One and Two dimensional NMR Spectroscopy: Concepts and Spectral Analysis

Prof. N. Suryaprakash

CSIR Emeritus Scientist, Solid State and Structural Chemistry Unit
Indian Institute of Science – Bengaluru

Lecture 24 13C-NMR-I

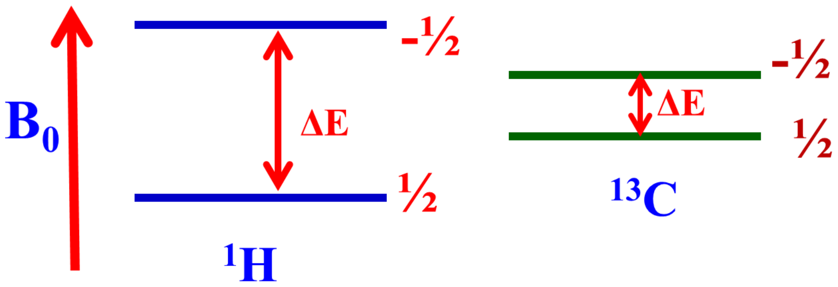
Welcome all of you. In the last couple of classes, we discussed extensively about the analysis of the proton NMR spectrum. We took various examples starting from simple molecule a CH2D2, we simulated the spectra for different nuclei, then we went to the real spectrum of molecules starting with small molecules simple spectra and went up to little bit complex spectra. We knew how to analyze the spectra based on chemical shift, multiplicity pattern and also intensity of the peaks, we understood all those things. Also we looked at the spectra of molecules containing phenyl groups and I explained to you about the multiplicity pattern that one expects in phenyl groups depending upon the substitution, whether it is a mono substituted, disubstituted, symmetrical asymmetrical disubstituted, ortho metta or para substitution, etc. various examples we took. And I showed you how the multiplicity patterns can be explained based on the substitution at the place where substituents are present in the phenyl ring.

Subsequently, we also analyzed variety spectra where we could determine heteronuclear coupling between proton and the abundant spins like fluorine and phosphorus, not only single heteronuclear sometimes where two heteronuclei are present. We extended further and analyzed the spectrum of molecules where some of the nuclei like carbon 13 and nitrogen 15 are present in the natural abundance. Such molecules will give rise to satellites spectra when protons are coupled to dilute spins like carbon 13, nitrogen 15, silicon, selenium, etcetera. And we could take a large number of such examples and we also analyzed such spectra and we knew how we can get the heteronuclear couplings from the satellite spectrum.

Further, in some symmetric molecules where the protons are chemically equivalent, the couplings are not reflected in the spectrum. In which case we wanted to get the coupling from the satellite spectrum. I also took an examples, not one several examples on how the proton-proton couplings are reflected in the satellite spectrum. The analysis of satellite spectrum also gives you not only heteronuclear coupling, but also proton-proton couplings. We also extended further and we wanted to see what will happen when we have magnetically equivalent spins. We do not get the coupling especially when the spins are magnetically equivalent. How do we get that? By what is called the isotopic substitution. I showed you how we can do isotopic substitution. For example, replacing proton by deuterium we get the heteronuclear coupling, ie. proton and deuterium. If I know JHD then I can calculate JHH by knowing the ratio of the gamma of proton over gamma of deuterium. This I explained to you and it is very easy in magnetically equivalent spins, where the couplings are not reflected in the spectrum. This is the way we can get the coupling by isotopic substitution. So, I explained all those things. Now, extensively we analyze proton NMR spectra, a lot of molecules and also further I showed you what happens when there is a complexity, we can do the decoupling. Selectively we can decouple particular nuclei and simplify the spectrum to get the coupling information. With all those things I think we have got sufficient information and I exposed quite a bit about the analysis of the spectra of proton. Now that you are going to be very comfortable in getting the information, we will further go further today and start working on carbon 13 NMR spectral analysis.

We look for carbon 13 NMR. Of course, carbon 13 is another NMR active nuclei which is extensively utilized, especially by organic chemists, because in most of the organic molecules carbon is invariably present. When I talk about the carbon we have several isotopes of carbon. Carbon 12 and carbon 13 are naturally occurring that is important for us. Carbon 14 is radioactive we do not have to worry about it. And if I look at the natural abundance, carbon 12 is 98.9 percent abundant whereas, carbon 13 is 1.1 percent abundant. Very interestingly the spin for carbon 12 is 0. That means, carbon 12 is NMR inactive, whereas, carbon 13 with a spin half is NMR active. And this is very important thing for us because if carbon were to be 100 percent abundant then our proton spectra in many of the organic molecule would have become extremely complex to get the information. So, nature has in a way helped us. So, that by making carbon 13 abundant 1.1 percent and though it participates in the coupling, but it does not complicate the proton spectrum. So, these are the general information. As for salient points of the carbon 13 NMR is concerned whatever the factors which affect the chemical shift of protons do affect 13C chemical shifts. We analyzed the chemical shifts of proton spectra; there are several factors I said steric factor, isomeric effect, and also the electronegativity, substituent effects, aromatic ring current various factors contribute to chemical shift.

All the factors which affect proton chemical shifts also affect the carbon 13 chemical shifts. That is another important point. So, that is the thing we should remember and the chemical shifts we are going to reference it with respect to tetramethylsilane in proton. Same reference is used for carbon 13 also. And the interesting point we should know is we discussed this when we explained about the chemical shift. Resonating frequency of proton is 4 times more than that of the carbon; or in other words resonating frequency of carbon is 4 times less than that of the proton. Meaning if I have an NMR spectrometer which is resonating at let us say 200 MHz, where does proton resonate in that spectrometer? and where does carbon resonate? Carbon resonates at 50 MHz with the proton is resonating at 200 MHz. The carbon is one fourth smaller than that because carbon gamma is 4 times smaller than that.



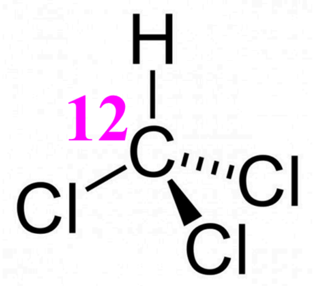
$$(\Delta E)^{1H} = 4\ (\Delta E)^{13C}$$

This also we discussed extensively even in the first or second class itself. Now, coming to the chemical shift range of carbon 13 NMR spectrum. In proton we discussed 0 to 10 ppm is the general scale and I also said it can go beyond TMS and also beyond 10 to 12 or even 15, 20 ppm in extreme cases. But by and large the range of chemical shift of proton is fixed it is between 0 to 10 or 12 ppm. Then what is the range of chemical shift of carbon 13? That is 0 to 300 ppm with respect to TMS; that is the range of chemical shifts of carbon 13. And we know different functional groups where do they resonate in the proton spectrum; similarly like where do they resonate in the proton spectrum there is also the table which is given in this chart you can see. The different functional groups where do they resonate. For example, if you have an aromatic it may come around, let us say 100 and 120 or 130 ppm, aliphatic, CH3, CH2, CHs will come from 0 to 40 or 50 ppm like that we know. Exactly whatever you see in the case of proton, the same thing can be more or less translated to carbon 13. Of course, slight modifications could be there, but not extremely different. It is more or less you can translate the chemical shift information what you do for proton also to carbon 13. And of course, salient points for carbon 13 if you look at it one thing is, always carbon 13 spectra are obtained with what is called broadband decoupling of protons. When we look at the carbon 13 usually all the protons that are coupled to carbon are completely broken; no coupling information will be there. We will remove the coupling of all protons to all the carbons. This is called broadband decoupling. This is a very important point otherwise spectrum would have been very complex.

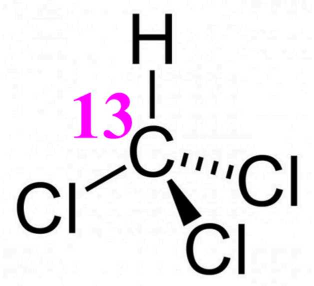
The result of that is, each peak is corresponding to a individual carbon, any chemical inequivalent carbon present in the molecule. So, if I have 10 peaks in the NMR spectrum of carbon 13; there will be 10 carbons in the molecule. That is all; very easy, each peak identifies a particular carbon in the molecule because of broadband decoupling. And if you continuing further the number of peaks always indicates number of chemically inequivalent carbons in the molecule. For example, there may be 10 carbons, but you may get 8 peaks that means, there are only 8 are chemical inequivalent; 2 of them may have chemical equivalence and they will be overlapping. So, similar to proton, number of peaks in the carbon 13 spectrum tells you the number of chemically inequivalent carbons present in the molecule. One point you must always remember, I said all protons are completely broadband decoupled from carbon, that is fine, we are removing only carbon proton couplings, but we are not removing the couplings of carbon with other heteronuclei. That couplings are still reflected in the spectrum.

Of course, nowadays in the present day spectrometers you can do the decoupling of 2 nuclei, or 3 nuclei at a time, that is a different question. But by and large generally when you record the carbon 13 spectrum, all protons are decoupled. Whereas, coupling of other nuclei with carbon are still present that gets reflected in the spectrum. With this let us now talk about the sensitivity of carbon 13 relative to that of proton. We already discussed about the sensitivity, I told you gamma of the carbon is 4 times smaller. As a consequence energy separation is 4 times smaller and I correlated this energy separation with the population using the Boltzmann distribution. I told you earlier, that means, the population difference between 2 energy states is much smaller here; as the energy separation becomes more and more the population becomes larger, that is what I said. So, as a consequence its sensitivity is 4 times less. But I also said gamma depends upon 3 other factors, it is gamma cube; it is 4 cube. So, as a consequence what is going to happen is, if you go to the sensitivity of the carbon 13 relative to proton, when proton resonates at 800 megahertz carbon resonates at 200 megahertz; and the energy gap is 4 times smaller, the sensitivity will be 4 into 4 cube times smaller. In addition, it is 1% abundant.

So, as a consequence it turns out to be 5672 times less sensitive compared to that of proton. Remember if I have a sample, keeping every parameters identical; including relaxation time, varieties of things, and if I take 1 minute to get a proton spectrum; in reality I assume that everything remains same, I must get I must take 5672 minutes, much more time consuming; that is because the sensitivity of the carbon is much much smaller. In fact, if I have to get the signal to noise ratio I have to take the square of this number 5672 X 5672. As a consequence, it is really time consuming; the sensitivity is very very low. Thus carbon NMR in a way more time consuming than proton NMR please remember this point. With this I want to introduce one parameter which already I discussed indirectly, in some one or two classes. Let us talk about isotopomers of the molecules. Let us consider an example of $CHCl_3$. In $CHCl_3$ there is only one carbon. And carbon has two isotopes; already I told you; carbon 12 and carbon 13. The carbon 12 is 99% abundant, and carbon 13 is 1% abundant that is what I told you. That means, if I have 100 molecules, only one molecule will be like this with carbon isotope as present as carbon 13.



99 molecules
NMR Inactive



1 molecule
NMR Active

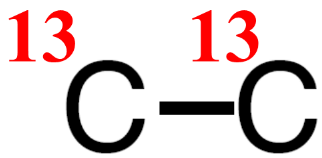
The 99 molecules will be present as carbon 12. Remember 99 molecules will be in the carbon 12 state, and only one molecule will be in its carbon 13 state. Consider isotopomers when two carbons are present in the molecule. Let us consider the example, let us say what happens if I have two carbons present in the molecule. We can think of possible combinations there is a possibility. I am taking a hypothetical molecule do not worry about other substituents of this molecule, an hypothetical molecule with the two carbons chemically inequivalent bonded by a single bond like this.

$^{12}\mathrm{C}-^{12}\mathrm{C}$ **98.9 x 98.9 (97.8%)**
NMR Inactive

$^{13}\mathrm{C}-^{12}\mathrm{C}$ **1. 1%**
NMR Active

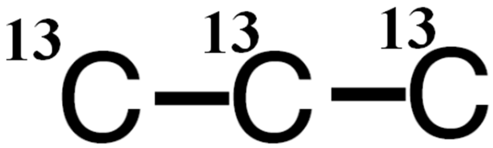
$^{12}\mathrm{C}-^{13}\mathrm{C}$ **1.1 %**
NMR Active

There is a possibility and large possibility, that this carbon this carbon both can be in carbon 12. Well probability is more because each carbon has 99% abundance; and the possibility of this is 97.8% of the time you will have this type of molecules. Unfortunately such molecules are NMR inactive it would not give you the NMR spectrum. So, but unfortunately they are present in the large quantity in the given molecule. This type of isotopomers is present almost 98%. Now, think of other possibility; one more possibility is this carbon can be carbon 13 and this can be carbon 12. Then what is the probability? only 1.1%. And this is NMR active spin. Think of the other possibility, this is carbon 12 and this is carbon 13. What is the probability of seeing this? what is the abundance of this? only 1.1% carbon is abundant, this is anyway 99% ruled out. So, it is 1.1% this is also NMR active. What is the last possibility you can think of both the carbons are present as carbon 13.



.000121 %
NMR Active

What is the probability of seeing both the carbons present as carbon 13 in the molecule? each one has a probability of 1/100. Because it is 1.1% abundant, forget about 1.1 make it 1%. So, is 1/100. The probability of both of them present jointly or simultaneously as carbon 13, is a joint probability it is 1/100 x 1/100. It is 1/10000. That means, the percentage of abundance of this type of molecule is 0.000121%. But this is NMR active; because both are carbon 13, and such type of molecules are extremely difficult to detect, because this already you know the abundance is so small; and this joint probability is 0.000121%. Extremely difficult to see such molecules and get the spectrum from such molecules. So, when there are two carbons present already you have lot of complications.

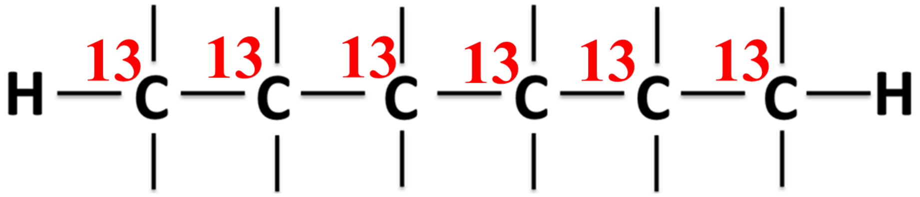


Probability of all carbons simultaneously present as ^{13}C

1/100 x 1/100 x 1/100 = 0.000001331

Neary one in a Million !!

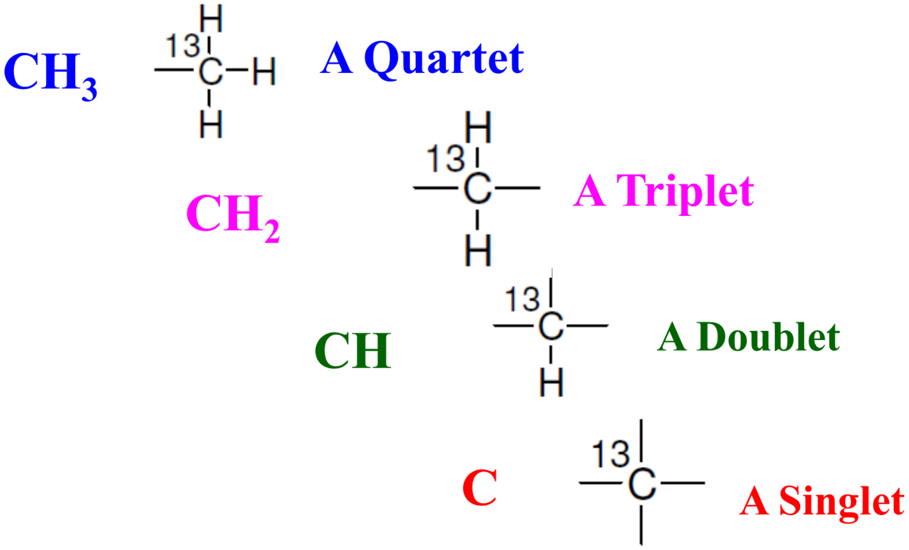
Let us go further, what happens if there are three carbons present. All the three in carbon 13 state, think of a situation probability of all carbons for simultaneously present as carbon 13. It is 1/100 X 1/100 X 1/100; which turns out to be 0.000001331. Look at it, this is a very very small number 0.000001331 is coming into the picture; and nearly this is 1 in a million. If there are 1 million molecules present in your sample there is only 1 molecule you will find where all the carbons are present as carbon 13; extremely difficult. Forget about it. Now, let us go to a complicated situation. In a given molecule, there can be organic molecule consisting of large number of carbons; 10 carbon, 15 carbons, 20 carbons, like that.



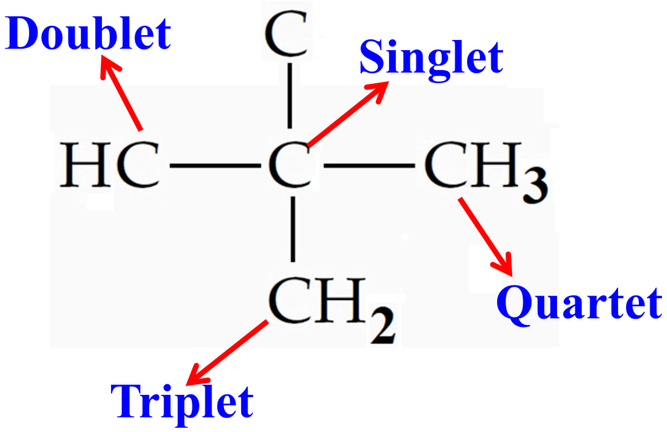
1/100 x 1/100 x 1/100 x 1/100 x 1/100 x 1/100
= 0.000000000001771561

One in a Quadrillion (10^{12})!!!

Take another complex example where a hypothetical molecule like this, there are 6 carbons present here. I have deliberately chosen this. What is the probability of seeing all the 6 carbons in carbon 13 state? all carbons being in carbon 13 state, what is the probability that is 1/100 X 1/100 X 1/100 X 1/100 X 1/100 X 1/100. That is 1 in 10 to the power of 12 . One in a quadrillion. Imagine such a very very very small number, almost points 8 or 10 zeros followed by this 1. That means possibility of seeing such molecule is one in 10 to the power of 12. In such a big number, 1 molecule you can find where all the carbons are present as carbon 13. How can you see that in 10 to the power of 12 molecules only 1. Already 10 to the power 23 is Avagadro number of molecules. If you take Avagadros number of moleclues, maybe 1 or 2 molecules will be there. How can you see a signal from such a thing; extremely difficult. Basically NMR is an insensitive technique; and if you have abundance like this how can you see the spectrum. It is really a very very difficult situation; and practically it is impossible to see the carbon 13 spectrum of such big molecules where all the carbons simultaneously present as carbon 13 ; very difficult practically impossible to see. Then the question one can ask is, how can we see all the carbon peaks in a molecule containing large number of carbons? Theoretically I showed you it is difficult. Go to any or take any organic molecule containing large number of carbons, 10 or 15 carbons; no problems; go and take the NMR spectrum; you will get it. Then how can we get that. I just now said even with six carbons, it is one in a quadrillion, but how can we get the carbon 13 spectrum of a big molecule contain large number of carbons. How all the carbons can be simultaneously present as carbon 13? which is impossible; not impossible, but it is extremely difficult to see, very very very low possibility. How? we will understand that now. This is the important point which you must know. The carbon 13 spectrum how we get it, because this is a concept is another important part of this course.



Before going further let us try to understand how does protons attached to carbon 13 influence the carbon 13 spectrum? Assuming we are not doing any decoupling. If I have a molecule like this a CH3 group is present where all the three protons are chemically equivalent coupled to carbon 13, then what is the pattern we get for carbon 13? Remember carbon 13 is a different nuclei; protons are different nuclei. When these two can couple to each other, these are all magnetically equivalent or chemically equivalent, doesn’t matter whatever you take, when this coupled to carbon 13 what is the pattern we expect? Same 2NI+1 rule. It should give rise to four lines. 2 into NI plus 1. You put it 2 into 3 into half plus 1; you are going to get 4 peaks and according to Pascal triangle the intensity pattern is 1 3 3 1. That's what we should get. So CH3 carbon if you look at it, when the protons are coupled to carbon you get a quartet with intensity ratio 1 3 3 1. What about CH2 you can think of a situation like this two CH2 protons are coupled to 1 carbon; For CH2 protons you are going to get a triplet; as usual like a CH2 proton coupled to CH3 makes it a triplet; here 2 protons are chemically equivalent; coupled to carbon makes it a triplet. 1 2 1 intensity. Next go further. If I have a situation where only one carbon is there, which is coupled to only one proton, what is the pattern you expect? it is only one proton, obviously like AX spin system, this carbon will be a doublet. That's what you are going to see. Go to a situation where there are no protons attached. There is no coupling of carbon with proton at all; they are called quaternary carbons. Each carbon has four carbon bonds; none of them are attached to proton. They are called the quaternary carbons. Such carbons give only a singlet, because there is no coupling with protons. So please remember if I look for the carbon 13 coupled spectrum CH3 gives a quartet; CH2 gives a triplet; CH gives a doublet and a quaternary carbon gives a singlet.



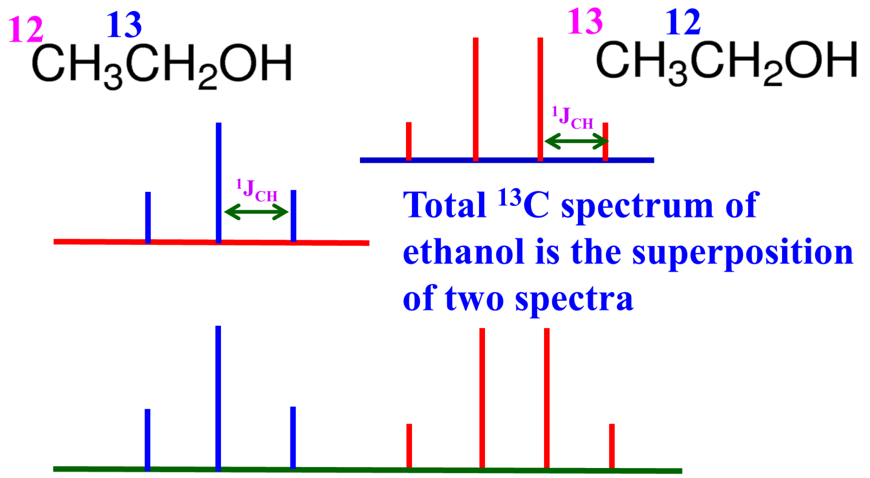
With this knowledge take an example like this, a hypothetical molecule. If I have a molecule like this, and if you take the record the spectrum of this molecule we'll get a quartet for this; triplet for this; doublet for this; and singlet for this. Hypothetically I have written this molecule just for you to get the feeling, for you to understand how the multiplicity of the carbons are there when they are attached to different number of protons. With this knowledge let us try to understand the carbon 13 NMR spectrum of ethanol. Very simple molecule. ethanol has two carbons; carbon CH3 group and CH2 group. How do we get the carbon spectrum?

$^{12}CH_3{}^{12}CH_2OH$	**97.8 %** **NMR Inactive**
$^{13}CH_3{}^{12}CH_2OH$	**1. 1%** **NMR Active**
$^{12}CH_3{}^{13}CH_2OH$	**1. 1%** **NMR Active**

First of all let us understand isotopomers of ethanol. How many isotopomers of ethanol you can think of. One way, both the carbons are carbon-12. That is what we said in a molecule containing two carbons, the probability is 97.8%. You will not get spectrum from this molecule at all. 98% of the molecule will not give you any NMR signal, okay, forget it. Come to other molecule this CH3 can give a peak 1.1% NMR active; see this carbon 13 spectrum can be obtained because of this CH3 group. Go to this situation this CH2 carbon will give a peak, but this will not give me the peak again this is only 1% active; 1% possibility.

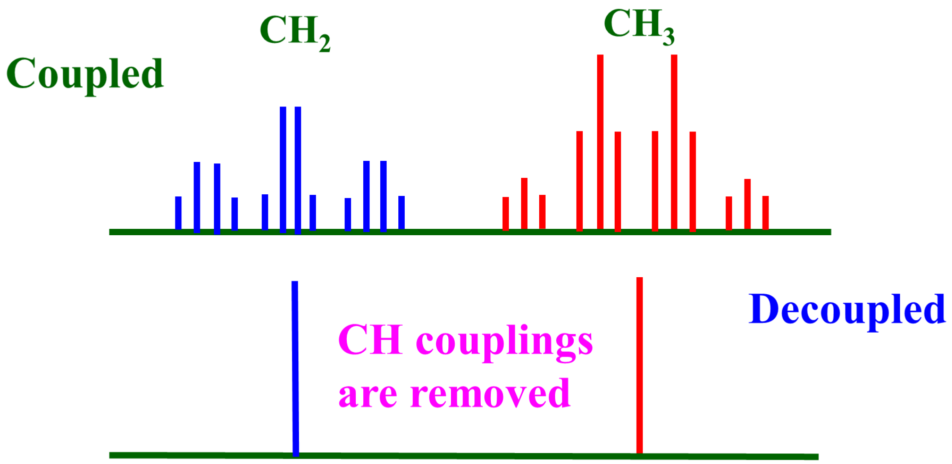
$^{13}CH_3{}^{13}CH_2OH$ **.000121 % NMR Active**

Go to a situation like this, the probability when both the carbons are present simultaneously as carbon 13, I told you, this probability is 0.000121, NMR active. But this is very difficult to see the signal from this isotopomer. It is not easy to detect, very difficult to detect so we will forget about this, because it is anyway difficult to detect so we have to worry about only this thing. We have to worry about only these two groups; these two molecules. Anyway this is NMR inactive. Do what we will do is we will not worry about first molecule, we will not worry about last molecule here we only worry about only second and third molecules. How does this carbon 13 spectrum come we will see.



Total ^{13}C spectrum of ethanol is the superposition of two spectra

Already I told you when carbon is coupled to protons 3 chemically equivalent, when I take this molecule, I am going to get only a quartet, that is all. This carbon is not contributing. Let us think of other isotope. Of course this separation corresponds to this one bond carbon proton coupling. Let us consider other isotopomer, I am sorry this should have been 13, this is a mistake; this should have been carbon 13. CH2 is in carbon 13 state. This is the carbon in which you are going to get a triplet here. So what is the separation? This is one bond; this is carbon proton coupling. This is one bond this carbon proton coupling; this anyway will not give you the signal in this isotope. You understood now for two isotopeomers, I got two different spectra. For this isotope of I got a quartet; and for this isotope I got a triplet. But in reality in a given sample, you do not have only these isotopomers, everything is simultaneously present. All four types of isotopomers are present. First one ignore, last one do not give signal, very difficult. Only these two will give the signal. Do in a given sample because all the molecules isotopomers are simultaneously present, only these two give the signal. Okay, let us worry about these two.



Then you may ask me a question what about long-range carbon-proton couplings? For example you consider a situation like this. When I am looking at this carbon I look for only one bond coupling with proton. Since I am not decoupling what stops me in telling this proton also can couple to this carbon, it is possible, it will couple. So in reality you will have the spectrum one bond coupling of this carbon gives a triplet. These three protons will give you a quartet. But at the same time these two equivalent protons CH2 protons will split each line of the quartet into triplet. Remember we understood the multiplicity pattern earlier so what is th type of spectrum you are going to get for this molecule? You are going to get for CH3 carbon a quartet and each line of the quartet will be split into triplet because of long range coupling. So you will get one bond CH coupling and three bond this carbon and this proton coupling. Okay, not HH coupling. One bond CH coupling and three bond CH coupling you are going to get. This is the thing you are you are going to observe. Go to this molecule what is the pattern you are going to get for this one? I told you, you will get a triplet because this carbon is coupled to these two protons and we can say triplet; but this carbon can also couple to this proton.

What stops us from preventing this proton coupling to this carbon? the long range proton carbon couplings are present. Then in reality there is a one bond coupling and also three bond coupling of these protons to this carbon; that is also possible. So CH2 carbon will be a triplet because of direct coupling with two protons, and each line of the triplet is split into quartets because of coupling with CH3 protons. So, there is one bond carbon proton coupling plus three bond carbon proton coupling, both are present. So what is the pattern you are going to get finally? of course this one you do not see you can ignore it. But remember that coupling exists. What about C13- C13 coupling? we do not see the spectrum of the molecule. But that does not mean you do not have the coupling. It is there, coupling is also there, but very difficult we do not detect it, that is all. Do what about the proton proton couplings? that is also possible. But remember when you are looking at the carbon spectrum carbon proton couplings are seen; one bond carbon proton and three bond carbon proton couplings, but not proton proton coupling. The CH3 protons and CH3 protons coupling you will not see. Why? protons are passive spins. I told you already passive couplings are not reflected in the spectrum of active spins. The active spin here is carbon 13. So at this carbon 13 resonance when you are looking at it proton proton couplings are not seen. So you do not have to worry about it. So in reality all spectrum of isotopomers are there; the first one do not give, the remaining three are there, the last one is not seen. So the total coupled carbon 13 spectrum of molecule ethanol is the superposition of the two spectra, two spectra coming because of two isotopomers. If you look at the carbon 13 spectrum, 1JCH coupling, the CH3 will be a quartet and CH2 will be a triplet. But there is also a long range coupling long range three bond CH coupling which makes this as triplets of quartet, this will become quartet of triplet. This is the complexity of the spectrum you get. In reality if you take the fully coupled proton coupled carbon spectrum, this is what you are going to get. But what we are going to do generally, in beginning itself I told, you all the carbon spectra are recorded with broadband proton decoupling. That means all these, 1 bond, 3 bond CH coupling will be completely removed. So, in reality it is a decoupled spectrum but what we get every day when you record the spectrum is like this two peaks, one for this, and one for this. Basically we are reduced the complex multiplicity pattern into a singlet. Similarly all these complexity will disappear and gives a singlet, so you are going to get two peaks for carbon 13; one for CH3 carbon, and one for CH2 carbon. See it is an ethanol molecule. But what do you understand from this? what do you understand from this is, for a molecule with N chemically non-equivalent protons and N chemically equivalent carbons, the proton detected spectrum is from a single molecule, where all are 100% abundant. Whereas, for a molecule with N chemically inequivalent carbons N isotopomers are there, each with a different isotope. There will be N spectra each with a different isotope having different carbons in carbon 13 state. So for example I have N isotopomers, each isotopomer will have one carbon in carbon 13 state. So if there are N isotopomers, you have N spectra overlapped. So carbon 13 spectrum is a superposition of spectra from N isotopomers. Please remember when you record the spectrum of any given molecule the proton spectrum is from a single molecule; but the carbon 13 spectrum is not from a single molecule, it is the super position of the NMR spectra from all isotopomers. The N different types of isotopomers together give rise to detectable carbon 13 spectrum. And that is what it is; and this is the simple example to show in a diethyl phthalate, we have CH3 group, CH2 group. This takes some more time to explain what I am going to do is I will stop here I will come back and discuss this in the next class. But so far what I was trying to tell you today is to introduce the carbon 13 NMR which is one of the exotic nuclei which is used apart from protonn by most of the chemists. And I gave the salient features of carbon 13; sensitivity is much low compared to proton and resonating frequency is 4 times smaller than that of the proton; always carbon 13 spectra are recorded in the broadband proton decoupled state in which case each carbon chemically inequivalent present in the molecule gives only one peak. If there are Nn peaks in the carbon 13, spectrum there are N chemical inequivalent carbons present in the molecule. That is what you should understand; and then I also said if I have molecule with N chemically inequivalent protons, the spectrum is from a single molecule. Whereas, if there are N chemical inequivalent carbons present, the spectrum is not from a single molecule, the carbon 13 spectrum is the superposition of the spectra of N different isotopomers where carbon 13 is present in different places, in each of these isotopomers. And that is the summary which I wanted to tell you. We will continue with the further discussion on carbon 13, in the next class. Thank you very much.