One and Two Dimensional NMR Spectroscopy for Chemists
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Lecture No: 44
Spin Echoes

Welcome back, so in the last couple of classes we discussed varieties of things about analysis of the spectra, spin nomenclature, multinuclear NMR, and varieties of interesting things we discussed. But there is one important thing we need to be understood, especially when we study Carbon 13 NMR, Silicon NMR and Nitrogen 15 NMR, they are low abundant nuclei.

We already discussed that the sensitivity depends upon gyromagnetic ratio and also on the natural abundance. So, how do you overcome some of these problems. Inherently the NMR technique, compared to other spectroscopic techniques, is insensitive. Added to that the detection of such lower abundant nuclei poses additional challenge. So there are many stalwarts and pioneers in NMR, who have come out with numerous methods and technologies to circumvent some of these problems to a large extent, and one such thing is polarization transfer.

What it means is, we take the magnetization from abundant spins and transfer to low gamma nuclei. what it means? Rob the rich and pay the poor. Then what will happen, the magnetization of low abundant spin goes up, and we can detect with better sensitivity. It is one of the ways. So and we can combine this thing also for doing spectral editing. This polarization transfer technique is invariably used in heteronuclear NMR, especially direct detection and inverse detection techniques, etcetera.

So, today let us discuss something about this polarization transfer technique and spectral editing both. So these are all some of the important things, especially when you are dealing with two dimensional experimental technique, which we are going to discuss later, where we use HSQC and others, where the polarization transfer is extremely useful when we do inverse detection.

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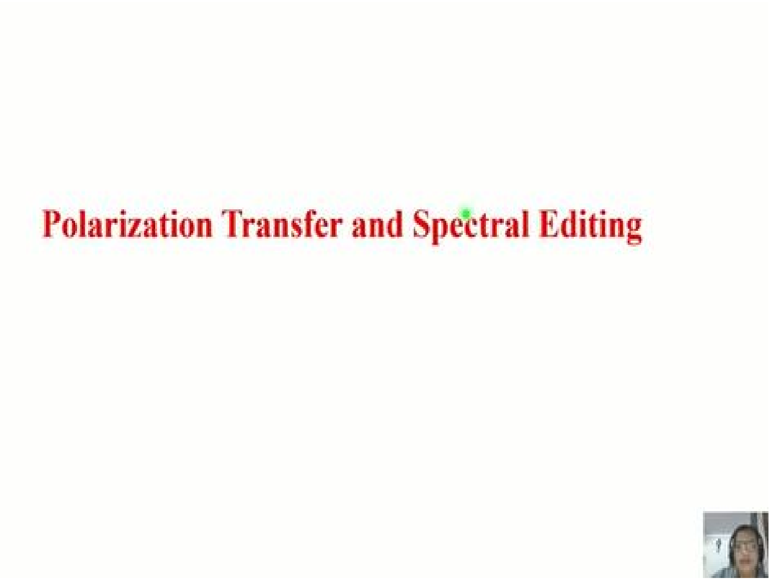

Polarization Transfer and Spectral Editing

So, today my focus is on polarization transfer and spectral editing.

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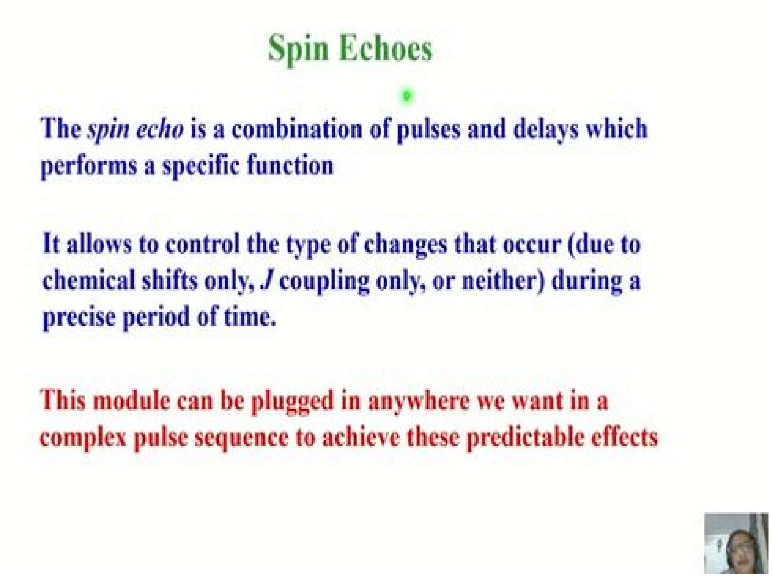

Spin Echoes

The *spin echo* is a combination of pulses and delays which performs a specific function

It allows to control the type of changes that occur (due to chemical shifts only, *J* coupling only, or neither) during a precise period of time.

This module can be plugged in anywhere we want in a complex pulse sequence to achieve these predictable effects

First thing we need to understand for this, the basic requirement; this is spin echo technique. This is one of important modules in NMR. What are spin echoes? If you understood the spin echoes lot more things about polarization techniques and spectral editing techniques can be a understood. And this spin echoes what I am going to tell you now is a small pulse sequence module, which can be incorporated in many experiments; and it does a specific job. It is a combination of pulses with some delays and performs a specific job.

And, we can have several types of experiments, we can do using spin echo, we can control what are the changes that occurs because of chemical shift only, we can get changes we can set a delay between the pulses; and during this delay what happens, I will explain to you as we go ahead. We can ensure and find out whether only chemical shift will refocus, and only J coupling will re-focus. What is going to happen during this delay? That can be understood, and such a module is so simple and is a small module you can put it anywhere in your complex pulse sequence. So that you can manipulate the evolution of magnetization in different pulse sequences, and you can get whatever the predictable information you want. See for example; I can use the pulse sequence I can plug it here into one of the pulse sequence, and, let us say it does a specific job what I want, it can be predicted and I know what is the desired output of the pulse sequence. So, these things we can manipulate by using spin dynamics and for many of the cases, we come across what is called spin echoes sequence.

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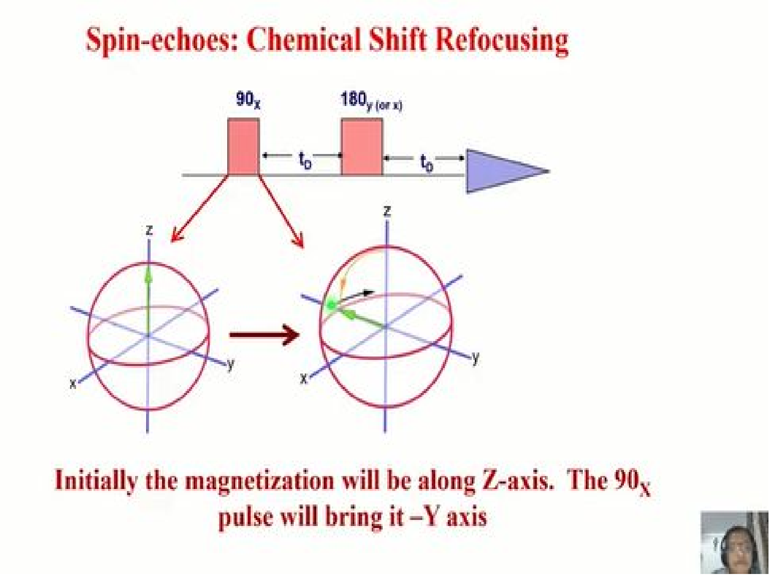

Spin-echoes: Chemical Shift Refocusing

Initially the magnetization will be along Z-axis. The 90_X pulse will bring it –Y axis

So, let us discuss something about what is the spin echo? The spin echo is something about chemical shift refocusing I am talking, what it means is look at this, I use very simple pulse sequence. We have a 90 degree pulse; we already discussed about what is a 90 degree pulse or 180 pulse and also discussed about the ways we can apply the pulse sequence pulses, you can apply pulse along X axis, pulse along Y axis, phases of pulses we discussed long back, in one of the classes.

So, 90 degree pulse when the magnetization from Z axis to XY plane, either X axis Y axis depending upon where I am applying the pulse. Then we give a small time delay called t_D. What will happen during this time we will see; and after that delay, we are going to apply another 180 degree pulse, give some more delay, exactly equal to this one, and then start collecting the signal. Remember, in a conventional experiment what we are going to do is apply 90 degree pulse, signal, that is the one dimensional experiment.

What will do here in spin echoes, we incorporate another 180 degree pulse, in between delays after 90 and after 180 degree and then after a sufficient delay, which we know we will decide what is t_D time delay, and then collect the signal; t_D is nothing but a time delay, that is all. Now, let us vectorially understand, with your vector diagram, what happens to this magnetization during this pulse sequence? Of course just before the application of a 90 degree pulse, what is the status of our magnetization? it is in thermal equilibrium and along Z axis.

We understood the bulk magnetization concept please remember, the bulk magnetization concept. The magnetization is along Z axis fine; now I am going to apply 90 degree pulse, what happens? the 90 degree pulse brings the magnetization, depending upon where I am applying the 90 degree pulse, along X axis then magnetization has to tilt or flip around -Y axis. This is because of the right hand thumb rule, I discussed.

Remember? Your thumb will point at the direction of the pulse, and the magnetization tilting is defined by the curling fingers, the direction in which the four fingers get curled. For example; this is the magnetization is here, now I am applying a pulse along X axis here, then what will happen? this is X; this is Y; this is X this is Y and this is Z, so I am applying a pulse along this axis.

So, the magnification is on Z axis. If I apply pulse on this axis what will happen? the magnetization will tilt in the direction of the fingers which get curled, so that is this is +Y, this is –Y. So this we have understood, I do not want to explain every time this thing, please remember. Now the 90 degree X pulse will bring the magnetization to -Y axis, okay, Now what is going to happen? As soon as depending upon the magnetization to -Y axis by 90 degree pulse, we keep quiet, we do not need anything.

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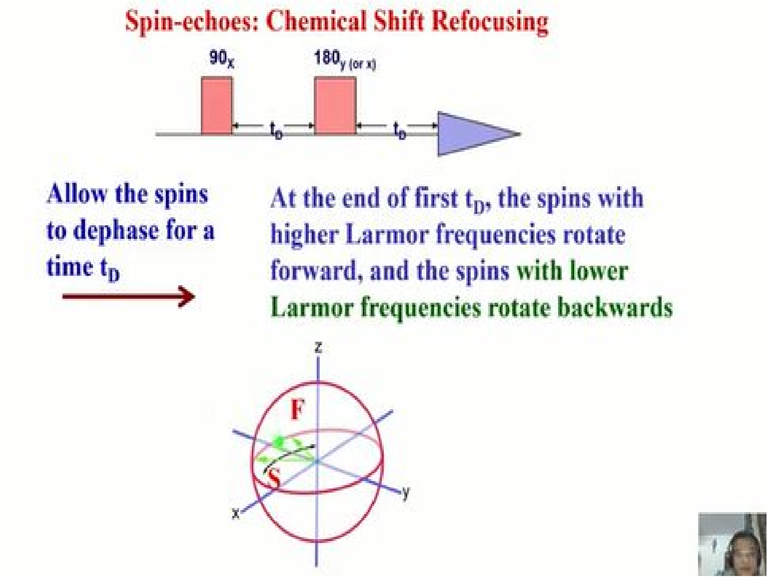

Spin-echoes: Chemical Shift Refocusing

Allow the spins to dephase for a time t_D

At the end of first t_D, the spins with higher Larmor frequencies rotate forward, and the spins with lower Larmor frequencies rotate backwards

What we will do is, we will allow this magnetization to start dephasing with the time t_D. Remember I discussed about dephasing. As soon as the magnetization is brought to XY plane, initially the entire magnetization will be in coherence. All nuclear spins, magnetic moments will be in phase coherence. There is a phase coherence, I told you. What happens due to various phenomena, the nucleus magnetic moments start dephasing, this we discussed I called this as a relaxation time T2, please remember.

So the spins start dephasing, how do they dephase? The diphase and start moving like this, like this. At the end of the first t_D, the spins that have higher frequency rotate forward and lower frequency rotate backwards. You remember, with respect to the rotating frame of reference we discussed, teh entire magnetization is initially along this axis, all the nuclear magnetic moments. Now because they start dephasing, some may start moving like this, some may move like this, with respect to the, on-resonance frequency, this is our carrier offset, let us say.

With respect to this now the spin packets or vectors in this direction start moving like this, the vectors here start moving like this, this is called a fast moving component, it is called a slow moving component. So with respect to this ω0, let us I call this as ω0, the spins with the higher

Larmor frequency moves faster, spins with the lower Larmor frequency start moving slower. That is what it is; you know in the rotating frame of reference we understood.

This one is $\omega0$, these are all different Larmor frequencies; so this is going with lower frequency, this is faster component, and it is a slower component.

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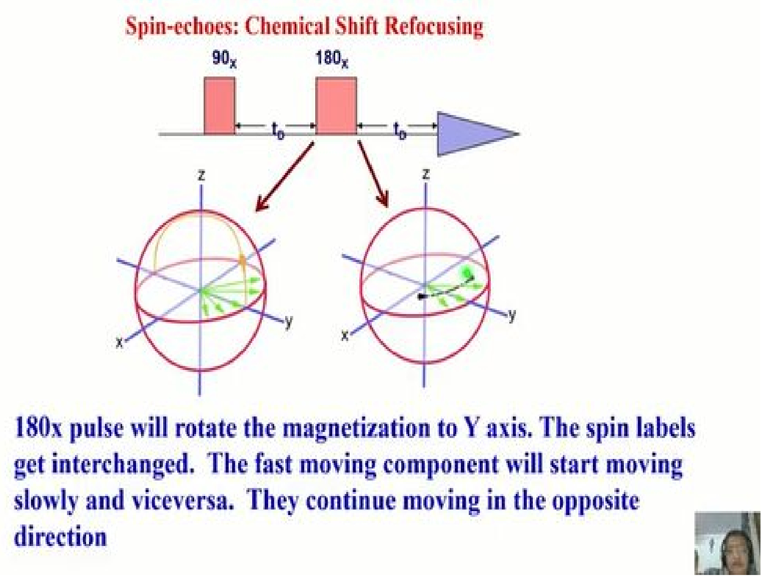

Spin-echoes: Chemical Shift Refocusing

180x pulse will rotate the magnetization to Y axis. The spin labels get interchanged. The fast moving component will start moving slowly and viceversa. They continue moving in the opposite direction

Now, after certain time t_D what is going to happen? So, what is going to happen is; after certain time t_D at this place, when it is completely dephased, I am going to apply a 180 degree pulse here; This one, at this at this point; when I apply 180 degree pulse what is going to happen? I am flipping my nuclear spins, which were in this axis, I will rotate it like this, remember it is like turning the pancake; you are turning a pancake, like this.

If was like this and we rotated like this; or in the Indian breakfast, this is dosa. You can turn the dosa like this; so this is what the role of the 180 degree pulse. It flips the magnetization from -Y axis to +Y axis, because I am applying the pulse along X axis. Remember I am sitting here; and I can rotate like this. When I rotate like this, it rotates by 1800 degree. Now what is happening they continue to move in the same direction. In that case what happens? the labels get interchanged.

So, by 180 degree pulse, when you completely invert like this, the fast moving components starts moving slowly, slow moving components starts moving faster, because they continue to move in the opposite direction now. The which was going like this, will continue to move in the same direction. Now with respect to this axis, the fast moving will be moving slowly and slow moving component will start moving fast. They continue to move in the opposite direction. 180 degree pulse, you understood, what happened?

So, what we did is you simply rotated the magnetization from -Y vector +Y axis, we interchanged the labels of the spin vectors; fast and slow moving components just gets interchanged. As far as the movement is concerned, the one which is moving like this, faster component will move slow now; the one which was slow, move like this move fast.

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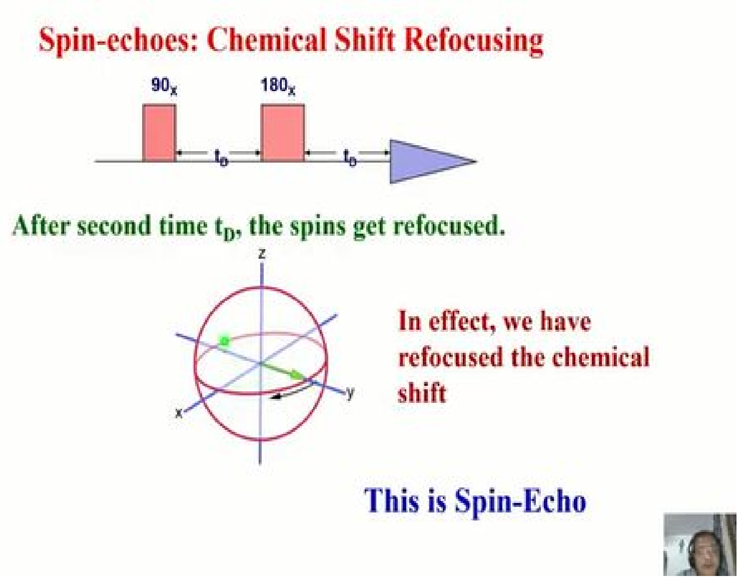

Spin-echoes: Chemical Shift Refocusing

After second time t_D, the spins get refocused.

In effect, we have refocused the chemical shift

This is Spin-Echo

Now, wait for another time tD, this is called a second time delay; after some time what happens? all the spins get refocused, imagine different spins will move with different speeds, one may be moving fast, one may be moving faster, one may be faster, similarly one will be slow, one will be slower, one will be slowest; and they are all moving with different frequencies. And what happens when you allow the same amount of delay, they come back and start getting re-focused here; on the Y axis. Imagine a condition like this. Now there are 10 people are standing in the queue or standing in a road. 10 people who are runners, A running race going on. As soon as the whistle is blown all 10 people who are standing in the road starts running. Of course all of them are not having the same speed, some will move faster, some will move slow. Exactly now what I said, like this spin vector components and, after some time immediately they are asked to stop wherever they are, then I apply 180 pulse, 180 pulse means I will invert it, I will rotate the spins. So I will ask them to turn back. All the runners wherever they are the turn backwards, that is the effect of 180 pulse. Now, I will ask them to run back, continue to run, what they will do? They will start running, their speed remains same, now one was fast, one was slow, does not matter, one was fast, fast was moved this much, one was slow, slow slower has moved this much. Now they continue to move in the opposite direction. Finally what will happen? It appears to you as if, the person who is slow has already move faster, and the person who is fasts is slow and is behind. You understand after some time what happens? after the same amount of time, although they were at different speeds, they come back to the same point where they started, at the same time. They started at this point, they come back to this point, at the same time. This is what is called refocusing, this is echo, this is called echo.

So, that means all the spins get refocused, this is what is called, in effect a spin echoe, where we are refocusing chemical shifts, understand? This is a concept of echo; 90, tau delay, 180, tau delay and collect the signal. It is an important module, called spin echo module. So, these spin vectors were moving at different Larmor frequencies, one moving faster, one moving slow like that, gets inverted as soon as 180 pulse is applied, simply turn it from -Y to +Y, spin labels get interchanged, they start going backwards and re-focus at the same point, where they started.

And these are all in phase, again the phase decoherence is disappeared, they all come back to the same phase; this is what is called spin echo.

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This is the animation of that which is taken from Wikipedia you can see that, this what is happening. Look at it, the spins are like this; you apply a 90 degree pulse, bring to X axis or Y axis, they start dephasing like this ; the 180 pulse rotates like a pancake, and then refocussing with the maximum signal, that is called an echo.

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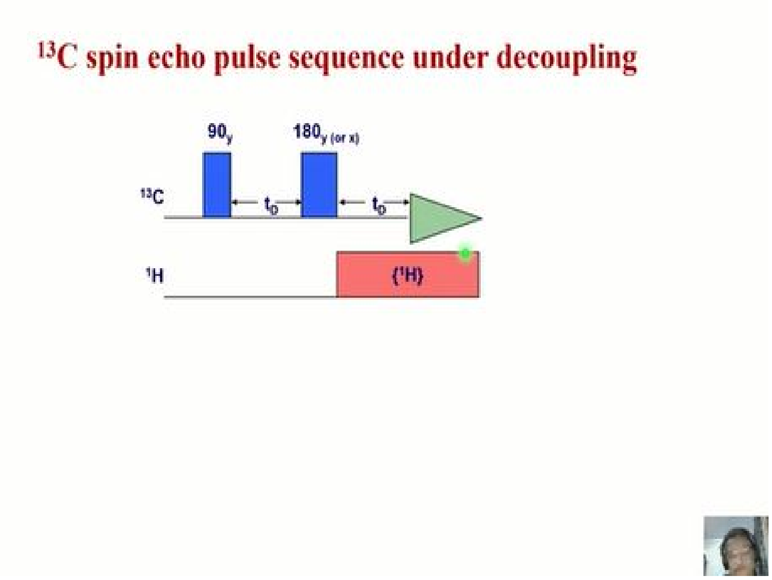

^{13}C spin echo pulse sequence under decoupling

So, now we will see the 13 spin echo pulse sequence under decoupling. We want to see, this is the 180 pulse sequence we understood what is going to happen. Now, I want to see 13C signal. This pulse sequence we apply, I am going to detect the signal by decoupling. I explained to you when I am looking for the carbon 13 NMR, normally we do the broadband decoupling; and detect this signal, right? This is the pulse sequence, this is spin echo sequence. And, then we are collecting the signal here, with proton decoupling. In this channel we are going to apply decoupling. what is a decoupling? I explained to you sometime back, but in any case what I am going to do is, quickly I will tell you what is decoupling, just in two or three minutes or five minutes; so that we can continue to go further, and then no need to come back to this decoupling.

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How to overcome some of these difficulties?

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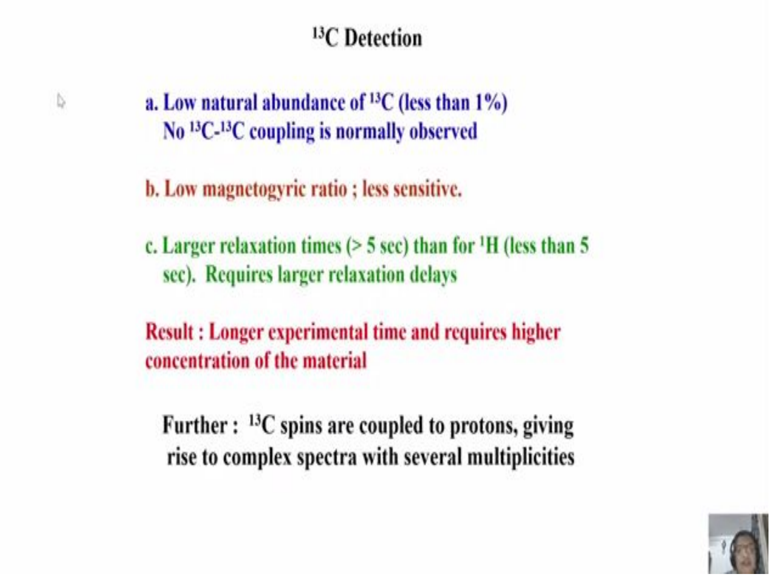

^{13}C Detection

a. Low natural abundance of ^{13}C (less than 1%)
No ^{13}C-^{13}C coupling is normally observed

b. Low magnetogyric ratio ; less sensitive.

c. Larger relaxation times (> 5 sec) than for ^{1}H (less than 5 sec). Requires larger relaxation delays

Result : Longer experimental time and requires higher concentration of the material

Further : ^{13}C spins are coupled to protons, giving rise to complex spectra with several multiplicities

One of the important problems I said in the carbon 13 NMR is the low sensitivity, it takes an enormous moment of time and especially when the carbons are coupled to protons. So first thing is, it takes larger experimental types because of various reasons, such as, long relaxation time and low sensitivity etc. And what we have to do is the decoupling, break all the couplings of protons with all the carbons simultaneously, so you will get only a single peak for each carbon.

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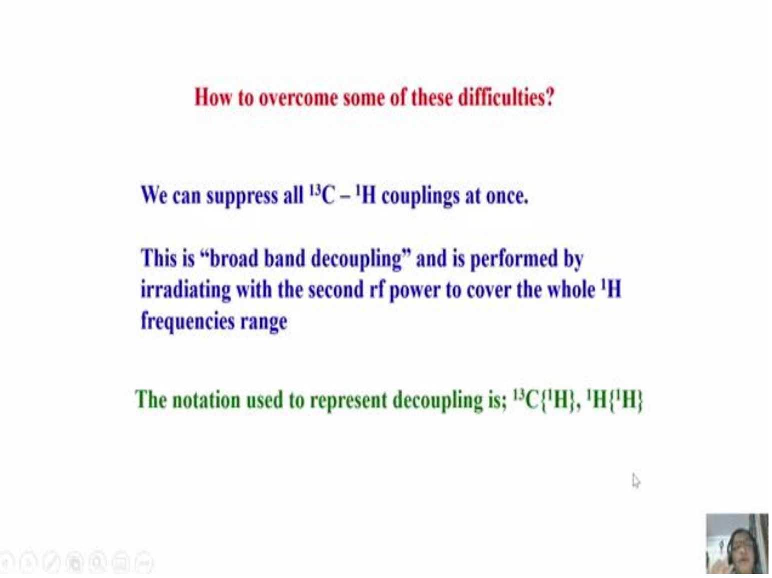

How to overcome some of these difficulties?

We can suppress all $^{13}C - {}^{1}H$ couplings at once.

This is "broad band decoupling" and is performed by irradiating with the second rf power to cover the whole ^{1}H frequencies range

The notation used to represent decoupling is; $^{13}C\{^{1}H\}$, $^{1}H\{^{1}H\}$

How we do this, we can suppress this by broad band decoupling where second RF power is applied to cover the whole range of proton frequencies. I told you when I explained carbon 13, just quickly I do not want to come back to decoupling again, so I am repeating it, I am telling you. Now let us look at the proton spectral width of 10 ppm, sit at the center of the proton spectral width that is at 5 ppm.

Apply RF pulse in such a way, it has a wide range of excitation profile. RF pulse you have to apply in such a way, the exhibition profile is so broad, at the same time has uniform power. You have to choose the rectangular pulse or square pulse you are applying, if you do the Fourier transformation, in one of the classes we showed. I discussed the Fourier transformation of that function is the sinc function, where we can see exactly at the center where we call it as ω0 or call it as a carrier frequency.

The power spectrum is more or less uniform, it is broad, it is really broad at the center, then take that region where the power is uniform, apply a pulse choose the offset in such a way, so that the excitation profile is uniform. Then what happens is, at a given instant of time, you are breaking the coupling between protons and all carbons and this is called heteronuclear decoupling.

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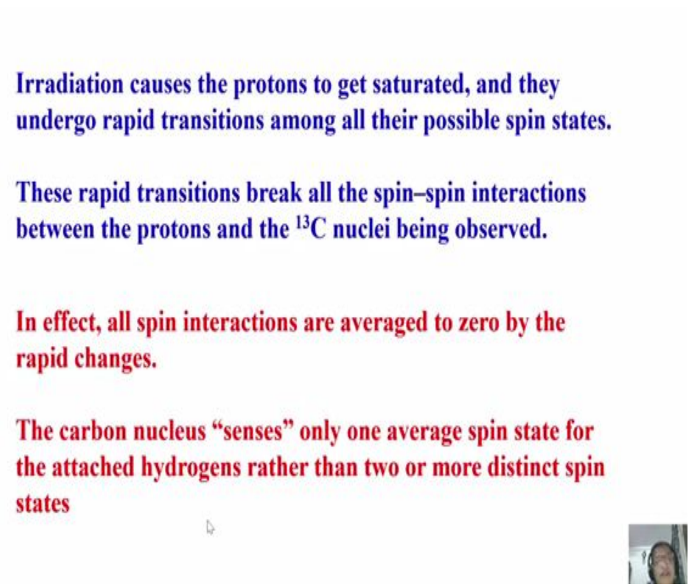

Irradiation causes the protons to get saturated, and they undergo rapid transitions among all their possible spin states.

These rapid transitions break all the spin–spin interactions between the protons and the ^{13}C nuclei being observed.

In effect, all spin interactions are averaged to zero by the rapid changes.

The carbon nucleus "senses" only one average spin state for the attached hydrogens rather than two or more distinct spin states

What happens is conceptually is like this, irradiation causes the proton to get saturated, what is the meaning of saturation? I told you this, the spin population become equal between two energy states. The proton energy states spin population become equal and they undergo rapid transitions among all the possible spin states. What does it mean? The coupled spin carbon 13 do not see the proton in either of the energy states, because of the rapid transition, carbons do not see the protons spins residing in either the alpha state or the beta state.

And you are applying the power in such a way, saturating the spin population equal and there is a rapid transition undergoing on. As a consequence, the detected nuclei carbon 13 will not see protons in either alpha state or beta state, so it breaks all the coupling, or it is as good as telling there is no energy separation at all like a degenerate state as far as carbon is concerned, there are no spin states for the proton.

That is what happens, in effect all the spin interactions are averaged to zero between carbon and proton; all I mean, between carbon and proton because of this rapid changes. And the carbon spins senses only an average spin state for the attached hydrogen. That is what I said, it appears as if there is degeneracy of the energy state and as a consequence we do not see coupling at all, each carbon does appear as a singlet.

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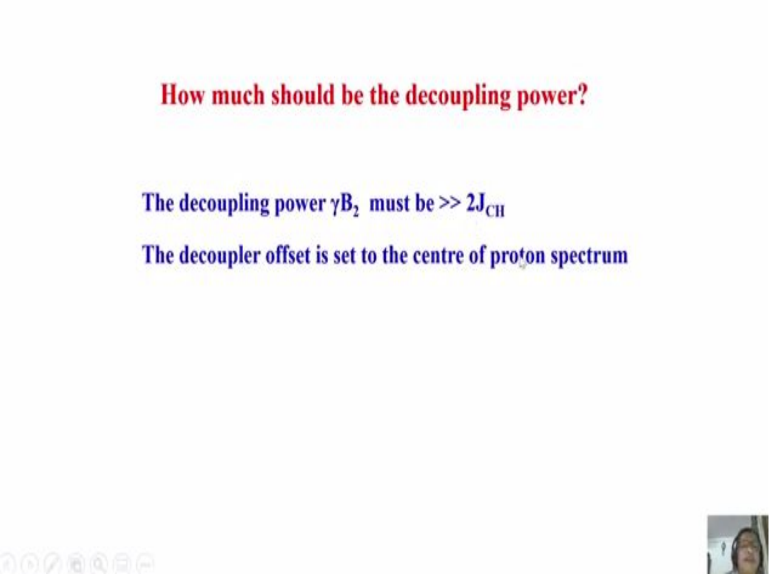

How much should be the decoupling power?

The decoupling power γB_2 must be >> $2J_{CH}$

The decoupler offset is set to the centre of proton spectrum

The question now is how much is the decoupling power? You have to apply the decoupling power in such away the decoupling power should be larger than the coupling strength. What is the coupling strength? If you measure the J coupling between carbon and proton, it can be 100 hertz and 150 hertz or 200 hertz or 250 hertz. Then which one do you choose? Choose the power such that the largest J coupling, the power should be larger than the largest J coupling between carbon and proton.

That means automatically that power is sufficient to decouple all carbons with protons. So what you have to do is, decoupling is simple, set the decoupler offset at the center of the proton frequency, apply another radio frequency pulse, it is called a decoupler pulse, in such a way the power of this pulse, set γB2 must be larger than the couplings strength, largest coupling strength.

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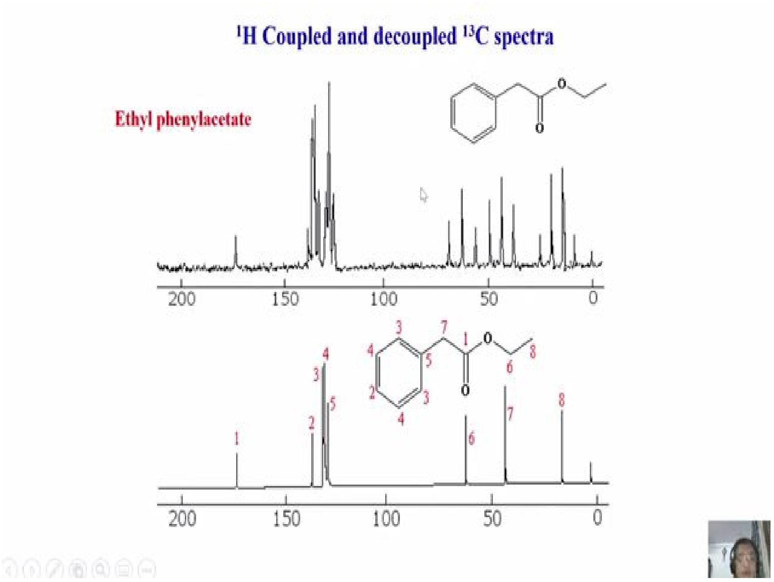

^{1}H Coupled and decoupled ^{13}C spectra

Ethyl phenylacetate

And, then when you do that, I showed this example all these multiplicities, for example, the CH3 is quartet reduced to a singlet, CH2 is a triplet because of this CH2; because of two protons becomes a singlet and this is CH2 is also a triplet, reduces to a singlet. So, in the carbon spectrum, you always see the coupling between carbon and directly attached protons. Again using the 2NI+1 rule you know the quartet, triplet, doublet, because of the directly attached protons, and this multiplicity reduces to singlets like this.

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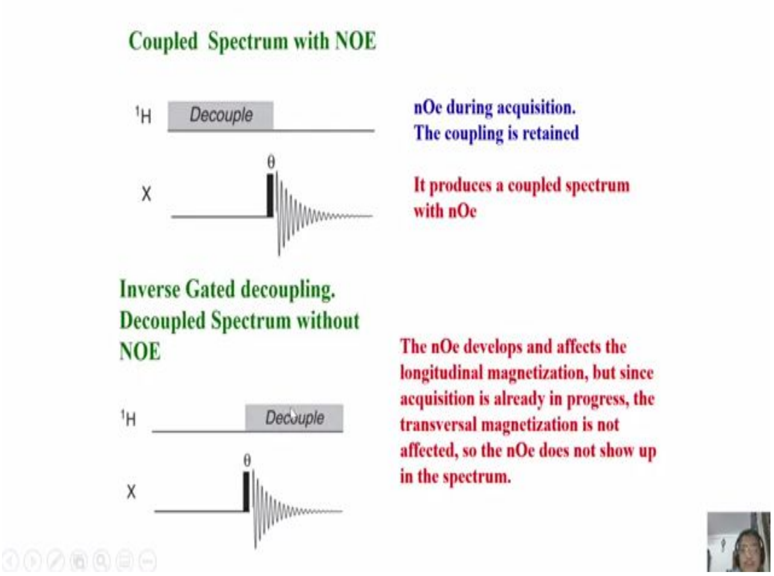

Coupled Spectrum with NOE

nOe during acquisition.
The coupling is retained

It produces a coupled spectrum with nOe

Inverse Gated decoupling.
Decoupled Spectrum without NOE

The nOe develops and affects the longitudinal magnetization, but since acquisition is already in progress, the transversal magnetization is not affected, so the nOe does not show up in the spectrum.

And this is what I said and there are various processes of decoupling also I discussed like Gated decoupling. I remember, I do not want to go again. Because now I am discussing the carbon 13 decoupling in the spin echo sequence; I just wanted to complete the decoupling topic so that I do not need to come back to it again. So you can apply the decoupling power before the rf pulse, where the coupling is retained, there is no decoupling during acquisition, it is a coupled spectrum. And when you apply the decoupling power before the rf pulse, this pulse creates what is called nOe. Next topic of my discussion after this is nOe. I will discuss more about nOe and we can do that here, you do not apply power, apply power only during acquisition, this is what we are going to get, there is no nOe, but only decoupling. This is a decoupled spectrum without nOe.

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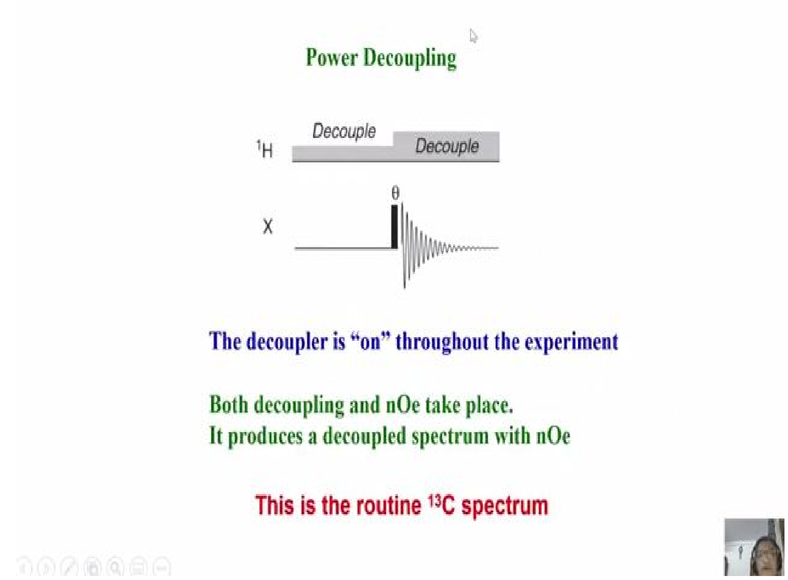

Power Decoupling

The decoupler is "on" throughout the experiment

Both decoupling and nOe take place.
It produces a decoupled spectrum with nOe

This is the routine ^{13}C spectrum

You can also do power decoupling, that is continuously keep applying the rf power, then you are going to get decoupling throughout the experiment. This is decoupling with nOe. This is the routine carbon 13 spectrum what you do.

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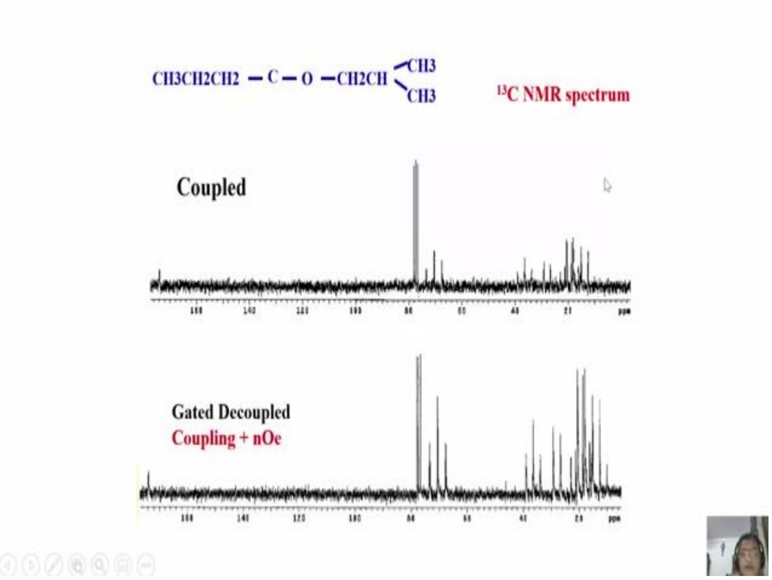

$CH_3CH_2CH_2-C-O-CH_2CH\begin{smallmatrix}CH_3\\CH_3\end{smallmatrix}$

^{13}C NMR spectrum

Coupled

Gated Decoupled

Coupling + nOe

And this is what we see and there are a number of ways of doing the decoupling.

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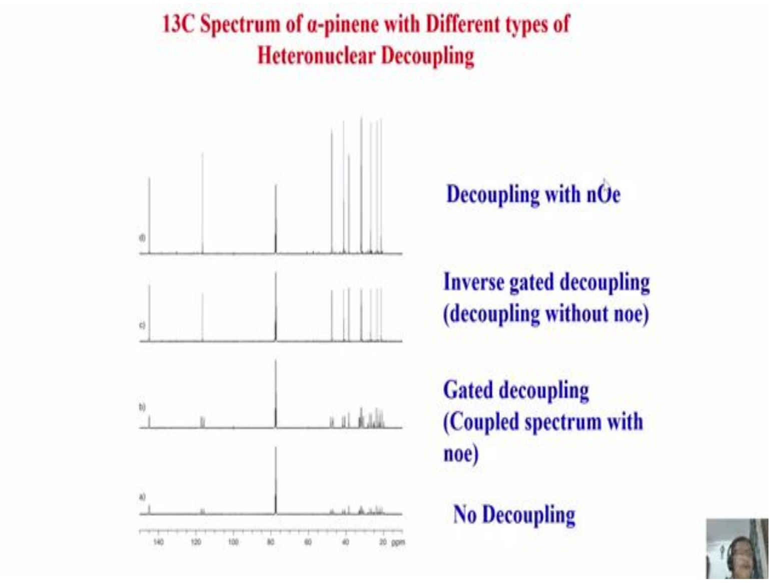

13C Spectrum of α-pinene with Different types of Heteronuclear Decoupling

Decoupling with nOe

Inverse gated decoupling (decoupling without noe)

Gated decoupling (Coupled spectrum with noe)

No Decoupling

And I do not want to tell you all those things, but decoupling nowadays can be done with lots of pulse sequences; which is possible.

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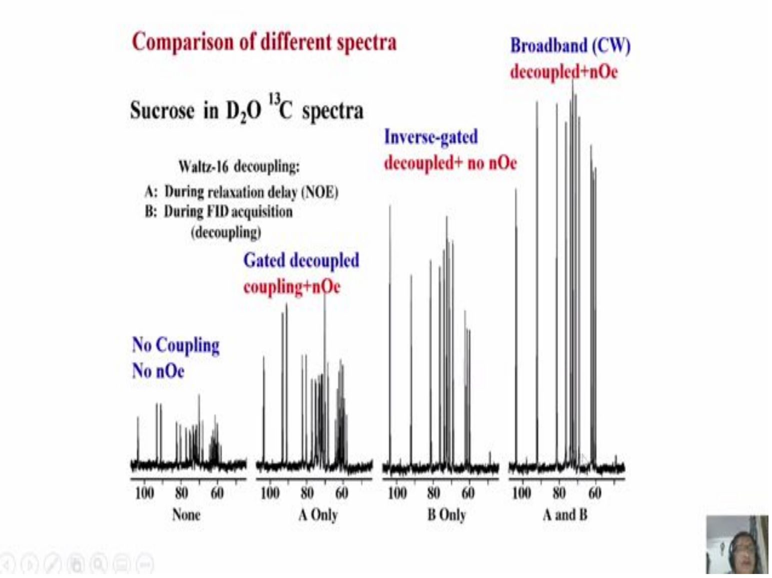

Comparison of different spectra

Sucrose in D_2O ^{13}C spectra

Waltz-16 decoupling:

A: During relaxation delay (NOE)

B: During FID acquisition (decoupling)

No Coupling No nOe

Gated decoupled coupling+nOe

Inverse-gated decoupled+ no nOe

Broadband (CW) decoupled+nOe

This I showed you how decoupling helps; when there is no decoupling, gated decoupling without nOe, with nOe and finally a broadband decoupling with nOe. Finally decoupling helps you with nOe to get a very good signal to noise ratio, compared to the one which is without decoupling and without nOe.

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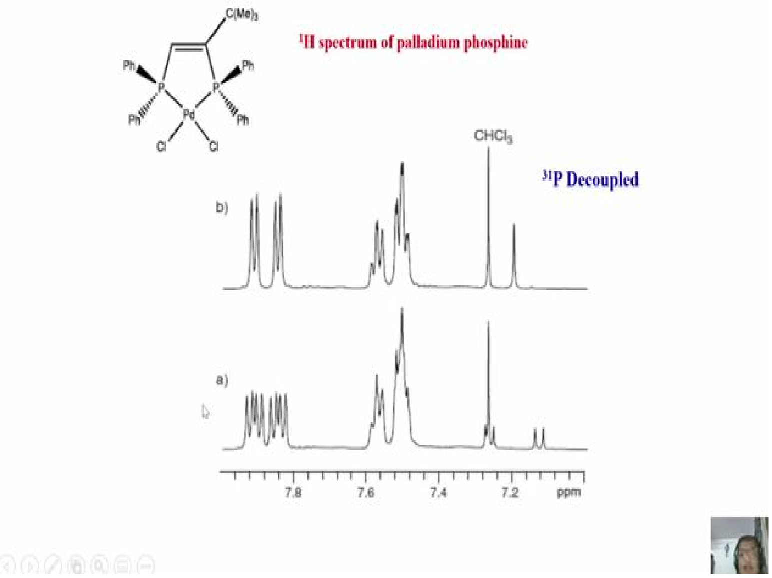

^1H spectrum of palladium phosphine

^{31}P Decoupled

And decoupling can also be done with pulse sequences; but a number of pulse sequences are available now. We use what is called;

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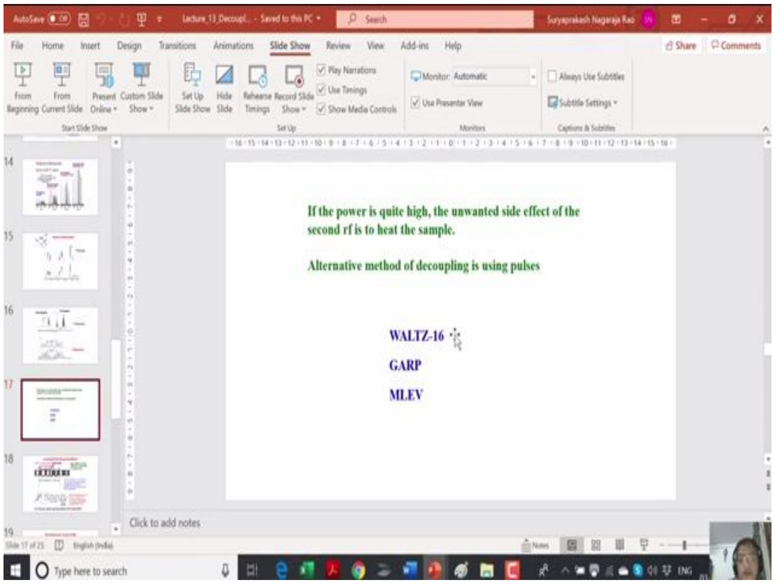

If the power is quite high, the unwanted side effect of the second rf is to heat the sample.

Alternative method of decoupling is using pulses

WALTZ-16

GARP

MLEV

WALTZ, GARP, MLEV, varieties of pulse sequences are utilized; and you can do decoupling by various methods like this, and I do not want to go into the details of the decoupling.

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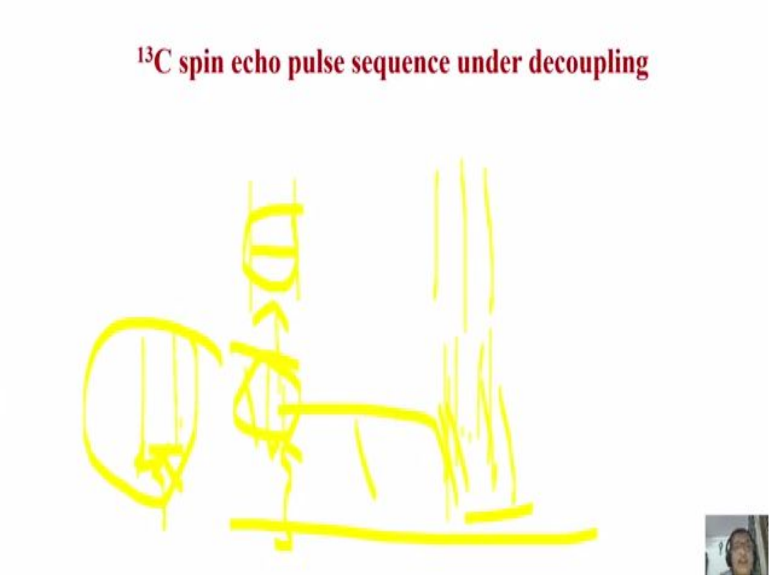

^{13}C spin echo pulse sequence under decoupling

Now we will concentrate on spin echoes. So that was the information which I wanted to give you as far as the decoupling is concerned. Now I do not want to come back to decoupling but the basic idea is simple; you can do broadband decoupling. Of course, you can asked me a question: I did heteronuclear decoupling? Can I not do homonuclear decoupling? Yes, it is possible but homonuclear broad banded decoupling is not possible.

Moment you bring the magnetization to the transvers plane and if we start applying rf pulse you are going to disturb the magnetization, it is not possible. But selective decoupling is possible. What happens is let us say the AX spin system is there, here is A and this is X, two spins are there. So what you can do is in the AX spin system, you know, we already saw that there are two doublets like this in the AX spin coupled system.

Now I can start selectively irradiating this proton, when I apply the power such that, it is just sufficient or more than this coupling strength, and exactly when I sit at the center of this doublet, then what happens? when this power matches this condition, this collapses into a singlet. This is what is called selective coupling. The broadband homonuclear decoupling we have to do it in a different way, that is called two shift NMR. Here this is two spins example I took. There can be more spins coupled; and a number of spins can be coupled and if they are all different spins coupled, peaks coming at different positions like here, there is a bunch here, there is a bunch of peaks here, then you can selectively irradiate here, apply rf power here, and see the effects somewhere in this spectrum. Let us say this is a four line pattern, if this proton is coupled to this and also coupled to this, no problem; it will coupled to proton A and proton B, let us say, when I hit at this frequency with a second radio frequency pulse, then coupling of this to this proton is broken, but then instead of doublet of a doublet, it will become only a doublet. So now I know this proton is coupled to this, and I can also do this selectively, I can decouple this proton, then if this and this are coupled, to this proton now when I hit separately this, again this coupling will get collapse and will get a doublet, you understand the logic?

So a very important thing; we can do in the case of protons is also homonuclear decoupling, and selective decoupling. So this is just a concept of decoupling, you remember. What basically we do is when we irradiate the spin is saturated, so that the coupled spin will not see the irradiated of spin in either of the two energy states, alpha or beta, and it is going to see only the average spin state or more or less it is a state, which is analogous to a degenerate state; so that no separation of energy levels. As a consequence it appears as if there is no coupling.

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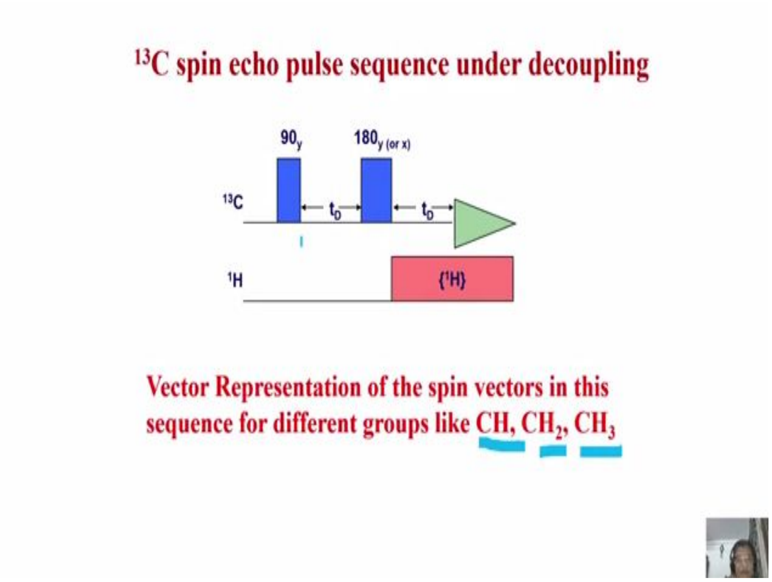

^{13}C spin echo pulse sequence under decoupling

Vector Representation of the spin vectors in this sequence for different groups like CH, CH_2, CH_3

With this now comes back to carbon 13 spin echoes. In this sequence with a decoupling this is what we do, this is what we understood, what is the spin echo. Now we are looking at carbon 13 at the same time I am trying to do the decoupling here. Now I will represent these spin vectors, what happens to different pulses sequences like we explained for the spin echoes in previous example, we can try this to find out what happens to different carbons which are attached to different protons like CH carbon, CH2 and CH3; all those things can be now discussed like this.

For example, if this is CH carbon, CH2 and CH3, we will see how they evolve in different parts of the pulse sequence, as a function of time.

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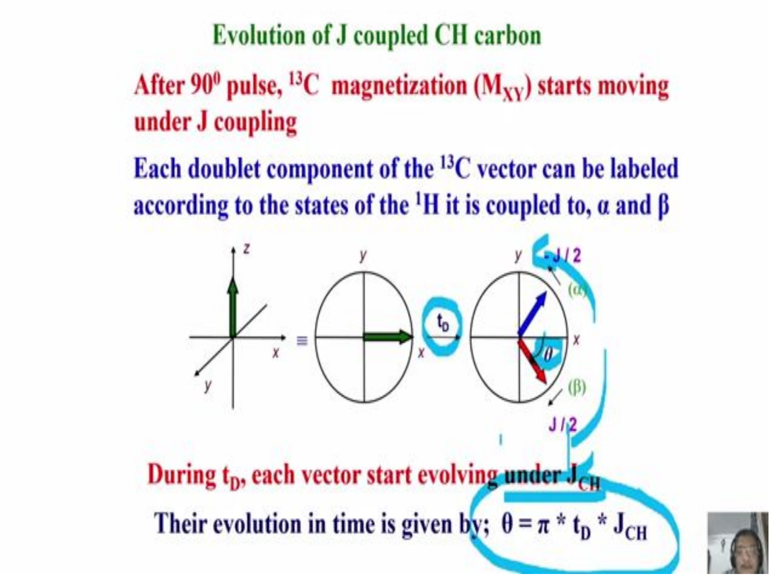

Evolution of J coupled CH carbon

After 90^0 pulse, ^{13}C magnetization (M_{XY}) starts moving under J coupling

Each doublet component of the ^{13}C vector can be labeled according to the states of the ^1H it is coupled to, α and β

During t_D, each vector start evolving under J_{CH}

Their evolution in time is given by; $\theta = \pi * t_D * J_{CH}$

Let us look at the evolution of J coupling for CH protons. What is happening to the spins after a 90 degree pulse, magnetization is coming to the XY plane. It starts moving. Up to this you know, in the pulse sequence, this we have already discussed. Now I am going step by step to see what happens to the CH vector. Carbon coupled to proton, it is is only a doublet, carbon will become doublet because of CH.

Now at this stage what happens, at this stage what happens for the CH vector, we will see. Now the moment you bring the magnetization to one axis, X or minus X or minus Y does not matter. Now just giving you a drawing. It is brought to one of the axes from Z axis to X axis, let us say, by applying a appropriate 90 pulse. Allow it evolve for time tD. Exactly what happens, as I said one is the fast moving component, and other is a slow moving component.

There are only two components for CH because it is a doublet, doublet comes because of alpha and beta spin states of protons right? So one alpha component of this carbon 13 starts moving like this; and the beta components starts moving like this, you understand. So one is going like this and other is moving like this; and then at any given instant of time, let us say, I can find out the angle theta by which these vectors are moved, I can understand. I can find out what is the angle by which this has moved, at any given instant of time. Now each vector starts evolving under the coupling of JCH. How much it has evolved at a given time, theta is given by this simple expression, please remember this. The theta, the angle in which these vectors are moved, the theta angle can be given by pi x tD x JCH. It depends upon 3 factors, Pi of course, one full rotation is pi; and it depends upon the delay; how much time you have waited in this pulse sequence, And then what is the strength of the coupling? Depending upon the strength of the coupling and the time for which you have waited, tD, I can calculate how much these vectors are moved.

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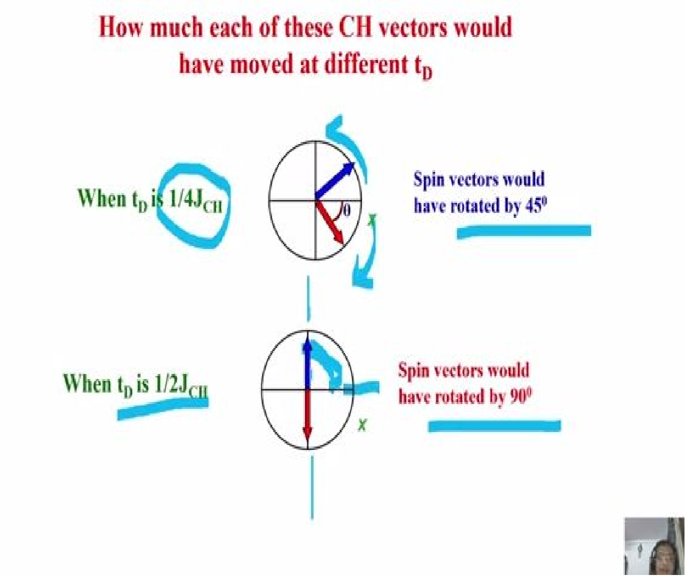

How much each of these CH vectors would have moved at different t_D

When t_D is $1/4J_{CH}$

Spin vectors would have rotated by 45^0

When t_D is $1/2J_{CH}$

Spin vectors would have rotated by 90^0

Now let us consider a situation when the time delay is exactly equal to 1/4 J, fit to this equation; now time delay is exactly equal to 1 / 4 J, then tD is equal to 1 /4 J, J will go, and it will become pi /4. Then what will happen? It has moved by 45 degrees, it is exactly moved by 45 degrees when tD is exactly equal to 1 / 4 J. just simply plug this value into this, 1/ 4 J; J, J cancel out. 1 over 4, it is pi / 4, pi radians is 180 degrees, pi / 4 is 45 degrees, it has moved by 45 degrees.

Now wait for some time, let us say the tD is now not 1 / 4, 1 / 2 J, put it into the equation, now what is going to happen? They are rotated by 90 degrees, so the fast moving components slow moving components, the one which is moving like this, the one which is moving like this, now one comes along +Z axis, one comes from –Z axis. They are now exactly moved by 180 degrees.

This is what happens, they have moved by 90 degrees with respect to the axis it was there. Between them, of course 180 degree difference is there, but each vector is moved away from the axis in which it was tilted by 90 degree.

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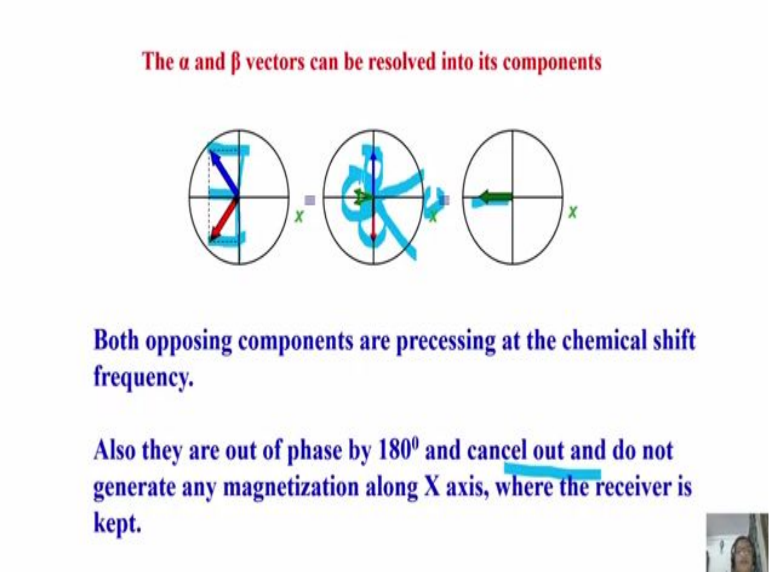

The α and β vectors can be resolved into its components

Both opposing components are precessing at the chemical shift frequency.

Also they are out of phase by 180^0 and cancel out and do not generate any magnetization along X axis, where the receiver is kept.

After the time delay of tD, a pi pulse is applied now. After this time delay of tD, a pi pulse is applied. This results in the rotation of the magnetization, along the X axis. Thus the pi pulse inverts the magnetization, you understand? I will show you the vector diagram like this. So either that was only the explanation I was giving you about what happened to the magnetization at different times.

Now, let us say, at any given time when it is rotated I will apply let us say, after a certain time that is when they are at pi/4, I apply 180 pulse. In the spin of echoes sequence we explained you know, it gets completely inverted. So it starts rotating like this, when we apply the 180 degree pulse what is going to happen? It inverts the magnetization, so the magnetization, the one which was here, which was here, now comes in this axis.

What was in the first and the third or fourth quadrant, now comes to the second or third quadrant. Now what is going to happen? They continue to move in the same direction. But now we will do one important thing, what we can do is, this is a vector this is a vector; can I not resolve these into two components? One component here, one component here, so there are two components: one along this axis, one along this axis.

So if I take the component along X axis and Y axis, this opposite components; this and this, what will happen, the vectors in the opposite direction, they just nullified. Only components along this axis start the adding up, it is a vector addition. They start moving with this direction and after the same time they start adding up. The opposing component will go, because they are out of this 180 degree and cancel out, and there is no magnetization generated by these two components because they cancel out. Whereas our receiver is kept here, let us X axis, it will not give magnetization, but only this component will give magnetization. Understand the point? Very clearly alpha and beta vectors now we resolved into two vector components, opposing components gets nullified, the components moving in one of the axis like these, non opposing components; they start adding up. And that is the one which generates signals in the receiver.

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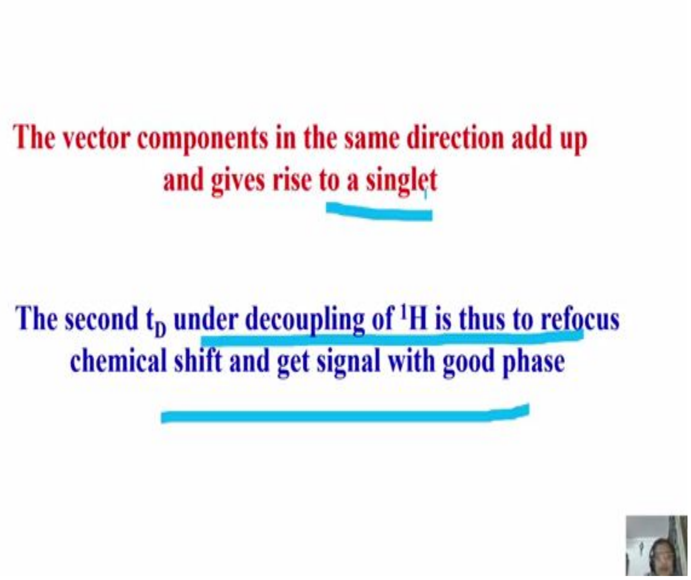

The vector components in the same direction add up and gives rise to a singlet

The second t_D under decoupling of ^1H is thus to refocus chemical shift and get signal with good phase

The vector component in the same direction add up gives a singlet and second tD and a decoupling of these two vectors is to refocus chemical shift and get a good signal phase. What it means is, now the second one, you give some time delay here, after some time now without giving a delay now after start decoupling they are all in different phases, so what you have to do is have to give another time delay so that all of them completely get refocused here.

That is what the thing you do; give a time delay so that all the spin vectors start getting refocused and the chemical shift gets refocused, then there is no phase distortion they are going to get the signal with a good phase, that is what is to be done.

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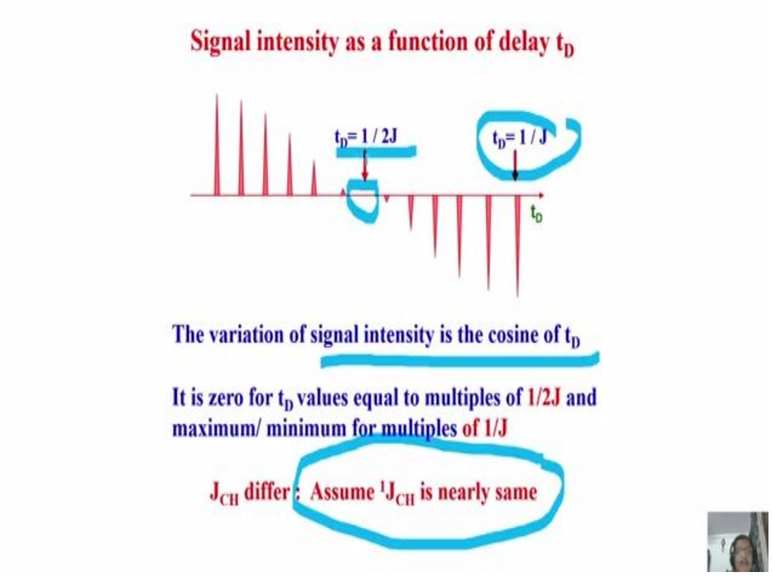

Signal intensity as a function of delay t_D

The variation of signal intensity is the cosine of t_D

It is zero for t_D values equal to multiples of 1/2J and maximum/ minimum for multiples of 1/J

J_{CH} differ : Assume $^1J_{CH}$ is nearly same

Now, let us see the signal intensity as a function of time. See when we measure the signal intensity as a function of time, when the exact time delay is equal to 1 /2J, signal intensity is 0, with the exact 1 /J, the signal intercity is negative maximum, what does it mean? The variation in signal intensity is following a cosine function. It is a cosine function starts, with the maximum comes here, 0, negative and then go on. It is an oscillatory function, follows a cosine function. Now this function is 0 for the tD values of 1 /2J and maximum for multiples of 1 / J. Of course, you may now ask me question, different carbons have different couplings with protons, how do I know which is J? Approximately we have to take the average, they are nearly the same, take the average value.

So this is what happens for CH carbons, remember, in a spin echo sequence with decoupling we observed, we saw that at exact time delay equal to 1/ 2J, the signal intensity is 0, at exactly 1/J, signal is negative maximum, this is an important point which you must understand. So what we will do is we will come back and understand what happens to CH2 pulse, CH2 vector, CH3 vectors, then we know how we can utilize this to identify, edit carbons depending upon the number of protons attached to it. We will come back and continue tomorrow.