Nanobiophotonics: Touching Our Daily Life
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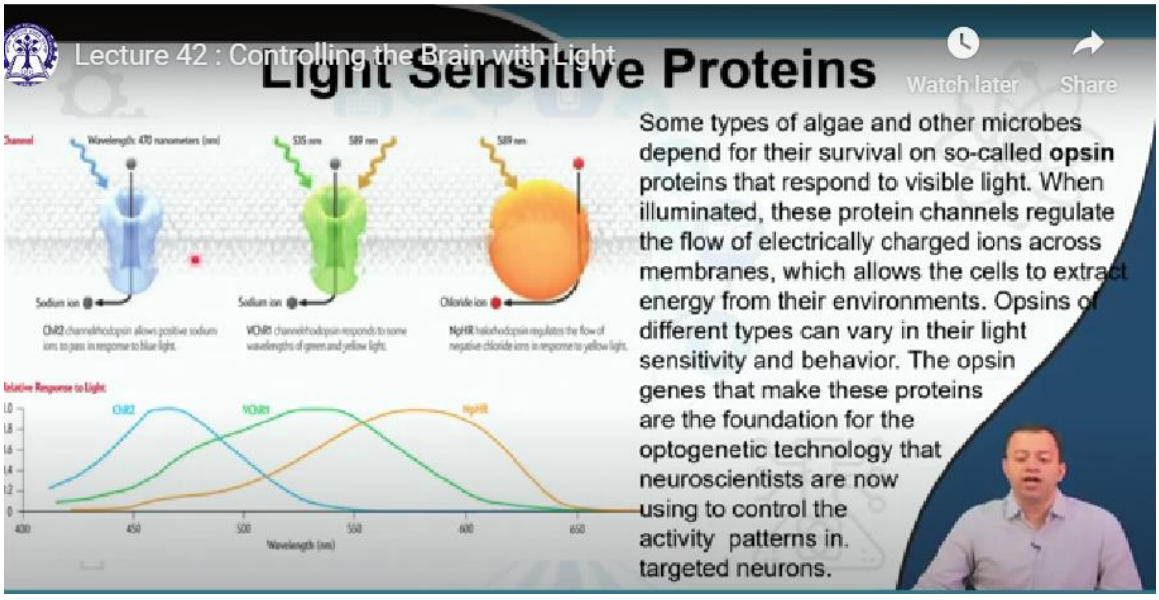
Lecture No. 42
Controlling the Brain with Light

Welcome back. We are still discussing optogenetic technology and today we will try to look into how to control the brain with light. So, welcome to lecture number 42, module number 9 principles of optogenetic technology and let us see if light could be utilized to control certain section of the brain. So, right I think it was in 2005 or 7 please correct me if I am wrong that at Stanford Karl Deisseroth and group of his students tried to did a revolutionary experiment that resulted in coining the term optogenetics. So, what they did is there are certain types of algae and other microbes which depend on their survival on certain specific proteins. Those proteins are called a group of proteins are called opsins.



What are the specific peculiarity of these opsins are that these proteins these proteins these specific proteins upon excitement by a specific wavelength of light upon excitement by a specific specific wavelength of light will result in flow of a specific electrical current. There is a group of protein, opsin group of proteins present in certain algae that are photosensitive and that result in somewhat very crudely speaking photoelectric effect i.e. you send light through at them of particular frequencies, particular energy, particular wavelength and they result in conduction of current through them.

Photoelectric effect naturally existing in proteins existing in certain algae and other microbes i.e. biocompatible. When illuminated these protein channel regulates the flow of

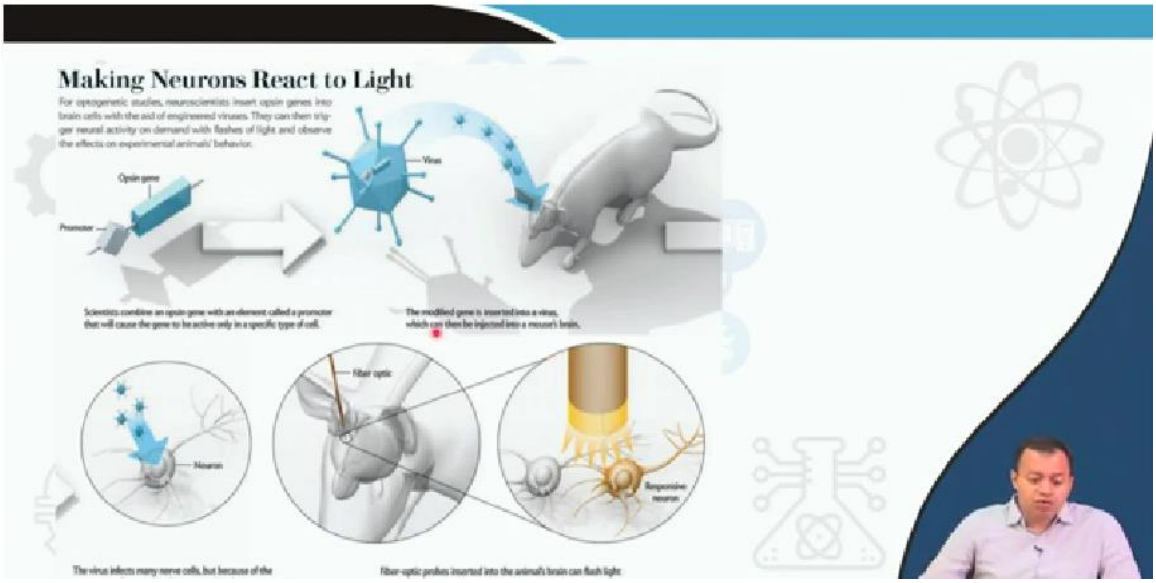
electrically charged ion across membranes which allows the cells to extract energy from their environment. Opsins of different types can vary in their light sensitivity and behavior.

The opsin genes that make these proteins are the foundation for optogenetic technology that neuroscientists are now using to control the activity pattern in targeted neurons. So, this is an example of the various types of opsin proteins that is channel rhodopsin, CHR2 then VCHR2 is another type of channel rhodosin then there is halorhodopsin. So, these are different types of proteins. If CHR2 channel rhodopsin is excited by a wavelength of 470 nanometer around indigo or blue this allows positive sodium ion to pass in response to blue light. Sodium potassium all those things chlorine is available in the body anyways.

So if this particular protein present in a cell is excited by 470 nanometer it allows a positive sodium ion Na plus to pass through it. If the light is not excited this particular protein will not allow sodium ion to pass through. VCHR1 another form of channel rhodopsin response to some wavelengths of green and yellow light. CHR2 channel rhodopsin is 470 nanometer another category. VCHR1 the input has to be green or yellow light and it will respond to some wavelength it will allow sodium ion to pass through.

Chlororopsin on the other hand gets excited by 589 nanometer yellow or orange light and it allows negative chloride ions to pass through. Now remember these ions these charge carriers this electrical current charge carriers will only be possible when you have excited with these specific wavelengths only photoelectric effect in proteins. Electrical effect in proteins only upon excitement by a particular particular wavelength of light a particular current will pass through it. So, you have relative response to light versus wavelength. So basically, this is current versus wavelength the different types of electric current that is that is being sent when excited by specific specific wavelength of light is actually shown.

So NPHR halorhodopsin will only be exciting or only be passing current when it is around 580, 590 nanometer radiation is present any other radiation either it is very less or none at all. Similarly, channel rhodopsin of different types of proteins allows the passage or blockage of different types of ions thereby either allow or disallow a particular electrical impulse a particular electrical current a particular electrical pulse to pass through it. Algons carry charges so they are charge carriers and they can be used to conduct electrical electricity basically. So what professor Karl Deisseroth's group did was something incredible. So, they isolated first the gene from this algae or microbe that produces this protein.



What is a gene? We have discussed this. If you have forgotten what gene is if you have forgotten what gene is go back to the previous lectures. So, Karl Deisseroth team isolated a particular gene which produces these photosensitive proteins that has a capacity to produce photoelectric effect. It was connected with a promoter, promoter is the beginning of a gene the start code the start code UAA remember the start code upon which the gene become functional the start code which the person receives from which the message become functional it is connected with the promoter which is then put inside a virus. This virus is then made to infect the brain of a mice.

This promoter and opsin gene then connected to specific neurons you can specify neurons by connecting it with a specific promoter the specific promoter attached with specific part of the gene. So, you know how viruses work right viruses affect the nucleus of the cell. So, this particular virus was made to attack the nucleus of the neurons presents into the into the mice. So, scientists combine an opsin gene with an element called a promoter that will cause the gene to be active only in specific types of cell. So, this promoter is specific to a specific neurons or specific group of neurons.

The modified gene is inserted into a virus this entire gene this gene that has been isolated from algae is inserted into a virus which can then be injected into the mouse's brain to infect it. So, this gene put inside a virus, virus put inside the mice brain. Here the virus is acting as a carrier acting as a transporter acting as a car or a plane which is carrying the cargo the cargo is this gene this is gene therapy they did it it is a fantastic almost Nobel Prize winning experiments there are few beautiful experiments of science I would rate this experiment as you know near the top. Gene you are extracted it from what is gene again come on group of DNAs' that has the capacity to create a protein that has to that can express proteins. So set of DNA's put inside a virus, virus contains DNA or RNA put inside a virus this virus is made to infect the mouse's brain.

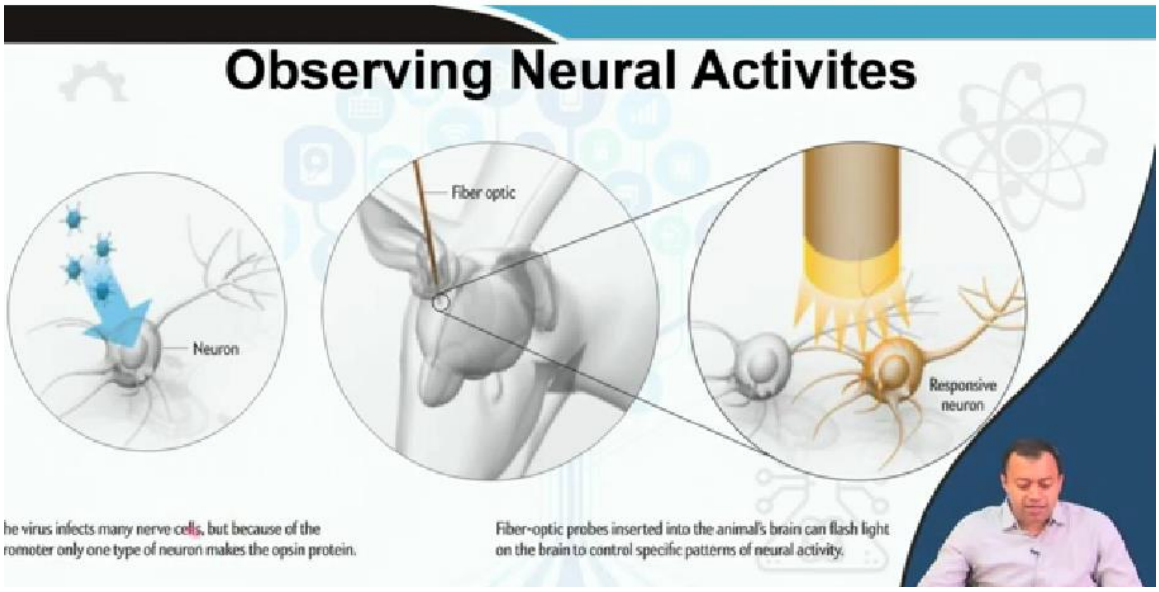
The virus infects many nerve cells but because the promoter only because the promoter only one type of neuron make the opsin proteins meaning, meaning using the virus as a carrier specific neurons of the body specific neurons of the mice's brain specific neurons of the mice's brain is made to connect with this specific opsin producing gene, opsin producing DNA understand it. The virus is attacking all over the brain but there is something called a promoter. The promoter is a specific DNA which has a specific code the start code attaches with specific neurons only. Yes, different neurons they will not be 100% similar DNA 99.99% similar there will be certain differences otherwise you would not be able to differentiate.

Now you have differentiated different brains, different brain cells a particular cell that promoter the particular cell which has a particular DNA which has a particular gene is connected with a particular promoter. Promoter is the beginning of the gene, promoter is the beginning of the start message. This start message is then connected with the opsin gene. So certain neurons are affected mature age, old age neurons or just baby neurons which are just forming or neurons with large number of dendrites or neurons with small amount of dendrites only those only specific ones are attacked. The virus infects many nerve cell but because of the promoter only one type of neuron make the opsin protein which type you decide old, young, specific this neuron present in this side of the body etc.

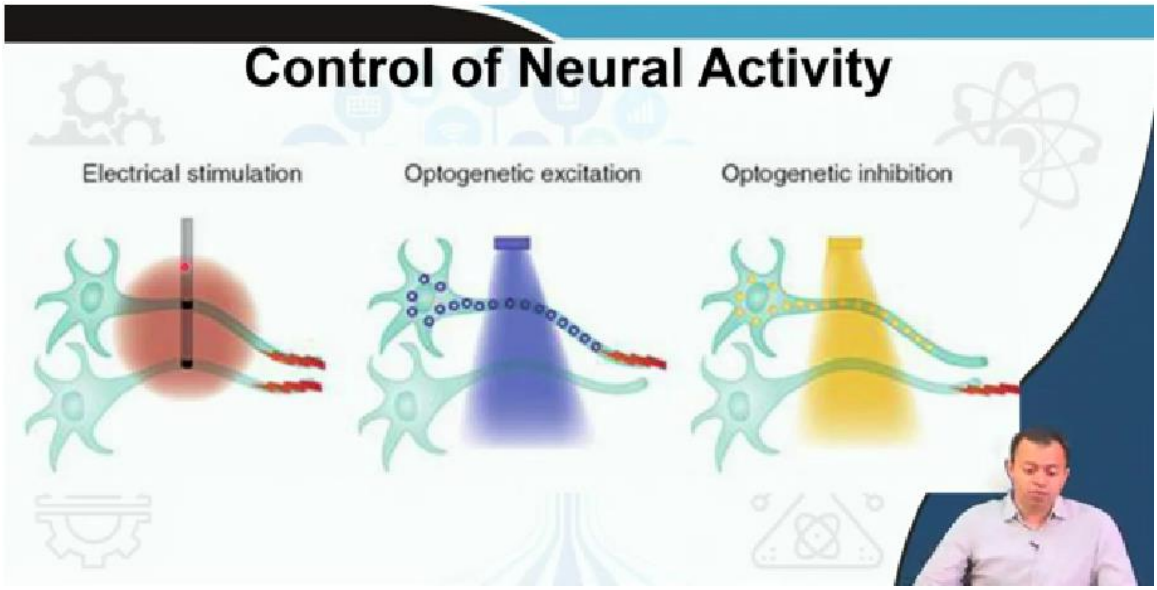
So the virus have now infected the nucleus of those particular neurons sooner or later the virus is benign so either it stays or your body flushes out but the nucleus of certain specific neurons have got the opsin gene meaning that neuron which contains large amount of gene one part of the gene one part of the gene one part one of the genes present in that you have large number of genes present in one single cell one of the gene present in that specific neuron is producing opsin proteins along with its normal function. Neuron containing large number of genes each gene produces specific specific proteins of that one particular protein is your opsin protein light sensory protein. Then they made a small hole a keyhole types on to the skull the cranium of the mouse inserted a fiber optic cable and started illuminating with a specific wavelength of light. Fiber optic probes inserted into the animal's brain can flash light on to the brain to control specific pattern of the neural activity. You have this much brain with millions and billions of neuron cells neurons certain neurons are actually containing opsin proteins halorhodopsin channel rhodopsin anything.

These opsin proteins are then excited excited with a specific wavelength with specific wavelength with specific wavelength they in turn then conduct electricity conduct electricity by sending sodium ions positive or chlorine ion negatives that result in certain specific activity of the mouse. The mouse becomes aggressive the mouse falls asleep mouse become hungry the mouse tries to you know behave differently you can map them

you can map them using light. The virus infects many nerve cells, but because of the promoter only one type of neuron makes the opsin protein and the fiber optic inserted into the animal brain can flash light to the brain to control specific patterns of neural activity. You have then thereby encoded certain areas only those which has this specific opsin halorhodopsin or channelrhodopsin will be affected no other nearby even if the neuron is nearby since it is not producing photoelectric photo sensitive protein it will have no effect it will not produce any electric current. Only this which will have a response which will have a gene that also produce photo sensitive proteins will be affected and that is it that is how you measure the neuronal activity or see the behavior.

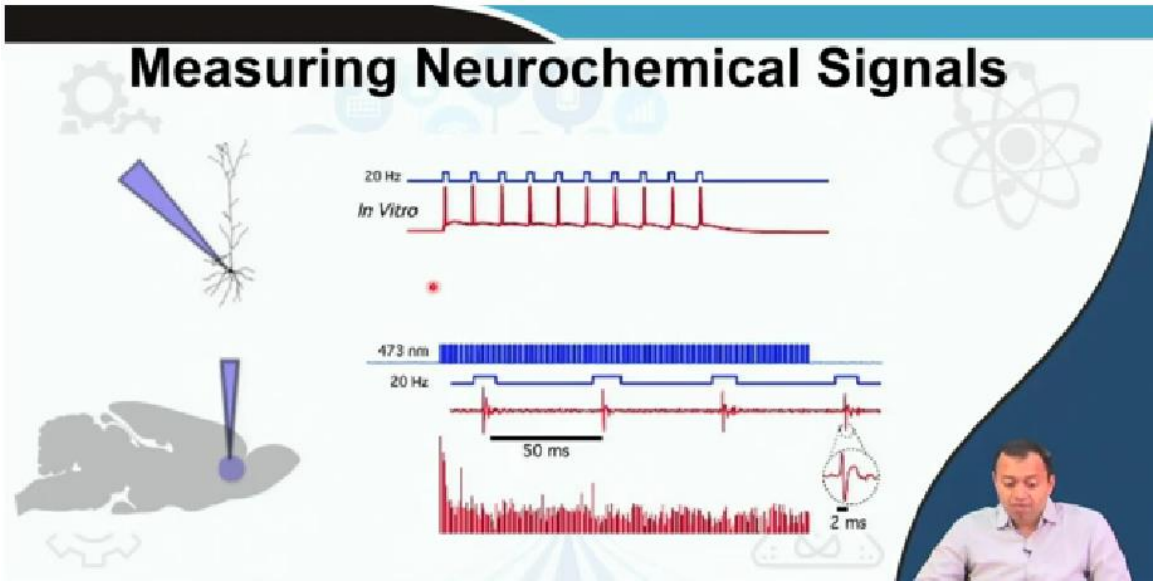


So, this is the entire entire mouse this is the ear and the mouth and this is the small small brain inserted through cranium a small keyhole surgery few micron thin is inserted and yes you can keep the mouse alive we will show you in the next I will show you in the next slides in a small keyhole like surgery it is possible to insert a optical fiber while keeping the mouse still alive and you measure the neuronal activity and map it with respect to behavior. So, see this is the biggest advantage of optogenetics or light based specific specific control of neuronal activity. If you have put a probe and electrical stimulation and electrode and electrode it will be affecting even if it is small large number of neurons and it will be passing same voltage to both of them or group of them

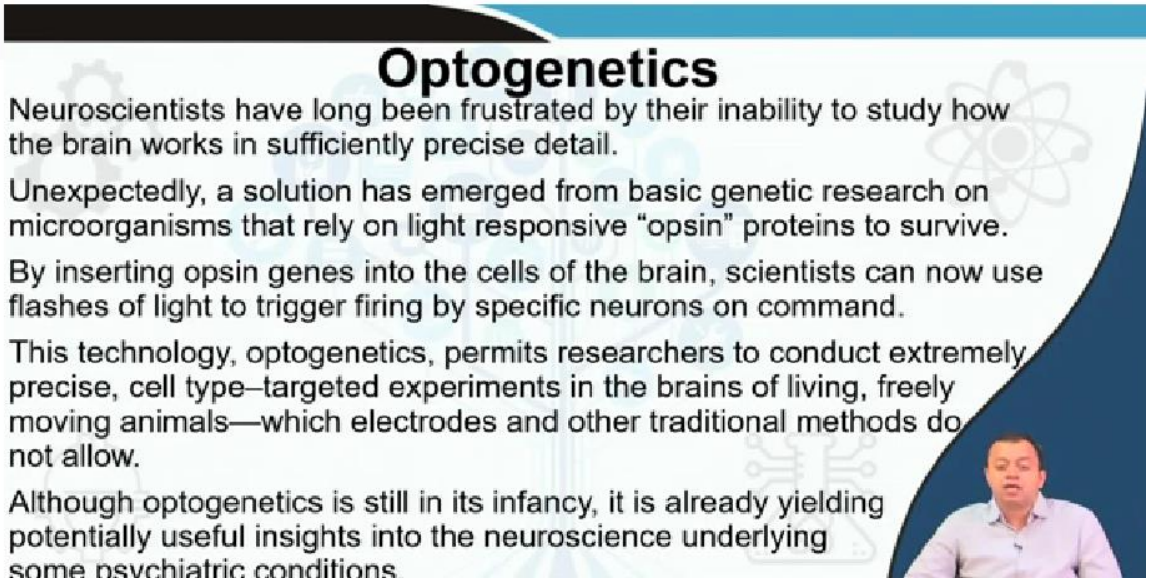


and you will be not able to distinguish between which neuron is trying to do what. With optogenetic excitation depending on the opsin protein that the neuron is encoded with depending on the blue light it will only result in sodium ion if it is yellow light it will result in chlorine ion negative ion flow positive ion flow direction is this direction is that negative ion flowing positive ion flowing sender receiver what is the resultant behavior what is the resultant behavior because of the difference flow of ions because of two different specific neurons talking among each other resulting in what specific behavior of the animal. Would you like to do something to your brain and let us probe it by inserting an optical fiber in your head in your skull and try to see whether you will start singing or dancing or become afraid or become very aggressive or become hungry or become asleep if blue light is excited in you inside your brain.

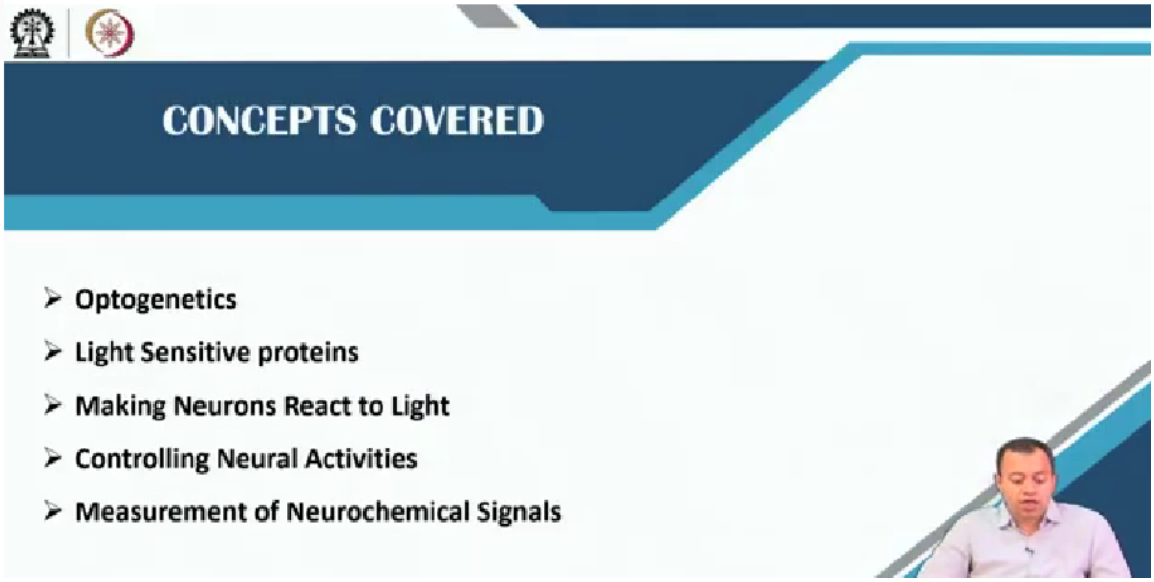
Do you think at certain time this would be possible in humans and you know primates and map different areas of the brain at this present moment this is very hot topic but we are still stuck to mice I am working on this on zebra fish the transparent fishes I am working on that this with these kinds of neuro photonic probes. So, they obviously they try to excite a specific area of the neuron so see this is the neuron with these branches this dendrites and exons forming they put some amount of light and then try to see the



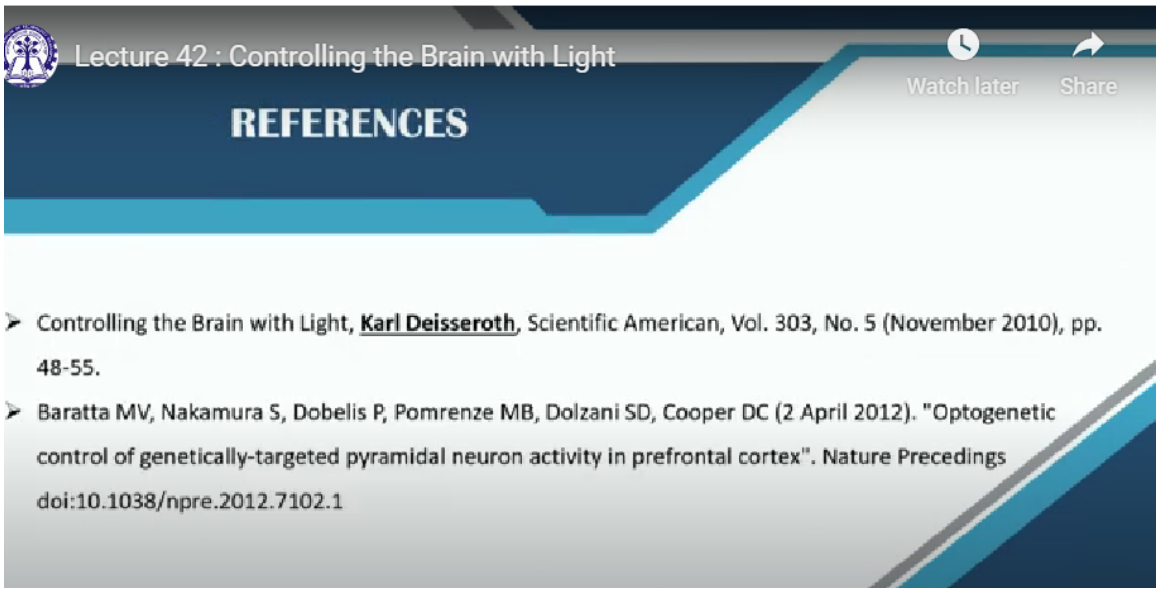
electrical activity and you see this 573 nanometer this is the laser pulse laser was given instead of continuous laser these were pulses and they were able to figure out the electrical measurement obviously you have to have some kind of electrical probe associated with it but that can be connected with a with a with the optical fiber which is a passive component like it is it will measure the electric current it will not send an electric current so it will it will just be there to measure the electric current passing from one neuron with another neuron or overall in the periphery it will not be sending any electric current by itself and they were able to see that yeah see this 2 milliseconds electrical pulses passing from one end of the neuron to another end by subjecting it to blue light basically photoelectric effect photoelectric effect of the brain electricity passes as light is being given as an input photoelectric effect. So please please read these references this is a fantastic new topic that has opened up I was actually debating whether this should be included in this or not but then I decided to double down and try to teach three chapters based on neuro photonics and optogenetics because this is state of the art there is nothing more complicated than brain and trying to analyze brain with light is the hottest new topic right.



So again optogenetics neuroscientists have long been frustrated by their inability to study how the brain works in sufficiently precise detail unexpectedly a solution has emerged from the basic genetic research on microorganism that rely on light responsive opsin proteins to survive proteins that produces photoelectric effect that passes electric current upon excitement by specific wavelength of light by inserting opsin genes into the cells of the brain scientists can now use flashes of light to trigger firing by specific neurons on command this technology optogenetics permits researchers to conduct extremely precise cell type targeted experiments in the brain of living freely moving animals which electrodes and other traditional methods do not allow although optogenetics is still in its infancy it is already yielding potentially useful insights into the neuroscience underlying several psychiatric conditions. So hopefully in 10-20 years with your help several of these mental health disorders mental diseases could be categorized and characterized to a cellular or a biochemical level as what exactly is wrong from a molecular point of view that is causing this particular disease and thereby probably we can cure it right.



So these are the concepts that I covered for today measurement of neurochemical signal controlling neural activities and I hope this is not too difficult of topic but this will continue for the time being please give me the feedbacks on the comment section and I will try to see what could be done if you find this very very difficult topic this is difficult I understand but is nevertheless very very refreshing and very very interesting.



So please go through my references fantastic new works are going on and I will see you in the next class. Thank you very much.