Nanobio Technology Enabled Point-of-Care Devices
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Lecture - 02
Biosensors and its Application

Ok. Dear students. So, today I will cover the second lecture in the Biosensors and its Applications. Let us come to the topic.

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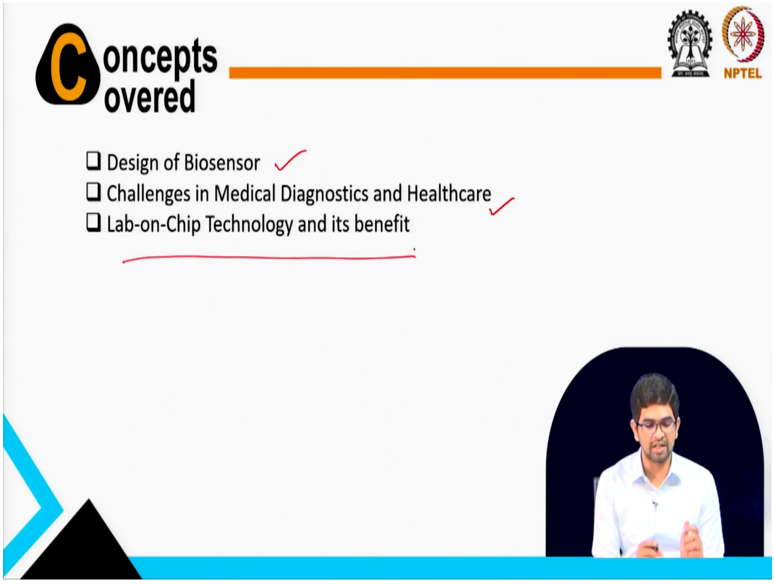

So, mainly I will cover here design and of and its applications for biosensing applications, for diagnosis. So, how you can design a biosensors? Right. So, 1st lecture I talked about like different kind of the biosensor it can be useful for this diagnosis, so how we can design them this is the very important part of the of this course.

Second topic is the challenges in the medical diagnostics in the healthcare. See in the medical industry they are facing different kind of challenges for the device development. As I mentioned we so many technologies if you see many publications available, but there is not all are in the market.

So, why? There is certain challenges definitely, so how we can solve these challenges? Let us address for solve the challenges and then I will let you know how we can bring this kind of technology on a single chip and then you can easily commercialize them. So, this is very important. So, lab on a chip technology this is the main solutions those challenges and its benefit that I will mention.

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So, main keyword of this lecture today is the biosensors and lab on a chip. So, this is very very important part of this course that I will start today.

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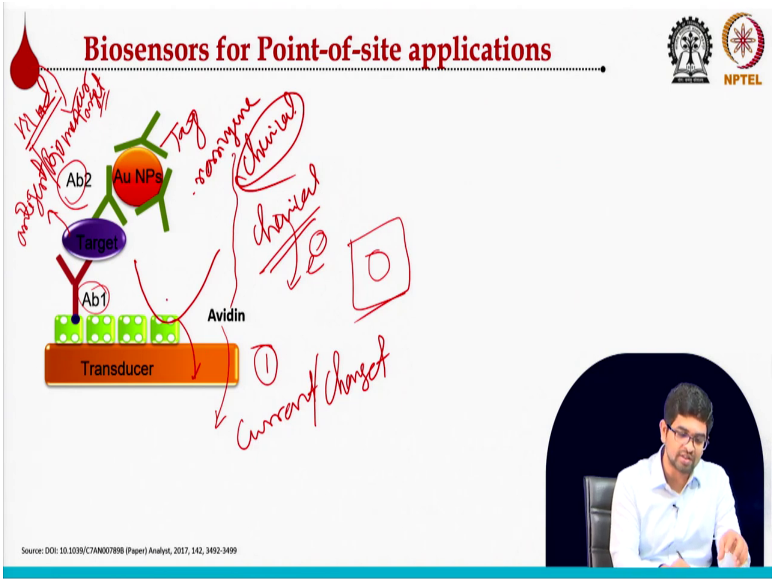

As I mentioned that biosensors is a analytical device that can detect specifically that can detect disease. And there is a different kind of recognition element I already mentioned in the first class like antibody, aptamer, enzymes, but as for example, I will always use the bioreceptor as for antibody. So, let us use antibody for development a biosensor.

I will show you a basic design now how we can develop a biosensor for point of site applications. Let us show you step by step. So, first we need a transducer this is the first steps we need a transducer. So, what is the transducer? Transducer just a you can say it is for the I mainly I will cover as I mentioned the electrochemical biosensor. In the electrical biosensor it transducer just an electrode that will convert.

So, here I will use some chemical for the device development. So, that chemical will react in this biosensor surface and it will transfer some electron that electron will go through the transducer surface and that will detect at the form of signal output in the form of current or may be charged.

So, so this is the role of the transducer it will change the signal from one form to another form like from chemical energy to the current or charge this electrical part and we will see the output value on a display. Maybe if you want to see like digital value even you can see the digital value for that we need some signal processing those things I will discuss later.

But this is the main part first main fundamental part of the biosensor is the transducers ok. Now, we will immobilize the antibody that is called the bioreceptor, so how will immobilize for that we need some material that can help to conjugate the antibody. So, again I will cover in the after few class. So, how we can conjugate the antibody on the sensor surface, but that is the another part.

But let us show you first part by part of the sensor development. So, we will use a Avidin and that Avidin actually help to immobilize the antibody. So, Ab1 where we mention because this is the primary antibody in first one antibody that will be immobilized on the sensor surface and it will help to target your analyte or that is called your biomarker.

So, what is this analyte or biomarker? That is a disease marker that you want to detect. So, first antibody is immobilized on the sensor surface with the help of Avidin. Now, see here this is the sample. So, suppose this is the blood sample right blood, so it can be urine sample, it can be saliva sample.

But as for example, I choose the sample is the blood samples ok. So, blood samples contain your target your disease marker right. So, that is called target. So, if you draw your samples on the sensor surface, then that target will be on your surface because of there is a antigen antibody interaction.

So, your target it is also called antigen also it is called your biomarker that is that you are going to detect this is the disease marker for cancer case it can be the cancer marker. So, this is the that is in general I am saying here target. So, because of the you know the antibody antigen they have a very specific interaction. So, your target only that specific disease that antibody which one you use that antibody can target that disease marker that is specific for this antibody.

Now, we will we can use the another antibody for this biosensor development that is called secondary antibody. So, the another antibody you can see here that is the secondary antibody. So, this secondary antibody can be conjugated with some tag. So, here Au NP you can see Au NPs means gold nano particles it can be any tag we can use here enzyme we can use here enzyme or even the nano particles even some chemical different kind of tag we can use.

Why we need this tag? Because this tag can react with some chemical as I mentioned know just, we can use some chemical and it will give some product or some another substrate and that will react on your sensor surface and it will give some electron that electron we can capture we can measure. And based on the number of electron transferring through the transducer surface we can get the signal output let us show you.

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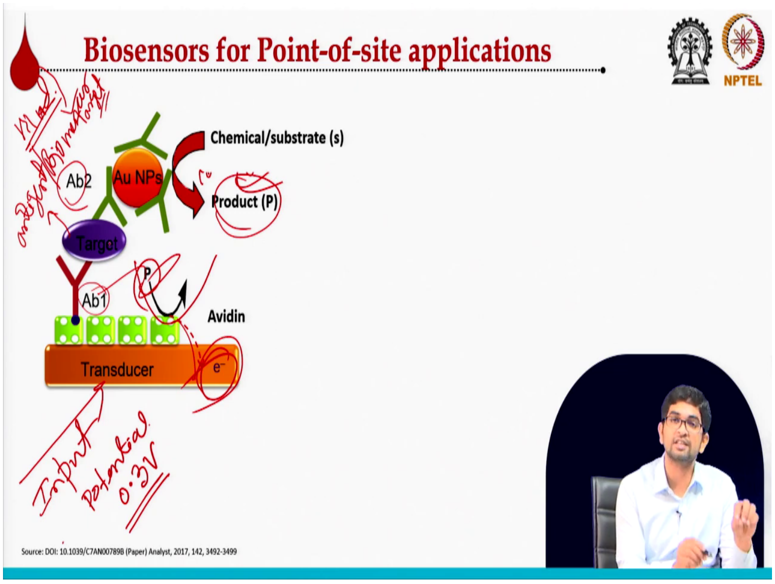

As I mentioned I can use some chemical that chemical can be useful that chemical can react with this tag and it will form some another molecules another substrate let us show you see. So, this chemical is reacting with this nano particle it forms some product, but some substrate and this product it can react on the sensor surface and it will generate some electron.

So, how this product will get? So, here we will give some input; input, what we can keep the input? We can apply some potential we can apply some potential may be suppose we are applying here 0.3 volt. So, as I mentioned that mainly we are discussing you know electrochemical biosensors, ok.

So, we will apply some input like 0.3 volt potentials. So, in this potential this P can be oxidized on the surface it will release the electron, this electron we can collect you know you can see the signal how much we you are getting current. See on your sensor surface if we increase the target concentration now.

So, this is the first the fundamental part of the sensor how we are developing the sensor and we are getting the output. Now, how we can change the output that can help you to determine the target concentrations in your sample, right.

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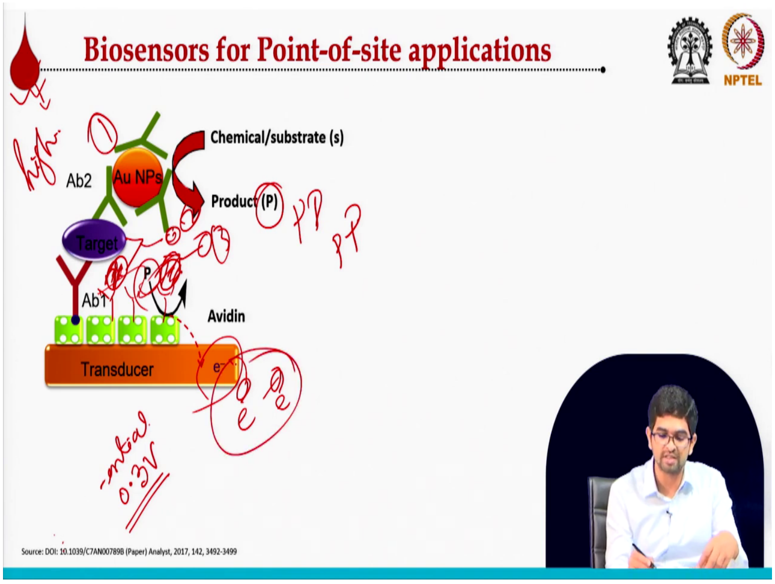

Suppose on your sensor surface you have 1 target right, but in your blood samples suppose your target concentration is very high. So, your target concentration is very high, but it is not only one antibody you know there is so many antibody actually present; there is so many antibody actually present. So, if target concentration there is another target can be on the surface may be another 2 target.

So, another two target means there can be the many gold nano particle can be on the sensor surface. So, not only 1 gold nano particle there can be 2, 3 or many gold nano particle can be there. So, there can be many product this P can be form like many. So, this all P now can be oxidized on the sensor surface and it will release the many electrons right.

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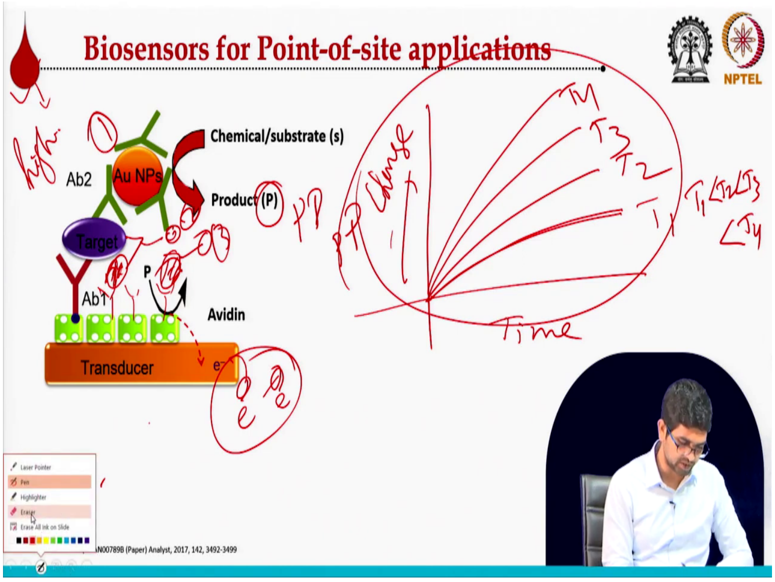

So, based on the number of the electrons your signal also can be increased right. So, at the very if the target 1s are very low suppose you are getting one signal this is suppose target concentration 1 this is suppose in this y axis this is the charge in the x axis this is the time. So, you this is called the chrono coulometry that all the technique name I will come later.

But just for your basic concept I am saying that I am taking the output in the form of charge with respect to time. So, if the target concentration is very low T 1, so you are getting this output. Now, if I increase the target concentrations. So, you also get like your C you charge like target 2 target 3 target 4, so T 1, then T 2, T 3, T 4 like this. So, your target concentration increase if slowly your charge also will be increased.

Now, based on this output you can make a calibration and that will help you to determine the concentrations of the unknown sample that I will show you now. So, let us erase this all. So, how we can detect the unknown concentrations in the samples?

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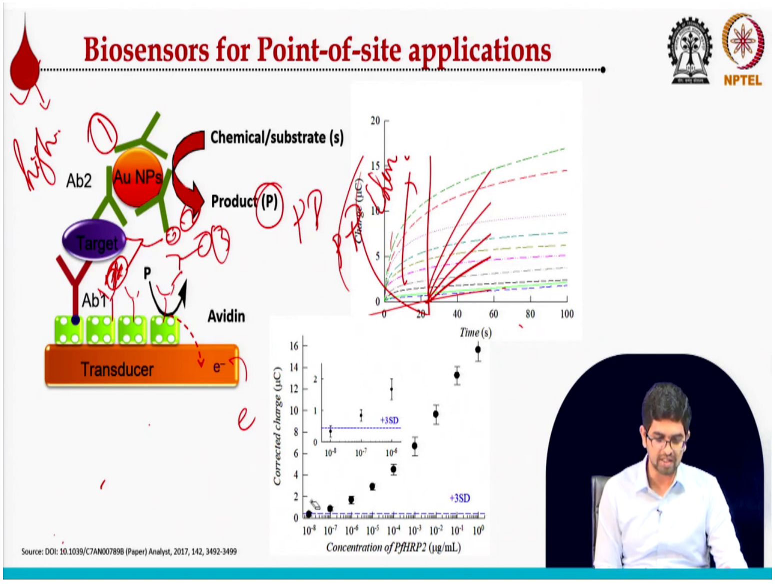

See this is the one example like we are detecting charge versus time with different concentrations right. So, you can see when we increase the target concentration we are getting high and high and high and high charge. Now, suppose here in a certain time suppose this 60 second time we can fix a time and we can measure the charge.

So, we can measure the charge and those charge we can plot here. So, here based on the concentration suppose here I use the concentration of PfHRP2. So, Pf PfHRP2 is just a malaria antigen, so just as for target I use the malaria antigen. Now, if you increase the concentration of the malaria antigen then you are getting high and high the charge right.

So, from this graph; from this graph in a certain time I am collecting the charge and I. So, this is a concentration increase charge also increase right charge also increase the concentration of the increase. So, this is called the calibration curve. So, in this calibration curve you can see that the increasing the target concentration charge changing. Now, if you have a unknown sample.

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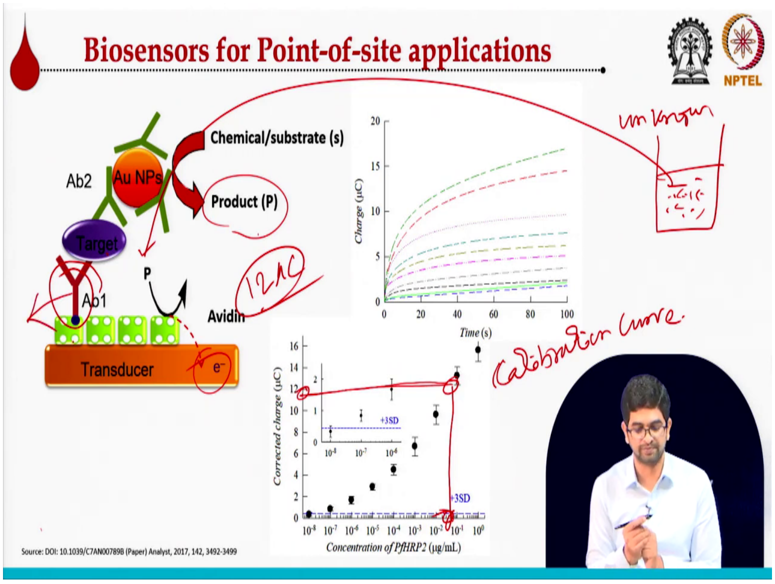

So, this is the calibration curve, so in this calibration curve we can determine the concentration of unknown sample. Suppose I provided you a sample unknown, but you do not know what is the concentrations of your target. So, what you will do?

You have to drop this sample to your sensor surface right just drop it and based on the concentrations your target will be immobilized on the surface and if the more target means more gold nano-particle will be immobilized on the sensor surface and based on this you will get this P product and your product will be oxidized on the sensor surface and you will get some current.

Now, this current or may be charged. So, this current was charged you can compare suppose you are getting from here suppose you are getting like 12 micro coulomb from the unknown sample. Then you have one calibration curve this is as I said this is the calibration curve, right calibration curve.

Now, here you can draw like 12 micro coulomb you got right, so 12. So, now you can, so here it is your charge. So, you have to draw is in line here then here you have to draw a straight perpendicular line and then you just see how much concentrations of your unknown sample suppose in this area.

So, based on this calibration curve then you can determine the target concentration right this is the very simple technique with the sensor you can determine any target concentrations and this is a very generic concept you may say sir why you use the PfHRP2 is it very specific or PfHRP2 yes this sensor is specific for PfHRP2.

But if you want to go for like dengue detection then this antibody you have to change and with dengue specific antibody you can use, then it can detect the dengue. So, it is very generic concept based on your bioreceptor you can change your biosensor ok.

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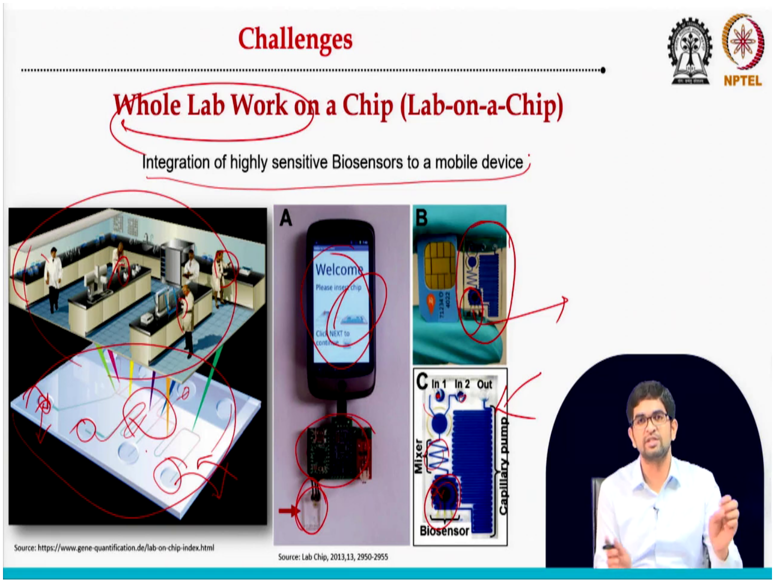

Now, come to the challenges. So, your sensor concept now this is ready now you can develop some different disease biosensor. Now, this sensor come to the device for the device development there is some challenges. As I mentioned the whole lab work, we are doing there is many stage when you collect the sample you have to go for this sample pre-processing stage right.

Like filtrations then for some chemical addition there is sample if you go for amplification then detections right there is so many stage that is called the lab work systems. But these all the lab work how you can bring to the on a chip see you can see in this image like some people they are processing the sample some people they are adding some maybe chemical here they are measuring. But these all the technique see you can bring on a single chip.

So, here you can see one inlet here maybe you can just you can insert you your some maybe chemicals here maybe you can drop your sample and that all the samples based on the microfluidic technology we can use. So, you it can go through this microfluidics and maybe some chemicals they also can come and mix with this sample.

And we all the sample processing, mixing everything can be done on a single chip, then it will come to maybe your detections area and here then you can apply some input that is I mentioned like potential then you can get the output in the form of current in the form of charge that you can see.

So, this is the lab on a. So, what is the lab on a chip? Then the definition what is the lab on a chip definitions means all the lab all the steps when you bring to the on a single chip and it there is no need any kind of like pre-processing sample amplification sample purifications everything can be done automatically.

Like you just drop the samples and you are done and maybe you have to just push a bottom there will be a bottom, then all the sample mixing can be happen automatically ha then it will come to the detections area and you will see the finally, the value. So, you then there will be a like a display like a mobile phone there will be a display you can see the value of the bio-marker concentrations.

So, you can see the lab on a chip there is you need some electronic part also why you need electronic? As I mentioned then you have to apply some electrical potential. So, that part actually will help you how much potential you need to do apply and this potentials will be applied to your this is the actually your chip your sensor chip.

So, this potential will come to your sensor chip and there will be some certain reaction based on your applications of the potentials and you can measure and you can see the value on the display. Now, come to the miniaturizations as I mentioned the lab on a chip is a very useful technology you can easily miniaturize very small chip you can generate that I already mentions no.

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So, you can see the one chip like this much see this is a very simple chip. So, here just you can drop like here sample and inside the chip all the processing system is there, but there is a already covered layer because for commercializations we cannot keep it open. So, this is the glucometer commercially available in the market. So, there is sample processing everything there. So, once you drop the blood samples it will go through this area then we will connect this one to a portable potentiostat here.

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If you connect the portable potentiostat then we done then we can see the digital value here. So, this is the concept of the lab on a chip. So, everything can be done near the patients and we do not need any very sophisticated expensive instrument right. This is the main concept for the lab on a chip technology.

See here in this slide we can make like a this kind of chip where there is some inlet there some microfluidics there is a sensor this is the sensor part there this all the part I will teach you again, but this is the actual lab on a chip all the integrated part, this is like just like a sim card very small.

So, you can see the like the like zoom here a part of this chip. See here we are actually dropping the sample here some chemical if you push this button all is come through the mixtures it will come to the biosensor part. This is because on this sensor surface you already have the antibody immobilized right. Now, your sample if you content the target then it will be it will bind here and it will show you the signal. So, this is the main concept.

So, integrations of the highly sensitive technology to any mobile device or Smartphone this is the main goal of our lab on a chip technology so, that we can plan to bring your technology near the patients that is called like bench to bedside applications. So, this lab on a chip technology basically help us to bring this concept.

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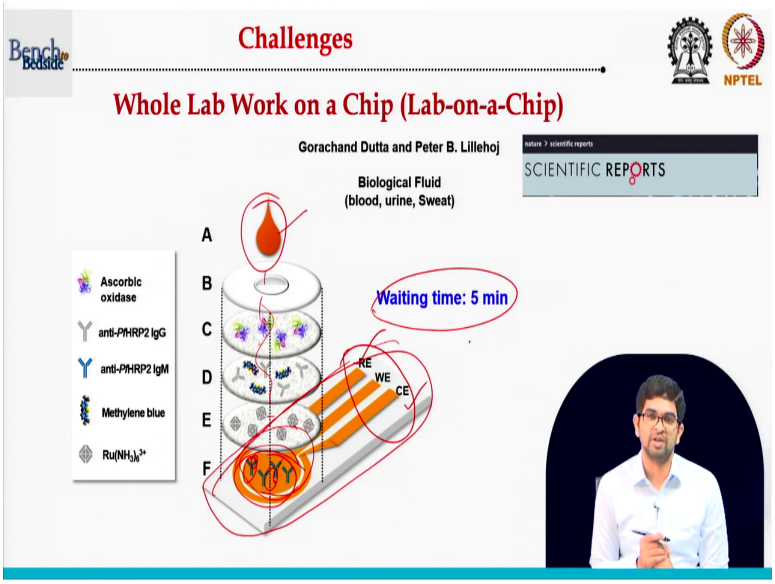

I will show you one fundamental again some example that we already develop in our lab that is the lab on a chip concept for detection of malaria. You can see this is a once very small chip there is a detection part that is called the working part this is called the working electrode and the in the electrochemical sensor, we need some reference electrode counter electrode that I will describe in the next class.

But just for your fundamental concept I am just showing you how we can develop a lab on a chip concept. So, we need a working area where generally we are immobilizing our bioreceptor for example, antibody. So, this is the antibody ready for the detections on the top of this sensors we are using some membrane like paper base membrane.

There we can use all the chemicals if you need the secondary antibody that we can use on this membrane. And on this top of this membrane now you can drop the your sample it can be blood urine anything any sample based on your interest. Now, your sample is content the target then it will go through the membrane it will dissolve the chemicals and even the secondary antibody also come then it will bind on the sensor surface then your sensor is ready.

Now, this part is the your this electronic part you can insert to the if it is Smartphone based you can use Smartphone or even any portable potentiostat that I showed you there you can insert. Then you can use some app using this app you can apply some potentials and you can see the value.

For this you may have to wait for some time because the sample will come it will dissolve some all the chemicals and it will come here an antigen will bind for the you need some time 5 to 10 minute sometime you may need the 15 minute, but for this work 5 minute also enough and after 5 minute you can see the level of the target in the sample.

So, that is if you think about the lab based work sometime you may need the longer time even the day, but here you can detect within a minute that is the main benefit for the lab on a chip concept.

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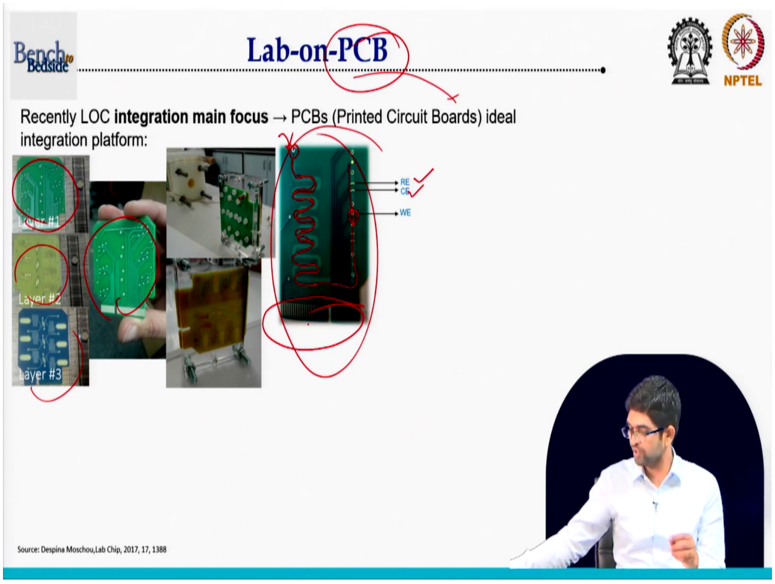

Now, I can show you the lab on a chip that is the main concept that you can see the bench to bedside that is the main part. So, some another applications some another substrate also different different substrate we can use for the lab on a chip development. So, we said lab on PCB you know the PCB means the Printed Circuit Board. So, recently PCB Printed Circuit Board are usually used for lab on a chip development.

So, because it is a very ideal platform. So, why PCB is the ideal platform? You know if you open a computer, I mean electronic part you can see is in the PCB right printed circuit board that part can be useful for the biosensor development. And not only in India every in all we have very good PCB fabrication setup we have.

Because you know the electronic part we are every day we are developing every country in India also we are developing a huge electronic part we are developing we have very very good setup and that PCB setup we can use for sensor development how? See suppose this is kind of the example you can see you already have seen maybe in your electronic device, so there we can fabricate our bio recognition molecules.

So, let us come to this part. Here you can see you can see the small small part those part we can use the as a working electrode as I mentioned as I showed you in the last here this is the working electrode where we can immobilize the antibody right whether the bioreceptors see.

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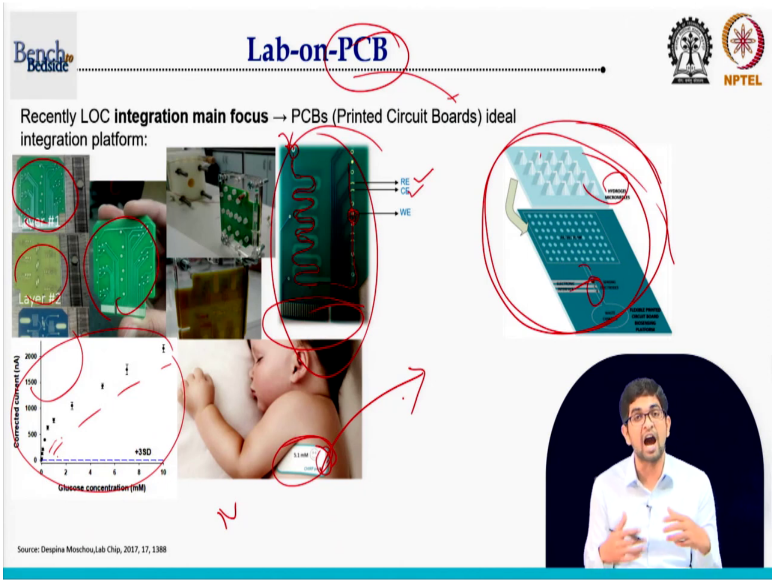

So, this part can be useful as a working electrode where you can immobilize the antibody and other as I mentioned I need some reference electrode counter electrode. So, other dot also we can use as a some reference or counter electrode. Now, I integrated here some you can see this is the microfluidic channel. So, if you drop the sample here with some buffer maybe some you may even if you drop one blood samples it may not go through this channel you may need some more you may need some more solution and liquid.

So, you can drop a buffer. So, blood now will flow through this microfluidics ok then it will go through the your working electrode where you immobilize already your bio sensor. Now, so this is the your electronic like you can develop a adapter that I will show you like this one see.

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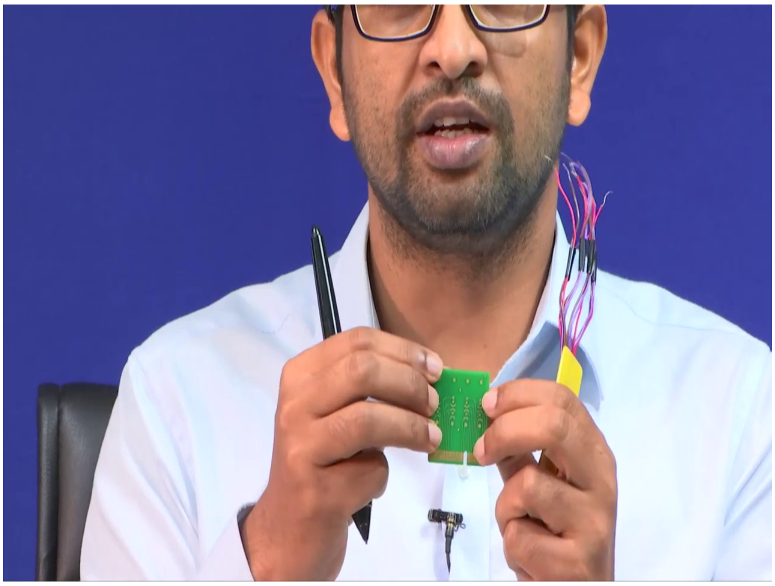

So, suppose this is the lab on a PCB one very good example right. So, here you can develop like your sensor here you can immobilize your antibody once your sensor is ready maybe you can integrate this one with the electronic part. There is some connector then this part you can now add to your detector or even the portable if you have any portable potentiostat and based on this your target concentrations, then you can detect easily your biomarker.

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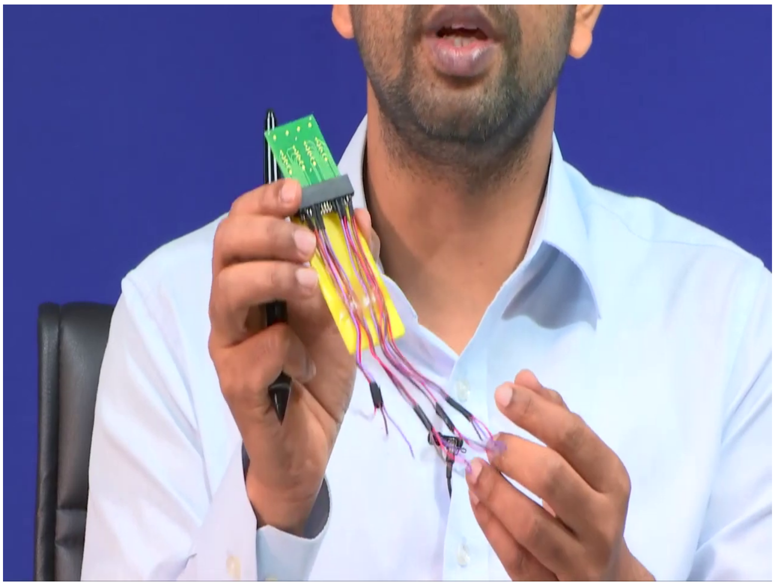

This is just a very simple this kind of portable device you can easily develop ok. So, this is the simple lab on a PCB technology you can use and this is the another part you can see we can easily integrate many part on your lab on a chip concept like here you can see some micro needle here you can see this micro needle why you are using? It can absorb the samples in non-invasive way also.

Although you can say that it is a needle, but it is very micro level it will not hurt your nerve system. So, it will not painful just you can put on your skin it can absorb the industrial fluid and then this fluid can go through the channel it will come to the working area or sensor area and it will show you the detections output.

So, some things you can even the integrate with your sensors you can see here this kind of patch also we can use this is just for the glucose detections patch just we can put on your skin and this kind of needle it can absorb the interstitial fluid from the interstitial fluid you can check the glucose concentrations.

And you can see a calibrations curve here based on the concentration you may get a different different current or charge output and you can measure how much glucose presence in the interstitial fluid. So, this is kind of non-invasive approach we can use. So, see the all the problems we can solve in a very simple miniaturized systems right.

So, like you are using here non-invasive approach, you are using a miniaturized small systems you do not need any expert persons just put on your skin and you can see the value on your you know mobile phone. So, maybe here you can use some electronic technology like Bluetooth they can send the value wirelessly to your Smartphone. So, this is the IOT enabled biosensor also we can develop.

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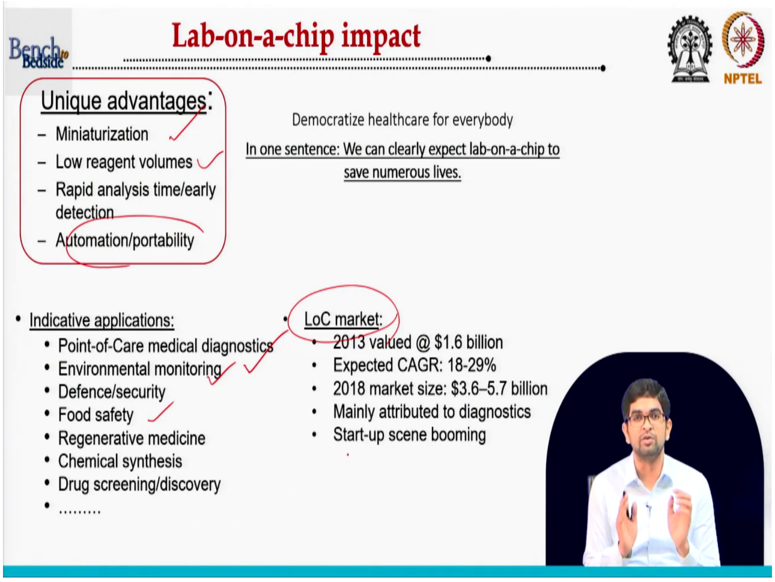

So, this is the impact of the lab on a chip technology that I want to mentions that I already told you that we can easily miniaturize we need very low volume of the samples we do not need like few ml of the blood just a drop of the blood is enough to detect the disease.

And it is very rapid method right within a few minute we can detect and it is also automatically when you just drop samples or push a button and wait for few minute and you are done. There is lots of applications in this lab on a chip concept like we can use for medical application point of care, we can use for environmental monitoring, in the defence case, food safety that I already mentioned. So, they have many applications.

And the lab on a chip concept is that is a very huge market I can ask my students who really want to see yourself in an industry or you want to make your own start-up your own company you can think about some new technology and you are I think you are done you can make you are your own business also like it has huge market.

You can say the billions almost to at the 2013 it was 1.6 billion market, but now in the recently that huge and it you have a very good scope that is the main thing that you have a very good scope to start your own like your independent your own business ok. So, that is the thing.

So, I think next class then I will cover again some lab on a chip concept and how we can complete this. And then I will slowly come the fundamental part like how we can develop the biosensor step–by-step ha like how we can synthesis the nanomaterial, then immobilize the sensor part then immobilize the antibody. So, step by step now I will teach you. So, this is just the fundamental part of the lab on a chip concept for today.

Thank you all.