Course on Analytical Chemistry
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Module 12
Lecture No 57
Applications (Contd.)

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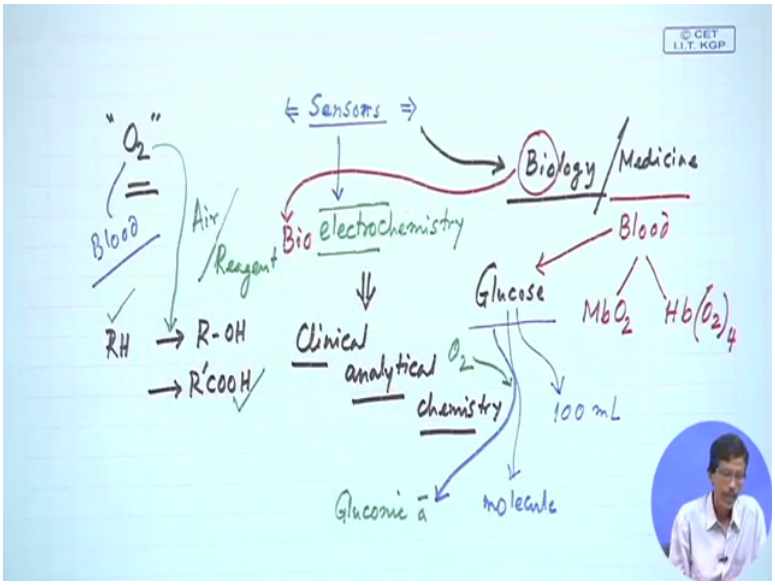

Hello, welcome back so we are talking about some sensors and as time passes on we should be now smart enough to understand these sensors in a different way, so the very basic thing what we require because all life processes are dependent on O2 but you have seen that the measurement of this O2 in anything any solution even within that environment also. Only thing that you have to have some saturated solution that means you bubble oxygen to some solution and you put that solution for your electrochemical testing or electrochemical analysis or electrochemical sensing of your O2 molecule.

So this is very important and a very straight cut one, now we slowly move to something where we can define it as a biological thing. How biology can benefit all these things and also the medicine or the medical people can also get benefit from this sort of measurements because we are linking this and this O2 molecule everybody should know that thing a chemists, a biologist, a doctor and everybody.

It is talking we are talking in terms of the corresponding O2 present in blood in any or any kind of fluid what you have in your body that means the dissolved oxygen is always very important but in terms of something that means if you go to a medical doctor that if you just

go for your blood to be tested and this blood can also be tested for the presence of your O2 molecule that means the dissolved O2 in blood that is why I told you that how your myoglobin molecules Mb are the hemoglobin molecules are involved because this we know that this can bind to O2 molecule and hemoglobin can bind 4 O2 molecule to give you species like this.

So these are very important and these are also interrelated to know now another components, so anything which can be analyte of interest for this thing that means if we talk in terms of the corresponding development of this sensors and we are talking about electrochemistry. The very basic thing what we are talking from our school days also that means electron transfer, electrodes and all things that is why we put electro.

Now we are bringing something will take out something from this biological part and we put here giving rise to study for bio electrochemistry. So not only your materials, solid materials in organic complex or organic molecule you can detect its presence as well as quantify because analytical chemist job is how to find out the amount presented at that means qualitatively you detect first weather oxygen is present or quantitatively you find out the amount present.

So the same blood if we can handle because we know that a common disease we are facing the aged people also the diabetes. So the presence of glucose in the blood we can detect why we are talking all this because this is not in that particular one bio electrochemistry for whom the doctors, the pathologist whom are responsible for making this measurements or quantifying the amount of all these species.

So they are looking for something we call clinical chemistry or clinical analytical chemistry which is very important, clinical analytical chemistry we should know. So if we have the glucose and if to a chemist particularly to a organic chemist, to an organic chemist what the glucose is a typical molecule it can come from a dextrose it can come from other other type of species maltose, fructose everything.

So is from so is a sugar molecule, so patience who are suffering from diabetes they are having we call as a sugar content or the sugar control of that. So we determine the presence of glucose say in 100 millilitre of blood. How much glucose is present, so glucose to us to any organic chemist is a molecule which is organic molecule and to a electrochemist why we are looking for this glucose for this determination, what we should know? Because the

sensors electrochemical sensor, so your electrochemical sensor should sense the electron transfer and we know that this is a nice molecule which can sense.

Now what you have to see you have to see 2 or 3 things together at whether this glucose molecules electroactive or not that may not be so easy to understand because most of this organic molecules we know if it goes for oxidation, you may not get the corresponding oxidised form like we get for the metal ion, the ferrous ion is oxidised to ferric ion. He can have the sufficient stability for ferrous ion as well as the ferric ion.

But if it is a glucose molecule whether that glucose molecule is oxidising to give you a oxidised form which is also sufficient and stable in terms of only electron transfer but to organic chemist, we all know that if they have some species like RH, they are happy to talk in terms of this formation as R-OH or if some carbon is coming out over here you can consider that means R (())(6:22) COOH formation that means the carbon center having this that means is a carbon hydrogen bond and go to carbon OH bond and the other carbon also if it has something means it is going from a alcoholic part OH or to (())(6:40) part CHO or a carboxylic part.

So what we are looking for here is that we are doing nothing but simply we are rationalising all these things, how you look at the problem, so is defining the problem to choose the problem also that your O2 is coming, that whether you get this O2 from a cylinder that oxygen cylinder or whether you get it from air only, that O2 is there and O2 is being inserted and sometimes if it is not coming from the air we say that no sir I have given some reagent for this organic transformation, so it is coming from some organic reagent also.

So is we know that a very good reagent for transferring those oxygen is your peroxide for organic peroxide uhh perbenzoic acid and all these. So the reagent but how is the reagents are uhh making, the reagent can also be making directly from this O2 and there are some other molecules also like this myoglobin, hemoglobin and hemocyanin and we know there are also bind oxygen.

So binding oxygen is something and this oxygen utilisation for your oxidation to convert RH to R-OH is something different, so if we can utilise this oxygen for a glucose thing at means this oxygen is utilised for glucose and glucose is your RH, you know that some other groups are also present that means uhh that CHO is there but term we should know that we know that glucose is oxidised, we get gluconic acid.

This is also an organic acid, so what does it mean? That means also that if you have something some groups like alcoholic function or a carbonyl or carboxyl function like (()) (8:34) it is there which can be oxidised straight way to its corresponding acid, so these 2 information whether we can develop some process such that we can go for your electrochemical measurement and bio electrochemical measurements and we can develop like your oxygen sensor, a glucose sensor. So the problem is that how you can develop a glucose sensor.

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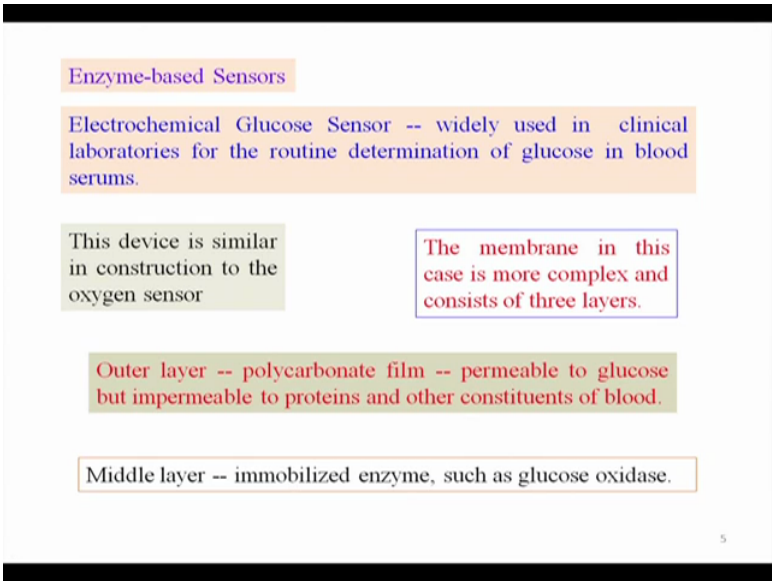

So it should be now we bring something that your life is much more complicated you believe that we are bringing something Enzyme. So you see a chemist should also know little bit about the enzyme because we all should be expert when much because we know that there are certain groups of enzyme which can be defined as metalloenzymes that means the metal ions are there. So who should know best about those metal enzymes, it is the chemists rather than organic chemist or the chemist who are dealing metal ions. So metal ions you know very much so you should not know the corresponding enzymes or the metal enzymes, so the sensors what you can develop or glucose is a enzyme base sensor.

So what we have seen for your case of O2 molecule the thing is very simple that you get this for at measurement of this O2 for other sensor but you have electrochemical glucose sensor and we can talk that is a organic molecule if you consider that glucose it can be a carbohydrate molecule is no carbohydrate chemist can also claim that it is a carbohydrate molecule.

So electrochemical behaviour and when you bring the glucose is the organic one so organic electro chemistry we are doing but when you bring the enzyme from the biology area, the field of biology is giving you the enzyme, so we can directly consider this as a study on bio electrochemistry. So in different clinical laboratories, the pathological laboratories they handling the clinical chemistry or clinical analytical chemistry they are routine day to day, it is not a analytical chemist job once in the standardise is the job of a chemical analyst.

So there are large numbers of this chemical analyst or the chemical analysts are there so they are handling all these thing in routine way at means 100 or 200 samples they can determine in a day, so glucose and blood serum, so you have to have the corresponding serum sample in hand and you take this. So how you develop the thing, so if you know nicely how oxygen sensor is been developed, you can develop other one that means the glucose sensor and be developed because basic principle or the basic idea behind the development of that sensor indicating sensor that is indicated electrodes and indicating sensors which can detect the presence of glucose in your blood samples.

Now what you have because already we have discussed a lot about the membrane, the porous part is fabrication with respect to the electrodes in this case it is more complex and consist of 3 layers now earlier we are talking about one particular layer and a porous part which is allowing the oxygen to enter. Now you have a outer layer, so as not much complicated what you go in a step wise fashion, you are moving from one after another that means you are going from one layer to the second layer to the third layer.

Due to the introduction of this enzyme because we have bought enzyme, where to keep their enzyme? So we brought that particular enzyme and we have seen earlier that how you can frame that thing that you have the electrode, then some mediator you can have, then the enzyme you can have, then the substitute you can have. So layer after layer you know already that how you can get that so now you bring the outer layer now you see some polymeric material earlier we are talking about the polypropylene cloth then the Teflon film now we are bringing another thing which is also (())(12:40) very important is your polycarbonate film.

So is the polymeric material of large number of carbonate (())(12:49), so the polycarbonate plane so the PC material is known as PCs like PP which is permeable that means which allows to pass something that means glucose is the bigger molecule compared to your O2. So glucose, it allows the glucose only to pass but is impermeable that is the selectivity comes into play that the size, the port size, so is a semipermeable membrane, semipermeable in

terms of allowance of the glucose to pass to that membrane but it retains proteins and other constituents of the blood.

So it is a typical filtration process which allows glucose to pass through that membrane, so this is therefore is the outer layer the polycarbonate film layer is there which allowing glucose to pass and in the middle layer we will trap the enzyme and the enzyme is immobilised because it is (())(13:47) it is trapped that is why you get you write it as the immobilised enzyme so there is no mobilization of the enzyme part because enzyme is the most costliest part only for several cycles only say 500 cycles or 100 cycles is a biologically or originating catalyst we all know.

So this enzyme should be immobilised on the electrodes surface trapping for the middle layer and the enzyme is nothing but glucose oxidise is now what is therefore we are talking about the enzyme what we are bringing is looking for some oxidation reaction in terms of oxidase that means oxidase. Oxygen will be one more extra oxygen from the air will be inserted in the glucose molecule by these glucose oxidase itself.

So glucose oxidase reaction will take place, so if electrodes is there so who will sense corresponding electron transfer or the current for this particular reaction, so who will develop that particular current because we have seen that the formation of water molecules from 2 can be detected by electron or the sensor.

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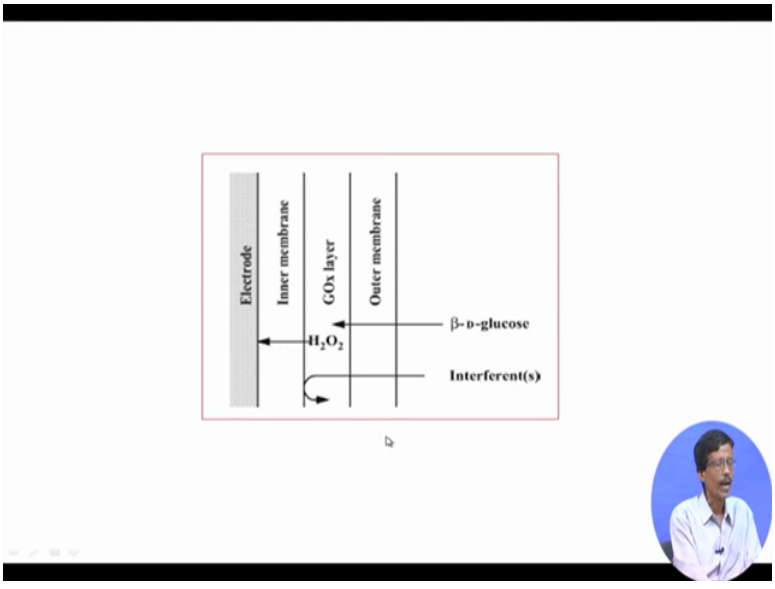

Now you get one after another everything that means the arrangement. So this arrangement is basically giving us all these things in terms of the electrode you have, then you have the inner

membrane, that inner membrane as we discussed earlier that you have a thin film of electrolyte so it is basically is no direct contact with that electron. So is a very thin film of electrolyte you can have then the middle layer and then the outer layer.

So this outer membrane which is made up of polycarbonate membrane, so the outer membrane is basically you have so this is the whole thing is your outer membrane is rejecting the interference that the protein part or the other species what can be present in the blood is basically the arrowhead is in the opposite direction that means it is basically rejecting, so when the beta D glucose, so one form basically what we basically find so this particular glucose that means we consider that a glucose can also be attached to your hemoglobin molecules or the myoglobin molecules that we consider as the glycated hemoglobin.

So hemoglobin percentage we determine their also how glucose is attached to the hemoglobin molecules also, so this is that beta D glucose molecule which is present in the blood, while is there in the blood definitely so that is why this it is attached to the hemoglobin. So this particular glucose then since it is going to semi-permeable outer membrane is passing through this and your interference are getting rejected, we do not allow the interference because otherwise everything will be hotchpotch.

So you reach their GOX layer, GOX is nothing but your glucose oxidase, enzyme is your glucose oxidase so enzyme is being trapped there, so enzyme cannot move away, so this can be considered as a typical enzyme electrode, so enzyme electrode will be responsible or showing your measurements or glucose which we can consider as a subject under your bio electrochemical measurements.

So where is passing through your GOX layer and is not directly reaching your electrodes, so nothing is reaching to your electrode that beta D glucose is not reaching your electrodes, it is reaching your enzyme area or enzyme layer or enzyme concentration or enzyme molecule but we are writing something that means you see now your O2 that means what we are talking in our previous class that O2 can be reduced by electron reduction to hydrogen peroxide and just now also in our previous as we have seen the ad rotating ring disk electrodes.

So rotating ring disk electrodes and have some sense for formation of your water molecule from O2 or formation of hydrogen peroxide from your O2, so depending upon the nature of this reduction reaction that we should exploit nicely to understand the path what is getting over there, so when you have this that means the enzyme what we are talking about.

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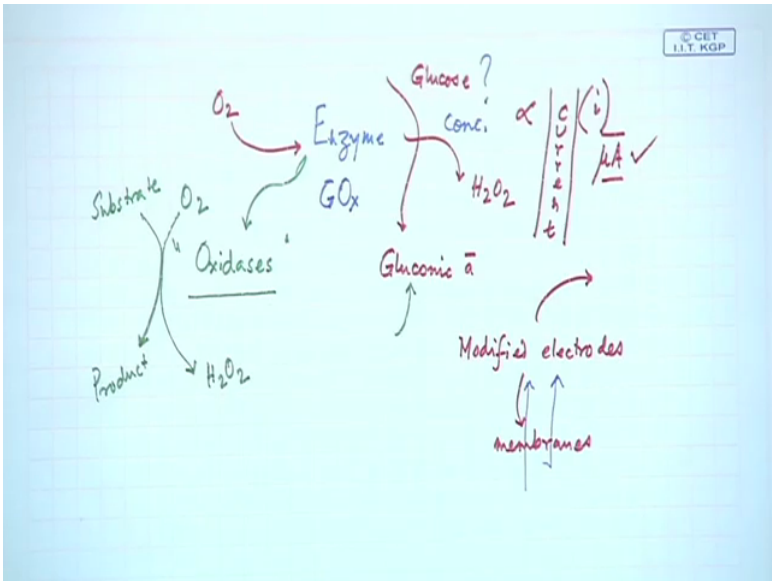

So enzyme is your GOX glucose oxidise enzyme, so this enzyme GOX is there and what we are getting we are looking for your O2 and we are going for this, so enzyme GOX is there so O2 is therefore so you have the glucose molecule forming to gluconic acid, so what you get therefore that the gluconic acid is forming okay fine that is the fate of this oxygen, so one of these oxygen can be inserted for your gluconic acid and the other oxygen molecule can give rise to the formation of H2O2.

So if the same amount of H2O2 is formed together with the formation of the gluconic acid, there is no need to detect this oxidised form of glucose that means the gluconic acid, you can only detect this presence of this concentration of this H2O2 as we know that this is also electroactive that hydrogen peroxide you can also detect by the electrodes for its concentration.

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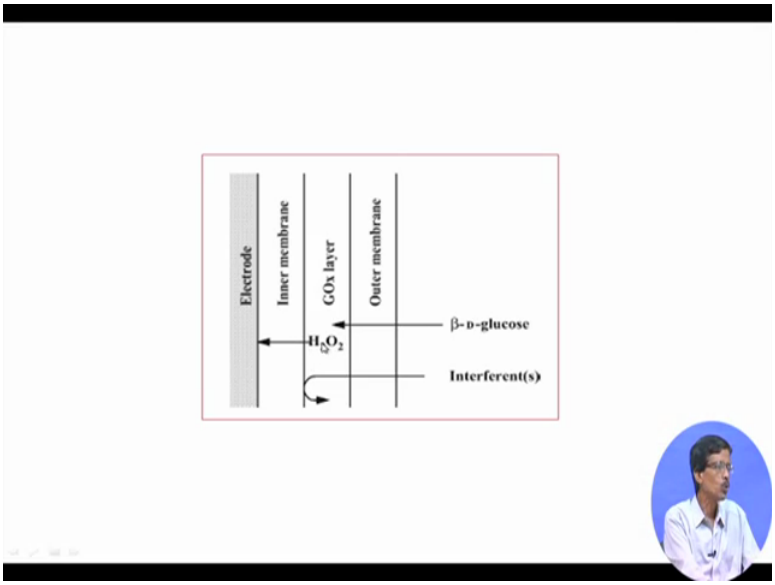

So this H2O2 will now pass through your inner membrane then again some selectivity comes into the picture that your inner membrane and only allow your H2O2 to pass through to reach the electrodes and electrodes now will work on your H2O2. So your chemical reactions in terms of the electron transfer that means electrodes are now doing your chemical reaction or the chemical transformations.

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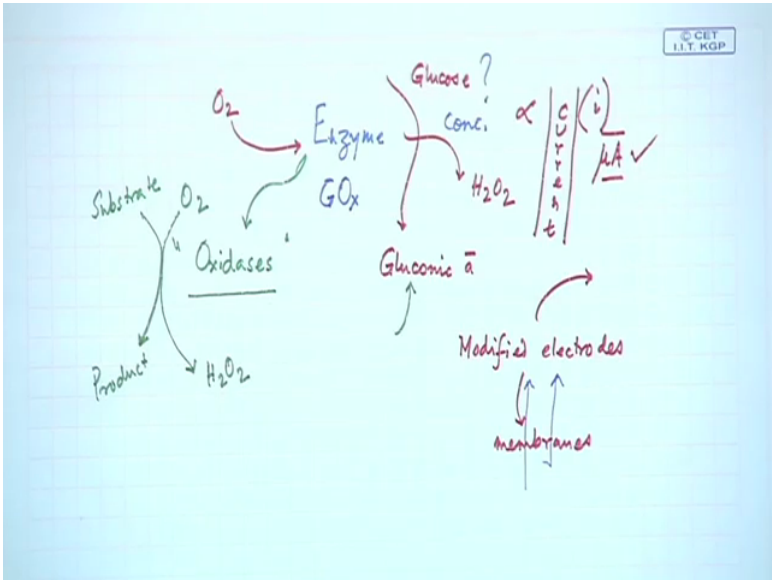

At the same time your enzymes is doing its enzymatic function working on your glucose molecules, so large number of species or substrates you can think of in a fashion. Similar to this that if you for something some oxidise reaction, so your general terminology or this is that we can study basically several oxidases this enzymes these are your oxidases, so is a

oxygenation reaction that means oxygen insertion reaction so oxygenation reaction on the gluconic acid.

So these oxidases are the so oxygenation reaction is the direct insertion of the oxygen on these oxidases, so large number of oxidases you can study where you have the substrates like this glucose to get the product and indirectly what we will be looking for, the fate of your O2, this O2 is giving your H2O2. So during that particular process that how much H2O2 is form because everything is selective, no other reduction process is taking place on your O2 molecule and this O2 molecule is giving only your hydrogen peroxide, so we will measure hydrogen peroxide in this specific situation.

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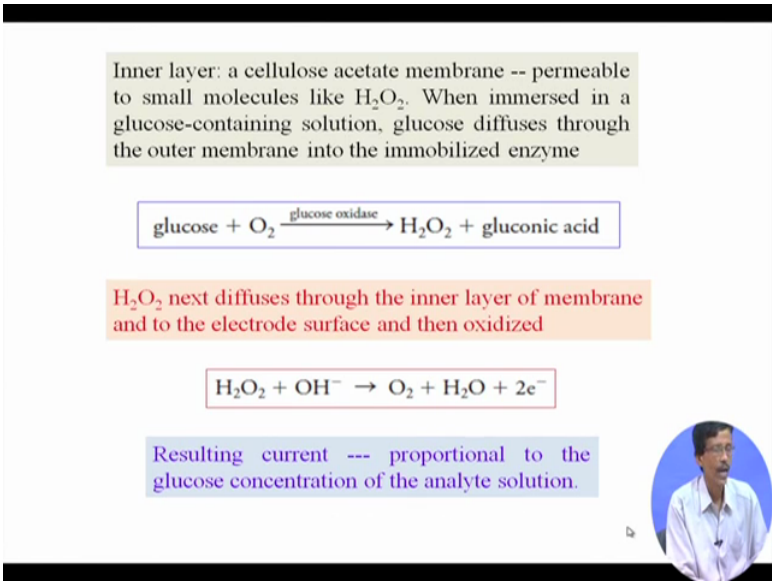

so this reaction we should see now, so the inner layer the last layer as we have seen that it can be a simple electrolyte layer part is made up of something some polymeric thing but is also a paper like thing is a cellulose acetate membrane. The papers we all know that is cellulose polymeric cellulose, so cellulose acetate is also very fine film which is permeable to small molecules like H2O2.

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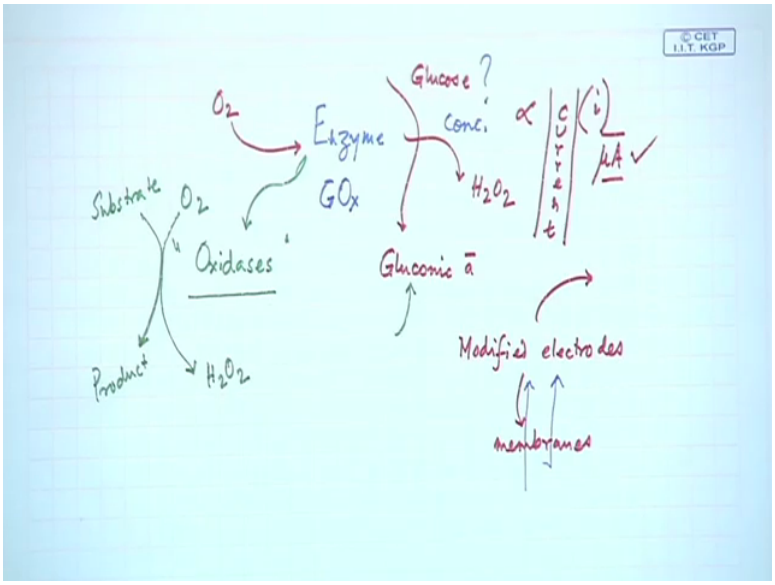

So everywhere the modified electrodes are there, so what we see that modified electrodes, it is the cover-up of the electrode but we are using some membranes and these membranes we are talking about their permeability, their type of permeability. So the type of permeability such that some molecules can pass through the membrane and reach the modified electron, so when you have the cellulose acetate membrane and we know is selective for your hydrogen peroxide, it allows hydrogen peroxide to pass in.

When immersed in a glucose containing solution, glucose diffusers through the outer membrane into the immobilised enzyme giving this reaction, so glucose plus O2 is giving rise to H2O2 and gluconic acid with respect to your GOX the glucose oxidase enzyme, so glucose oxidase enzyme is working to convert glucose to gluconic acid and hydrogen peroxide, then this hydrogen peroxide is next diffusers through the inner layer of membrane and the inner layer of membrane is goes that the inner layer is the cellulose acetate membrane which allow your H2O2 to the electrodes surface and then your H2O2 this oxidised.

So the current as well as the required potential for the oxidation of hydrogen peroxide will be judged now and you will be oxidising your hydrogen peroxide back to O2. That means it is not a any kind of other disproportionation reaction or any kind of side reaction. Then hydrogen peroxide can also give rise to water and hydroxide also, hydroxide ion but this basically go for a oxidation reaction in presence of some OH minus if the medium is not alkaline, it gets from the Aqua centers at means the water molecules to give back your O2, water and to electron.

So you get this so that in one way is basically a sensing mechanism where we talk in terms of the corresponding O2, so you will now co-relate these two sensors what we are talking from our previous class that in one case we are we were having the O2 sensors that oxygen electrodes. Now the product is your oxygen now so this basically the previous one is oxygen sensor or oxygen sensing electrode use and this is a sensing mechanism or the sensor for hydrogen peroxide.

So this can also be used for any hundred reactions enzymatic uhh hundred reactions where hydrogen peroxide is produced so do not try to learn all these in a fashion that you will be learning only glucose but it is not that, you have to apply this if you know the basic understanding that what is forming and how we are detecting electrochemically is the formation of hydrogen peroxide.

So if in the nature in the laboratory in the industry or in any other activity, if hydrogen peroxide is produced in the medium you should be able to detect that and you can follow up why this hydrogen peroxide is forming even for in water also something that somebody can tell you that due to water pollution we are getting some other obnoxious things and together with that we are getting some amount of hydrogen peroxide.

So what thing you should do first okay see let us find out the hydrogen peroxide concentration over there then only you can try to find that which is responsible for the generation of production of hydrogen peroxide in that water medium, so you have to sense that so we are bringing so many things that means enzyme we are bringing, we are bringing membranes the different types of membrane that is the part of the fabrication of the electrodes.

So for fabrication of this electrodes or making all these things are very much related to the corresponding formation of this hydrogen peroxide detection, so it is sensor which is giving you the hydrogen peroxide, so what you sense then? Since the electrons are coming out from the hydrogen peroxide or giving you the oxygen and water molecule and 2 electrons so you will measure the current.

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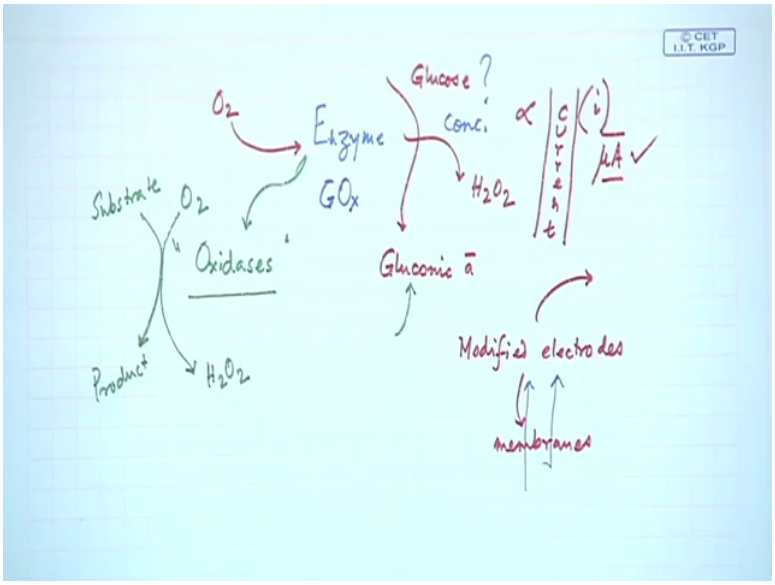

So you measure the current so the resulting current which will be proportional the glucose concentration of the analyte solution, so what we are going for that, we were looking for the unknown concentration of this glucose, so what should be your concentration and any substrate suppose an industry is making your Glucon d for us, so if they also want to measure because you require a very small sample the amount of sample because our next classes we will discuss that how you make or how you get the real samples and how you prepare those real samples for analysis.

So this concentration is very less and there also you can determine that (())(27:23) what this glucose so this concentration is proportional to your current, should remember it is the current this i value. So this is the thing that when you talk in terms of the electrochemistry always remember that either potential or the current now is a very simple way that you determine the corresponding current.

The amount of current will be proportional to this and as I told you earlier that whether you have this particular mechanism or any kind of amperometric measurement, you can go for a nano MPL level of current. So whatever small concentration of glucose can have there is no requirement for your enrichment of the analyte concentration for glucose, you can directly use the blood sample for that.

So for the typical activity for the last 30 or 40 years industry people are making this glucose sensor very nicely uhh having some disposable electrodes definitely, for some measurements the electrodes is working otherwise you have to discarded. So this nano MPL level very low

level of concentration of glucose can be determine because you can detect nicely you can amplify the current so the resulting current will be very much proportional to that of your glucose solution, okay. Thank you very much.