Inorganic Chemistry of Life Principles & Properties
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Lecture – 51
Inorganics in medicine – PDT, MRI & Barium tests

Welcome you all to the next class on Inorganic Chemistry of Life Principles and Perspectives. Just in the immediately previous class we have started looking at the therapeutic aspects of inorganic compounds, and we have looked at the details of apoptosis that happens which is pertinent to the cancer. Another technique that we will look at another therapeutic method which is also equally well suited for the cancer therapy is the photodynamic therapy. Photodynamic therapy in the short form is known as PDT - photodynamic therapy ok.

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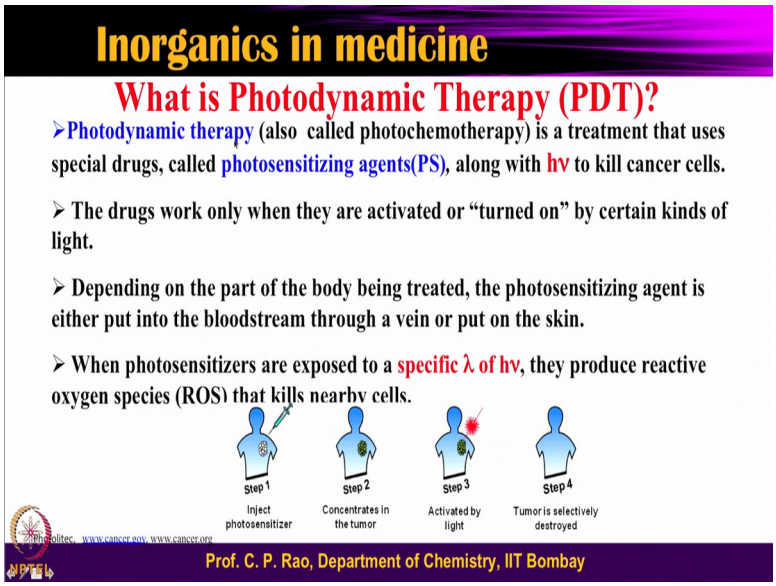

Let us look at what is the photodynamic therapy? So, it is basically in the photodynamic therapy what will would be there, there will be a molecule a compound which is which is sensitized by the photons. In other words, it can be activated it can be activated under the photons. So, and thereby the excited or activated compound would release certain things and which in turn can kill the cells.

So, therefore, so it is photodynamic therapy is nothing but is also called photo chemotherapy. What we have seen earlier is the just the chemotherapy, is a

photochemotherapy is a treatment that uses special drugs. And these drugs are photosensitizing agents or photosensitizing compounds ok or sometimes shortly known as a PS.

So, and these will act when you shine light on them, and they will kill the cancer cells. So, that means, when you shine light something is happening in these compounds, because these compounds are being sensitized by the light that means they go to the excited state. And when the excited state returns back something happens and as a result of that you can kill the cancer ok.

So, therefore, in this case the work the drug works only when the compounds are activated or you can say the activity is turned on when light is incidented on this particular compound. And that light not any kind of a light, the light suited to activate that particular compound. So, therefore, it is not a fixed wavelength for every compound, it is the compound decides what is the wavelength required? Primarily these are all into the visible radiation, visible range ok. So, depending upon the part of the body that is being treated, the photosensitizing agent is either put into the bloodstream through the veins or put into the skin. So, first you have to give the drug.

The drug is nothing but the photosensitizer so attached to that sometimes you can it can be a combined two. So, you have a sensitizer plus additional drug to that or sensitizer itself is a drug, all these kinds of things are possible. So, when photosensitizers are exposed to a specific wavelength lambda of the light, these produce certain active species, these active species called the reactive oxygen species.

And these reactive oxygen species you will see in the next slide or slide later the more details what are the kinds of things like the superoxide radical, hydroxy radicals, oxo species all these kinds of things are called oxy reactive oxygen species which I have already explained to you earlier when I was talking to you on regarding the superoxide dismutase enzyme catalase enzyme those kind of things that when I talked about. And this will bring the depth to the cells ok.

So, you have a patient. A patient is being injected with drug; and drug is allowed to go through the body. And then it gets it gets absorbed by tumor regions, and take plenty of the fluid, the fluid will remove the unbound, and then once this is stuck are bound to that then you will shine with the light. And when you shine with the light, and the photo

sensitizer will get activated and the activated complex when it returns back to the ground state, it will activate the oxygen to form. So the reactive oxygen kind of a species, so then the tumor is basically removed or destroyed.

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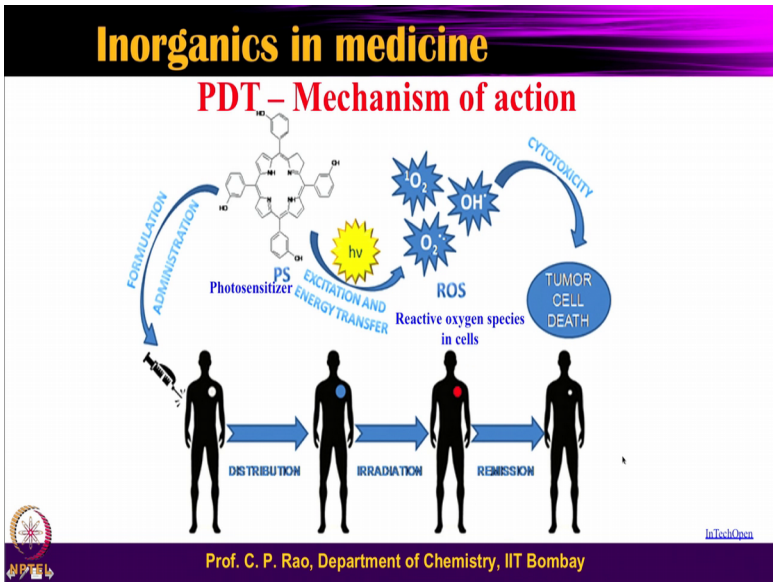

So, whatever I talk to you now, let us say in a more schematic approach, so that we can understand better. So, for this example is shown the porphyrin compound, but in couple of slides from now I will be showing non porphyrin compounds, which are good also for the PDT. Generally the PDT is started as the PDT is started with the porphyrin always the first example comes as the porphyrin based.

So, that does not mean only porphyrin based compounds will do the photodynamic therapy, but there are many other compounds. What kind of compounds? Wait a while, I will show you in a while. So, when you inject this compound through some kind of a formulation administered to the person having a tumor, and this is the region, where the tumor is shown like a white hole, and this distributes through the body, and then gets into the particular region also.

And this upon irradiation with the light whatever you the light is required, and then that will create this the ROS kind of a species. And how that happens? This compound when it is inside, when the light is shine, see you see the singlet oxygen O 2 minus dot O H dot, all these kinds of reactive oxygen species. And those reactive oxygen species are toxic to the tumor cell, and tumor cell dies, because of that.

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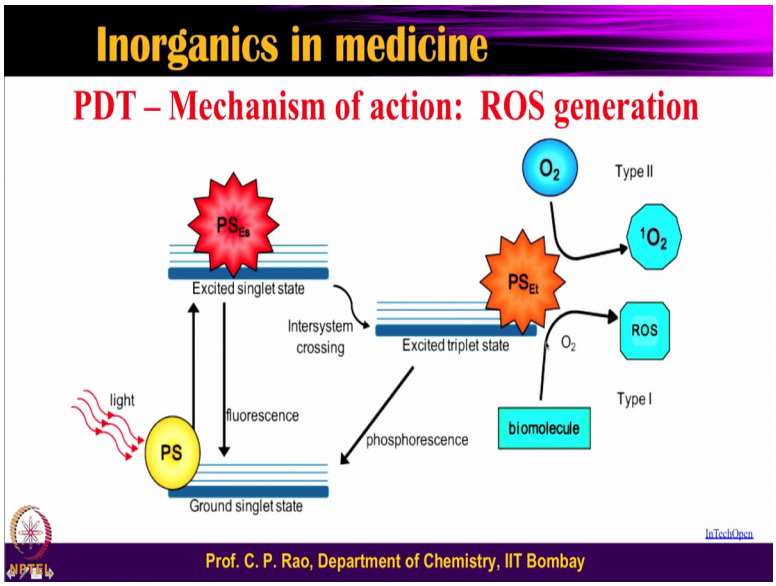

Now the same thing let us look at the mechanistic, how the ROS generates. I will show you two different ways, one way one way to understand from this the same thing shown in a different manner. The PS is nothing but photosensitizer, and this is the ground electronic state, and these are the vibrational states, ground, excited, second excited etcetera.

And this is excited electronic state, and the vibrational states of the excited electronic state. Now, this photosensitizer is shine with light appropriate wavelength. Then this will take a transition electronic transition to the excited state, and could be any of these any of these vibrational states. And these vibrational states, it can return back to the 0 vibrational state, and release the energy in the form of fluorescence. O in some cases, it can even go into a intersystem crossing. Intersystem crossing is because this is a single state, and this is a triplet state.

So, singlet or triplet is not allowed, so is forbidden kinds of a transition, so such a forbidden transitions are made available for this. And such then you get into the triplet state. From this triplet state of your molecule, then you can get the light, or which is phosphorescence that can activate the oxygen from the biomolecule, they will create ROS, it can activate O 2, and create singlet oxygen.

So, therefore, all of these are being generated, so the type 1 conversion, type 2 conversion all of these. So, at the end the end result is that the excited triplet state of the

sensitizer P S E s is the photosensitizer with the excited state ok. And here, E t excited sorry E s excited singlet this is excited triplet, and from there you get all this ROS generation.

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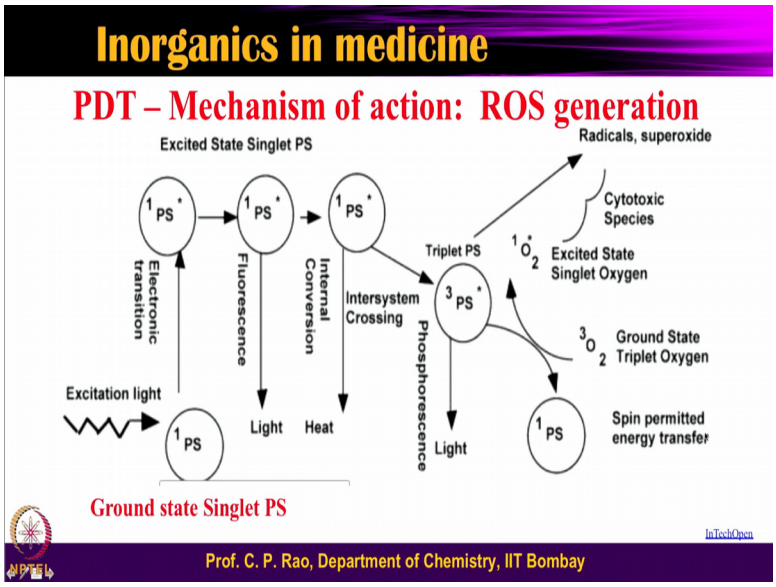

Exactly the same what I talked earlier is shown in a slightly different whichever one you understand is better is this is the ground state or the photosensitizer. And now shine light, it will go electronic transition, electronic excited state shown with this star. But still singlet, and this can as I said can go to the 0 vibrational level in the excited electronic level, and release the energy in the form of light, which is a fluorescence, O it can do some kind of a changing from one vibrational to the other vibrational to the other vibrational, which is called the internal conversion.

And then can that kind of thing can deactivate the vibrational state, O it can do inter system crossing as I mentioned in earlier, and it will go from 1 PS to 3 PS this is stories excited, so excited triplet state. So, excited triplet state can come to the ground state by the light, which is called the phosphorescence, and the excited singlet state coming to the ground state is called the fluorescence. And now this will react with the oxygen, to convert into the singlet oxygen, to convert into the other kinds of ROS, cytotoxic species, the radical superoxides. So, there is a lot of energy transfer that can make ground state triplet will come to that.

So, so therefore, what we have seen, that there is an electronic excitation, and the conversion to the intersystem crossing conversion to a triplet state. And from triplet state, when it returns back to the ground state, this energy being utilized to convert into the oxygen as well as a lot of biomolecules, cytosolic components can get oxidized, and all of them turn to give some reactive oxygen species.

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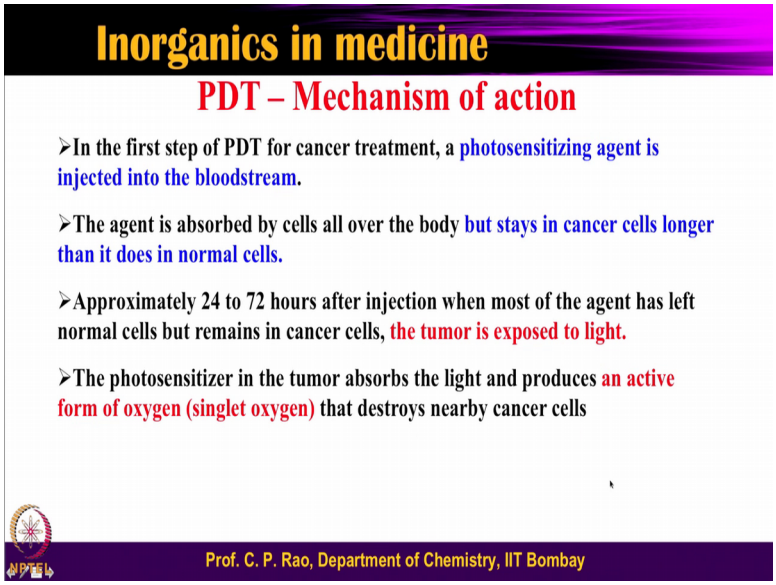

So, this is what put into the form of a statements is nothing much to do that. So, in the first step the PDT for the cancer treatment photosensitizing agent is injected into the bloodstream, the agent is absorbed by the cells all over the body. But the cancer cells it stays on and normal cells it comes out, that is why you have to flush with the fluid ok. And allow for 1 to 3 days, after the injection is given, then the other things will go out. So, the all other unbound un species will go out, and only the cancer cells will remain.

The tumor is now exposed to the light. So, the photosensitizer in the tumor absorbs the light, and produces the active form of the oxygen, singlet oxygen superoxide radical, hydroxy radicals etcetera, and these destroys the nearby cells, that means, it destroys the tissue.

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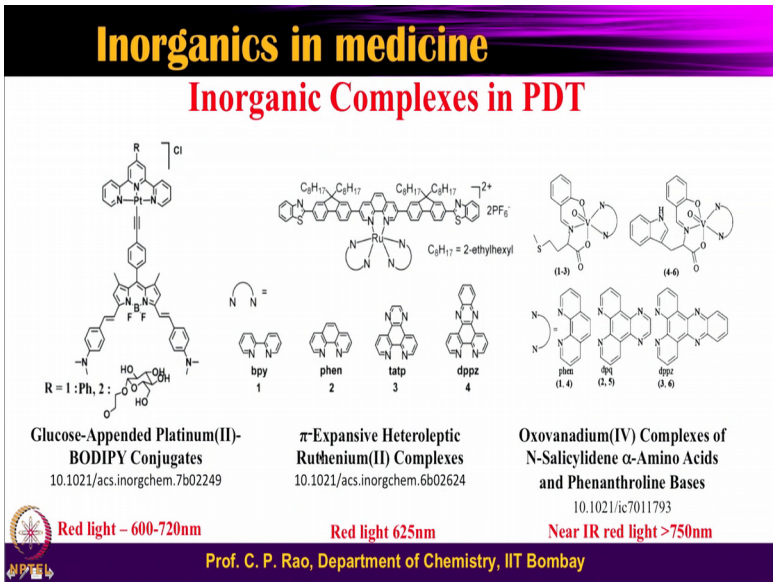

Now, let us look at the compounds as I mentioned to you earlier, whenever we say PDT, every for everyone what comes in mind, is the porphyrin. But you see huge number of molecules here, and these are all metal bound species here, ruthenium and all that you require you see huge. You require a huge organic molecule, where there is a conjugation, why, because the light goes into the visible region, and the metal ion will activate this part of it.

So, you have a platinum base with all this, and the O group could be either phenol or the carbohydrate. So, in fact, to any of this molecule you can attach a group, which will find their receptor sites. In fact, in cancer cells there are certain receptor sites are move over expressed therefore, you can take advantage. The PDT drug is derivatized with the molecule that is received by the cancer cells, selectively not the other one. For example here, carbohydrate moieties there, you also have a huge series of molecules here.

So, these are the two nitrogen containing, you call it is a polypyridyl or bipyridyl, this is b py, phenanthroline, ta tp, and dppz. So, different things, what is happening more and more of the conjugation is growing here. So, and all these can occupy this, then you have already a conjugated 1, 2. So, it is excellent system for acting as a PDT, it is not necessary you need to have only performance, and you can also make a compounds of these kinds using the vanadium compound. So, this is oxo vanadium, so this is also 6-coordinated, this is also 6-coordinated.

So, these oxo vanadium complexes are also useful in the PDT by decorating these with the huge species. So, this is oxo vanadium 4 complexes of salicylidene alpha amino acids; and phenanthroline bases ok. And these are pi expansive heteroleptic ruthenium 2 complexes, these are glucose-appended platinum two BODIPY compounds.

So, all of these are suitable for different kinds of activities particularly these are for the PDT in cancer treatment, but you can also put different kind of molecules, and then make it more diverse as the drug. So, this particular molecule works around 600 to 720, this molecule needs to be excited around 625, and this molecule needs to be excited around 750. So, all of these are visible to near IR. So, therefore, you are looking for invisible to near IR kind of thing

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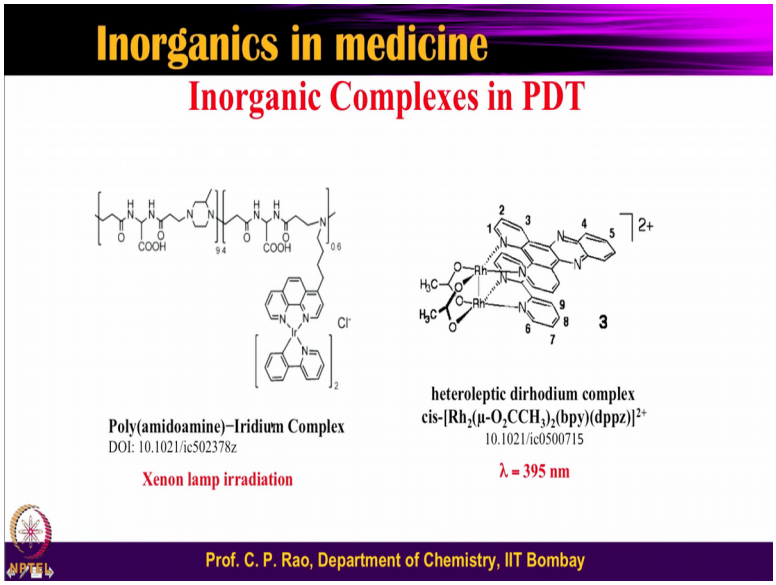

Now, two more compounds are shown over there, the iridium and rhodium, so it is a mononuclear, it is a dinuclear kind of thing. So, you have attached all this poly amino amidoamine-iridium complex, and this is heteroleptic dirhodium complex ok. And this can be by the xenon lamp, this can be by 395, which is more or less at the beginning of these visible or at the end of the U V light radiation ok. So, you have seen how these things happen ok.

Now, let us look at the inorganic complexes, just look back into this. So, what I have talked about the photosensitizer as a compound, and then giving this compound to the organ, or to the body, or to the skin, or to the blood etcetera, allowing it to deposit on to

the cancer cells. Where the drug, which is which is adhere to the remaining regions will come out, when you flush with the fluids, whereas, the one which is stuck to the cancer cells will not come out. And then that when you shine light with appropriate wavelength, that will activate, the photosensitizer is activated, and the oxygen is converted to oxygen radicals, hydroxy radicals, reactive oxygen species ok.

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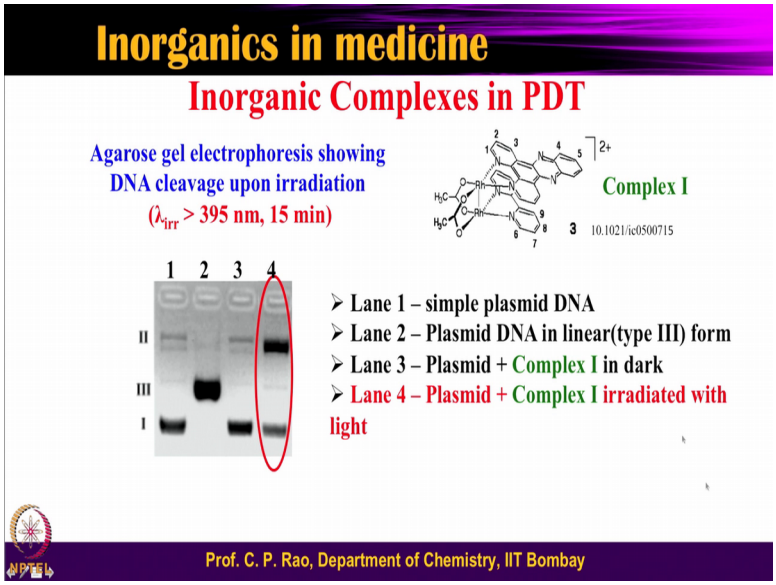

Now, let us look at how when it happens how does it look like? So, here is some picture shown here, this is from agarose gel electrophoresis, which is done for generally agarose gel electrophoresis is used for the DNA kind of a systems or plasmid kind of systems. So, agarose gel electrophoresis showing DNA cleavage upon radiation. So, what should happen? When you cleave the DNA, DNA will become into the fragments, and you will not see the original DNA band or you may see smeared DNA band too.

So, number 1 is simple plasmid, plasmid in its fully in its form 1, and then you have lane 2 the plasmid DNA in type 3 form linear type 3 form this is also standard, you can take it as, now in lane 3, this is the lane 3 the plasmid plus complex 1, whatever it is. And this is the complex 1, and this is done in dark, when you do in the dark nothing much is happening. Now, when you do in the line, half of it is converted to that, so this is irradiated. So, which means this compound upon activating with the appropriate light in this case 395 nanometer, this will cleave the DNA even in the in vitro. So, this kind of a picture is referred as the electrophoresis.

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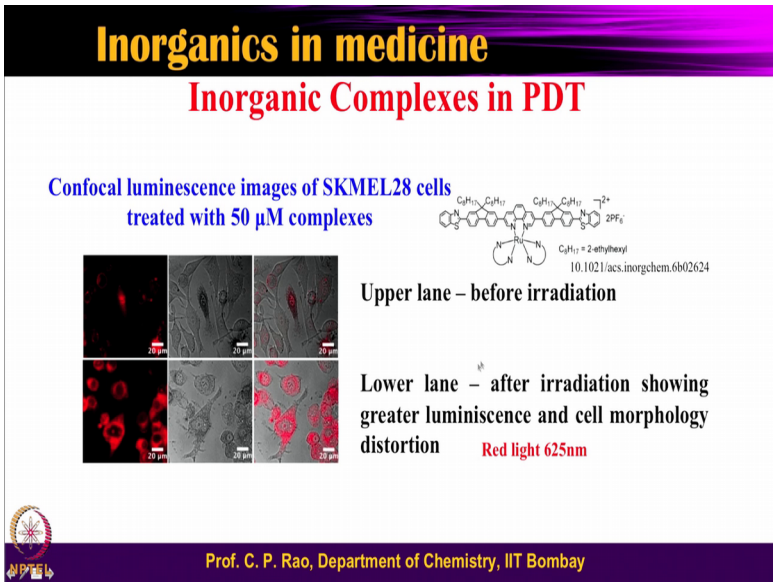

And let us look at the next slide. Next slide shows the confocal luminescence image of a particular type of cells, then they name is SKMEL28 cells ok. These are treated with the complexes, and of course, the control as well as this one ok. Now, so upper is without the irradiation, this is your normal or fluorescence microscopy, this is the bright field case, and this is upon the overlay of these ones.

So, similarly cells now treated with the PDT agent, and then this you can see the overlay, because it gives an emission band that so after irradiation showing greater luminescence and cell morphology on distortion. In this case, the excitation is at 6 625 nanometer not the other cases. So, there is how the PDT compounds work, and cancer may be in many other kinds of things.

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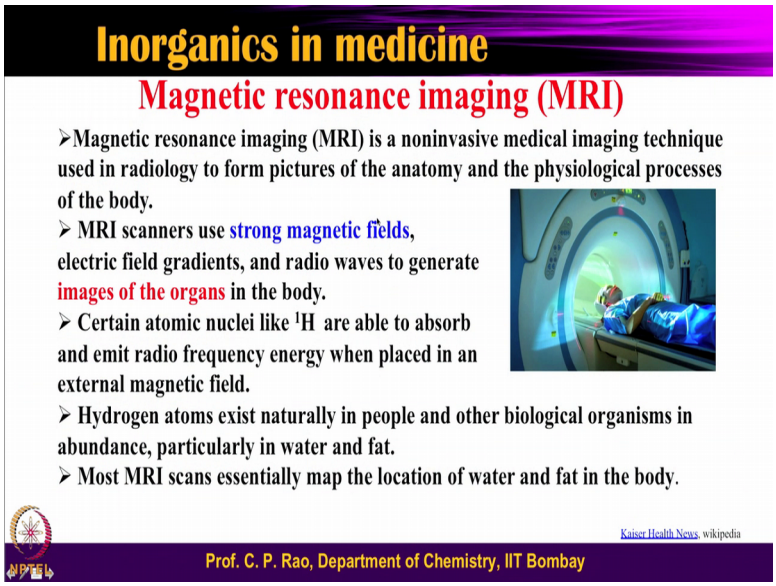

Now, let us look at another technique, which is called the magnetic resonance imaging, this is called MRI. And we all know very well without studying any of these things, MRI is taken for the body, MRI is taken for the bearing, MRI is taken for the different parts of the body we know very well ok.

So, so this is a kind of an imaging technique, wherein the one can identify the defect parts, or whether the physiological problems are coming or other kinds of things. So, magnetic resonance imaging is a non invasive, it does not affect your body. Medical imaging technique used in radiology to form pictures of the anatomy and the physiological processes that occur in the body.

So, certain physiological changes will bring certain kind of responses. So, therefore, we can find indirectly even the physiological responses to all these MRIs are taken in very strong fields, magnetic field, electric fields, gradients, and also you use one more thing is called the radio wave energy, so and in order to generate the organs. So, without any of these component you will not get your image at all, so this entire body can be imaged ok.

So, what is actually happening? So, you have a body, in the body you have a molecules, the molecules like water molecules fat facts etcetera. All of these have got protons, and you know that the proton nucleus spin already we are aware of the proton nuclear spin going from one kind of a nuclear spin to other a 1 plus half to minus half minus half to plus half that kind of a change will be caused by a lower energy radiation, is basically

gigahertz frequency of radio frequency. And this radio frequency will cause transition ok. So, this that is what you are using.

So, hydrogen atoms as we know that there the nuclear are present in people in the biological systems in the organisms ok. And they are lot in abundance, they are there in the water, they are there in the fat, all of these things you can utilize. Most of these MRI scans, essentially map the location of water, and the fats in the body. So, if that is the case, how do you find out, the difference between the tumor or any lesion all these kinds of things you can find. You can find by using some kind of a contrast agents ok.

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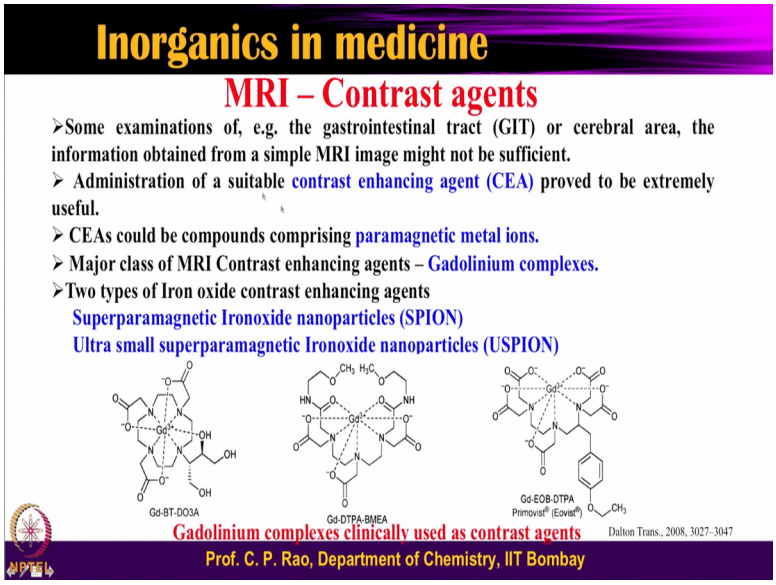

Some of these tests are the like gastro internal inter intestinal tract. You know many people have got some problems with the gastrointestinal tracts, because there are some reasons, and that gives a really pain in this in their intestines, or in their stomach, so when they eat other kinds of things. In such cases, you can get the information of MRI, which you can interpret in terms of what is the problem occurring in the body?

So, the administration of a suitable contrast enhancing agent CEA is proved to be extremely useful. Otherwise what happens is different parts of the body, the protons will have very close relaxation times, because the very close relaxation times you cannot differentiate one region versus the other, therefore, we or at the disadvantage of the MRI. To take the advantage of the MRI, then you use contrast enhancing agent, how do they do, explain this, and what are the things suitable for magnetic resonance contrasting

agent, and what can be seen magnetic resonance agents, that we have here, and how do they actually function in that.

So, this is contrast agents are basically a paramagnetic systems, superparamagnetic systems, and like gadolinium compounds, like ironoxide contrast agents, superparamagnetic ironoxide nanoparticles, ultra small superparamagnetic iron oxide nanoparticles all of these. All of these a maximum studied was the gadolinium compound, you can see one gadolinium compound, nor the gadolinium compound. And these as you know very well, demands higher coordination, and they are all 7, 8, 9 etcetera. So, these are basically used as MRI.

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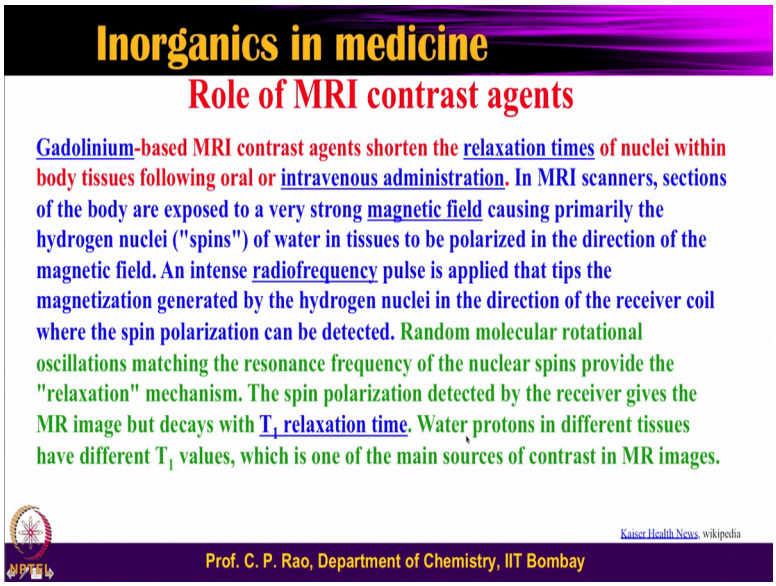

So, what is the principle here? So, what you need is a contrasting agent, which shortens the relaxation time. So, there are two types of relaxation times are their longitudinal, which is the T 1 for the proton, nucleus, and transverse, which is in the X Y plane. And longitudinal is a vertical or Z plane.

So, therefore, this kind of a gadolinium kind of reagents will shorten the relaxation times particularly of the T 1. So, of some of the nuclei in the body tissue, where the gadolinium is attached, so therefore, you get differential way of the tissue being imaged, and when you do intravenous or other kind of things.

In fact, using MRI scanning, you can scan different portions of the body, and you can integrate them, and get a total image too. And these are all used in a very high magnetic field, and in this magnetic field the nuclei will spin, and these nuclei the spin nuclei can be polarized by supplying the radio frequency of this energy, and that is received by the receiver call coil, and the spin polarization can be detected.

When you have a system a random molecular motion oscillating, when these oscillations or moments or rotations of this match with the resonance frequency of the nuclear spin, then you will see that there is a relaxation happening. So, the spin polarization detected by the receiver gives the magnetic resonance imaging, but decays with the given relaxation time.

So, the water protons in different regions tissues, tissue in the healthy region, tissue in the region, where there is a lesion, tissue where there is a damage, tissue where there is a kind of a deteriorated, all these things have different T 1 values. So, which is one of the main source for contrasting the once.

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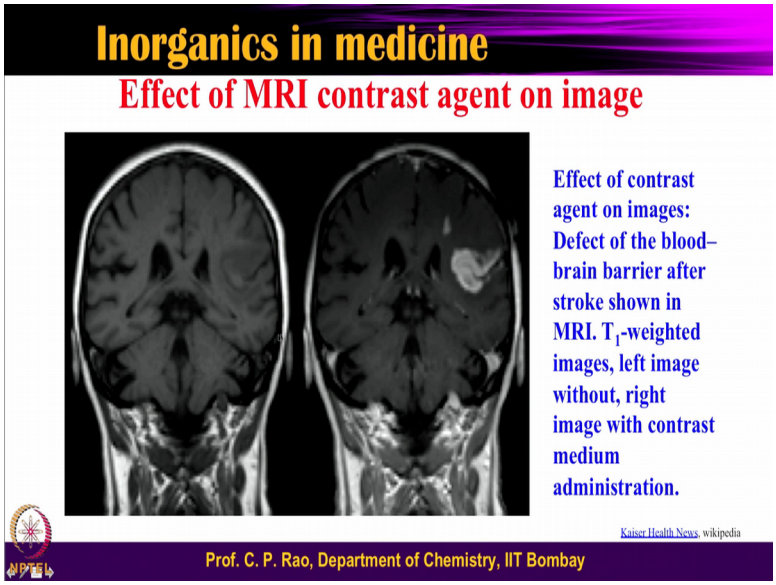

So, here is an example shown, and this is taken for to find out a defect, which is present in the blood brain barrier after a stroke shown in MRI. And this part is without the contrast agent, and this part with the contrast agent here you see, you cannot see that much difference in the contrast, this you can see a lot of contrast difference.

So, wherever there is a problem that is being seen much clearer on the right hand side, which is basically done using the MRI contrast agent, the left side one is without the MRI contrast agents. Of course, this is the magnetic field, without using the magnetic field certain regions of the body can be tested particularly intestinal tract, stomach etcetera.

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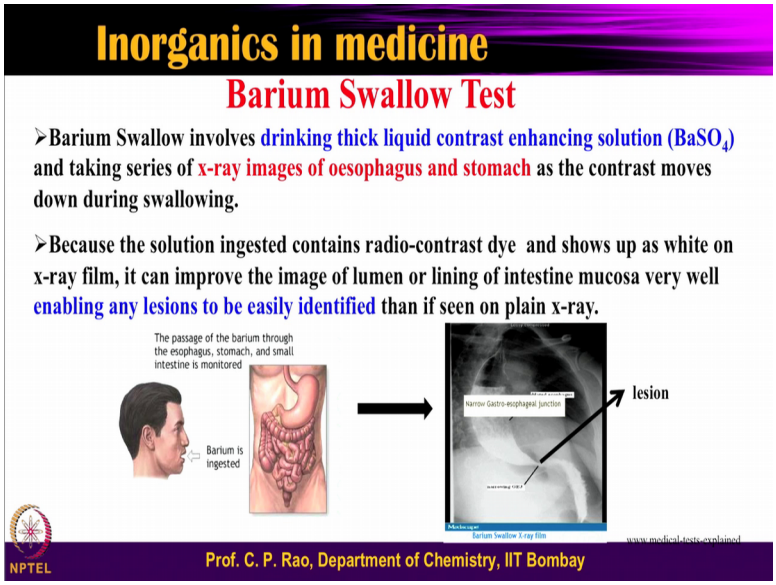

Many times you have ulcers, and this is because of the lesions in the intestines. Such kind of thing is done by a little different mechanism, different method, where you take the barium containing compound. So, this barium containing compound is taken as a barium sulfate ok, and taken into the body I mean you, drink it basically the guy, who is undergoing this test also known as the barium meal test ok.

And then you take, you allow for some time the passing through the body, then you take x-ray of these oesophagus and stomach, and the contrast moves down the during the swallowing. So, because the solution ingested constraints radio contrast dye and shows up as a white on the x-ray film. So, you will start seeing the white patches versus the dark patches you can find. So, it can improve the image of the lumen lining of intestine mucosa very well, so enabling any lesions to be easily identified than if seen on the plain x-ray.

So, if you take x-ray as such for a person who is having the lesions, in their stomach in their intestines, you can see nothing. But if you make him or her to take the barium meal,

drink the barium meal, and then you take the x-ray, then you will find the contrast kind of things ok. So, we have does we have shown the method of a PDT, and the method of them in a highly magnetic field, and the barium meal test in a x x-ray method that one can identify. So, these are some and a few more will be covered in the next class in terms of the in terms of the therapy by the inorganic chemistry, inorganic compounds as well.

Thank you very much.