

**Nanobiophotonics: Touching Our Daily Life**  
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**Lecture No. 16**  
**Interaction of Light with Cells**

Hello, and welcome. We will continue discussion on ah nanobiophotonic nanobiophotonics. I think I have given you enough preliminary information thus far. So, let us jump straight on to this topic of Fundamentals of Biospectroscopy and Bioimaging. This will form the basic the very basic or the very first chapter of the actual nanobiophotonics. Prior to that it was preliminary, it was basically high school stuff I was just revising it.

So, that student from different backgrounds medicine engineering physics can come together. This chapter onwards we will go on to the heart of the ah nanobiophotonics. So, we are in this module going to discuss about ah biospectroscopy and bio imaging. We would like to image biological material and also we would like to detect understand the chemistry that forms the vast plethora of biological materials per say.

So, in order to understand what biospectroscopy or bio imaging is all of you have heard of spectroscopy or at least all of you have heard of imaging any type of imaging your camera is an image your eyes perform the function of imaging telescope a microscope anything a lens performs imaging. But here we will be talking about bit specific items where imaging and spectroscopy are utilized for ah biological matters and thereby it is biospectroscopy and bio imaging. Imaging in itself is a huge science spectroscopy is the bread and butter of several chemists instead of going into a vast area in this module we would like to see how biological imaging and biological spectroscopy could be utilized for ah nanobiophotonics applications for understanding biological material for detecting for classifying and perhaps even certain times curing a biological material that is not functioning as it is expected to do. So, welcome again and let us start this module. So, in today's class we will be trying to discuss about the interaction of light with cells.

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**Photobiology**

Interaction of Light with living organism, to sectional tissues, to *in vivo* live specimen.

Interaction of light with matter with sizes ranging from 100 nm to macro sizes.

These types of interactions often induces a chain of events.

Can induce Physical, Chemical, Thermal and Mechanical effects.

Forms the basis of several diagnostics and therapeutic processes.

So, cell is the basic unit of life cellular structure is substantially complicated and its various part of the cell as well as biological matter ah of which cell is a primary example interacts with light and performs several different functions. Of course, light of a specific specific frequency falling on to cells or cellular components or everything else that cell create tissues organs organ systems etcetera. Light gets absorbed light can get reflected light can get refracted light can also get scattered. So, various functions are there and all of these interaction of light with biological matter for a long time has been studied and they gave it a term photo biology right. Photo biology is the study of effect of light on biological matter I think we have discussed this in the first one or two class.

Let us go little bit more deep into this why because it will allow us for ah imaging as well as spectroscopic information on biological matter. So, again what is the difference then photo biology with nano bio photonics. Photo biology is simply the study we try to understand what it is photo biology. Biology interaction of biology with light nano bio photonics or bio photonics is application its technology wherever photonics is coming up as compared to optics photonics is a technological standpoint not only we are studying the effect of light on ah biological material we are trying to apply it for something. We are either trying to image we are either trying to find out what it is and we are trying to control it manipulate it modify it change it that is what nano bio photonics is.

So, for the time being let us return back to photo biology. So, last time I gave you a very very simple ah ah definition from Wikipedia. Now, let us go into a little bit detail interaction of light with living organisms to sectional tissue to *in vivo* live specimen. *Vivo* is life *in vivo* is inside the life i.

e. a living organism how it reacts how it acts when a specific light frequency fall onto it.

We are going to see a detailed discussion of that in the optogenetics and neuro photonics well you will see an optical fiber is inserted inside the brain of a mouse and how the mouse's behavior changes because of different frequencies affecting the brain. But overall photo biology deals with interaction of light with living organism for various tissues or even specimen and interaction of light with matter that sizes from 100 nanometer hence nanometer to even higher sizes. And herein lies the crux of the matter why we need to study photo biology because these types of interaction often meaning not always significant times large number of times often includes a chain of events. I will come to that let me finish this through can induce physical chemical thermal and mechanical effects and form the basis of several diagnostics and therapeutic processes read that at your own leisure.

But this point number 3 is the most important point it often induces a chain of events a chain reaction starts when light of a particular frequency falls onto several biological materials cells are just an example of such biological material. Now, let us try to understand this. Light falls on any material we discussed in I think chapter number 2 light falls into any matter some of it get transmitted some of it get reflected back some of it is refracted some of it is absorbed some of it is scattered you all by this time know  $t + r + a = 100$  percent. So, why are we interested in photo biology or why why are we what is special about light interacting with biological matter they also go through the same processes some light get transmitted some light get refracted absorbed scattered etcetera etcetera. So, the most interesting part of photo biology or light interacting with biological matter is that it starts several different sets of reaction chain reaction as light falls on a particular biological matter.

What you ask what what are those let me try to give you an example. So, for example, photosynthesis yes, the sunlight of very specific frequency sunlight contains a huge huge electromagnetic spectrum, but within the visible spectrum 400 to 800 nanometer roughly certain bands 580 560 check if I am wrong they can actually interact directly with the chlorophyll pigment present in the leaves of the plant a biological matter and induce the process of photosynthesis resulting in formation of glucose byproduct is oxygen which we all breathe right. A simple light photon or group of electromagnetic waves etcetera falling into an inorganic normal matter chair table piece of silicon etcetera will not induce this complicated process that it happens in photosynthesis as an example in biological matter think about it. Light falls and yes it does some amount of light is being absorbed by the proteins the electrons move from lower level to upper level the molecules rotate etcetera all of those things happen, but the result is a chain reaction result is creation of some the photosynthesis reaction the curve cycle as they call it ah if you do not know curve cycle do not worry about it. The photosynthesis is a complicated biochemical

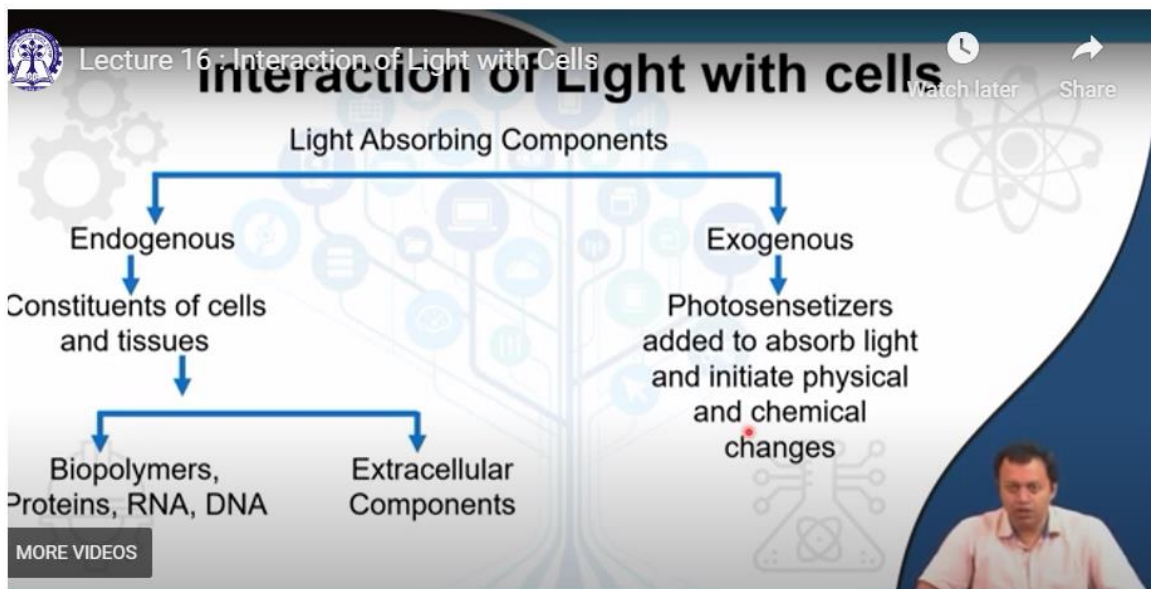
process that starts glucose for the plants it creates glucose for the plant's oxygen is released out right.

So, a chain reaction starts and that is quite important for us to study several other examples are there several other examples are there. For example, ah in in in in in western ah Europe in Europe as well as ah North America people like to go to tanning saloons yeah where they expose their skin to UV radiation to darken it. On the other hand, ah here in the subcontinent region ah people ask ah small children or vulnerable ah men women not to go out too much in high sun because the color of the skin will change the skin will take a dark and shade. So, remember you need ah sunlight exposure ah and sunlight exposing your skin when your skin is exposed to sunlight ah several different reactions takes place yeah melanin formation can actually change the contour or can change the texture or can change the shade of your skin you can get few shades darker, but also it induce ah formation of vitamin D yeah sunlight creates vitamin D induces the reaction in which your skin starts producing vitamin D which is very very essential. So, it is not simply a piece of matter is exposed to light and then some amount of reflection absorption scattering has happened like anywhere else.

Here in this particular case here in this particular case something else is happening apart from simple absorption scattering refraction reflection a set of biochemical reaction often not always often starts often creates often starts which results in some sort of some sort of ah modification in what it previously used to be. So, the matter the biological matter in certain way modifies or its function a normal function changes a bit a plant which is not exposed to light when exposed to light in the evening it is not exposed to light in the dark is not exposed to light it is not causing photosynthesis it is not creating photosynthesis, but in the morning its property changes its property changes the photosynthesis starts to happen same with skin color. Vision human vision or any vision per say is another example when light falls into our retina a complicated process which we will discuss in subsequent chapters light falls into the retina the overall polarity of the rod cells and cone cells changes and that results in formation of ah electro optical signal that passes through our our our ah optical nerve into the visual cortex in our brain and we are able to see. So, it is not simply light falling into something and there is absorption and that is it unlike any other material here a biochemical reaction starts. So, chain reaction is quite important and this chain reaction falling light and thereby step 1 step 2 step 3 step 4 can form the basis of diagnostics and therapeutic processes.

We can diagnose if a particular step A or step B is missing or not or if step A is fine, but step B part or step C part after light has been exposed is not working properly or if there is a problem and thereby if we can add something if we can modify something and here the crux of the matter is these reactions our vision ah photosynthesis this photo biology

works on specific specific light frequency only you cannot expose your ah plant to microwave radiation which is also part of electromagnetism terahertz radiation and ask it to do photosynthesis. Similarly, you cannot see things if infrared light is falling onto your eye you can only see the visible spectrum. So, a specific frequency is needed a specific frequency only allows this chain reaction to perform. So, keep that in mind you cannot just randomly send any frequency onto a biological matter cell or ah tissue or organ and then decide that why it is not working as as as as you have expected. It has to be some specific photon some specific light that will perform a specific biochemical reaction that we would like to understand and then utilize it for the basis of diagnosis as well as therapeutic processes.



Now within the cell what exactly are the light absorbing components within the cell what exactly are the light absorbing components. So, the light absorbing components within the cell can be divided roughly into endogenous and exogenous. Endogenous means inherent something that is already present endo inside. So, it is cellular component exogenous is something that has been artificially brought in artificially brought in which is not directly part of the cell, but now you have artificially induced into it. Within the endogenous that is the constituents of cells and tissue all of these biopolymers, proteins, nucleic acids, carbohydrates, fats etcetera all of them absorb light light all of these these components that is inside the nucleus absorb light.

Extracellular components cell organelles extracellular components are those matrixes that allow a cell to adhere with another cell and form large tissues. So, the glue-like substance that make one cell stick with another cell and they then combine with another cells and then this thing builds up and form tissues several different tissues can combine together to form organs. So, this this interface between one cell and another cell extracellular

component all of them absorb light. Whereas, exogenous are fluorophores fluorophores remember from your fluorescence ah they can be nanomaterials as well other type of non-fluorescent nanomaterials like metal nanoparticles. They are specifically added inside the cell they are attached with the cells you can artificially create them or they can be ah present naturally you just simply extracted by a chemical process and add them and they instead of these you instead of these indigenous endogenous components these components these additional components this foreign bodies they absorb another specific wavelength of light another specific wavelength of light and they starts some sort of a physical chemical change.

They start some sort of a physical chemical change within them within them photosensitizer sensitive to photons sensitive to light which will then react with the cell inside which these nanomaterials are injected right. For example, labeling in fluorescence material you can put some sort of a quantum dot ah that absorbs a particular frequency of light that is put inside a cell upon ah ah absorbing a particular frequency of light that quantum dot starts emitting a particular frequency another frequency of light which you can detect using your microscope and that can form the basis of identifying where the cell and the surrounding area from which light is emitting. It is not the cell that is emitting light it is the quantum dot the nanomaterials which is inside the cell that is emitting light yeah putting a torch inside or putting a bulb inside a room the room is not emitting light the bulb is emitting light the bulb is outside from the room somewhere else you have put it inside the room bad analogy I know to illuminate the room something like that happens.

Lecture 16 : Interaction of Light with Cells

## Light Absorption in cells

Constituent of Cells	Absorption Wavelength
Aliphatic Amino Acids e.g., Glycine (Gly)	<240nm
Aromatic Amino Acids e.g., Phenylalanine (Phe)	>240nm
Hemoglobin	280nm, 420nm, 540nm
Melanin	Entire visible region (and some UV)
DNA bases	230-300nm
Water	3μm

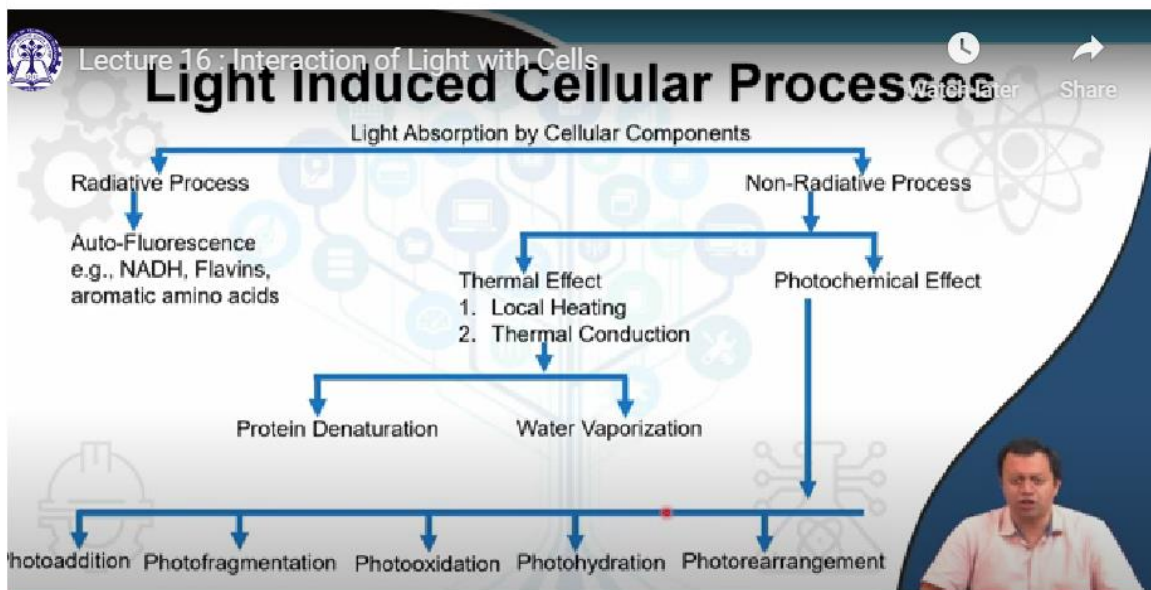
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So, these are a table of ah what are the different frequencies that are absorbed by different areas or different parts of ah cells amino acids glycine etcetera aromatic amino acids etcetera they all absorb ultraviolet hemoglobin which is part of your blood ah absorb 280

nanometer in ultraviolet 420 nanometer 540 nanometer ah do they fall in visible red if hemoglobin absorbs these what are the color of these these falls under the ah visible spectrum light 420 nanometer is blue blue shade ah ah violet indigo or something like that 540 nanometer could be green. So, a particular light hemoglobin does not absorb and reflects you can understand what color that is reflects and then you can see hemoglobin.

Melanin as I was seeing the part of the skin cells ah it absorbs the entire visible region and some amount of UV. So, when your mother asked you not to go out into the sun ah maybe she was actually right because your body will absorb your skin will absorb the entire visible spectrum and some amount of UV and a chain reaction will start maybe you want that chain reaction there is nothing wrong in that ah or maybe you do not want that chain reaction to happen. Similarly, DNA bases adenine guanine etcetera they all absorb this particular wavelength of the ultraviolet hence we use ultraviolet certain times this is close to visible frequency we use ultraviolet sometimes to generate mutations water is present in every biological compound and they absorb in the infrared mid infrared 3 well mid to near infrared depending on what is the definition of near infrared ah 3.3 micron is usually where water absorbs. So, all constituents of cells from nucleic acids to proteins to ah other components ah all of them absorb some sort of light and if it is absorbed then you can expect 9 out of 10 times or 8 say 8 out of 10 times if absorption has happened in a biological material some sort of biological change or biological biochemical reaction have induced 8 out of 10 times yeah this is biology we are not talking about 2 plus 2 equal to 4 right.



So, what are the biological processes what are the exact biological processes or what are the basic biological processes that we can expect when light or when cell absorbs light right. Particular frequency, particular frequency is the most important thing I am telling

you repeatedly enough that you cannot simply randomly expose some amount of light and expect the cell or the biological material to react. Specific frequencies specific wavelength specific colors specific energies of light will perform specific function specific biochemical function onto the biological matter. So, the basic functions the output are these types of cellular processes. Let us divide into 2 part radiative process and non radiative process radiative process when light is emitted after absorption after a particular frequency of light is absorbed by a particular biological compound it emits another frequency fluorescence basic of fluorescence.

And then there is non radiative process which no light is absorbed ah when no light is emitted per say the photon is simply absorbed the photon is destroyed and instead of it the energy is dissipated in several different form it can form a thermal effect instead of light heat is emitted heat is emitted or some kind of photo chemical reaction takes place this light gets into the cell and as I said a photo chemical biochemical reaction takes place. The radiative process is pretty straight forward auto fluorescence NADH I keep on memorizing the name I think check it out nicotinamide adenine dinucleotide flavins are also this group of riboflavin you must have heard these are groups of this yellow organic compounds they are part of. So, both of these are enzyme type of things and they help in production of energy medical student's ah write your comments on how I am wrong, but to the best of my knowledge they they form parts of enzymes and as soon as a particular frequency of light with especially the visible ultraviolet and etcetera they absorb they start emitting light of a lower frequency. So, if they absorb visible blue they start emitting visible green or something like that. So, you can simply put a piece of ah cell or tissue that contains NADH flavins etcetera aromatic amino acids which are part of proteins and shine it with some sort of a laser a particular frequency of lasers a visible blue and if green or red comes out without adding anything extra additional exogenous maybe you can be sure about the presence of this flavins are there are much more nuances to that, but overall several part of cellular structures absorb a particular frequency of light and this absorption leads to fluorescence effect another frequency of light is emitted.

In the non radiative process thermal effect is there thermal effect is local heating the light is absorbed the infrared light is getting absorbed by a particular ah part of your ah ah cellular component it absorbs that light and then it produces local heating that area heats up you all get ah boils or ah sunburn perhaps you have heard this happens mostly in caucasian European ah white population if they go too much into open sun white people ah caucasians their skin starts burning up. So, local heating so, the light that falls onto their skin because less amount of melanin helps you protect against ah harmful rays of the sun they can they get ah sunburned probably and even several other people can also get sunburned it is not ah specifically to caucasian population, but where there the effect is prominent ah you expose your skin to large amount of ah sunlight and it it it burns off you



get boils and etcetera on your skin. Then there is thermal conduction where light has resulted in creation of heat because molecules have started vibrating molecules have absorbed light sunlight and started vibrating sunlight is just an example you can also burn somebody skin by sending high frequency laser light, but anyway ah how many of you have access to high frequency laser light sunlight is much more accessible there is a thermal conduction of heat meaning the heat is not localized it has travelled it has travelled from one area of the cell to another area of the cell my body contains larger number of cells I can a combination of cells so and so forth and that results either in protein denaturation the protein breaks down somewhere else or water vaporization dehydration have you thought of it too much exhaustion into the sun you are working in high temperature full exposed sun for 4 hours 5 hours continuously nonstop ah the amount of water in your body can evaporate in the form of sweat in the form of your breathing several different forms and that is thermal conduction maybe light has fallen just on to your head or exposed areas, but you still feel thirsty right. So, the light heat has actually travelled resulting in ah water vaporization from your body. Then come the photochemical effect these are all those different types of photochemical effect photo addition photo fragmentation and we will go all of them one by one.

Lecture 16: Interaction of Light with Cells

## Light induced Cellular Processes

- **Photoaddition:** Addition, e.g., photodimerization of Thymine.
- **Photofragmentation:** Fragmentation, e.g., decomposition of Riboflavin.
- **Photooxidation:** Excited molecule adds oxygen, e.g., Cholesterol.
- **Photohydration:** Excited molecule adds water molecule, e.g., Uracil.
- **Photorearrangement:** Rearrangement of bonds occurs; chemical formula of molecule remains unchanged. Example 7-dehydrocholesterol in skin producing Vitamin D3 upon exposure to UV.

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So, this is the light induced cellular processes which is non radiative photochemical effect what they are well as the name suggest photo addition light can result in ah a particular chemical compound to attach to another particular chemical compound the light acts as catalyst light acts as you have learnt about Frank Condon principle why exactly that happens dimerization takes place dimerization is monomerization or polymerization. So, dimerization another another one of the same things can attach thiamine can attached with another of similar species photo fragmentation the opposite of addition this is subtraction fragmentation is breaking down decomposition of riboflavin riboflavin is a chemical compound some part of it absorbs light the molecules swell up the intermolecular forces

the attractive forces break down repulsive forces win and they subtract. So, addition and subtraction photo addition photo fragmentation are a better way than subtraction. So, call it ah photo fragmentation photo oxidation photo hydration similar thing excited molecules add oxygen. So, when light is exposed ah molecule absorb it starts exciting it it gets excited and in that excited state it marries an oxygen an oxygen molecule right again if it does not like oxygen it can get very excited and start adding water it has excited itself.

Now, it wants to return back to its original state may be the intermolecular forces have destroyed itself or it it is no longer balanced. So, I am still allergic it is no longer balanced and there there is nearby a water molecule which will probably not ah form the chemical reaction under normal circumstances, but now a particular part of the molecule is having higher energy. So, this higher energy is allowing it to perform another set of chemical reaction while being part of a larger biomolecule. So, they can start adding oxygen or they can start adding water molecules photo oxidation photo hydration photo rearrangement is another thing that is quite cool where rearrangement of bonds occurs like the chemical formula of molecule remain unchanged, but the ah how to say the arrangement how a molecule is arranged in three-dimensional space changes yeah it changes. Again, Franck Condon principle intermolecular forces changes and thereby there is a reorientation reorientation which stays it does not return back to its original position it stays.

Cis and trans remember in organic compound you have a particular molecule with similar molecular formula everything benzene ring or it can be. So, suppose the particular molecule is attached diagonally upon interaction of light it will now arrange symmetrically. Same thing same thing it is a big chain molecule two dangling bonds hydrogen hydrogen or carbon dioxide or methane methyl group is attached at two separate ends of the molecules upon light excitement one changes its position and they are now close to one another. So, ah example is skin producing vitamin D<sub>3</sub> upon exposure to UV. So, the overall property remain as it is inside your body, but upon exposure to ah light the process changes and it can give rise to several reaction this is very ah common in vision I keep on talking about vision we need to we need to discuss.



# Photochemistry Induced by Photosensitizers



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- Exogenous Chromophores that mostly performs photoaddition or photooxidation processes.
- When excited by light absorption, covalently bonds to a constituent molecule of a cell.
- Often used for production of Reactive Oxygen Species (ROS) such as singlet oxygen  $^1O_2$ , superoxide anions  $O_2^-$  or free radicals.
- Forms the basis of Light-activated cancer therapy, called **Photodynamic Therapy**.



So, what are the photochemistry induced by photosensitizers external endogenous agents exogenous chromophores are mostly performed photo addition or photo oxidation process either they add. So, these are external materials that are put inside cell biological matter per say and they either use photo addition photo addition as I said ah more than one molecule start adding or photo oxidation they start attracting oxygen towards them. When excited by light absorption covalent bond when excited by light absorption covalently bonds to a constant molecule of a cell they attach themselves with cellular structure by covalent bond not hydrogen bond not Van der Waal bond. And they often used to produce reactive oxygen species ROS they are singlet oxygen species highly highly reactive and forms basis of photo dynamic therapy what does that mean? It means that you have produce or you have injected some sort of a chromophore something that ah absorbs a specific specific frequency of light and does it function what type of function it does. So, suppose you have a tumor right bad cells they are producing something that is unwanted you have deposited some sort of chromophore into them.

These chromophores these chromophores absorb a specific frequency of light something in the infrared or something in the visible spectrum. So, this part of the tumor where which has been injected which has been injected with nano materials quantum dots or something like that they are then exposed by a torch light to a particular frequency of light. The chromophore absorbs this light they get excited and they start ah attracting or producing photo oxidation or we can produce a singlet species of oxygen. This is a highly highly reactive oxygen species reactive oxygen species this this this oxygen species is singlet oxygen remember singlet and triplet species singlet oxygen species they for a lack of better word burns through the oxidizes. Oxidization is burning oxidation is the burning process they produce heat they burn through the tumor destroying the bad cells destroying the cancerous cells right.

Quite similarly quite similarly when exposed to light these chromophores they can they can be exposed to ah light which is in the visible spectrum and they can fluoresce in the infrared spectrum infrared itself is heat. So, photons which are infrared and this localized heat production of infrared photons localized heat can burn or destroy the cells nearby right. Two things photodynamic therapy this form the part of photodynamic therapy they can either create heat by themselves they can ah emit infrared photons that burns away their periphery these chromophores this exogenous ah photosensitizer or or or they can get excited and start ah attracting oxygenated species near them oxygen species burn through the ah tumor and attach with them. Result is formation of heat either they produce it directly or they produce they ask oxygen to come to their rescue and thereby produce heat and that burns away the tumor. So, that is form of part of photodynamic therapy.

So, thereby you know what are the processes by which light can interact with cells and biological compounds. So, if we know this and then apply our technology to understand what are the reaction what are the photochemical reaction happens because of what specific absorption emission refraction scattering etcetera we can fully comprehend this entire process and make our technology make therapeutic make cure. Overall idea here is that biological matters ah interacts affect and ah modify themselves when exposed to particular frequencies. If you can understand the modification process interaction process we can perhaps control them and that is the crux of the matter.

The image is a screenshot of a video lecture interface. At the top left, there is a logo and the title "Lecture 16 : Interaction of Light with Cells". To the right of the title are icons for "Watch later" and "Share". The main heading in the center is "CONCEPTS COVERED". Below this, there is a list of three bullet points:

- Interaction of Light with cells.
- Light absorption in Cells.
- Light Induced Cellular Processes.

At the bottom left, there is a button labeled "MORE VIDEOS". In the bottom right corner, there is a small inset video of a man speaking.

So, these are the concepts that I covered today in this class and these are my references



## REFERENCES

- **Introduction to Biophotonics, Paras N. Prasad, 2003, Wiley.**
- **Molecular biology of the Cell, Bruce Alberts *et al.* 5th edition, 2008, Garland Science.**
- **Photobiology: The Science of Light and Life, Editor L.O. Bjorn, 2015 Springer.**

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I will see you in the next class. Thank you very much.