Principles and Applications of Electron Paramagnetic Resonance Spectroscopy
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Lecture - 19
Photochemistry and EPR Spectroscopy

Today we are going to study the application of electron paramagnetic resonance spectroscopy to photochemical research. So, that is photochemistry and EPR.

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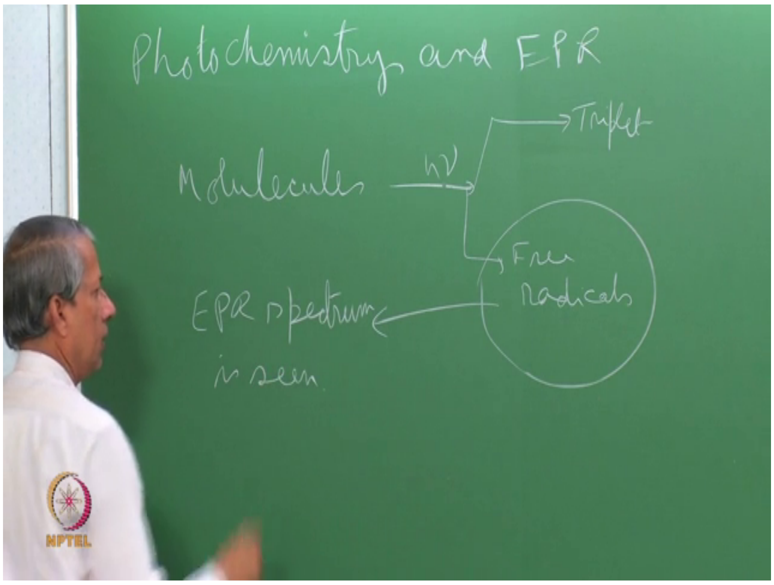

So, what happens when molecules absorb light, molecules are exposed to light what happens?

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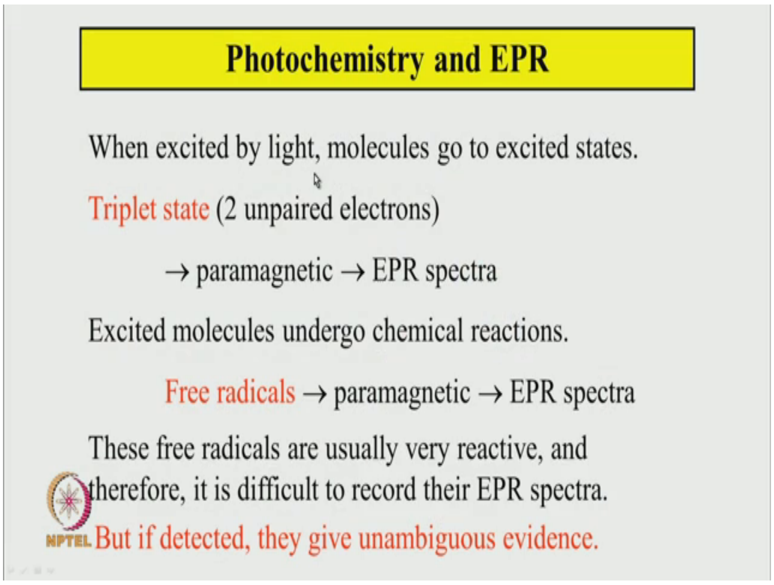

Photochemistry and EPR

When excited by light, molecules go to excited states.

Triplet state (2 unpaired electrons)

→ paramagnetic → EPR spectra

Excited molecules undergo chemical reactions.

Free radicals → paramagnetic → EPR spectra

These free radicals are usually very reactive, and therefore, it is difficult to record their EPR spectra.

But if detected, they give unambiguous evidence.

So, when excited by light molecule will go to excited state provided the light is absorbed by the molecule.

Now, most of the molecules in the ground state are singlet all electrons are paired. So, the first excitation usually brings the molecule to the singlet state and from singlet state the molecule can go to triplet state and then triplet state there are 2 unpaired electrons and you know that unpaired electrons means the system is paramagnetic. So, here 2 electrons are unpaired so it will be paramagnetic species. So, the triplet can give rise to EPR spectra.

Similarly, the excited molecule can undergo chemical reaction and chemical reaction sometimes produces free radical, free radicals also have unpaired electrons and they are paramagnetic. So, they can give rise to EPR spectra. So, here I have 2 types of possibilities 1 is triplet, other is free radicals and both can be studied by EPR spectroscopy. Today we will concentrate on this aspect and not discuss the EPR spectroscopy of triplets, now free radicals because they have unpaired electrons usually they are very reactive they always try to reactive something else. So, it is very difficult therefore, to capture them and see their EPR spectrum.

So, it is not going to be easy task to capture this thing in a EPR spectrometer. So, it is possible that molecules absorb light and does some photochemistry and they just spectrometer is not able to detect them. So, then not seeing EPR spectrum does not necessarily mean the reaction is not taking place, but if you do see this thing suppose EPR spectrum is seen, then there is unambiguous evidence that radical is formed and whatever reaction mechanism 1 can think of at least partly supported by the observation of this. What I am trying to say is that, if you do not see the then EPR spectrum of any radical it does not mean it is not taking place.

So, observation is a definitely proof not observation is not the proof that it does not happen. So, that is why I have written in this slide that if detected they give unambiguous evidence.

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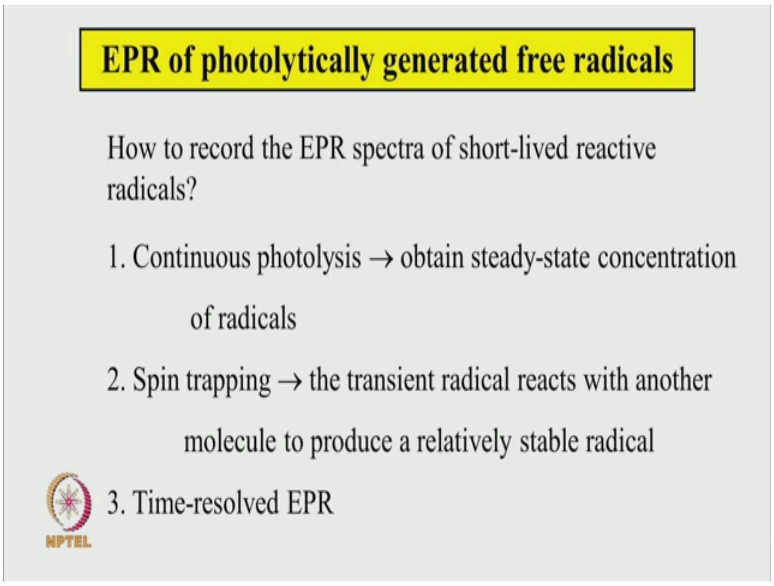

EPR of photolytically generated free radicals

How to record the EPR spectra of short-lived reactive radicals?

1. Continuous photolysis → obtain steady-state concentration of radicals
2. Spin trapping → the transient radical reacts with another molecule to produce a relatively stable radical
3. Time-resolved EPR

So, because these are very short lived reactive radicals, how does 1 really detect them and try to capture them even if they are short lived. So, we have to find some way of doing the experiment, unlike the examples that we have seen in the EPR lectures almost all the radical that we showed are stable reasonably stable radical. So, that you can scan the magnetic field very slowly and get the spectrum, radicals do not die during recording here, because they are short lived some special techniques need to be adopted.

The 3 ways one can do the experiment here shown in the slide 1 is that continuous photolysis, let the reaction be carried all the time and light is on the sample and we can a hope to get some steady state concentration of the radical and that can be seen in a steady state EPR spectrometer, another possibility is called spin trapping in this spin trapping technique this transient radical, that is radical which do not live for long time they react with some other molecule to produce a relatively stable radical.

So, this stable radical can be seen in the steady state EPR spectrometer and 3rd one is called time resolved EPR spectroscopy, where we try to capture the radical as soon as they are created and will see later how that is done, this continuous photolysis.

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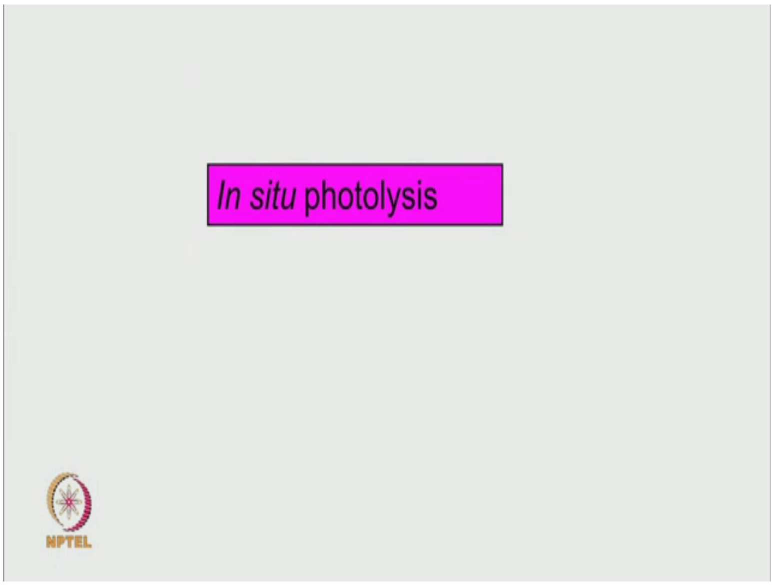

In situ photolysis

So, here to capture the radical as they are formed, we have to keep on continue the photolysis and try to record the spectrum. So, how that is done?

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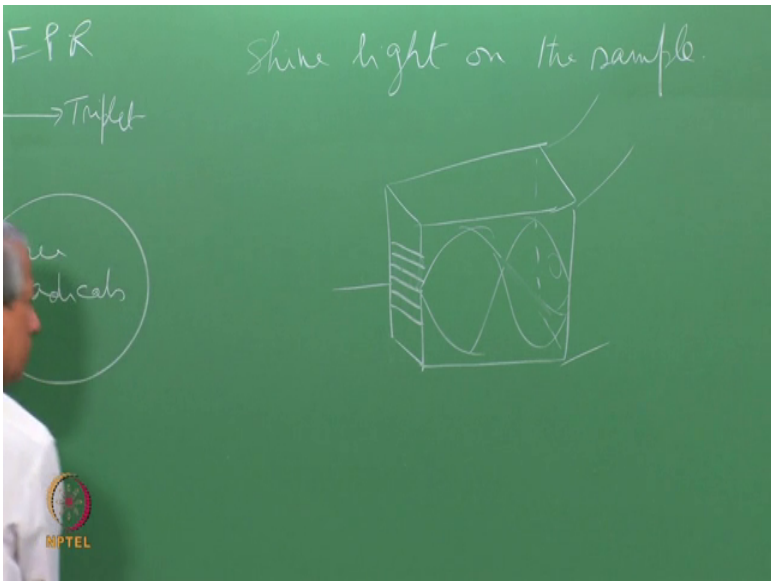

So, here first I need to Shine light on the sample and this will be done in shine to that is we cannot carry the this excitation somewhere else and bring the sample to the spectrometer and hope to get the spectrum that is not possible it has to be done inside the spectrometer to a specific, it is has to be done inside the microwave cavity of the spectrometer. So, how is it carried out?

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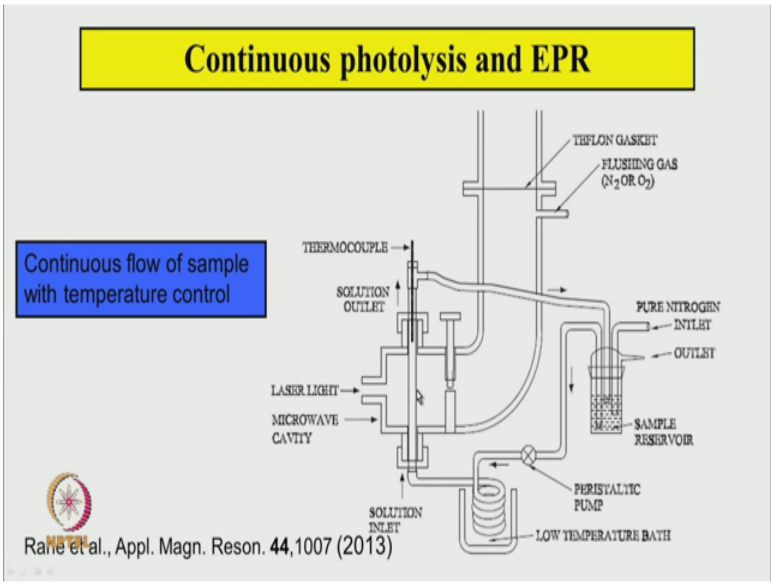

Continuous photolysis and EPR

Continuous flow of sample with temperature control

Rane et al., Appl. Magn. Reson. **44**,1007 (2013)

For that here is a design this is the microwave cavity and this is the iris tuning screw. So, we could put a sample tube and let the sample which is kept here reaction mixture can be taken through a pump and this is a low temperature bath and through this liquid goes out here and then again it can be brought back to the reservoir.

Now, here there is a hole which is kept here in front of the cavity and light can enter through this either laser light or it could be steady sate lamp could be inserted here. So, you know cavity looks like this, a iris hole and this goes to the waveguide here. So, for this end was blocked, but we could open a hole here and allow the light to go through, but trouble is that if you allow light to go through and hole is drilled here and microwave can also leak out of that. So that will reduce the key of the cavity. So, that is not very desirable so what is done here is that some manufacturer instead of opening a hole they make a grid type of arrangement here, is tiny gaps are kept there. So, that. So, far as the microwave is concerned, which wavelengths are fairly beamed they find the almost a continuous metal plane so it will not escape.

So, this is one way of, but the allowing the light to go through this holes are thin, but the trouble of doing this is that much of the light is not penetrating, they are blocked by this grills here.

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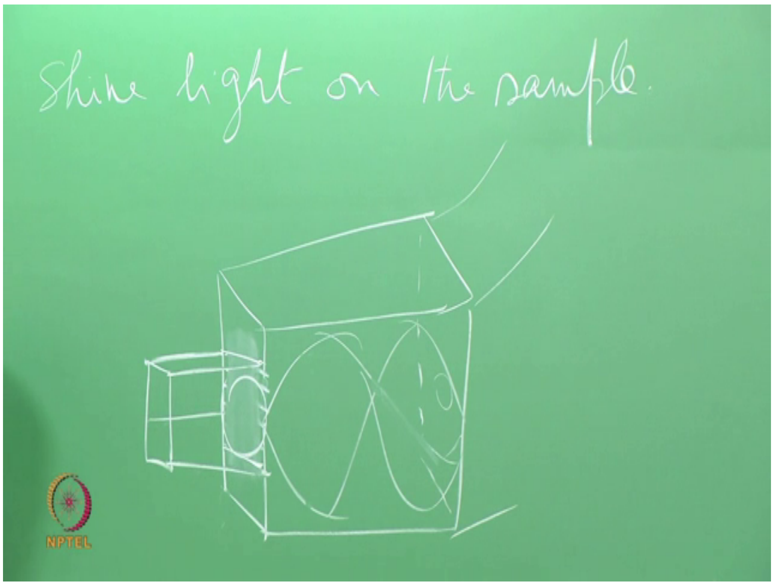

So, another technique is to actually have a hole here and let another piece of small waveguide which is mounted here. So, that the wavelength of this is such that this really cannot sustain within this, this is much smaller dimension this one. So, this almost act like a like a tube which blocks the wave from entering here, that way there could be a small slight loss of the cavity cube**,** but never the less this will work very well particularly because, see the hole is clear all the light will go through the microwave cavity. And this thermostat arrange low temperature bath and having a liquid to go through this is to have some control in the temperature, because at lower temperature radicals could live for some more longer time given, making it easier to record the EPR spectrum another possibilities is that.

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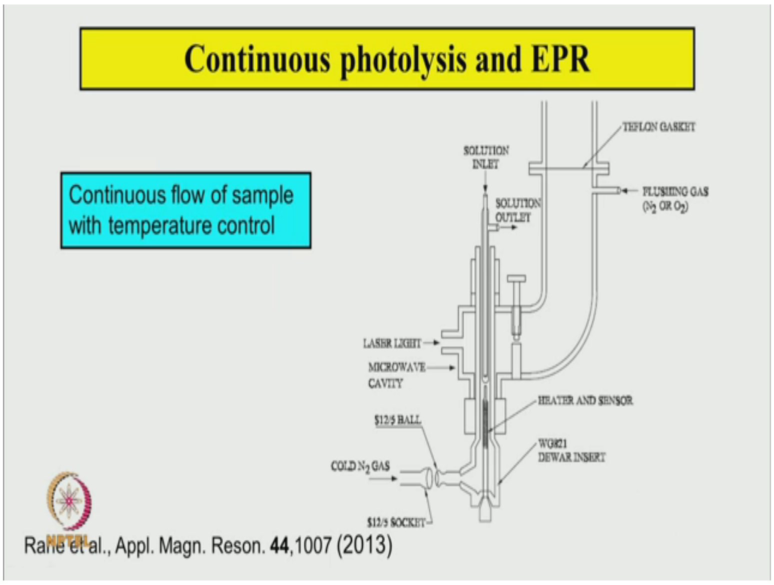

Continuous photolysis and EPR

Continuous flow of sample with temperature control

Rane et al., Appl. Magn. Reson. **44**,1007 (2013)

Here the there is a special tube which is used here which looks like this.

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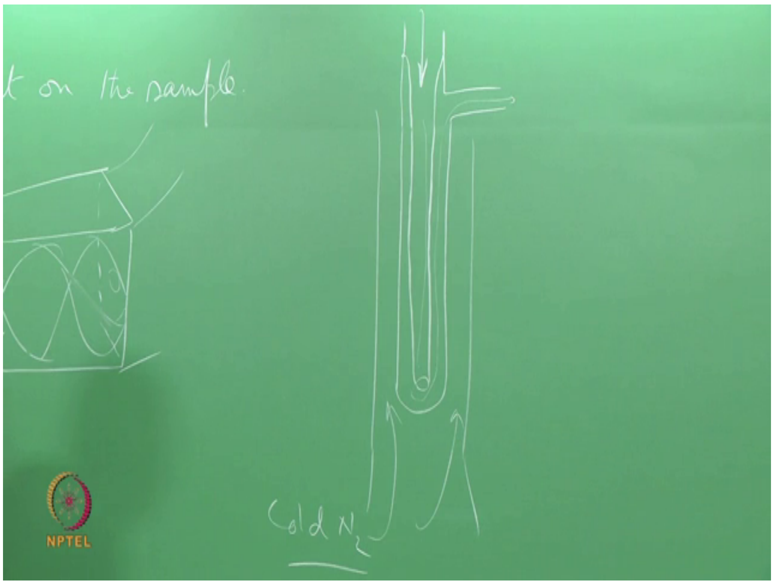

So, sample the liquid sample enters here and then comes out and through this it goes out here and this is kept inside a standard variable temperature Dewar insert, through which cold nitrogen gas is passed through that. So, that sample is cooled by this cold gas here and if you flow this liquid reasonably slowly, then it will acquire some sort of steady state temperature which is decided by the flow of this cold nitrogen gas. And the temperature is measured by this heater sensor assembly here and previous one temperature is measured by the thermocouple here.

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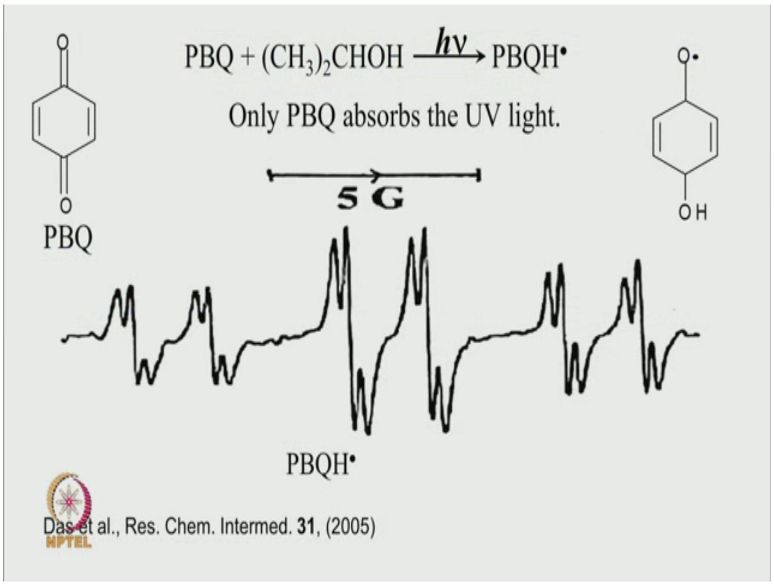

$PBQ + (CH_3)_2CHOH \xrightarrow{h\nu} PBQH^{\bullet}$

Only PBQ absorbs the UV light.

Das et al., Res. Chem. Intermed. **31**, (2005)

Here is an example now having described how the experiment is done, this experiment involves exciting para benzoquinone in this and we shine light and see what happens.

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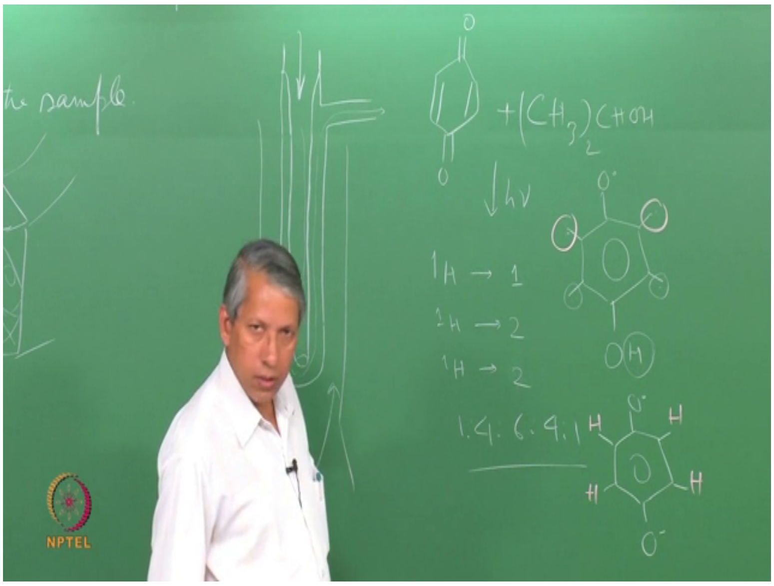

So, the light is on all the time. So, sample is flowing through this and we are recording the steady state EPR spectrum, the way we have described earlier. The experiment is very simple now except for this special modification on the cavity and the flow system that is used to continuously replace the sample. So, here signal that is seen here is given in this slide. So, this is triplet another triplet this triplet overall you can see the this gap is same as this gap, we can now guess there without making any careful measurement this gap is also same as this gap, this will is individual triplet also have similar hyperfine splitting and overall this intensity of these and these follow 1 is to 2 is to 1. So that means, this radical has one spin with one proton hydrogen proton because, the some the chemicals are made of basically proton magnetic nuclei, so this gives one proton of one number 1 another proton, which you know two of them and another one pro which is 1.

So, this gives a triplet that is these one is to 2 is to 1. This one gives doublet. So, this is mistake because this also gives triplet. So, this will be triplet. So, this is the type of a radical, which the radical should have this type of protons. So, here is the structure shown which is consistent with this spectrum and that is so you see that this one proton which gives a doublet splitting and these and these are equivalent protons. So, they also they give rise to a triplet and another proton here and another one here these two are also equivalent, but they are different from this. So, they give rise to a triplet. So, here this coupling constant due to unpaired quite different for the coupling constant to other one. So, that is why the spectrum looks like.

So, these definitely shows that radical must be same this is the only the possible radical that is consistent with the spectrum and knowing the chemical nature of the reactants nothing else can possibly give rise to similar EPR spectrum. So, the observation is therefore, very definitude.

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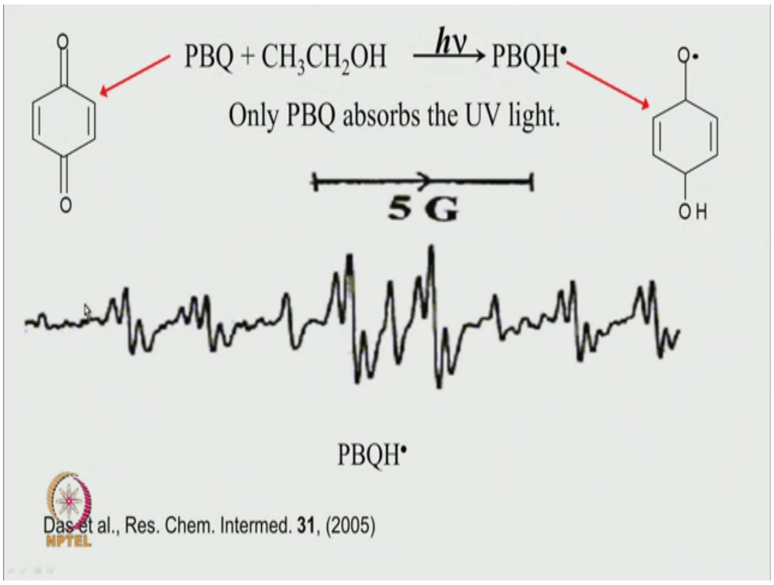

$PBQ + CH_3CH_2OH \xrightarrow{h\nu} PBQH^{\bullet}$

Only PBQ absorbs the UV light.

PBQH•

Das et al., Res. Chem. Intermed. **31**, (2005)

So, instead of now is isopropenon if you take ethanol, it is what happens it again looks that a similar pattern triplet this triplet. So, this must be same radical as this 1, but in addition it is some smaller lines here this one. So, what are these things, little hump is here. So, here then one can get some idea by measuring the relative heights of these intensities, so even if the now that I say little bit here and gap between these and these same as between these and these. So, there should be a partner of this little line somewhere here, which even if I do not see it I have to assume that is there and the intensity ratio of these turns out to be 1 4 6 4 1 . So, this some other radical is also formed here along with this radical and knowing the again whatever chemicals are there the most intelligent guess will be that this corresponds to here.

So, this is are neutral semiquinone radical, which is there when these dissociates to make it anion radical then it will have this one pair of protons and another pair of protons, now here because of the delocalization all 4 protons become equivalent and then I can get this sort of spectrum which is consistent with this. See for isopropanol I get this radical only, but in ethanol this radical is predominantly seen, but this also seen to a certain extent.

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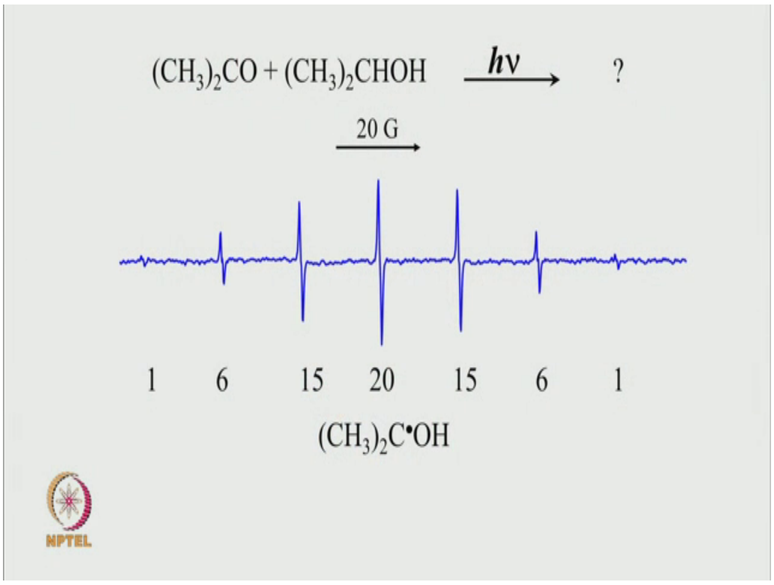

$(CH_3)_2CO + (CH_3)_2CHOH \xrightarrow{h\nu} ?$

$(CH_3)_2C^{\bullet}OH$

Another example here we use this acetone plus isopropanol and shine light.

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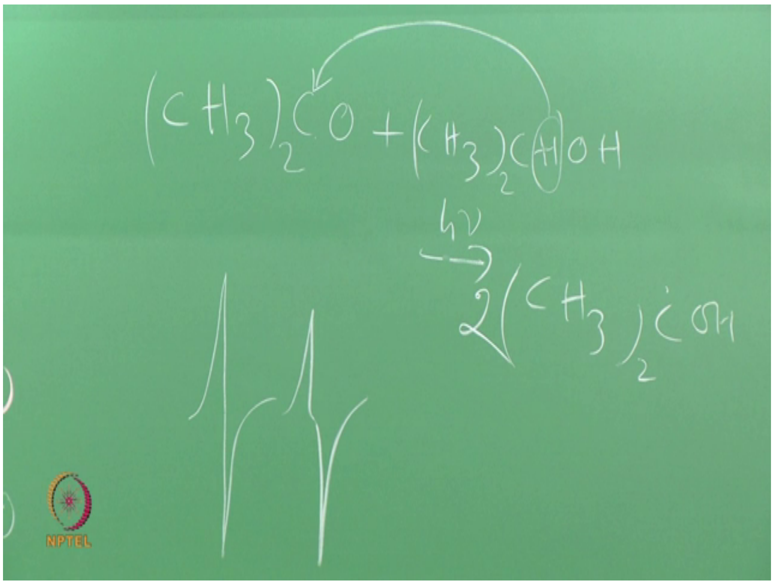

$(CH_3)_2CO + (CH_3)_2CHOH \xrightarrow{h\nu} 2(CH_3)_2\dot{C}OH$

So, by the way here the light that is used here that light is absorbed by this one, this is the one which absorbs near UV region this does not absorb. So, these go to excited state and does photochemistry and produce this radical.

Similarly, here this carbonyl group here of acetone, that absorbs u v light and that goes to excited state reacts with this and produces some radical and this is a result of that. So, here this lines are seen now 7 lines 1 2 3 4 5 6 7 and if you see it is maybe it is not very perceptible, but they do not quite look like one transition there, this is what I discussed in one of the earlier lecture see this is the one should be careful if there are overlap lines are there or not. So, here one can see these sets of discontinuities are there. So, there is a indication that some other proton is also involved in coupling that. Of course one can improve the resolution and get a more direct evidence of that. So, here up to measure in the intensities of relate all the lines one gets this set of numbers and that is consistent with 6 equivalent protons and if you consider this little doublet, then the another proton which gives this small doublet and the radical which is consistent this EPR signal is this one.

So, here naturally the here one can write the reaction mechanism, that this excited state of this takes this hydrogen atom from here and produces this radical. It is so often if you remove this hydrogen atom now the protons bring it here, then this also becomes free radical. So, 2 of this thing form there. So, that is way the steady state photolysis and we can look at the radical directly if of course, the circumstances are such that there favourable then we can get this radical directly there. Of course, the condition for this is that we are doing the continuous photolysis, by shining light all the time and getting this spectrum, but radical slipping short live they also dying at the same time. So, what about the steady state concentrations that are build up here, they will contribute to the absorb spectrum. So, if the radicals do not have subsistent steady state concentration, it may be almost impossible to see the spectrum in this fashion.

So, here comes the other technique that even if that do not live for long time, if I allow the radical to react with something else to produce some other radical and at that radical has subsistent long life time, then we could probably detect those radical, the secondary radical and try to understand the reaction mechanism and probably make some guess about what the original radical that was. So, this technique is called spin trapping EPR you trapped the free radical.

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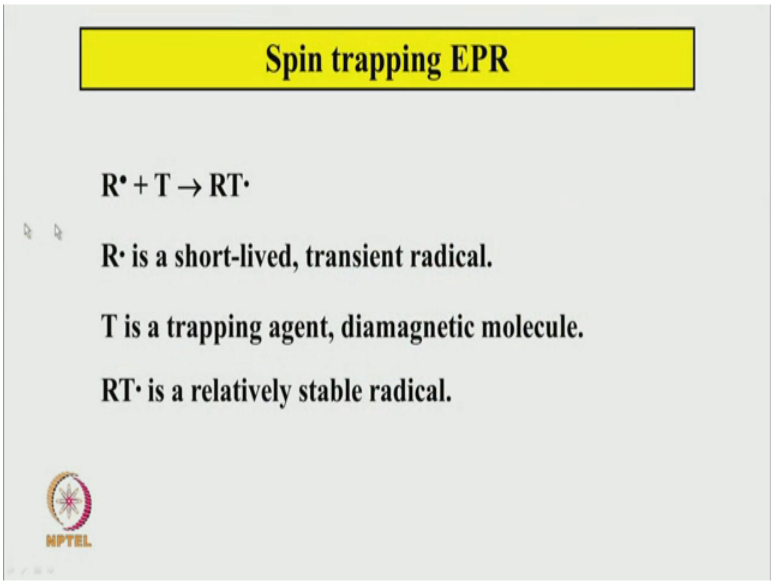

Spin trapping EPR

$R^{\bullet} + T \rightarrow RT^{\bullet}$

$R^{\bullet}$ is a short-lived, transient radical.

T is a trapping agent, diamagnetic molecule.

$RT^{\bullet}$ is a relatively stable radical.

So, it idea is very simple that r is transient radical the some trapping agent. So, this reacts with this to give another species, which is trapped radical and this is has subsistent has subsistent long life time and may be easy easier to detect.

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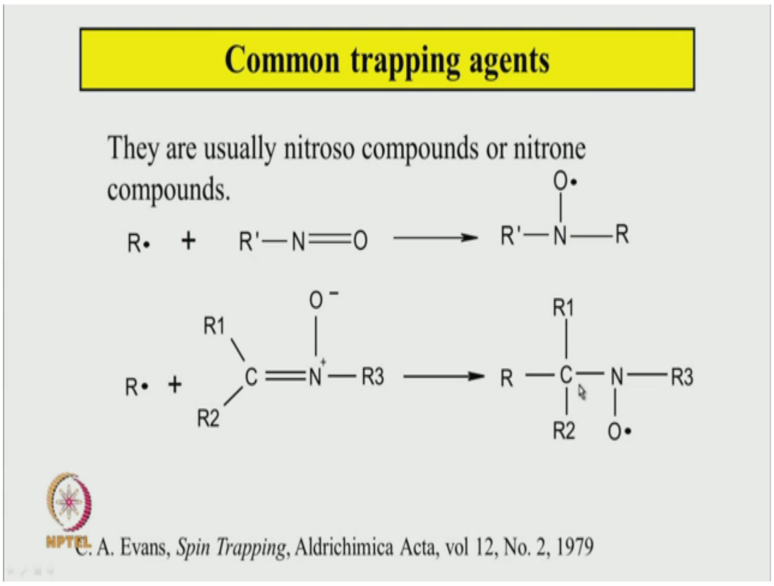

Common trapping agents

They are usually nitroso compounds or nitrone compounds.

C. A. Evans, *Spin Trapping*, Aldrichimica Acta, vol 12, No. 2, 1979

These are some common trapping agents and often they are the nitroso compounds NO group, this is diamagnetic phase the radical comes here and attacks here to form a radical, these could premium will be should have subsistent long life time and easy to detect another one is of this kind. So, this radical comes and attacks here produce a radical of this kind, then this could have long life time then you can see it.

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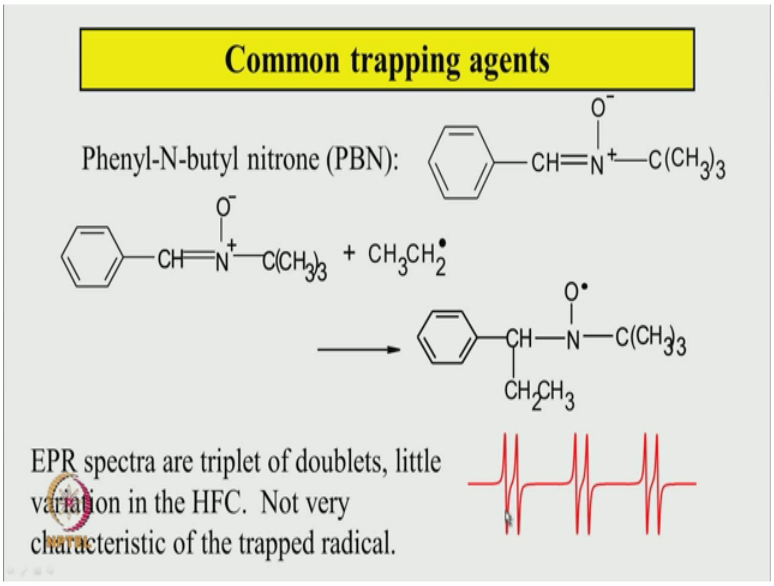

Common trapping agents

Phenyl-N-butyl nitrone (PBN):

EPR spectra are triplet of doublets, little variation in the HFC. Not very characteristic of the trapped radical.

So, one of the common trapping agent is this molecule called Phenyl N butyl nitrone or called PBN it is similar to this.

So, phenyl group is here so here is a example, so if ethyl radical is coming attacking here then the radical that form is NO dot. So, what will be the spectrum of that this oxygen has the radical centre. So, nitrogen nearby so this will be triplet because of nitrogen spin one and then adjacent that is this single proton, which is coming from this trapping agent. So, that gives doublet this is type of splitting one gets for this set of trapped radical and since the actual radical is sitting somewhere here which is rather far from this radical centre, this spectrum does not depend very much on the type of radical which is seen in the spectrum. So, will be often all almost always be a triplet an is spin by doublet because of this one, there could very small difference of the coupling constant here. Never the less by enlarge all the trapped radical gives similar EPR spectrum.

So, this is not very characteristic evidence of what radical was trapped, all one can say is that some radical is trapped and one can think of forming a reaction mechanism.

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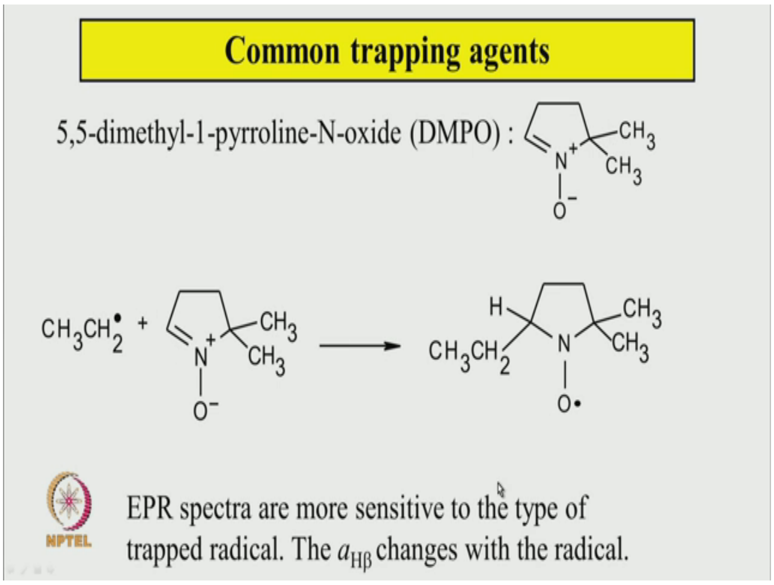

Common trapping agents

5,5-dimethyl-1-pyrroline-N-oxide (DMPO) :

EPR spectra are more sensitive to the type of trapped radical. The $a_{H\beta}$ changes with the radical.

The another common trapping agent is called DMPO, this is structure of this here it is again same ethyl radical comes and attacks here, this place then this is the trapped radical this can be detected on EPR spectrum, here again the radical centre is near nitrogen. So, this is the triplet line then there is another proton here which is also give doublet, but here this paired constants due to this is somewhat more sensitive to the presence of this radical because this is very near, that one that is hydrogen coupling constant of beta proton is does not sensitive to this.

So, it will be easier to make better individual guess about the type of radical by seeing how much of this splitting constant has changed.

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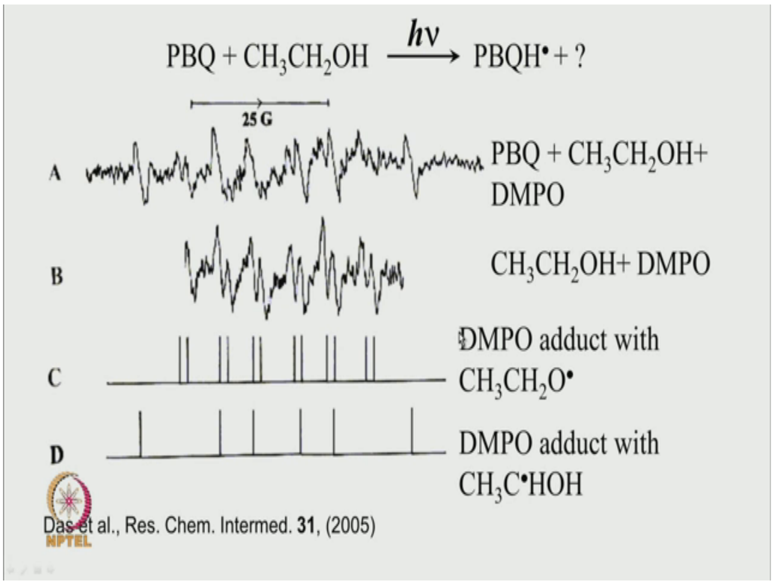

$PBQ + CH_3CH_2OH \xrightarrow{h\nu} PBQH^{\bullet} + ?$

Das et al., Res. Chem. Intermed. **31**, (2005)

Here is an example para benzoquinone, the same experiment that we did in the steady state 1 and we got only evidence of this radical without having any knowledge of what the other radical was para benzoquinone ethanol and DMPO. So, you get some lines of this kind here.

So, here see this doublet some say or may not be there some here to make some careful observation and now see the these are signals coming from this adduct, this is the CH 3 CH2O radical which are forming a adduct with these two give a radical of this kind, other one is this one. When we use ethanol sorry this is isopropanol, so these splitting constants this is the last splitting constant due to this for one trapped radical and some are smaller splitting constants to this radical so these values I write here.

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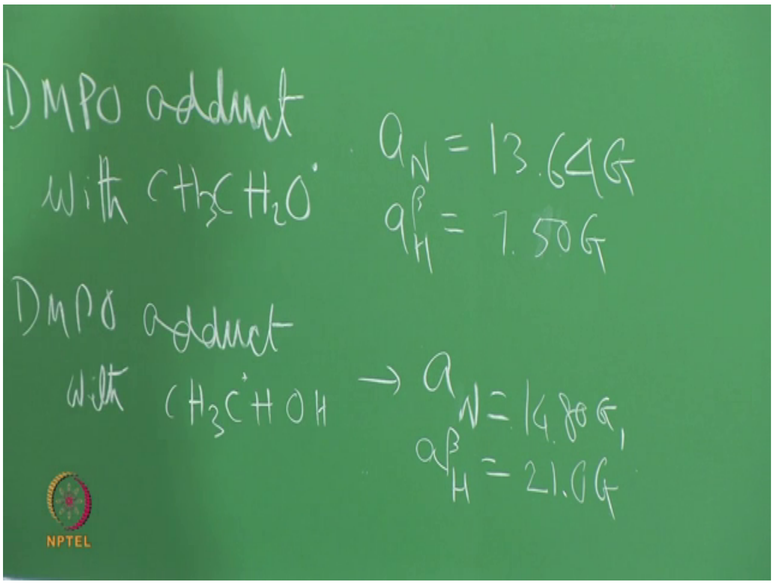

DMPO adduct with $CH_3CH_2O^{\cdot}$ $a_N = 13.64G$

$a_H^{\beta} = 7.50G$

DMPO adduct with $CH_3\dot{C}HOH$ $\rightarrow a_N = 14.80G,$

$a_H^{\beta} = 21.0G$

Adduct with this nitrogen splitting constant 13.64 gauss this beta coupling constant and similarly, adduct with 14.80 gauss and a H beta is equal to 21.0 gauss. So, here you see the difference in the hyperbolic coupling is quite appreciable, when this adduct is formed and these adduct is formed.

So, that way DMPO is much better trapping agent for identifying the radical that is trapped there.

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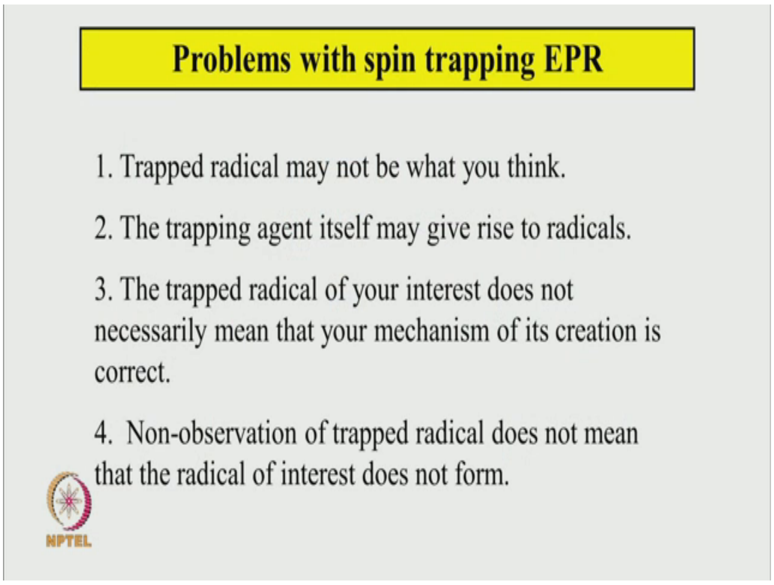

Problems with spin trapping EPR

1. Trapped radical may not be what you think.
2. The trapping agent itself may give rise to radicals.
3. The trapped radical of your interest does not necessarily mean that your mechanism of its creation is correct.
4. Non-observation of trapped radical does not mean that the radical of interest does not form.

But problem with spin trapping is that as you say the earlier is very difficult to often identify the radical that is trapped unambiguously, some time the trapping agent itself may react with the light and then produce some radicals with solvent and some time the trapped radical may not be radical that one has in mind, while formulating the reaction mechanism and finally, even if the spin trapping technique yields no signal it does not mean the radical is not forming.

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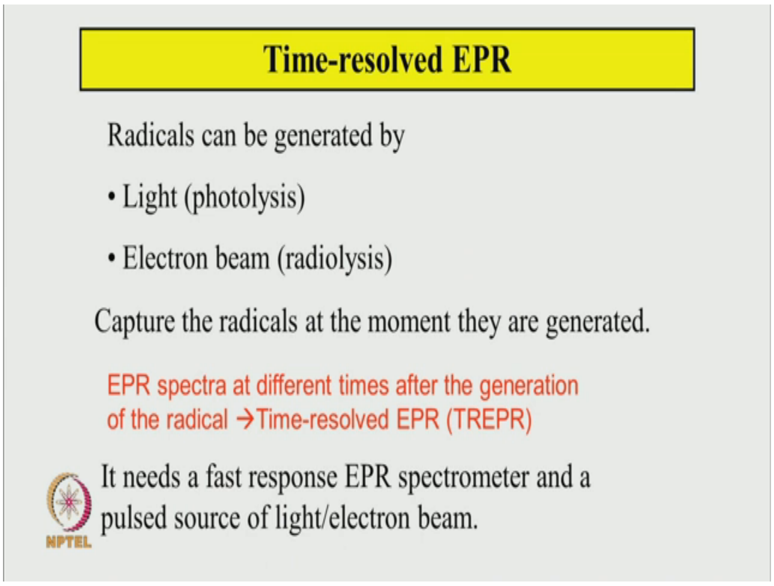

Time-resolved EPR

Radicals can be generated by

- Light (photolysis)
- Electron beam (radiolysis)

Capture the radicals at the moment they are generated.

EPR spectra at different times after the generation of the radical →Time-resolved EPR (TREPR)

It needs a fast response EPR spectrometer and a pulsed source of light/electron beam.

Finally quickly go through this time resolved EPR spectroscopy, where we tried to capture the EPR spectrum of radical which is right at the time of their formation and before they react. So, for that we usually pulse laser light or electron beam pulses and spectra are detected at different times after the generation radical, that is way it is called time resolved EPR. Naturally, it needs a fast response EPR spectrometer and a pulsed source of light or electron because; you have to compete with the reaction of the radical

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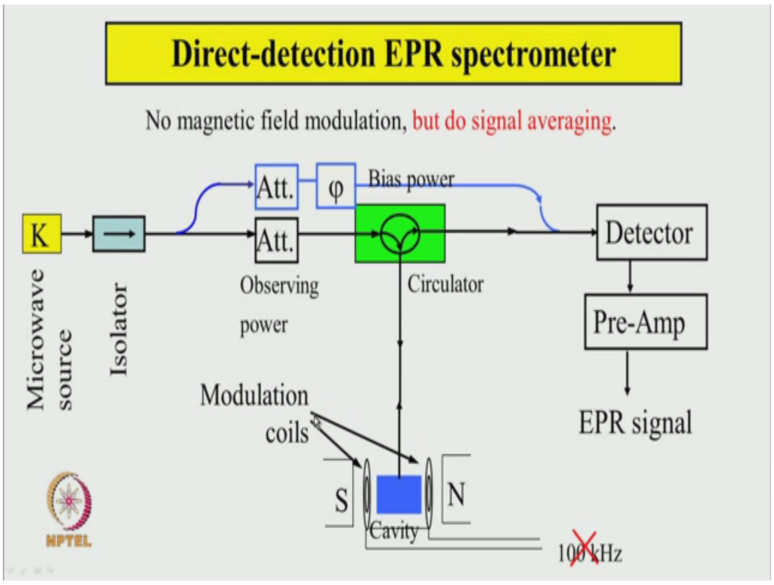

Direct-detection EPR spectrometer

No magnetic field modulation, but do signal averaging.

So, this is the one I said earlier is that we use direct detection EPR spectrometer and we do not used the field modulation technique, simply we get the output from the pre amplifier, but to improve the signal noise ratio we do signal averaging.

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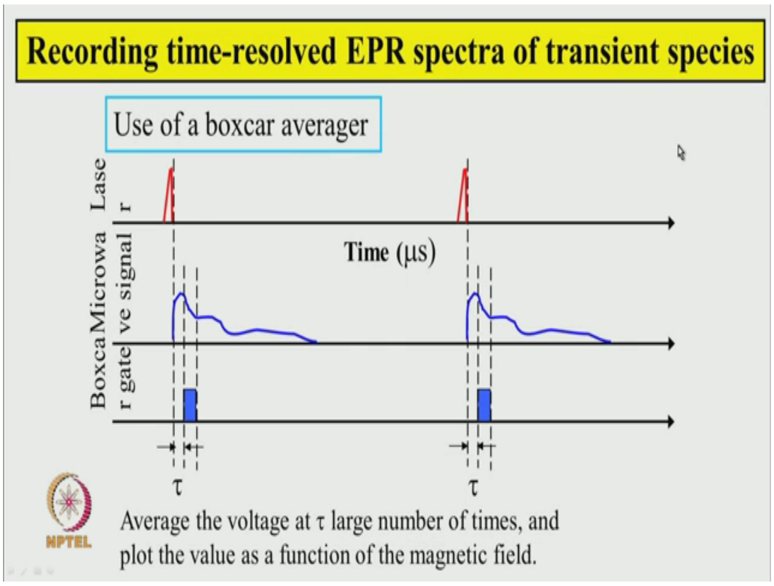

Recording time-resolved EPR spectra of transient species

Use of a boxcar averager

Average the voltage at τ large number of times, and plot the value as a function of the magnetic field.

So, signal averaging is done by this way, suppose the laser is operated and repeated more and every time microwave signal looks very similar. So, we try to get les a small portion of the voltage at given time after the laser, here given blue block here and average this many times there is a device call boxcar average, it is job is just that take a sample of the voltage at given time and then average it out.

So, if we plot this voltage at this time after the laser pulse whilst scanning, the magnetic field I can get the time resolved EPR spectrum of this species at this time after laser pulse.

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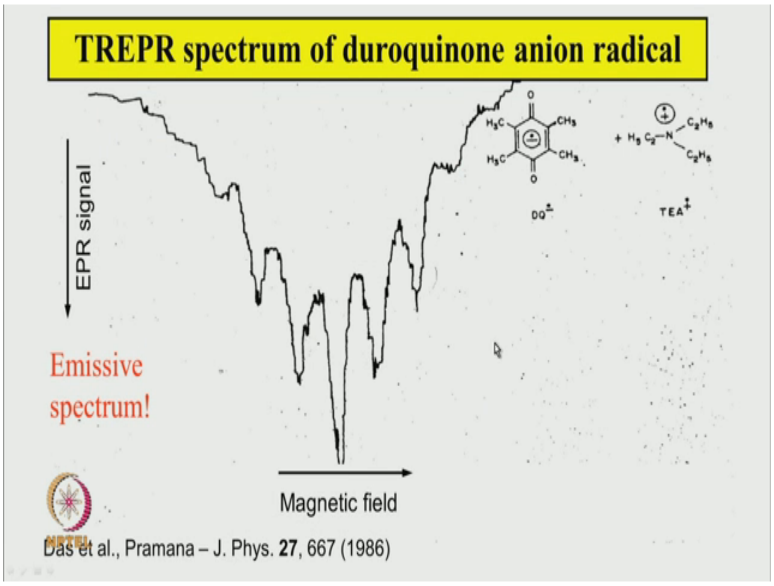

TREPR spectrum of duroquinone anion radical

Emissive spectrum!

Das et al., Pramana – J. Phys. **27**, 667 (1986)

Here is an example in duroquinone is dissolved in this triethylamine solvent and then a laser u v laser light was shown on that and this is the type of radical spectrum I get. So, this corresponds to the duroquinone anion radical of this, this is the duroquinone anion radical. Here I said earlier I direct detection technique is the absorption spectrum.

But here the way we have adjusted the spectrometer, that this is supposed to give the absorptive signal, but everything comes in a opposite direction. That is very strange they are achieving the signal is in the emissive form it is indeed very strange. So, will not try to explore why it is so, but we will postpone it for later discussion. We can do another experiment that instead of getting the spectrum as function, magnetic field we can hold the magnetic field at anyplace we like and then see the microwave signal as a function of time now and average the whole thing by using a very fast transient analogy to digital converter.

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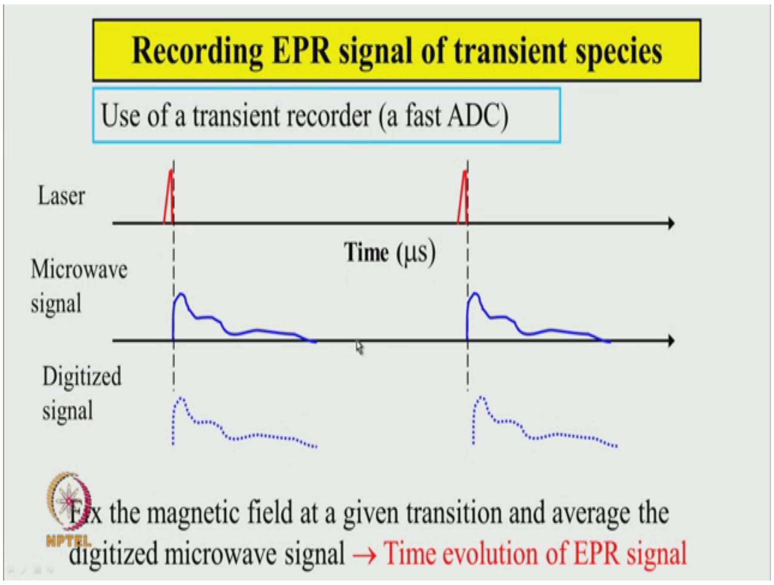

Recording EPR signal of transient species

Use of a transient recorder (a fast ADC)

Fix the magnetic field at a given transition and average the digitized microwave signal → Time evolution of EPR signal

So, transient digital converter will capture the signal in digits at sort of sampling rate of given by the user. So, this various digital values are added to generate the average value of signal by adding several of this transient.

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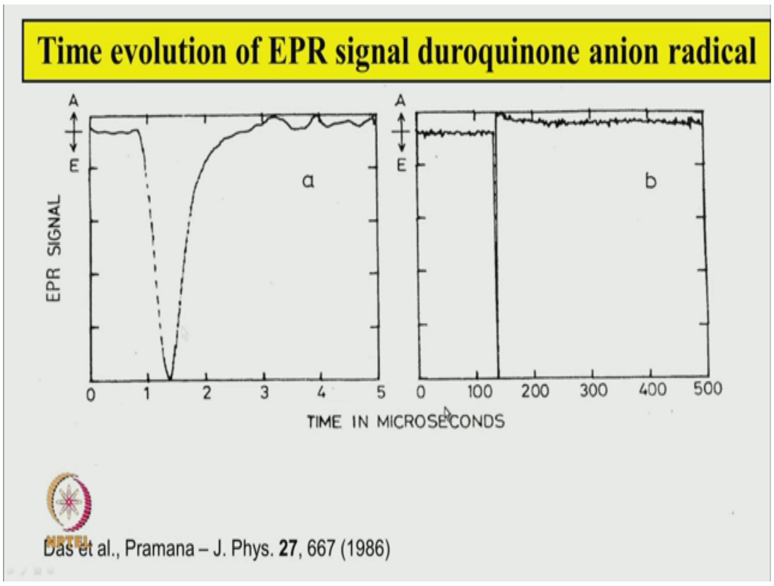

Time evolution of EPR signal duroquinone anion radical

Das et al., Pramana – J. Phys. **27**, 667 (1986)

So, here is an example so again duroquinone signal a function of time, it forms in the negative direction again it slowly goes to near 0. So, this time is of the order of few microseconds if we increase this time to. So, will 100 microsecond one can clearly see that signal first, starts emissive and then goes to absorptive and then goes to 0. So, presumably something is happening here and then radical is decaying in this fashion.

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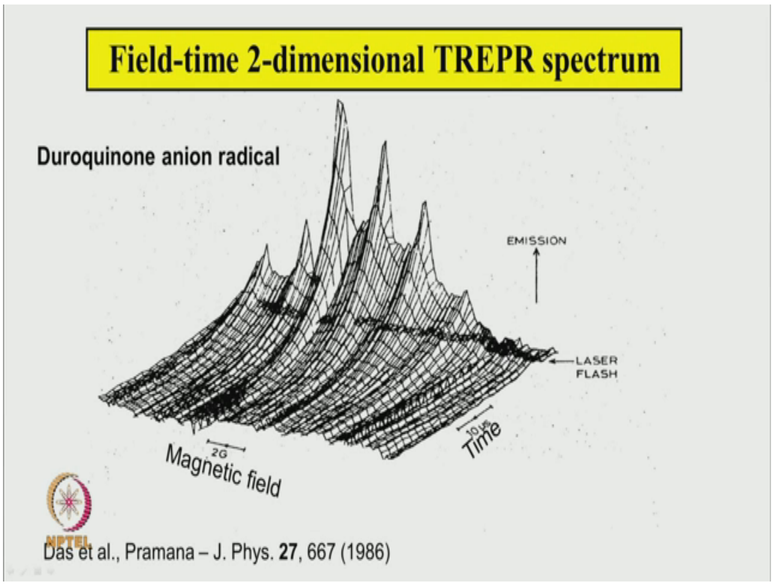

Field-time 2-dimensional TREPR spectrum

Duroquinone anion radical

Das et al., Pramana – J. Phys. **27**, 667 (1986)

So, this is the 2 dimensional spectrum, one can create by getting similar spectrum at different magnetic field as a one axis magnetic field, axis other time axis.

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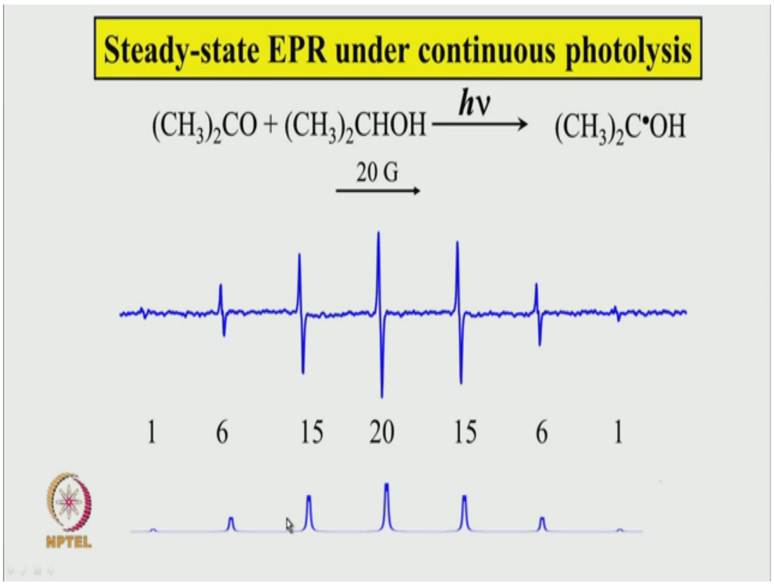

Steady-state EPR under continuous photolysis

$$(CH_3)_2CO + (CH_3)_2CHOH \xrightarrow{h\nu} (CH_3)_2C^{\bullet}OH$$

So, the example that we saw earlier in steady state photolysis, acetone and isopropanol gives a other spectrum of this kind this is the derivative signal.

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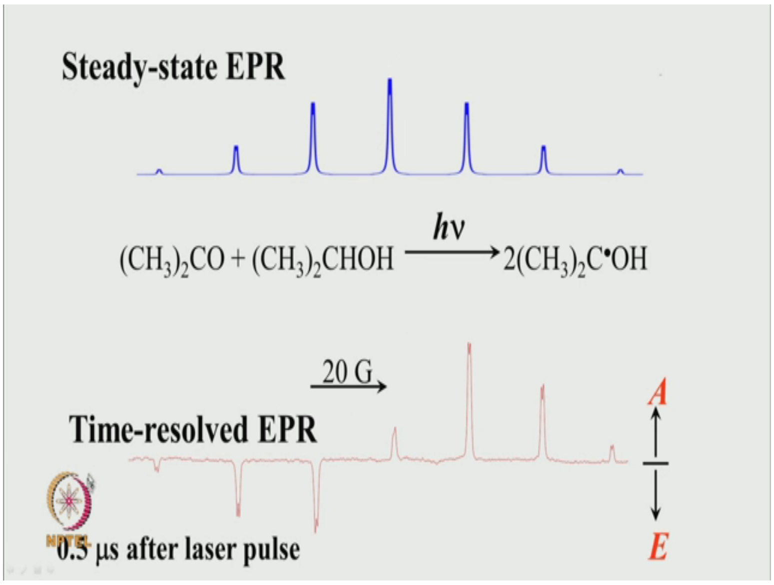

Steady-state EPR

$$(CH_3)_2CO + (CH_3)_2CHOH \xrightarrow{h\nu} 2(CH_3)_2C^{\bullet}OH$$

Time-resolved EPR

0.5 μs after laser pulse

But if we a imagine that this is the way absorptive signal and this experiment is in time resolved spectroscopy gives complete different type of signal, a intensities this is steady state spectrum that the time resolved spectrum. You see some of them are emissive some are absorptive and doublet signal much seen much more clearly here. So, naturally something unusual information is contained in this time resolved EPR spectroscopy.

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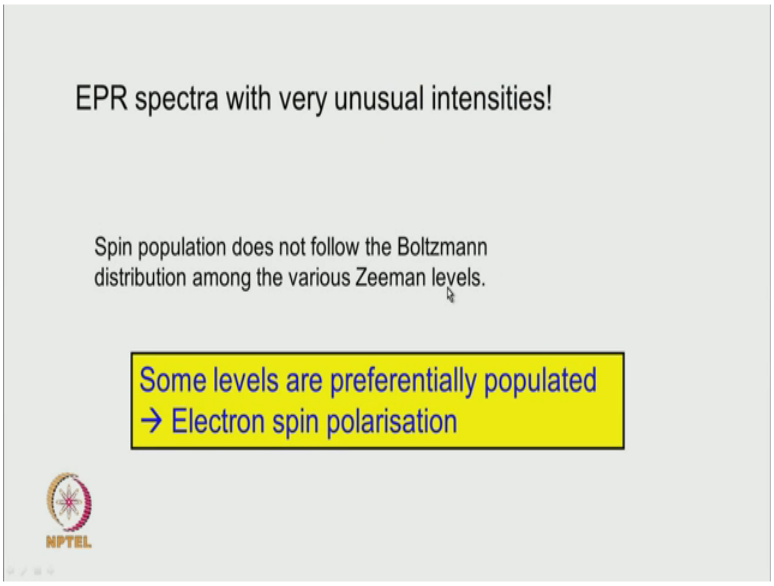

EPR spectra with very unusual intensities!

Spin population does not follow the Boltzmann distribution among the various Zeeman levels.

Some levels are preferentially populated
→ Electron spin polarisation

Therefore, the spin population does not follow the Boltzmann distribution. Boltzmann distribution ensure the lower level more populated in a higher level, but here things are all getting quite different. So, this set of information one can therefore, get in experiment of this kind and this phenomenon is called electron spin polarisation.

Right now we simply learned that it is possible to detect such phenomenon, and why it is so we will the therefore involve a further discussion which we will take up in a subsequent lecture.