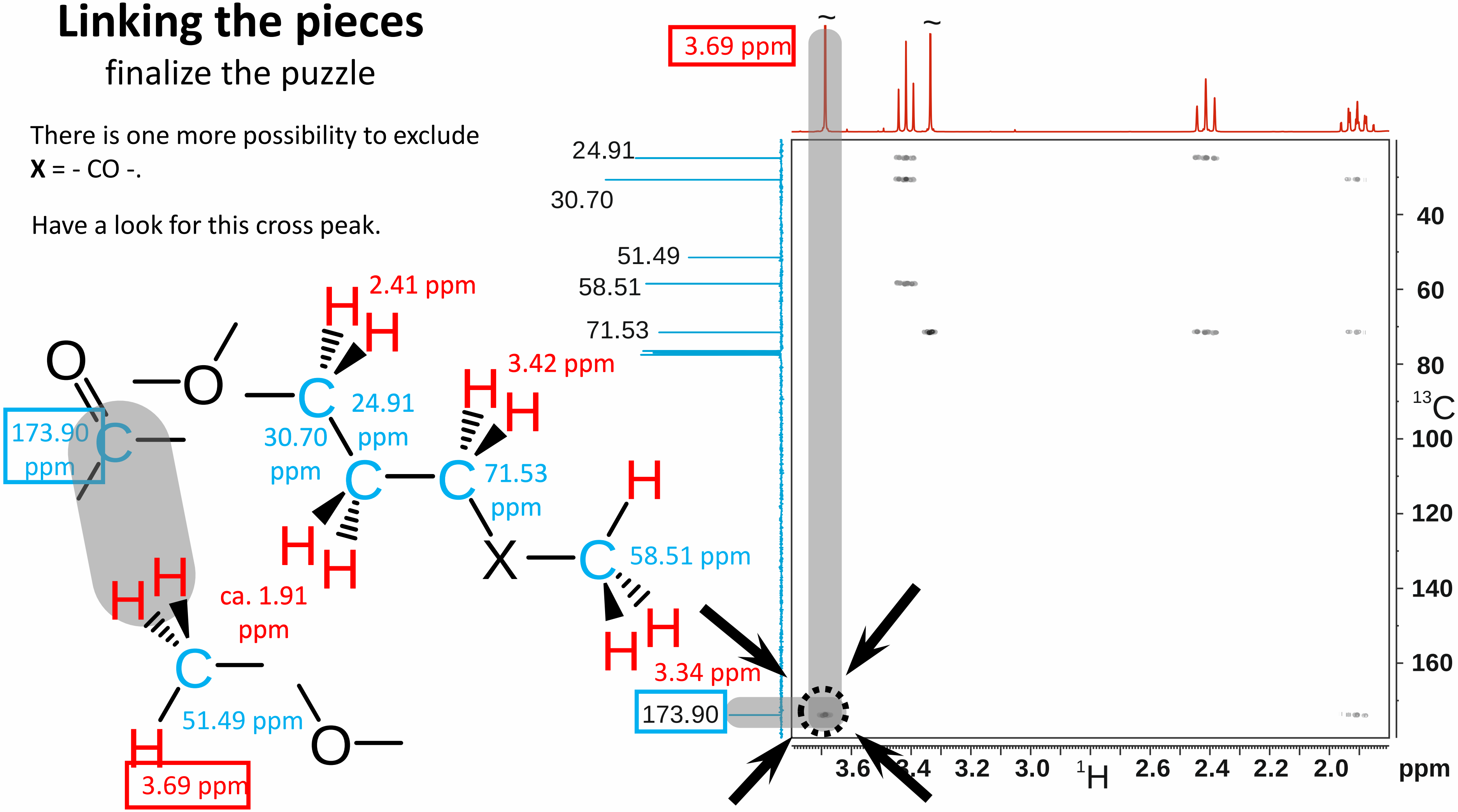


Linking the pieces

finalize the puzzle

There is one more possibility to exclude **X** = - CO -.

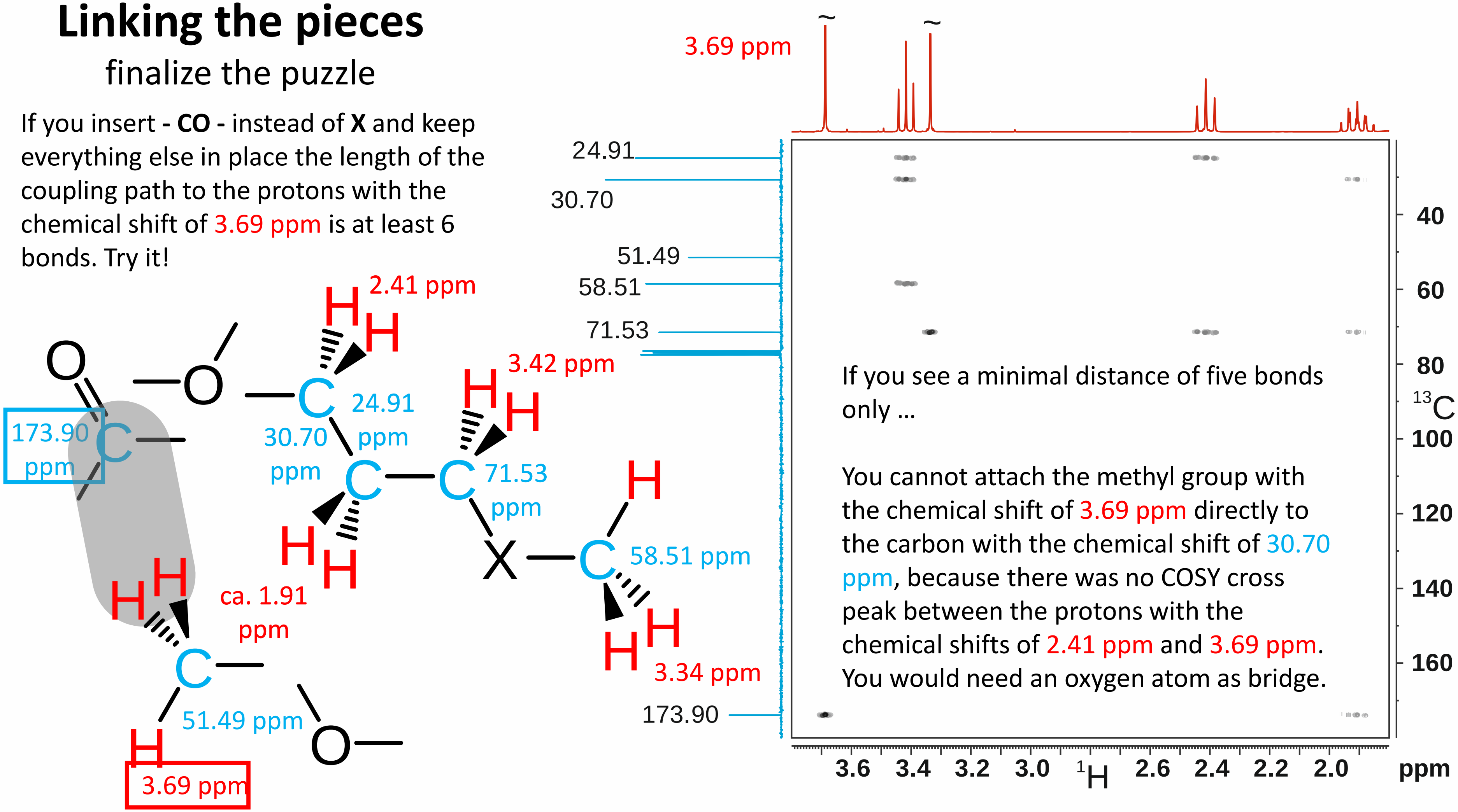
Have a look for this cross peak.



Linking the pieces

finalize the puzzle

If you insert **- CO -** instead of **X** and keep everything else in place the length of the coupling path to the protons with the chemical shift of 3.69 ppm is at least 6 bonds. Try it!



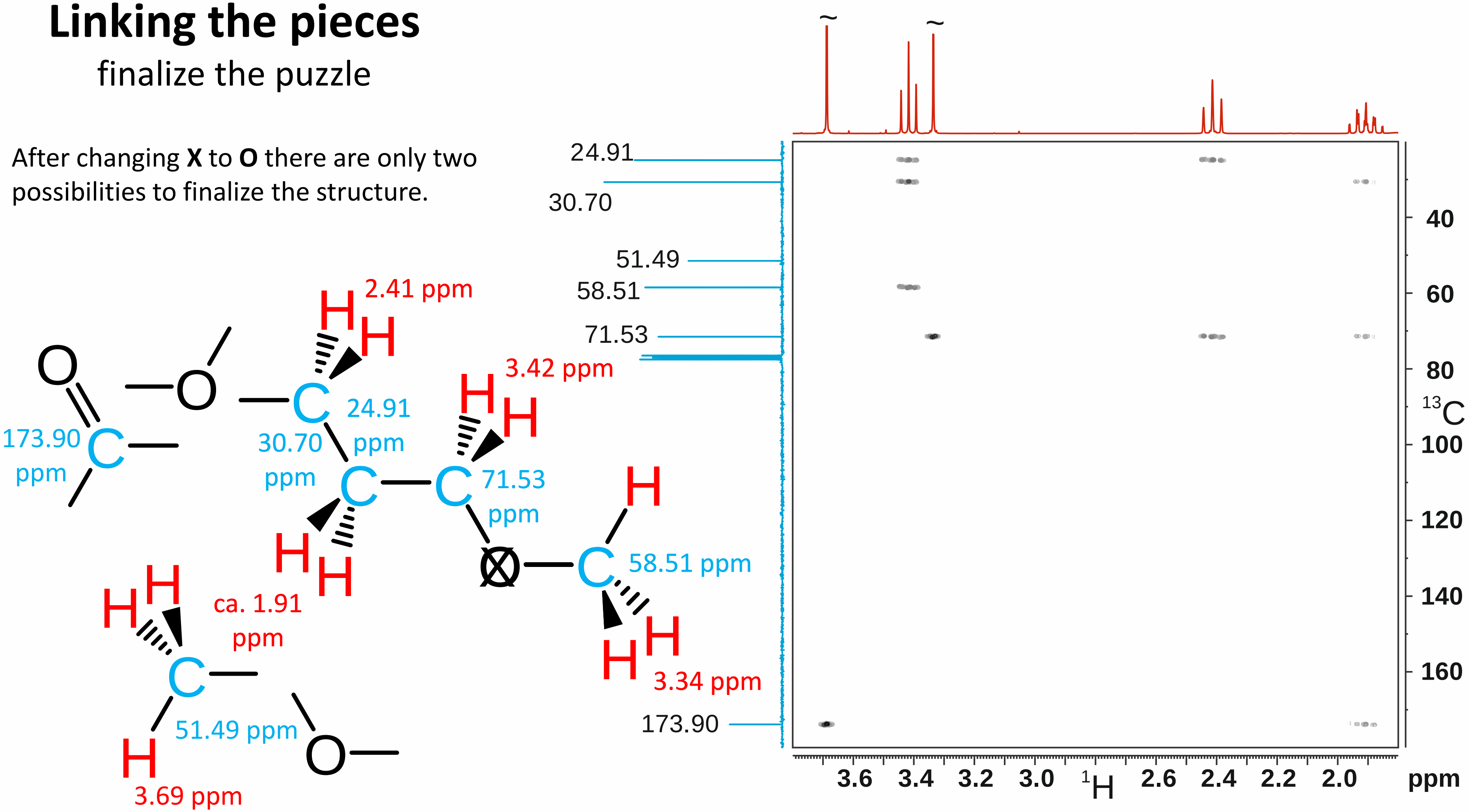
If you see a minimal distance of five bonds only …

You cannot attach the methyl group with the chemical shift of 3.69 ppm directly to the carbon with the chemical shift of 30.70 ppm, because there was no COSY cross peak between the protons with the chemical shifts of 2.41 ppm and 3.69 ppm. You would need an oxygen atom as bridge.

Linking the pieces

finalize the puzzle

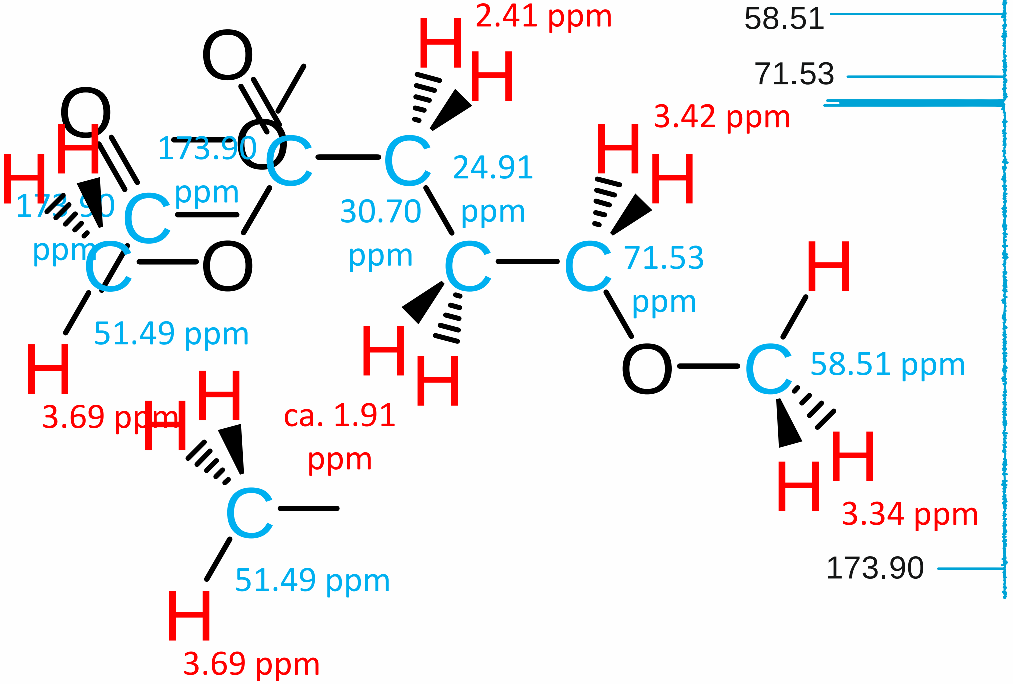
After changing **X** to **O** there are only two possibilities to finalize the structure.

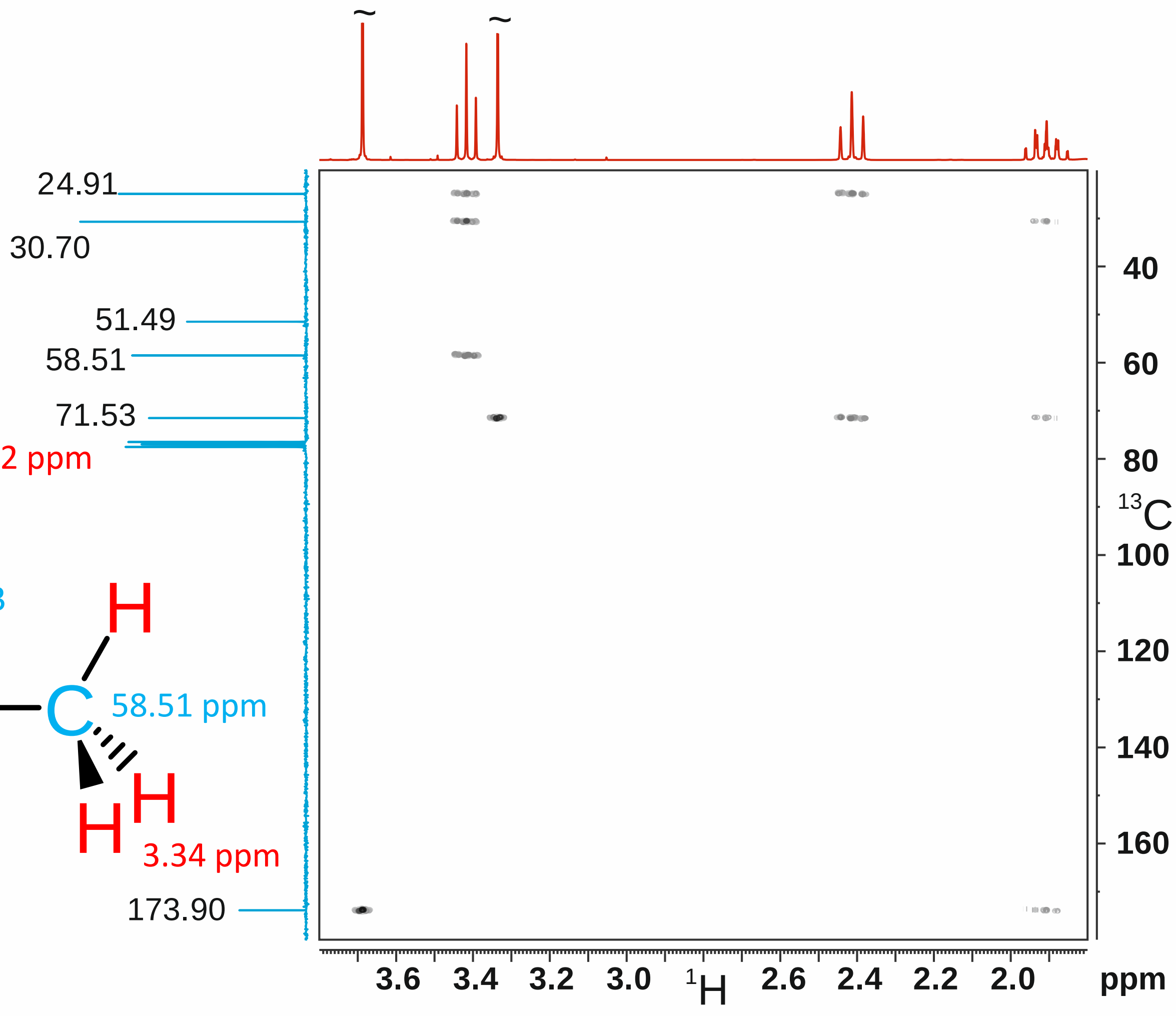


Linking the pieces

finalize the puzzle

Let us try one of them.



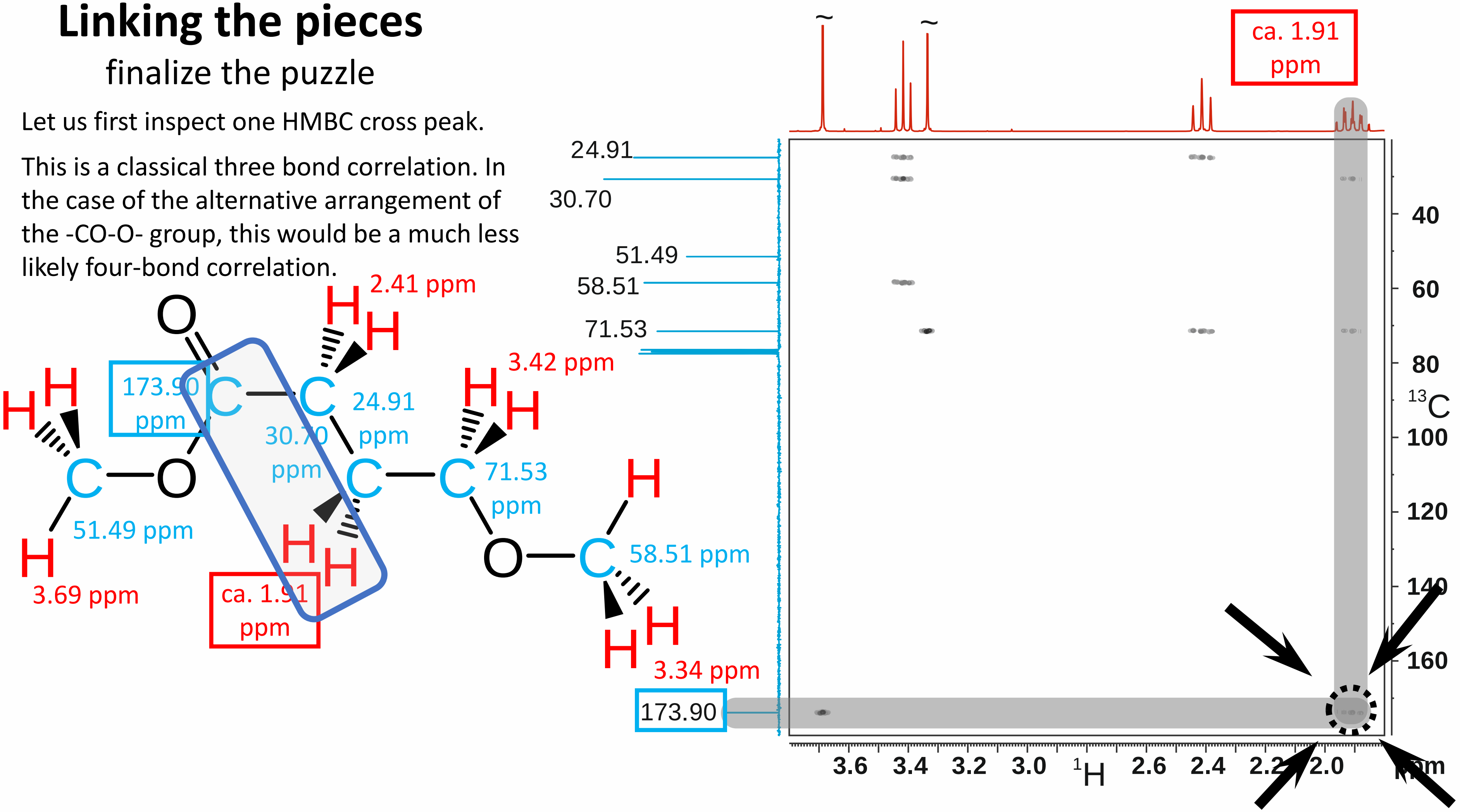


Linking the pieces

finalize the puzzle

Let us first inspect one HMBC cross peak.

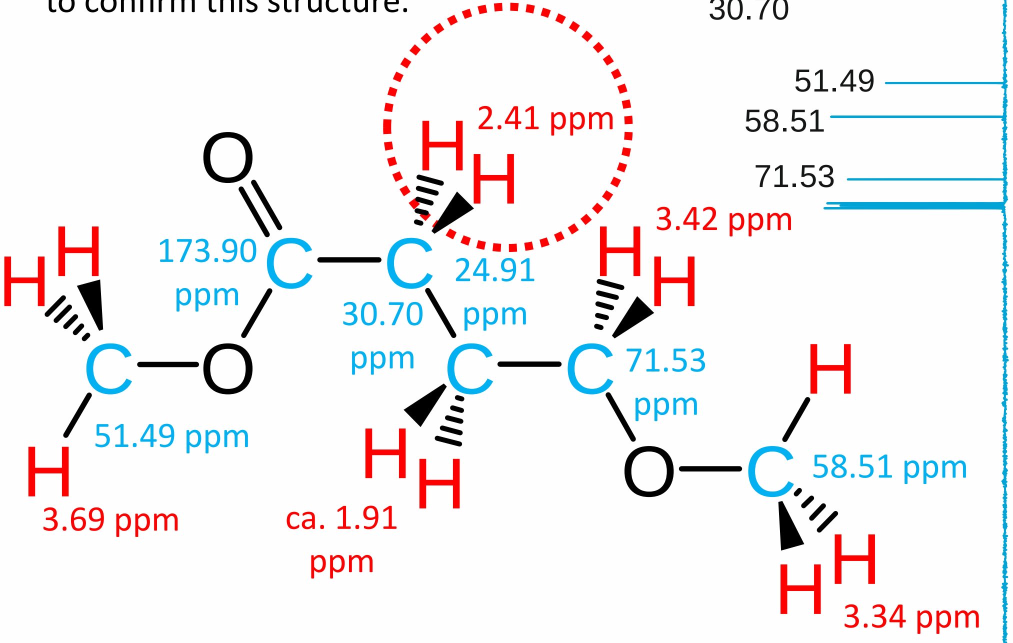
This is a classical three bond correlation. In the case of the alternative arrangement of the -CO-O- group, this would be a much less likely four-bond correlation.

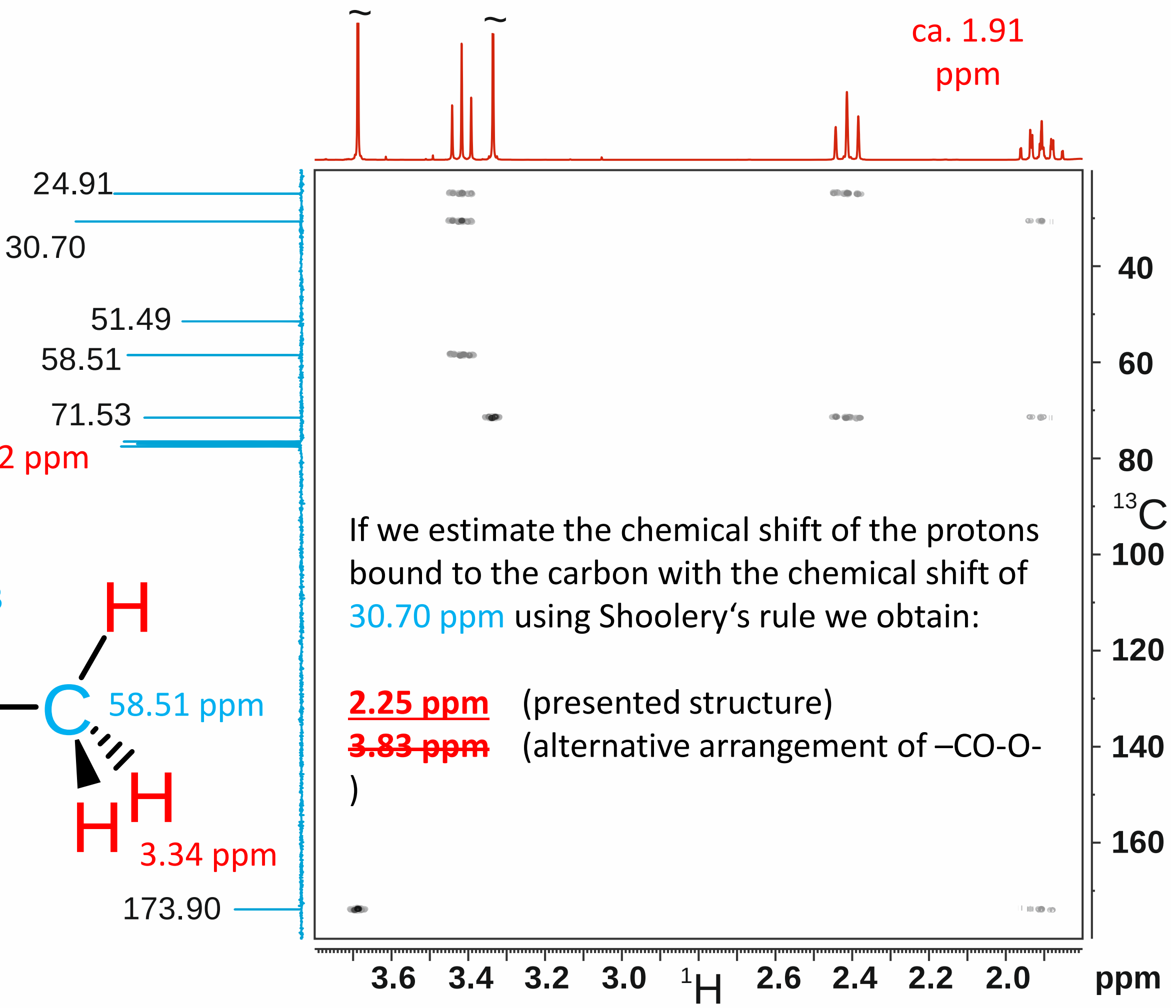


Linking the pieces

finalize the puzzle

Good old Shoolery‘s rule is another way to confirm this structure.





If we estimate the chemical shift of the protons bound to the carbon with the chemical shift of 30.70 ppm using Shoolery‘s rule we obtain:

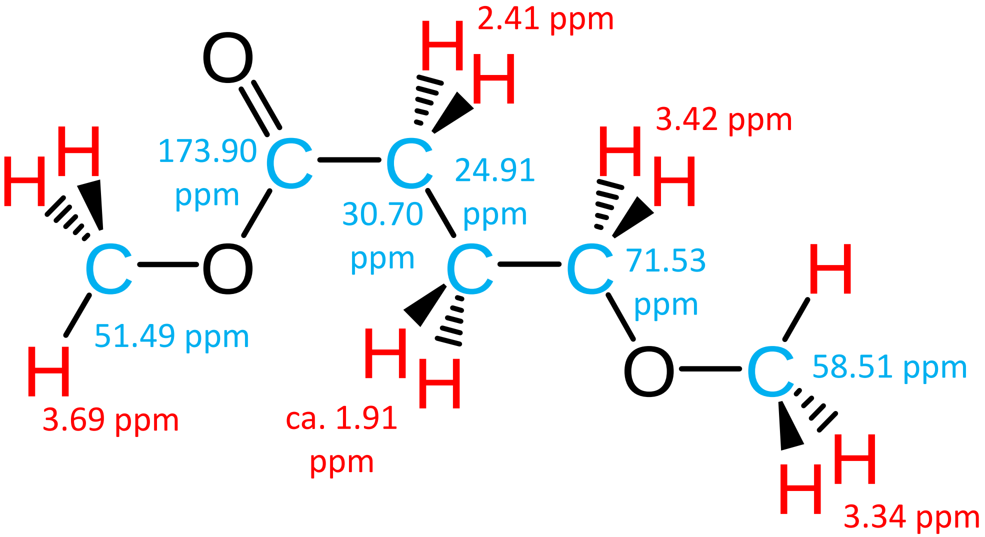
2.25 ppm (presented structure)

~~3.83 ppm~~ (alternative arrangement of –CO-O-)

Coupling constants

It looks simple, but it is not

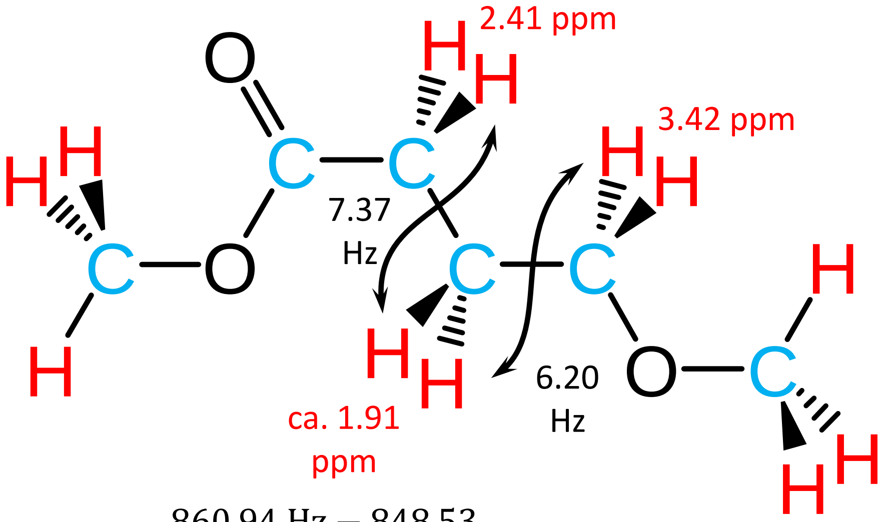
For the sake of clarity let us remove all carbon assignments and the proton assignments of both methyl groups. We don‘t need them anymore.



Coupling constants

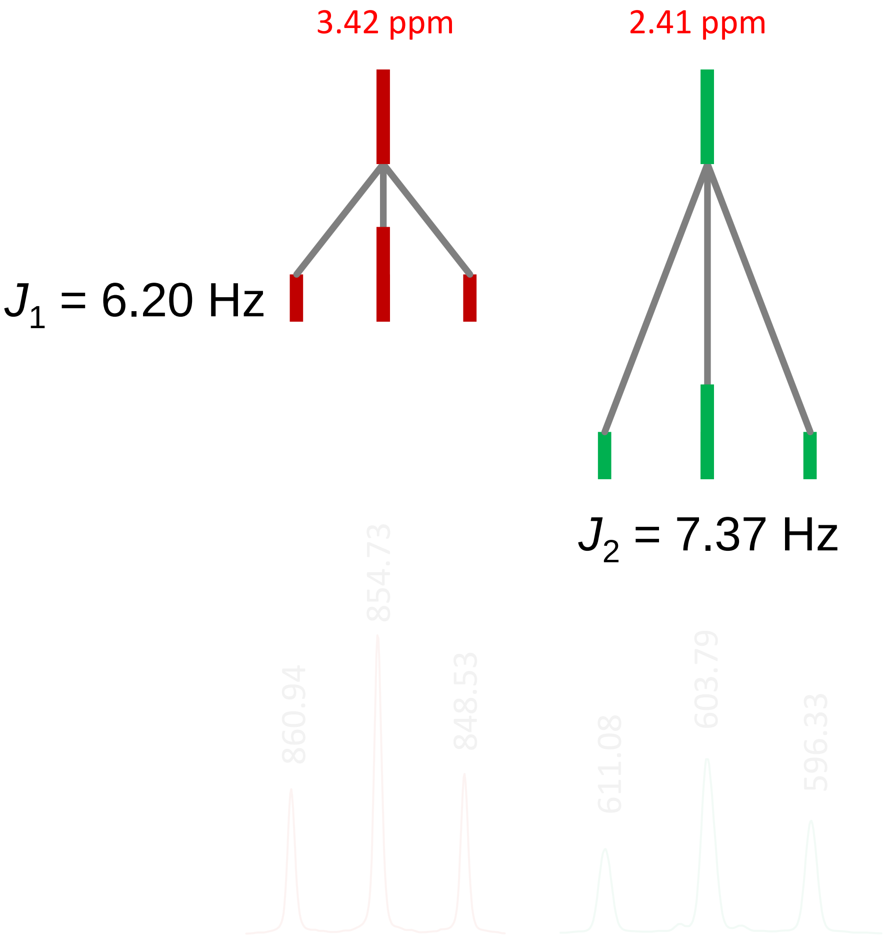
easy start

Both the chemically equivalent methylene protons at 2.41 ppm and 3.42 ppm have two chemically equivalent protons at about 1.91 ppm as the only vicinal coupling partners. We expect a triplet in both cases.



$$J_1 = \frac{860.94\ \text{Hz} - 848.53}{2} = \mathbf{6.20\ Hz}$$

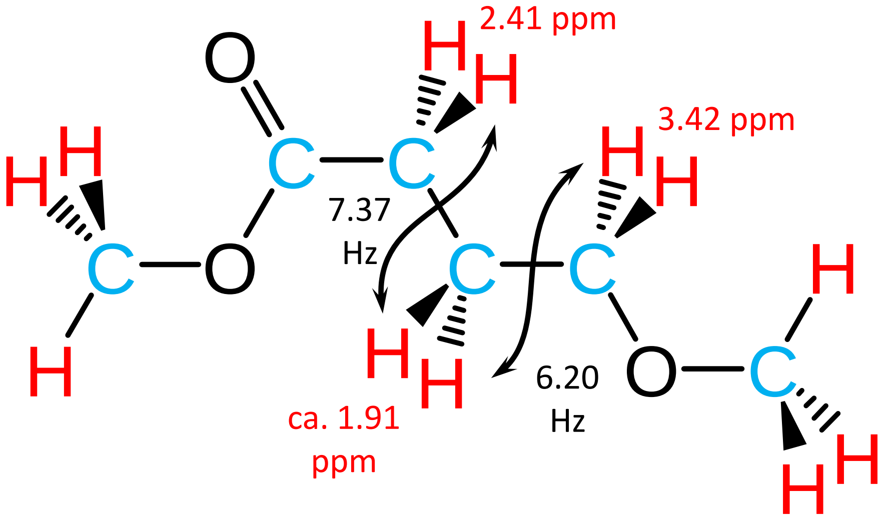
$$J_2 = \frac{611.08\ \text{Hz} - 596.33}{2} = \mathbf{7.37\ Hz}$$



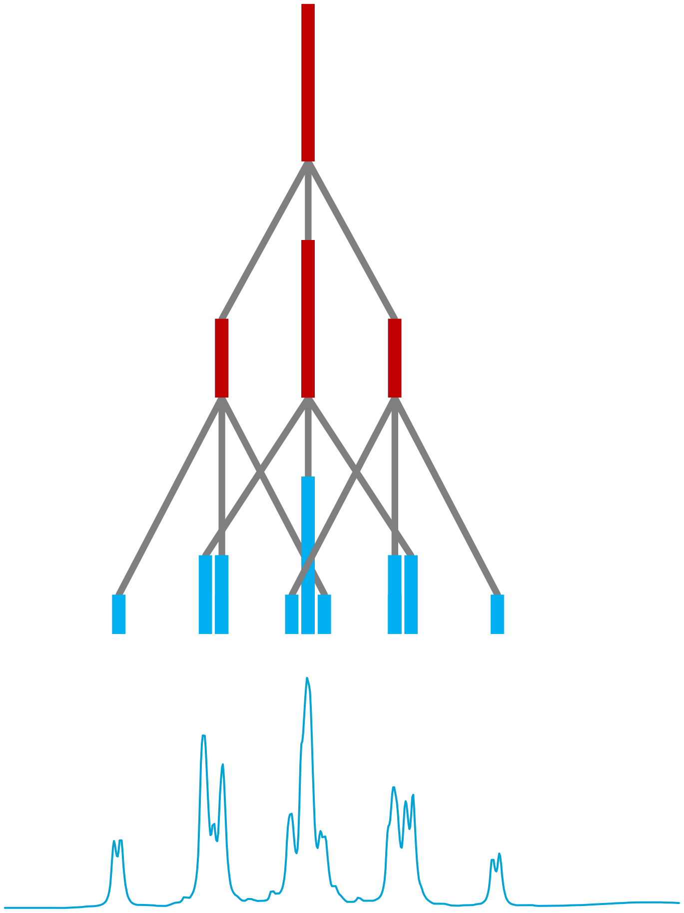
Coupling constants

done

For the methylene protons at 1.91 ppm we expect a triplet of triplets. Because 1.91 ppm and 2.41 ppm are not that different in size we expect very first signs of higher order, but in principle the triplet of triplets looks fine.



$$\frac{\Delta\delta}{J} = \frac{(2.41\text{ ppm} - 1.91\text{ppm}) * 250.13\text{ MHz}}{7.37\text{ Hz}} = \mathbf{16.97}$$



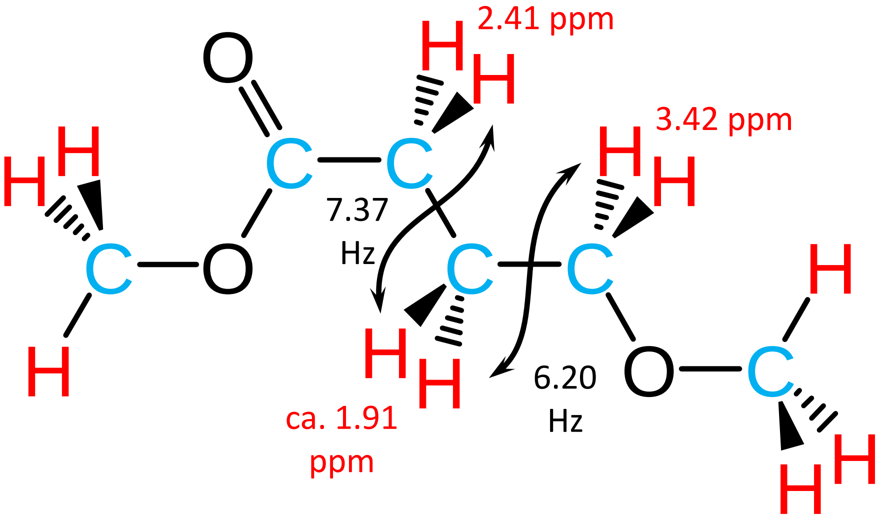
Coupling constants

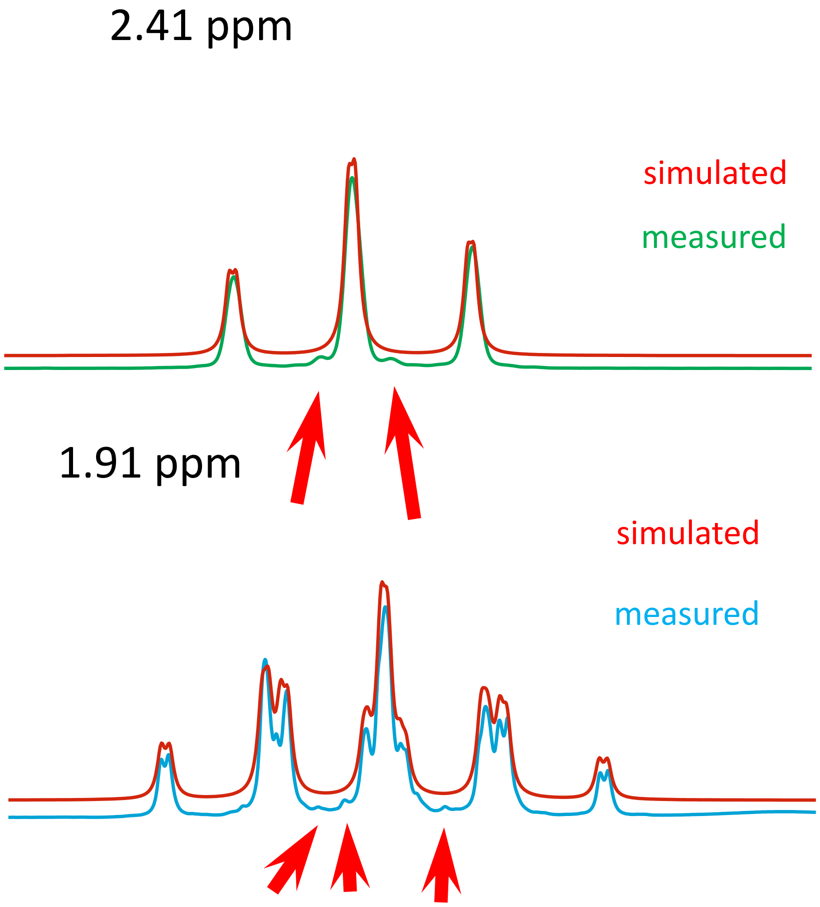
A last check

The simulation of two multiplets looks fine.

But … Have a closer look.

Some experimental details are missing in the simulation. Noise?

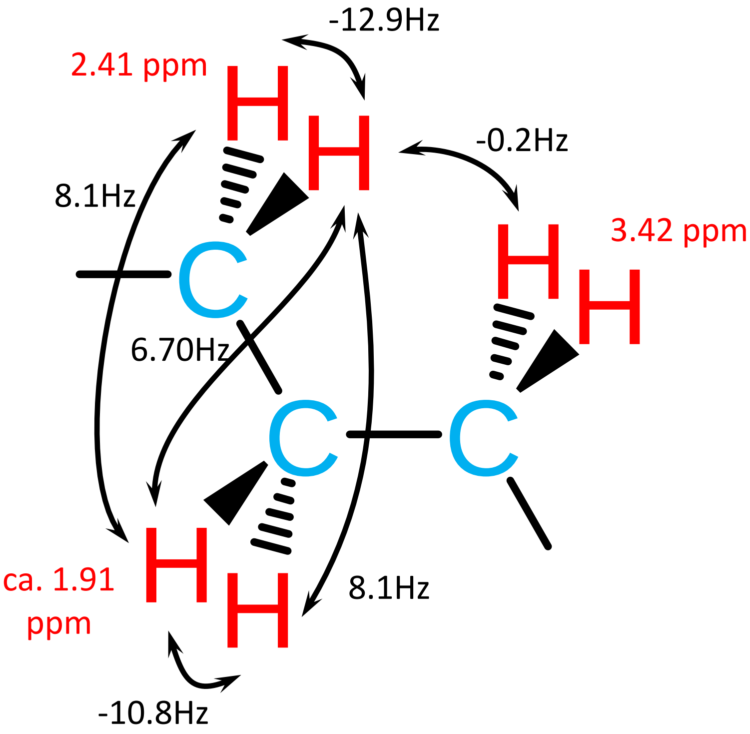




Coupling constants

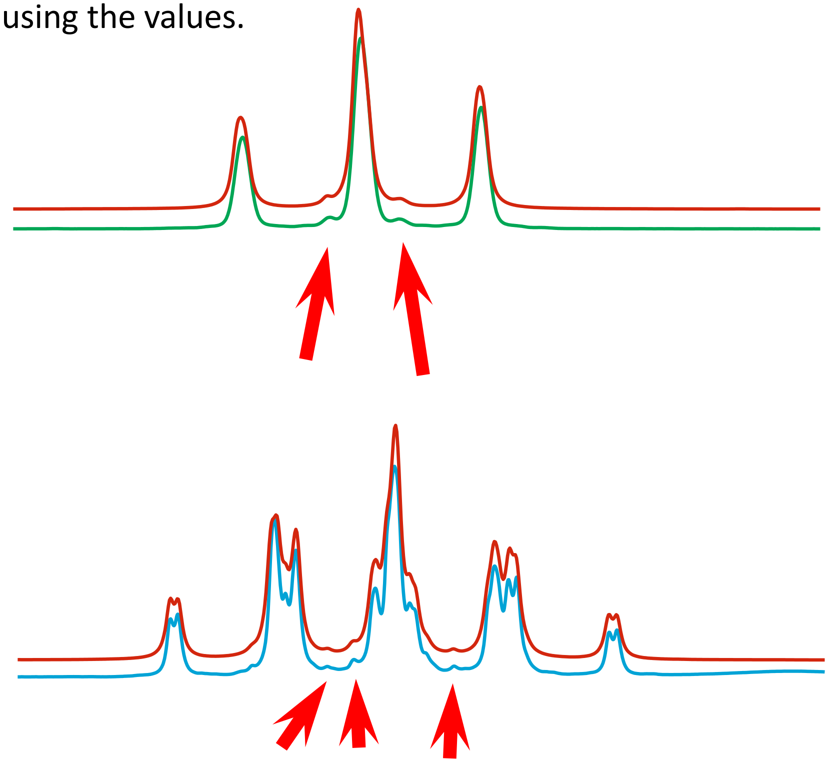
Some refinement

The second coupling pathway with a coupling constant of 6.70 Hz is not shown here for reason of clarity.



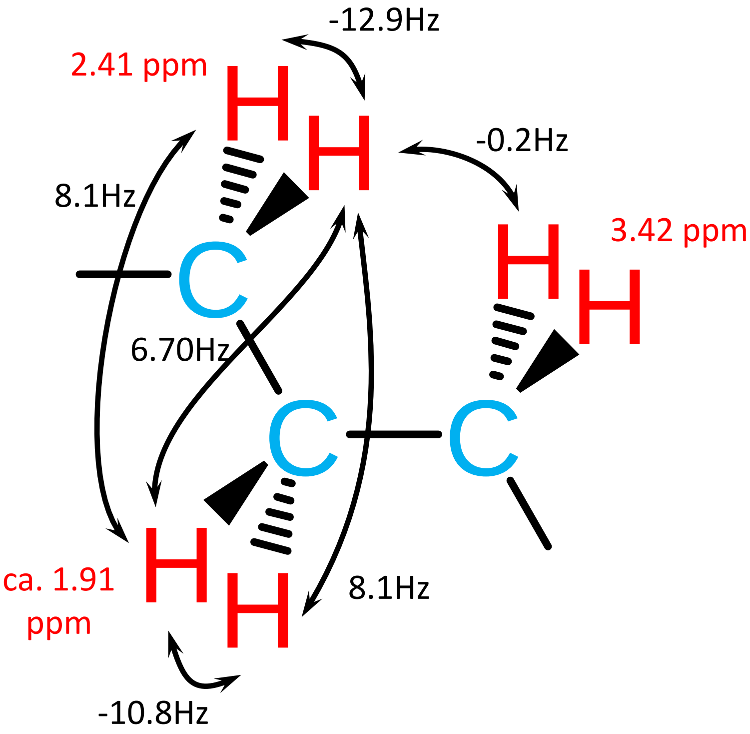
After changing some coupling constants and adding a long rang coupling constant the „warts“ become simulated nearly perfectly.

It is not possible to get these values from the spectra presented here, but you can repeat the simulation using the values.



Coupling constants

Some refinement



But …

Why should vicinal coupling constants between chemically equivalent protons have different values? There is the possibility of free rotation around the single bond between the carbon atoms. The vicinal coupling constants, of course, depend on the dihedral angle following the Karplus equation, but this effect should be averaged out by the fast rotation around all possible dihedral angles between 0 and 360 degrees.

Coupling constants

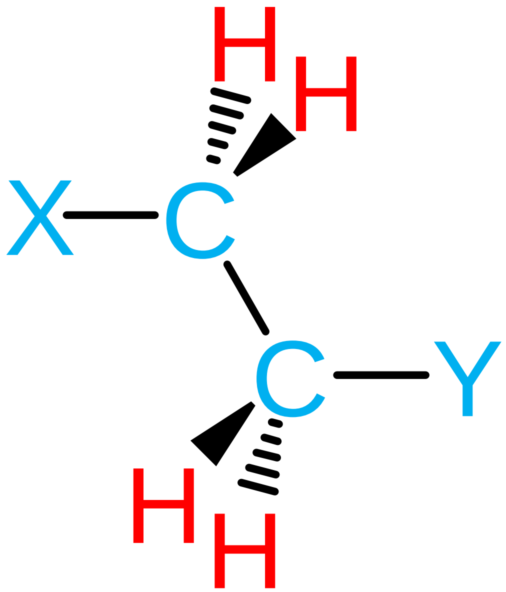
An explanation

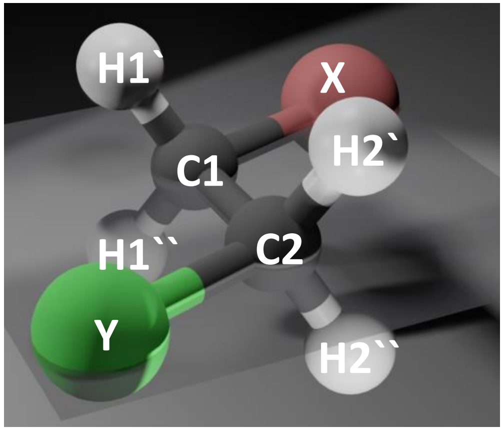
Let us reduce our molecule to a bisubstituted ethane derivative with two different substituents

X = -CO-O-CH$_3$

and

Y = -CH2-O-CH$_3$.

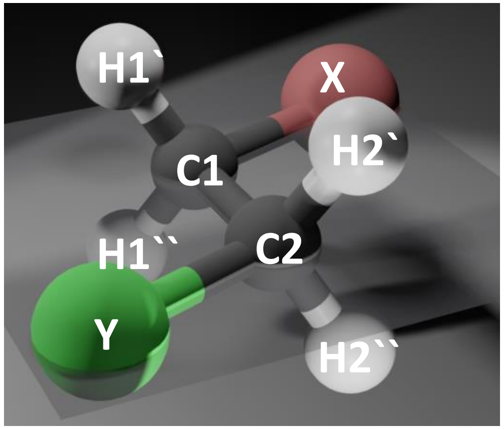




For the moment don‘t worry about the hydrogen atoms with four different labels shown here, although you expect that H1` and H1`` or H2`and H2`` should be equivalent.

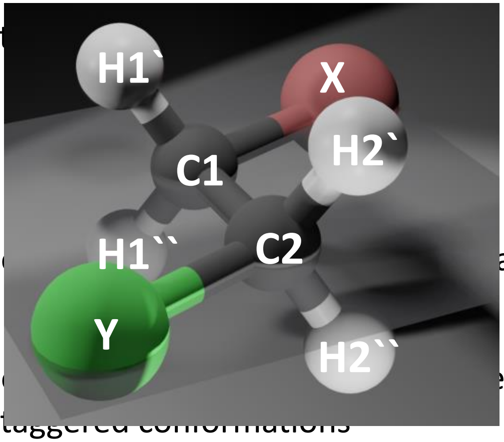
Symmetry

Are H2`and H2`` chemically equivalent?



If you see the static structure of our unsymmetric ethane derivative there seems to be no question.

There is a symmetry plane inside the molecule, which makes both **H1`/H1``** and **H2`/H2``** chemically equivalent.



But there is free rotat possible and the rotamer shown here

Let us introduce som around the **C1**-**C2** bond.

- The first assumpti steric hindrance will favour the three staggered conformations
- The second assumption is to assume bond rotation is so much faster than NMR dwell time and we are seeing the average of the three staggered rotamers.

These assumptions are only necessary to keep the mathematics simple.

Symmetry

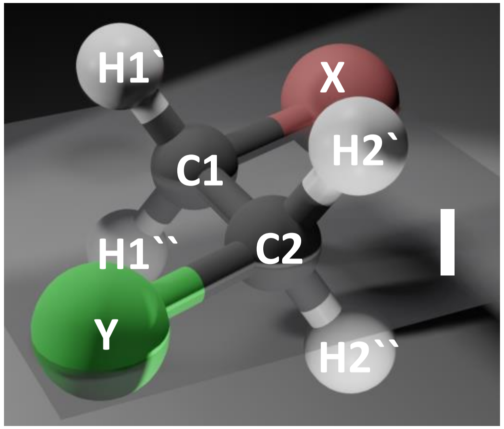
Are H2`and H2`` chemically equivalent?

First let us create the three rotamers (**I**, **II** and **III**)

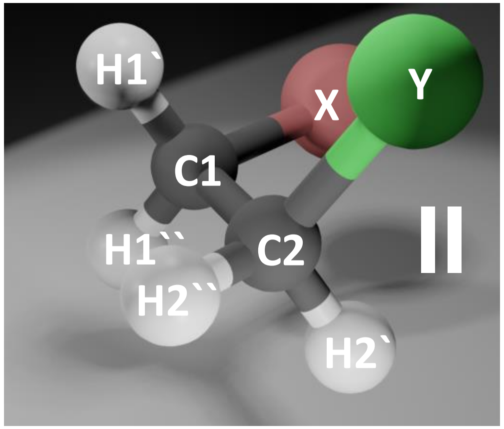
If we turn **C2** in rotamer **I** clockwise by 120 degree we get rotamer **II**.

Turning once more by 120 degree results in rotamer **III**.

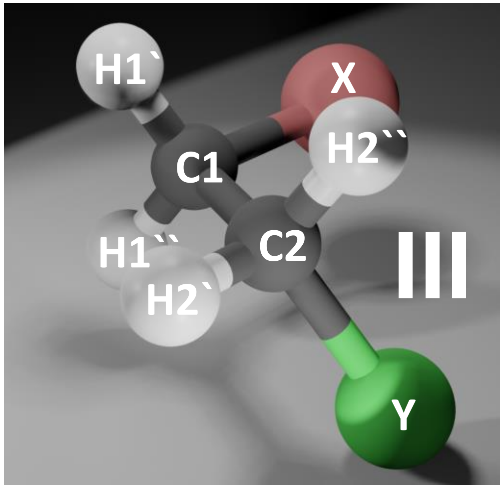
For rotamer **III** it is recommendable to change the viewpoint. Turn the whole molecule around the **C1**-**C2** bond by 180 degree and have a view to the molecule from the right side instead from the left side.



C2
120°

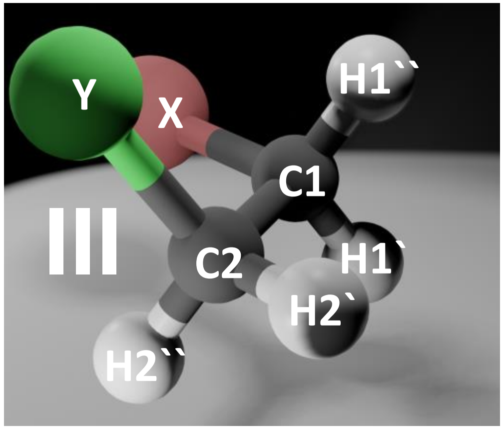


C2
120°



C1-C2 180°

change view
to right side



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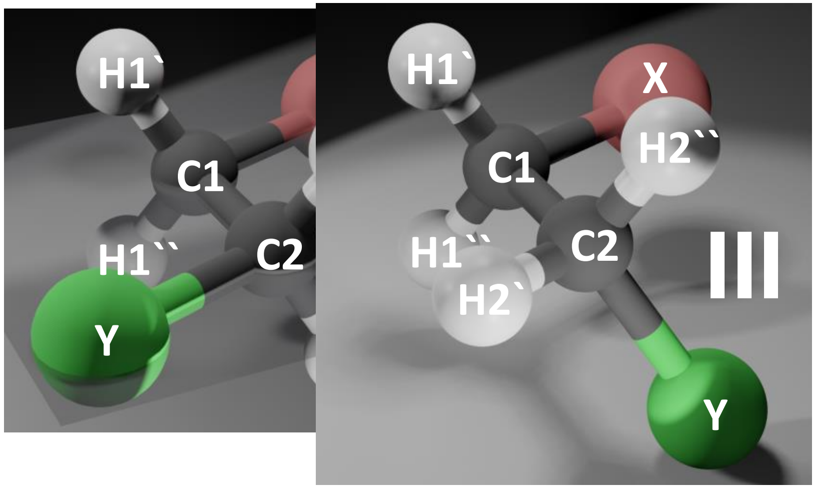
Symmetry

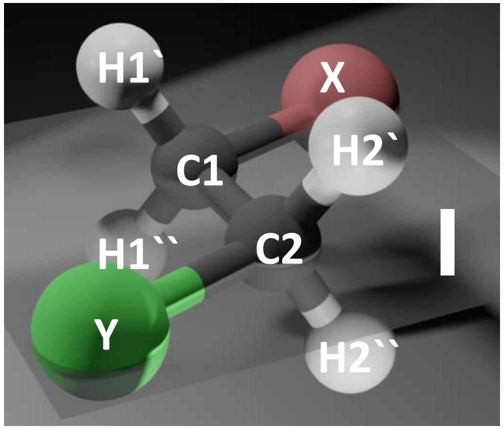
Are H2`and H2`` chemically equivalent?

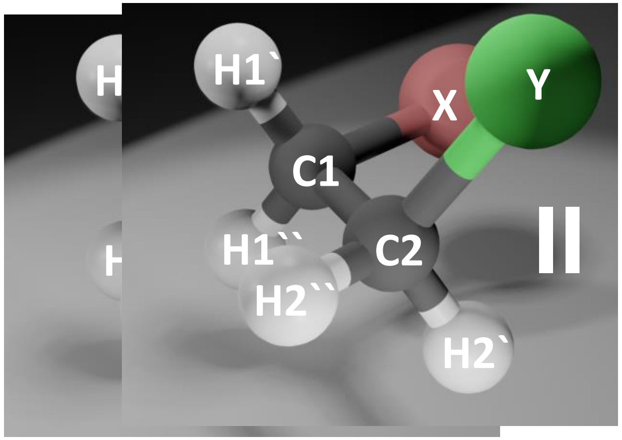
Let us reorder the three rotamers a little bit.

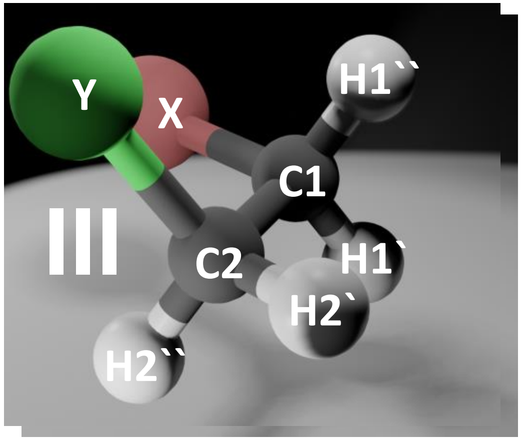
As you see there is a mirror plane inside rotamer **I** and no symmetry element inside the other two rotamers.

But on the other hand rotamer **II** and rotamer **III** are mirror and mirror image.





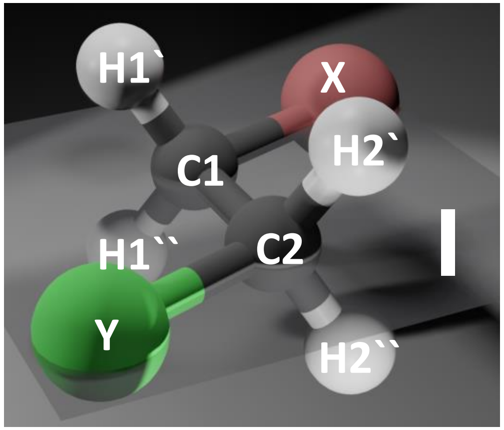


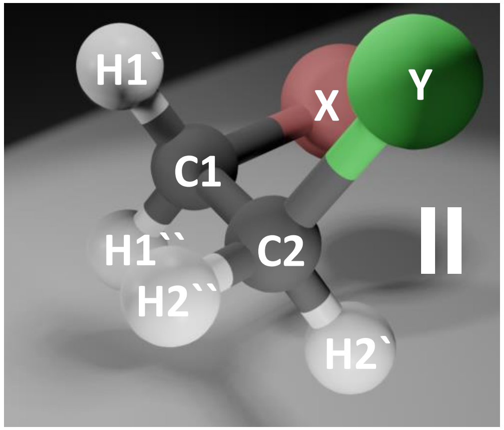


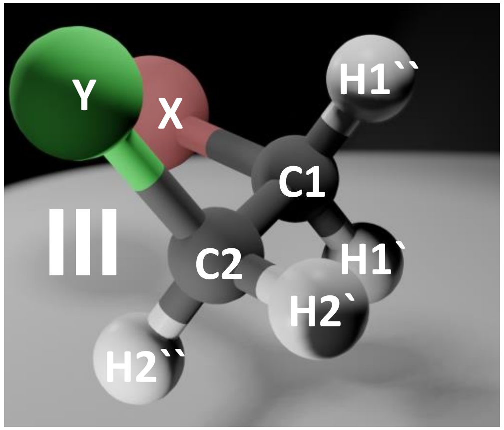
Symmetry

Are H2`and H2`` chemically equivalent?

And now let us paint the protons a little bit.







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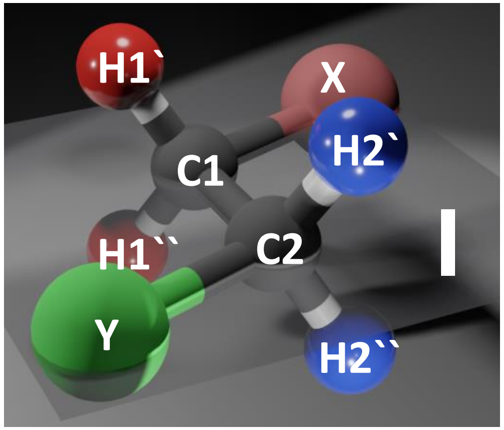
Symmetry

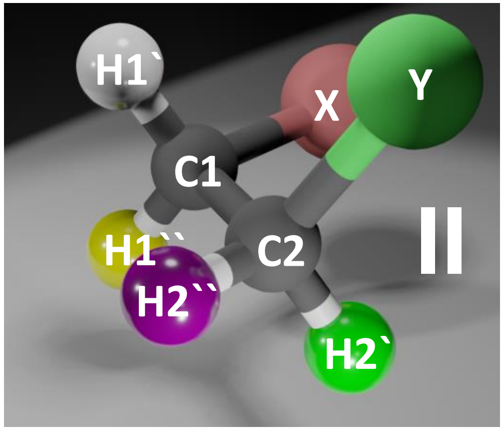
Are H2`and H2`` chemically equivalent?

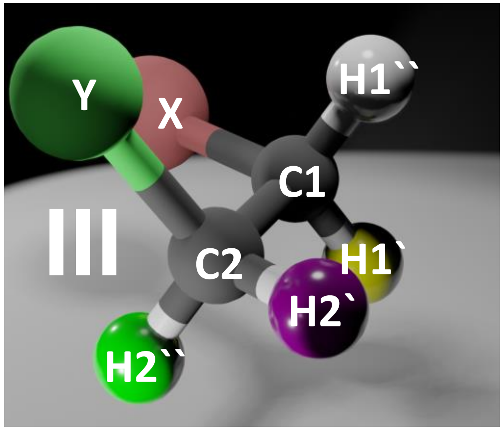
Different colours mean different chemical shifts, identical colours represent identical chemical shifts. Alltogether we have **six** different chemical shifts for the four protons inside the three rotamers.



As an example **H1`** and **H1``** in rotamer **I** are identical due to the internal mirror plane.

H2`` in rotamer **II** and **H2`** in rotamer **III** are identical, because rotamer **II** and rotamer **III** behave like image and mirror mage.





Symmetry

Are H1`and H1`` chemically equivalent?

The population of the rotamers is p_{I}, p_{II} and p_{III} with

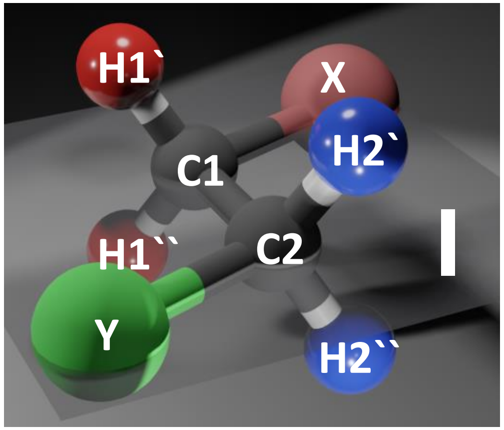
$$p_{II} = p_{III}$$

and

$$p_{I} + p_{II} + p_{III} = 1$$

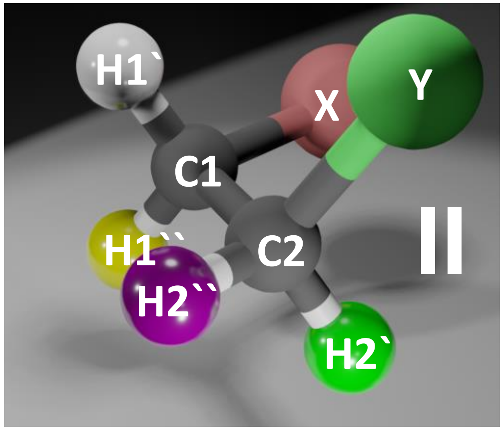
To keep the following equations short, we use single letters for the six different chemical shifts as follows:

$\delta_{H(red)}$ = **R**
$\delta_{H(blue)}$ = **B**
$\delta_{H(green)}$ = **G**
$\delta_{H(yellow)}$ = **Y**
$\delta_{H(purple)}$ = **P**
$\delta_{H(white)}$ = **W** *(you wouldn‘t see a white letter)*

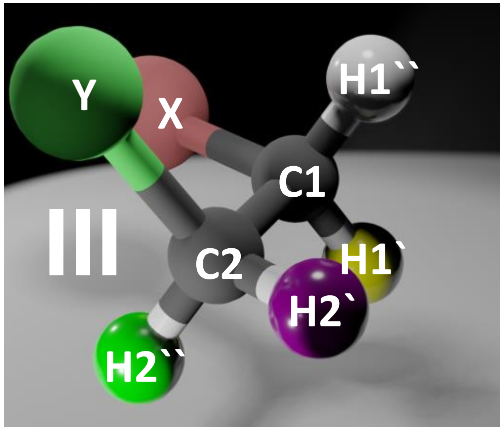


$\boldsymbol{p_{I}}$
(rotamer population)

$\boldsymbol{p_{II}}$



$\boldsymbol{p_{III}}$



Symmetry

Are H2`and H2`` chemically equivalent?

Now we get for the four protons

$$\delta_{H1`} = p_I * R + p_{II} * W + p_{III} * Y$$
$$\delta_{H1``} = p_I * R + p_{II} * Y + p_{III} * W$$
$$\delta_{H2`} = p_I * B + p_{II} * G + p_{III} * P$$
$$\delta_{H2``} = p_I * B + p_{II} * P + p_{III} * G$$

With the boundary condition

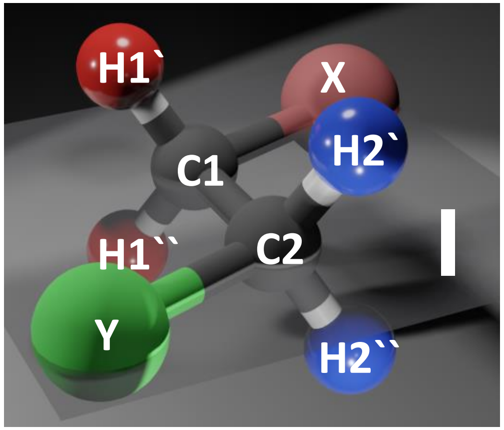
$$p_{II} = p_{III}$$

we get

$$\delta_{H2`} = \delta_{H2``}$$

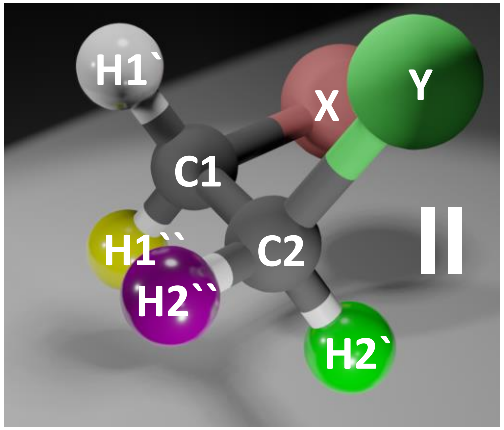
and

$$\delta_{H1`} = \delta_{H1``}$$

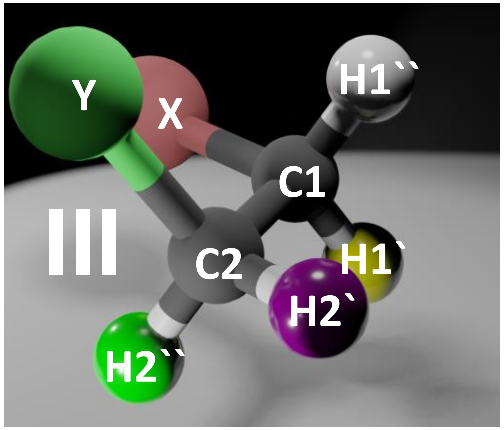


p_I
(rotamer population)

p_{II}



p_{III}



Symmetry

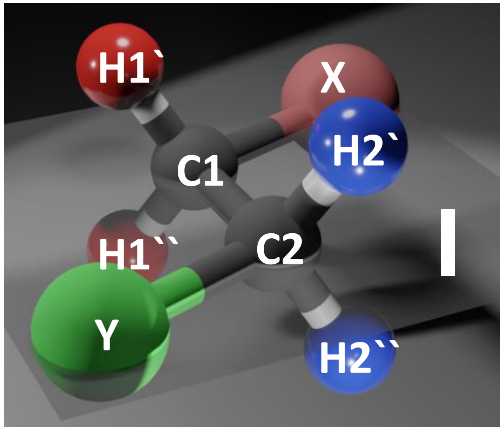
Are H2`and H2`` chemically equivalent?

We are finished with the result you expected from the beginning without all that difficult considerations.

But, just for your curiosity, try to repeat the calculation after replacing **H1``** with a third substituent **Z**, different from **X** and **Y**. In this case, there is no symmetry, no mirror plane inside rotamer **I** nor a mirror plane between the rotamers **II** and **III**.

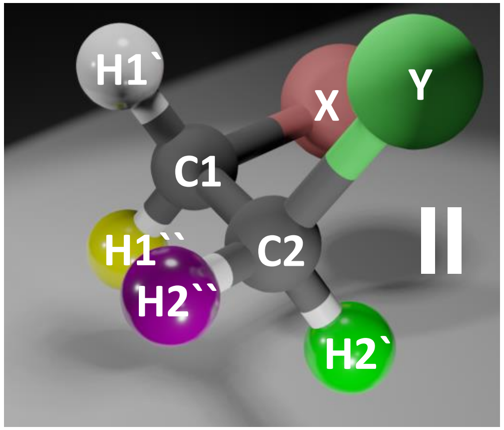
But beside this consideration let us return to the main question:

are **H2`** and **H2``** **magnetically** equivalent?

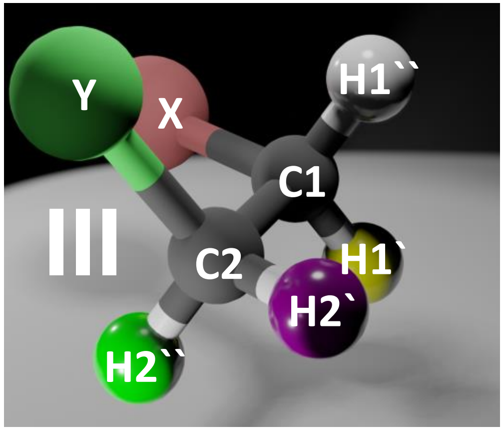


p_{I}
(rotamer population)

p_{II}



p_{III}



Symmetry

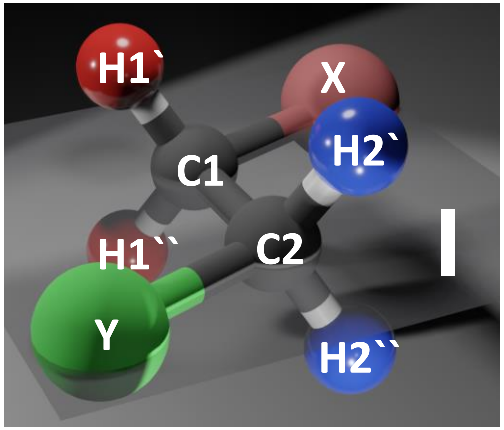
Are H2`and H2`` magnetically equivalent?

As we have seen, the protons **H2`** and **H2``** are chemically equivalent. They are magnetically equivalent as well, if the condition

$$ {}^{3}\boldsymbol{J}_{\text{H1`,H2`}} = {}^{3}\boldsymbol{J}_{\text{H1`,H2``}} $$

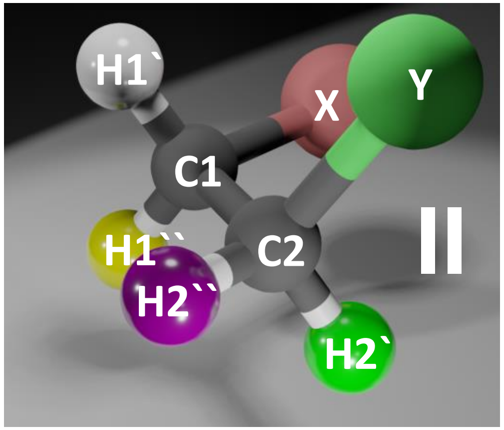
is fulfilled.
Of course the same has to be valid, if we replace **H1`** by **H1``** on both sides of the equation.

Let us see the geometric relations between **H1‘/H2`** and **H1`/H2``** one after the other for all three rotamers.

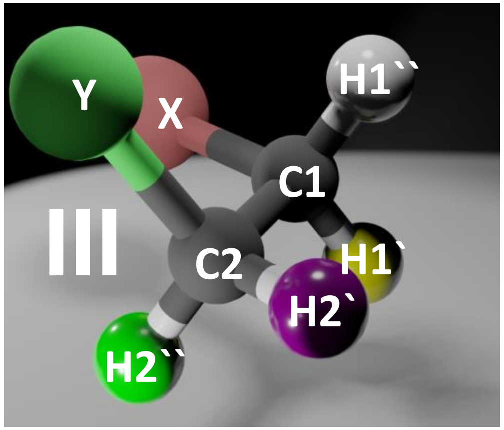


$\boldsymbol{p}_{\mathrm{I}}$
(rotamer population)

$\boldsymbol{p}_{\mathrm{II}}$



$\boldsymbol{p}_{\mathrm{III}}$



Symmetry

Are H2`and H2``
magnetically equivalent?

Let us start with the geometry between **H1`** and **H2`**. In all three rotamers **H1`** is labeled in black and **H2`** labeld in red.

We always have to focus on two planes. The first one is created from the atoms

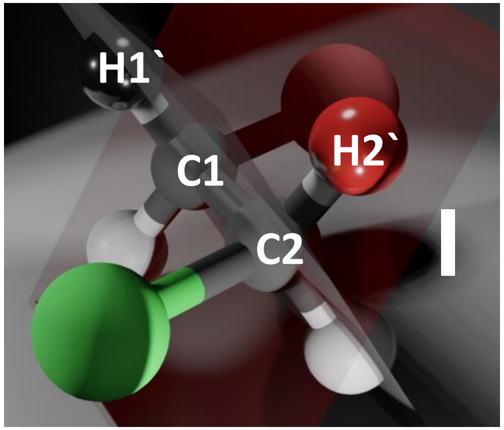
H1`, **C1** and **C2**,

the second one from the atoms

H2`, **C2** and **C1**.

The dihedral angles between these planes are

rotamer **I** – 60 degree
rotamer **II** – 180 degree
rotamer **III** – 60 degree

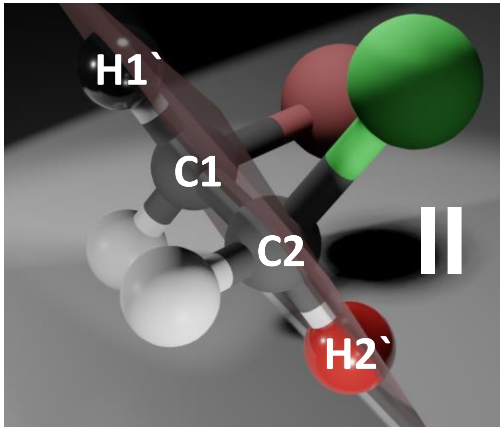


p_{I}
(rotamer probability)

dihedral angle
(H1`-C1-C2-H2`)
60°

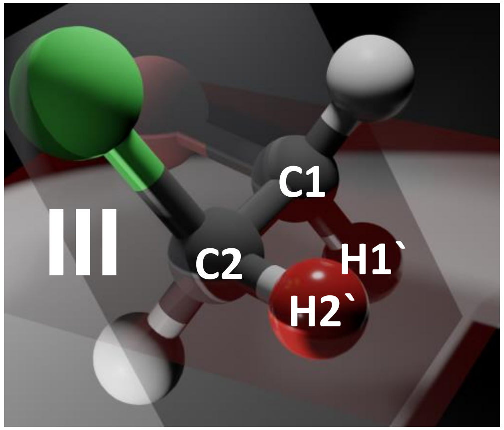
p_{II}

dihedral angle
(H1`-C1-C2-H2`)
180°



p_{III}

dihedral angle
(H1`-C1-C2-H2`)
60°



Symmetry

Are H2`and H2``
magnetically equivalent?

According to the Karplus equation, the vicinal coupling constant for a dihedral angle of 180 degree is significantly larger than the vicinal coupling constant in the case of a dihedral angle of 60 degrees.

Let us write for the coupling constants between **H1`** and **H2`**

$J_{L\text{(arge)}}$

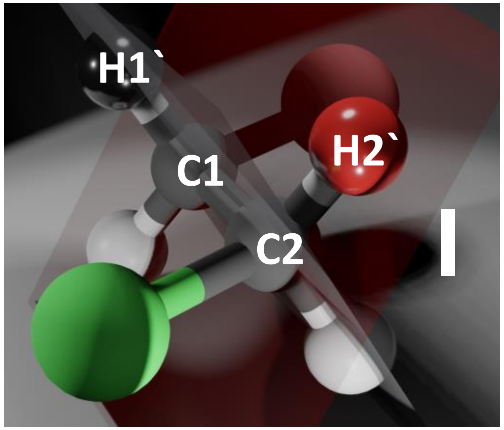
If the dihedral angle is 180 degree and

$J_{S\text{(mall)}}$

in the case of 60 degree.

$$J_{H1`,H2`} = p_I * J_S + p_{II} * J_L + p_{III} * J_S$$

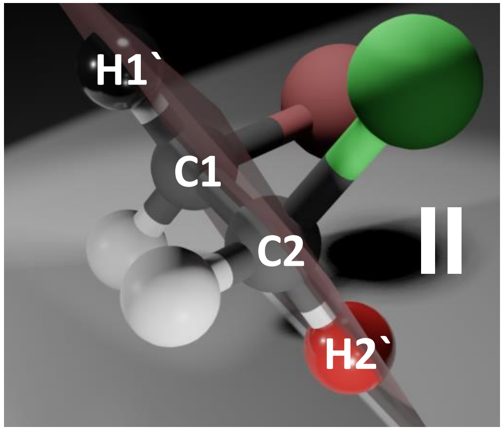
J_S



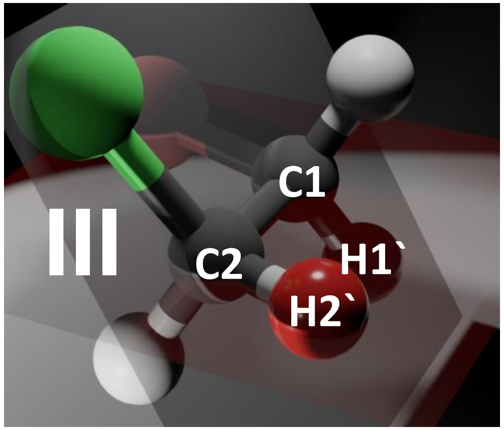
p_I
(rotamer
probability)

dihedral angle
(H1`-C1-C2-H2`)
60°

p_{II} dihedral angle
(H1`-C1-C2-H2`)
180° J_L



p_{III} dihedral angle
(H1`-C1-C2-H2`)
60° J_S

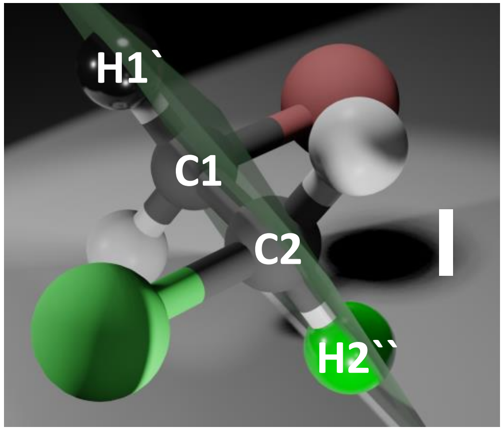


Symmetry

Are H2`and H2``
magnetically equivalent?

Let us repeat the same considerations for **H2``**, labeled in green.

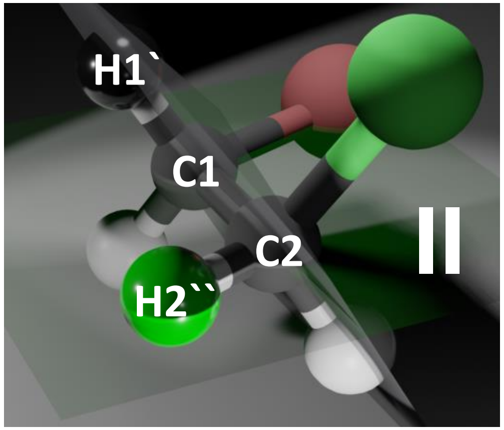
J_L



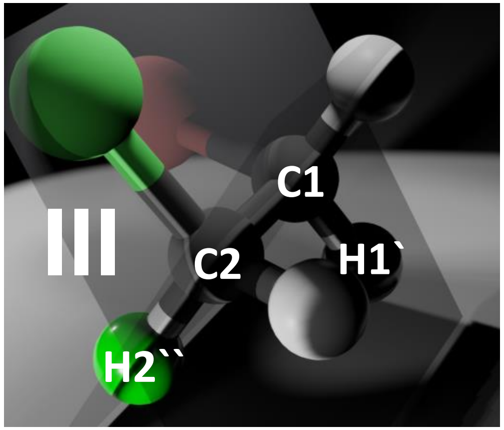
p_I
(rotamer probability)

dihedral angle (H1`-C1-C2-H2``)
180°

p_{II} dihedral angle (H1`-C1-C2-H2``) **60°** J_S



p_{III} dihedral angle (H1`-C1-C2-H2``) **60°** J_S



$$J_{H1`,H2``} =$$
$$p_I * J_L + p_{II} * J_S + p_{III} * J_S$$

Symmetry

Are H2`and H2``
magnetically equivalent?

Finally we have

$$J_{\text{H1`,H2``}} = p_{\text{I}} * J_{\text{L}} + p_{\text{II}} * J_{\text{S}} + p_{\text{III}} * J_{\text{S}}$$

$$J_{\text{H1`,H2`}} = p_{\text{I}} * J_{\text{S}} + p_{\text{II}} * J_{\text{L}} + p_{\text{III}} * J_{\text{S}}$$

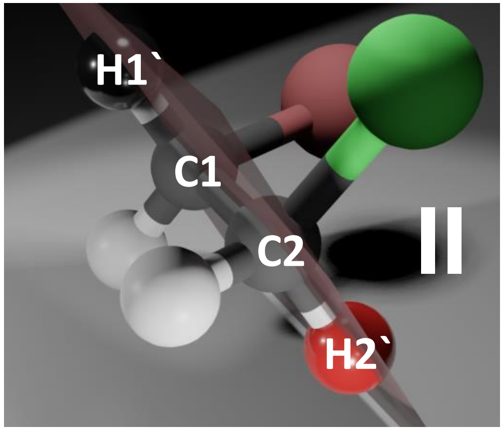
Now, please keep in mind the already know relations ($p_{\text{II}} = p_{\text{III}}$ and $p_{\text{I}} + p_{\text{II}} + p_{\text{III}} = 1$) and play around a little bit with the population of rotamer **I**. Start with $p_{\text{I}} = 0.333$.

You will see, how both coupling constants depend in opposite direction from the population p_{I}.

But even in the case of $p_{\text{I}} = p_{\text{II}} = p_{\text{III}}$ the coupling constants are only identical by chance.

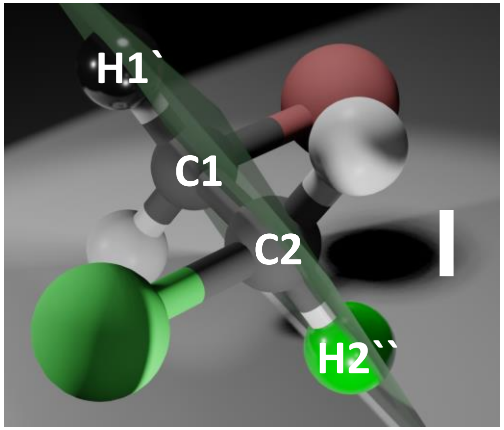
dihedral angle
(H1`-C1-C2-H2`)
180°

J_{L}



dihedral angle
(H1`-C1-C2-H2`)
180°

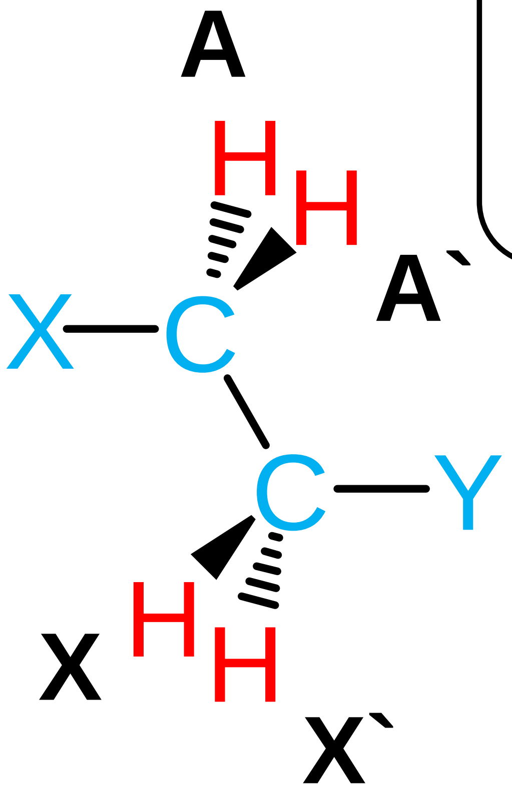
J_{L}



We made some simplifications. Indeed, no pair of the six rotamers result in identical coupling constants. As an example see the environment for the two rotamers with dihedral angles of 180 degree between the coupling protons. In spite of an idential dihedral angle the coupling pathway is clearly different.

Symmetry

Conclusion



As soon as an asymmetrically substituted ethane is recognized as a structural fragment within an *achiral* compound, the methylene protons of this ethane fragment are **always** chemically equivalent and **always** magnetically non-equivalent.

Why *achiral*?

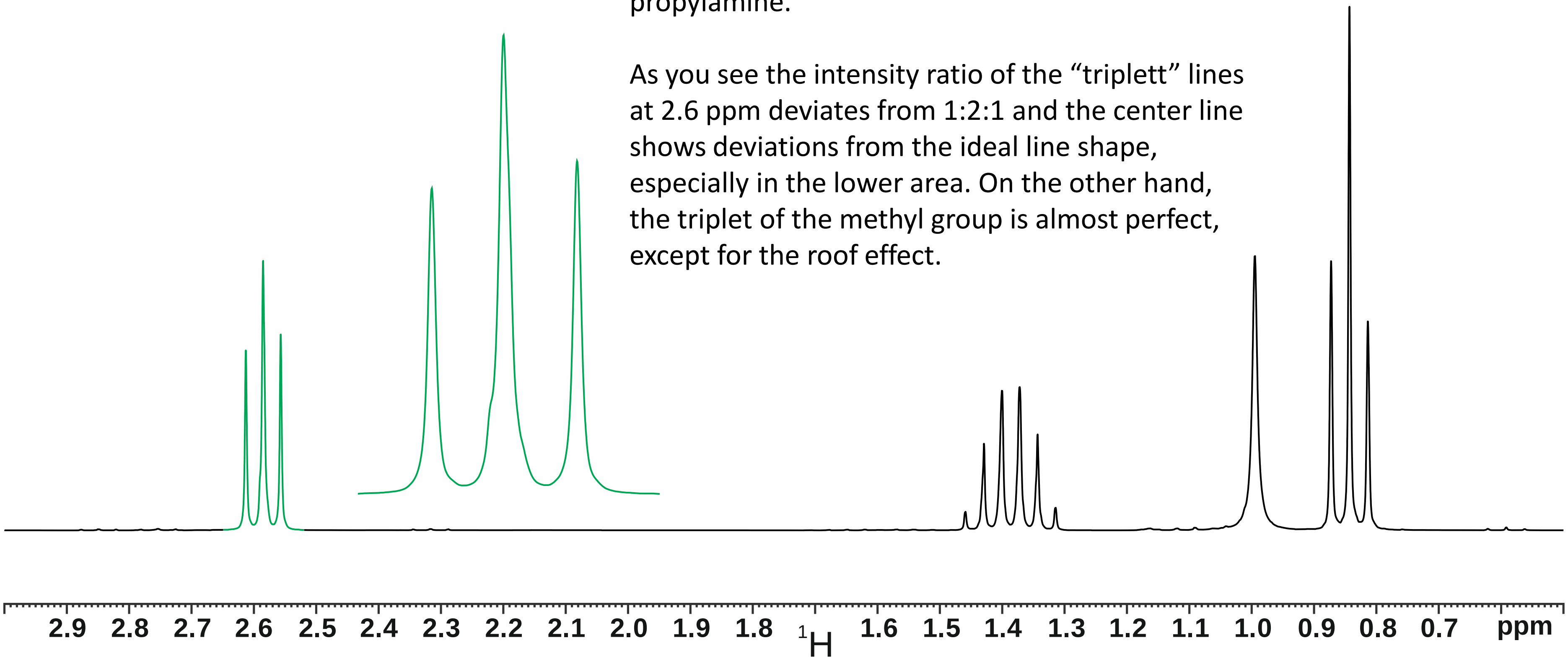
That‘s very simple. Within chiral compounds the methylene protons are chemically non-equivalent, which means, the question of magnetic equivalence doesn‘t appear.

Symmetry

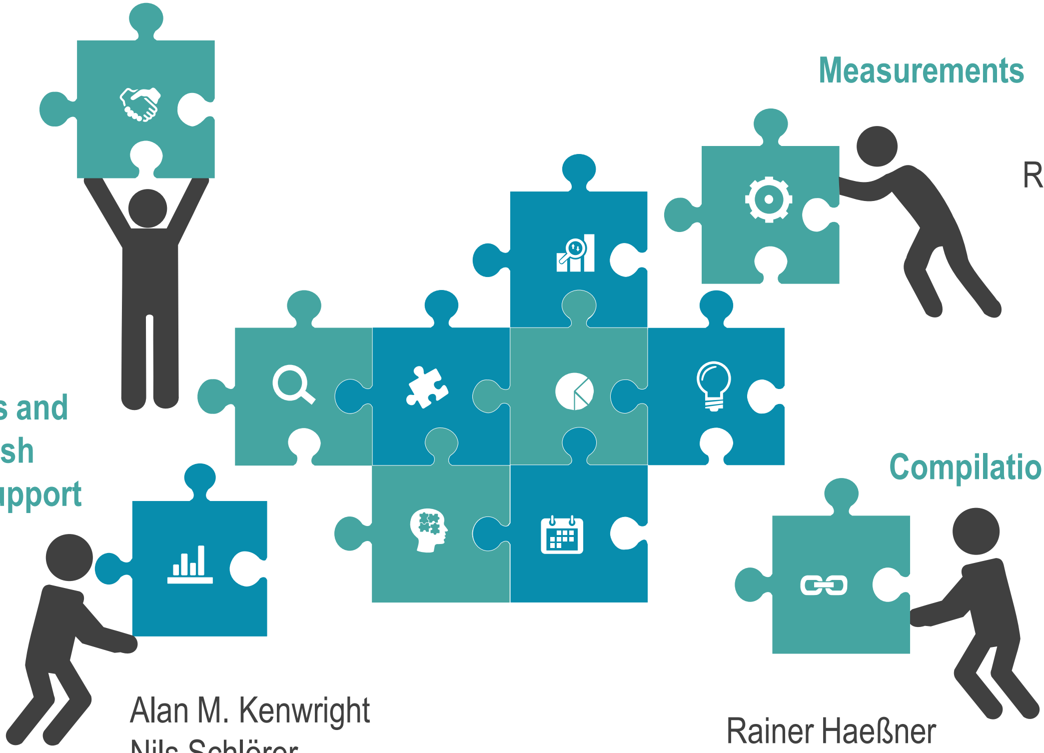
Keep your eyes open

Magnetic non-equivalence in alkyl chains is often not visible at a first glance. But with open eyes, you can see the effect almost everywhere, such as for example in the methylene group of propylamine.

As you see the intensity ratio of the “triplett” lines at 2.6 ppm deviates from 1:2:1 and the center line shows deviations from the ideal line shape, especially in the lower area. On the other hand, the triplet of the methyl group is almost perfect, except for the roof effect.



Contributions



Spectrometer time

TU Munich

Measurements

Rainer Haeßner

Discussions and native English language support

Alan M. Kenwright
Nils Schlörer

Compilation

Rainer Haeßner

Contributions

Some special thanks.

This *problem of the month* is the result of an exciting discussion within the AMMRL mailing list.
It is not possible to mention all of the valuable feedback here - sorry - but some special contributions should be mentioned, I believe.

Svetlana Simowa provided an easy to understand explanation.

Novruz Akhmedov extracted the coupling constants from the raw data.

Hsin Wang contributed some text building blocks for the explanation using only a few words to focus to the essential details.

Karel Klika pointed out, that there is no perfect average at all even in the case of idential population of all three rotamers.