Nanobio Technology Enabled Point-of-Care Devices
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Lecture - 26
Tutorial on Biosensors Fabrication (Continued)

Ok dear all. So, as I propose that I will again start some tutorial. So, let us see in this class I will show you again some problems. As I said this is not just a discuss some of the questions and answer. This is just kind of the problem that I am throwing you and try to answer that you can think independently some new problem you can try to design some new concept for biosensor design that is the main goal to teach this all the tutorials.

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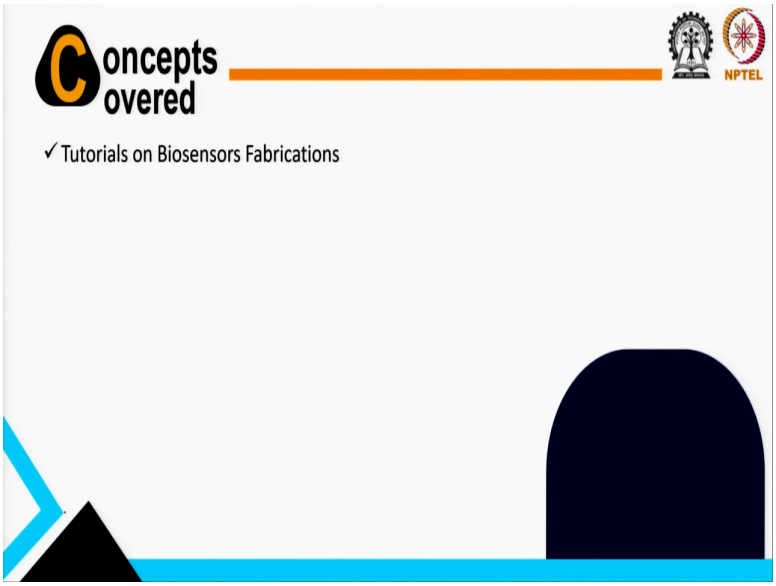

So, that us why todays main concept that I will cover. Let us show you some basic tutorial the problems and then try to solve in the different way. So, whatever I taught until now let us summarize everything in a single slide so, that you can solve these basic issues. Let us see.

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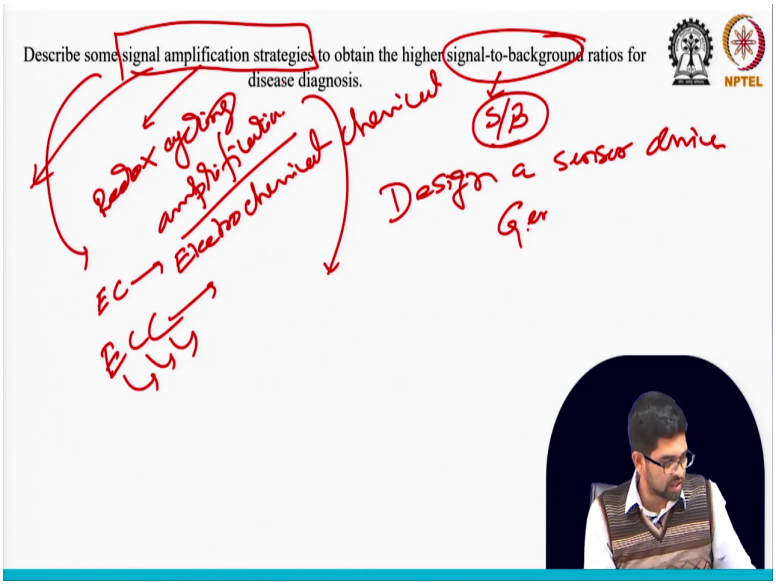

So, first questions for today describe some signal amplification strategies to obtain the higher signal to background ratio for disease diagnosis say. So, when this kind of questions may come how we will start to solve this? So, here I mentioned describe some signal amplification strategies right.

So, just think the signal amplification strategies I taught you mainly please try to think here I taught here redox cycling redox cycling amplifications. So, describe some signal

amplifications means, you can start some EC means electrochemical chemical right or maybe you can start ECC. ECC means, electrochemical chemical redox cycling.

So, this kind of redox cycle is may help you to amplify the signal. So, you solve the first part. Now, here the mission to obtain higher signal to background ratio. So, signal to background that I already told you in the last tutorials also. Like all the strategies you have to keep in mind while you will design. So, I mean the main goal to give this problem is design a sensor device, but that is very generic that is very generic thing not like very specific.

So, here that is why do not mentions any specific disease just for disease any kind of disease like this way if you start to think first. Then you can try to design some very specific biosensor ok. So, those things just try to remember.

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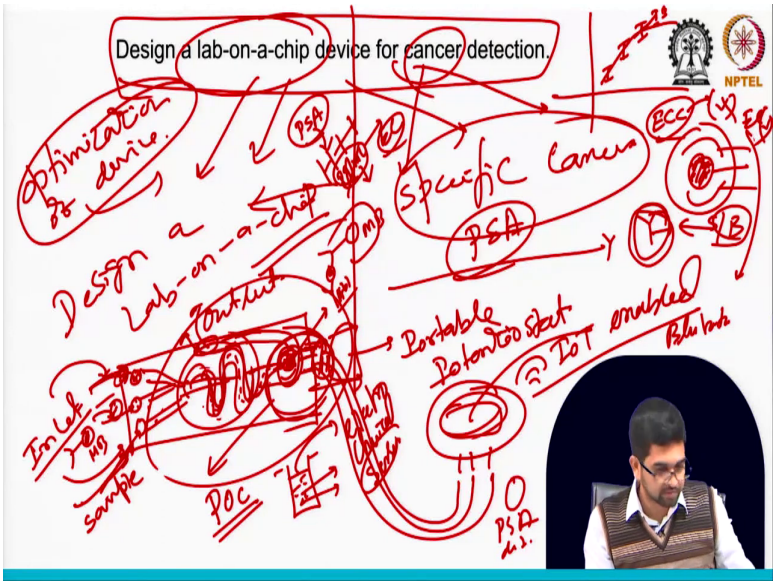

Now, here see in this problem this is again some generic concept so, that whatever you learn let us bring all together here in this problem. Suppose this is also can be a one project for you who are the students who are thinking for your higher studying you want to go to another university for internship or maybe you want to go for may be one-year project like you can think about like a small project like how to design a lab on a chip for cancer detection.

Means, if you go for the whole optimization of the lab on a chip it can be a PhD all of your four to five years you may take, but you may because it is very interdisciplinary design a lab on a chip then its optimizations, then cancer detections, but also it can be like just go for a specific cancer specific cancer like prostate specific antigen suppose for example, I choose.

Then this is the biology part prostate specific antigen and here is the your engineering part engineering part is the design a lab on a chip. So, this chip design. So, first what you will do, let us make your chip right here. So, on chip it should be useful for point of care that definitely you should keep in mind. If you want to because in this course is designed the main concept is the is for point of care.

So, that is why I will I am teaching you such a way. So, it should have a inlet right inlet means where you will drop your sample and outlet there will be outlet, outlet means, excess solutions may come out ok. And there is no need many steps sample handling, sample purifications, sample amplifications those things we do not need.

But definitely we need that that will be automatic everything on your chip it will be automatic. What lemon will do? They just drop the sample and they will wait and they wanted to see the value that I said digital value I wanted to say like the glucometer what we are doing in the glucometer just you prick the finger one drop blood then we take this blood on a chip, then just you can see after few second you see the digital value on yours detector scheme something like this lab on a chip comes away.

So, such a way you can design your project. So, inlet means here. So, you can definitely you have to integrate this one with the micro fluidics why we need micro fluidics? Because you

can handle easily. Maybe in this inlet will be your sample right maybe you need another inlet where you can add some reason maybe you will be provided a tube that will contain some basic reagent right.

For your I mean amplifications of the signal that I taught you know like some chemicals that some very basic chemicals and that you can provide. So, one thing you can ask me sir, if we provide this chemical maybe they may not be stable for longer time because once I prepare and then you are means when you commercial in this maybe this solution you have to keep it for the longer time that time you may be destabilize it may not. So, the very stable data day to day.

So, what you have to do maybe you can make some solutions with a stabilizer so, that it should be stable or you can give some very basic buffer solutions, but some reason you can keep on the chip in a lyophilized condition so, that they will not be unstable that you have to keep in mind. The all the details again I will show you how to store all the reagent and how to go for packaging of your final device, but that is very important part.

If you not follow the in packaging part may be if you keep in the air maybe it can be oxidized maybe somehow your reagent can be damaged right packaging is very important part that you just try to think here maybe after this tutorial I will show you again very details where how to go for the packaging that is very important for the stability without good packaging you cannot commercialize your device.

So, here your chemical, here your sample you may add some more inlet also based on your requirement and there may be serpentine like this way you can design your micro fluidics right because you need proper mixing of the sample. Maybe see here may be your some reagent, here you are adding the sample, but you may think you may add one by ones also maybe sometime first add the sample then wait for few minutes.

So, the sample may come to your sensor may be here you will be your sensor your working electrode your reference electrode your counter electrode. So, and it will be connected to through your micro fluidics then your micro fluidics channel will be here in the outlet. So,

excess solution will go out from this outlet ok. So, your solutions your reagent may come like this way, then it will come to your working electrode, then your target if you have the cancer target like prostate specific antigen. So, it will bind.

So, on your working electrode your prostate specific antibody that is antibody one will be here right and now if you have the prostate specific antigen thing it will bind then another part maybe you can give like a secondary antibody that conjugate with methylene blue right that I taught you already then it will come then methylene blue will be bind then it will form the sandwich right.

Then another way you can send some chemical, but I am saying very simple way, but you need a optimizations that you can go for a project that I am saying like a how long you want to like a when you mix the sample everything and how you want to start like first maybe your sample, then it will bind then this there is a some incubation time that you have to think, there is a design of this micro fluidics this serpentine structure that you have to optimize right.

Your nature of the substrate of the micro fluidics that also you have to optimize and you have to check which one is the best size of the micro fluidics channels everything lots of factor that that is called optimization of device that is very important point it may take some time that some time.

It may be your PhD whole project also if you go for a very systematic way if you want to develop really a good point of care device and you want to commercialize it then definitely you have to go very systematic way. Then your this design of the sensor I mean here is a sensor main electrochemical sensor part your electrochemical sensor are here.

So, your working electrode reference electrode counter electrode then here see your connector. So, here you can connect with a small portable potentiostat. So, here you can design a portable potentiostat portable potentiostat just like glucometer potentiostat you have seen right something like this.

So, your potentiostat. So, maybe here will be a some your connector. So, this will be connected here this will be connected here then here you can see your digital value or else what you can do? You can transfer this data via wirelessly you can transfer this data that is IoT enabled IoT enable a device you can make that is something this whole part is the engineering part you can make.

So, IoT enabled means via bluetooth via bluetooth you can transfer this data to a smart phone right you do not need any extra detector. So, on your smartphone you can see the data. So, how maybe on your smartphone there will be app like PSA detection app just turn it on and via Bluetooth it will transfer the data and you will connect your mobile phone and you will see.

So, such a way we can develop a smartphone integrated nano biosensor this is the engineering part that you can do. This whole things will may take longer time also and biological part that is the things we can handle there is a sensor part right like working electrode counter electrode reference electrode right.

That here means, how you want to modify this sensor working electrode and then you want to modify your antibody then which concentrations of primary antibody you are immobilizing on the working electrode right that concentration you may need to optimize.

Because sometimes signal to background ratio is very much close to this antibody concentrations on the surface if you use very low amount of the antibody on the surface then easily your surface can be saturated why suppose you are making a calibration curve right.

So, may be few pictogram, nanogram, nanogram then your then your sensor can be saturated, but if you can really optimize your surface with antibody then you can go further nano to maybe microgram also it will not be easily saturated. So, your detection range of your sensor will be very high right may be femto, pico, nano, micro you can detect.

But if your optimization not done properly maybe within nanogram it will be saturated. So, that is why this part also we have to take care then again, the concentrations of your reagent. So, many for if you want to use ECC redox cycling there is many chemicals, but I will favor if you want to really commercialize and you want to make very simple chip and people let us go for very simple reactions not like very complex ECC reaction means, very little bit complex right there is lots of chemical there.

Let us go for first very simple reactions for that you have to go for some review like if you go review like prostate specific antigen you want to detect what is the clinical range actual clinical range in human body. If you see if you go the like normal patients to like who are the critical stage what is the range of the PSA. If you can check it will start from picogram per ml to then again it will go to slowly higher concentration I mean nanogram something like this.

Picogram per ml means, you dont need very ultrasensitive biosensor you do npt need actually really to use the electrochemical chemical redox cycling maybe just simple redox cycling like EC redox cycling just simple redox cycling can help you to detect the picogram ml I mean. So, if you can detect like the healthy range to the like who are in a critical stage in a patient this PSA range, then that is fine they no need to make them more complicated right no need to make the complications on in the reaction side.

So, do not use the ECC let us try EC only like this way you can design. So, and further EC you need maybe like two chemicals let us try to optimize their concentrations right that part also you have to take care of ok then you may need washings you may need some here pump like valve you may need that also you may think like manually you want to just inject it or there will be some automatic the just press on just push this button then automatic it will start something like this you can I mean design I just giving the concept you may think.

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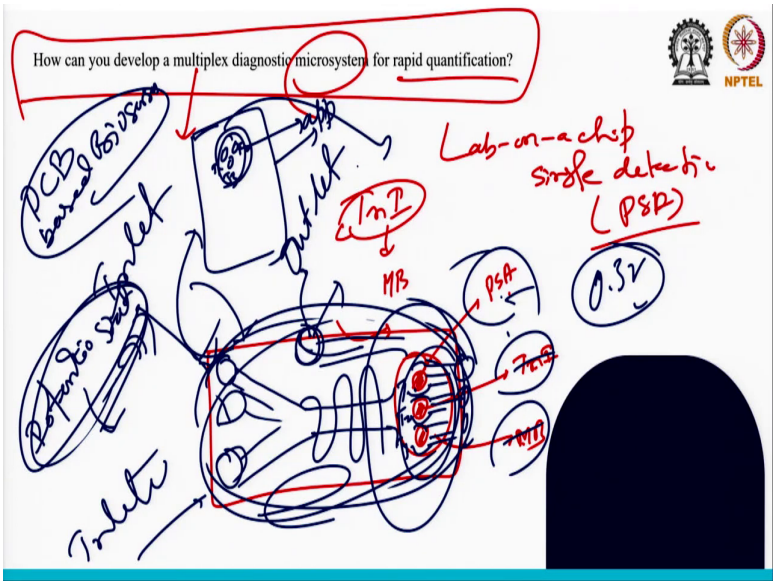

Now, let us come the see again some this is do not think this is the problem, this is kind of a like a small small project I am teaching you how can you develop multiplex diagnostic microsystem for rapid quantifications means last slide I we developed a lab on a chip; lab on a chip that is for single detection right single detection just like PSA.

But we can go for multiple analyte like you may need to detect like cardiac troponin. I like some another like this kind of biomarker related for the heart attack or myoglobin. So, myoglobin or cardiac troponin I. So, they are related to the this is for the heart attack. So, in the like when age like more than like 50 and nowadays you may see the younger age also this kind of level may come and we are facing the different cancer or heart attack.

But if we can a; if we can a design a device like here your whole sensor part one sensor can detect prostate specific antigen, another cardiac troponin I, another myoglobin. So, when the

age become 50 or more than 50. So, we may keep this kind of small device in home and regularly we can regular means not like every day means with months or after few months like six with every six month like we can check this level that this kind of disease may come like slowly not suddenly.

But if we check it regularly maybe we can take the precaution we can take the doctor's advice and definitely we can save the patient life. This kind of device definitely will be helpful for society that is why and for that nobody will come to doctors every time to check my prostate specific level troponin level or MB level if you have something in your home may be it can go for the yourself means self-checking and you can do this kind of some data that you can predict you can take some your own decisions that is really useful.

So, that we can minimize may this kind of death. So, this is the motivation. So, same way let us design a lab on a chip for multiplex diagnosis. So, one for PSA one for troponin I one for myoglobin two way you can make one this is they are the working electrode here you have to immobilize your specific antibody for PSA here troponin I here MB here.

One way just working electrode separately common counter common reference or another way you can design another way is that maybe you can make like separate a working counter difference working counter reference your working like this way also you can drag.

So, then you need may connector here that is all and same way the you will have inlet here maybe here you will have one outlet here inlet here also inlet and you can design what like I taught you in the last slide then what you will do like. So, in this case you may need like separate inlet you can I may say as it is the multiplex you do not know that all are present or not together.

So, let us use only one single sample and then this sample may come it will go through this or whole surface right you will go through the whole surface then there will be the outlet and then will come will come here excess solutions and you will have like others like reagent or if you may need some buffer that you can try right here you may need some buffer those the

things you may need here serpentine structure because to mixing of the all the things like your reagents your sample that you can design that like single like I taught you last slide.

Like this way you can make a very innovative and important lab on a chip, but just try to remember there is a two part as I told you in last slide also one is your engineering part that is the design another is a biological part the biological part is you have to properly review like which marker you want to go for the multiplex development right like based on the current scenario I mean there is a the winter time then you are facing like fever right.

So, you may develop a device cause for this fever like dengue or a right now COVID. So, dengue, COVID, malaria maybe you can combine together right or this is because of this like viral fever or some bacterial fever that also you can make right well you do not know sometime it is if you because of viral fever you are giving the antibiotics. So, what happened nothing and your body would be coming antibiotic resistance right.

So, in the future your body become resistance and if you add the antibiotic, it will not work. So, you actually it is more dangerous that is why you may decide I mean doctor also can check in his chamber that he is her chamber that yes this is for viral that do not prescribe him the antibiotic or it is antibiotic do not go for the viral treatment something likely this you can design some lab on a chip see that is why this tutorial elements are giving the concept.

So, that you can think about this kind of problem and design something new idea ok. So, now this see. So, here sensor part ready one is the your engineering part that is the design of the sensor and another is the biological part that selections of the target. You selected all the target now if you want to make the more handy's more smart a device what we will do? On the back side of this chip, I like to I taught you know already the PCB based biosensor right.

That concept now you can bring here what we will do on the top of the PCB you just design your all the multiplex sensor in the back side of the PCB you can integrate your electronic part that is the utility of the printed circuit board you are using one part sensor another part the electronic well easily electronic small small part you can integrate with the PCB.

So, there you can make a small potentiostat itself potentiostat because you have to apply some in input right some potential like for the oxygen scale like 0.3 volt you want to apply.

So, this potentiostat may help to apply the potential and all the like yield to chemical technique that we want to apply here that maybe cyclic voltammetry maybe impedance may be chronocoulometry, amperometry whatever those programming you can set you can set here and that you can make a app on your smartphone and in that app you can connect this one wirelessly through your smartphone and your device.

So, your device will be connected through your smartphone wirelessly via Bluetooth. So, it is IoT enabled biosensor. So, that what happened. So, this chip back side you have all the electronic part and via Bluetooth by controlling your app once you start then your chip will get the signal then let us run then run means it will start apply the 0.3 volt automatically.

Then how much current it is generating that much current equivalent to how much target concentration for like target 1, target 2, target 3 everything will transfer to your smartphone and here you will get the 3 data T 1, T 2, T 3, data you will get then that data also can be stored to the I to the cloud so, that doctors also can access this data means in any emergency case doctor also can give you immediate advice to you yes you need you need maybe some treatment immediately.

So, it is kind of that is why IoT enabled smart device is really important in this current time this technique you are applying on the smartphone this whole the device that is why it is very very important this technique you should use.

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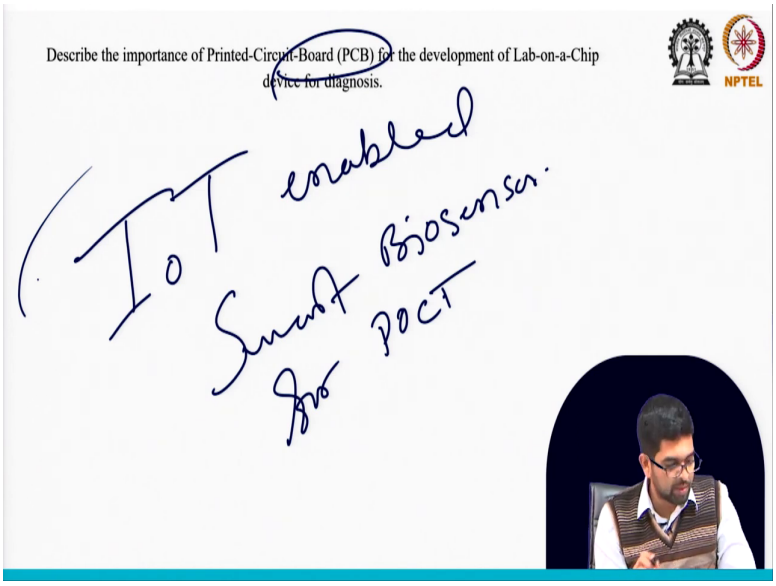

Yes, that problem that I told you in the last slide like describe the importance of printed circuit board for the development of lab on a chip that I taught you know let us use PCB for biosensor development why you need the PCB because you can easily integrate your electronic part on your biosensor you can make a IoT enabled smart biosensor for POC point of care testing POCT that help you ok that is why you need the printed circuit board.

And another things the PCB that you know already I taught last class when I taught the PCB easily you can fabricate we have very good fabrication technology that is why you need the PCB ok. So, that is you have to keep in mind. So, that I taught you that how to develop the new concept please try to remember this all the concept that is why this as I told you at the very beginning introduction class this class is not only the for your examples.

So, you will get all the techniques all the concept for your future study also for like you can start your own startup your own career you can start like this way that is why this tutorial is very very equally important and want and easily you can pass your exam because if you have all the this concept this can same questions may come in the exam also. So, let us go through all the tutorial I may start some more tutorial in the next few classes then I will start again some new topic ok.

Thank you students, thank you.