Cell Culture Technologies
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Lecture - 17
Surface Chemical Analysis

Welcome back to the lecture series in Cell Culture Technology. So, just start off with.

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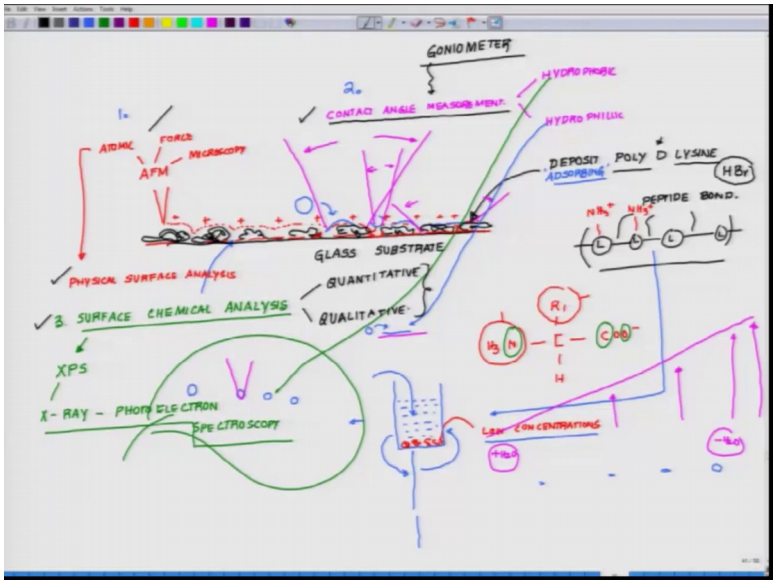

Now, doing a surface analysis and one of the easiest and most thorough surface analysis what you can do is X P S, x p s stands for x ray photo electron spectroscopy. So, this is one form of a spectroscopy where what you do the emerging beam. So, you try to understand in simple language you shine an radiant energy on the substrate and it has its certain level on you do not go very high on it ok.

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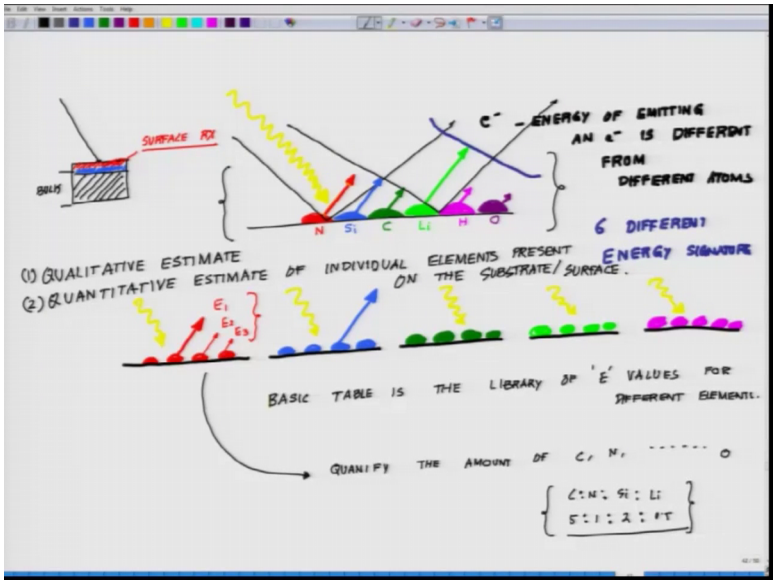

Now, what this does this ejects out it say for example, let us let me draw it the remain more sense say for example, you have a substrate like this and this substrate has these different colour of elements which are present red say, blue, green, light green, pink like this, just to distinguish a kind of making its slightly darker. So, this is how the surfaces mix up, now essentially speaking you have 6 different colours or 6 different atoms or elements which are present on the substrate. Now let us give them some different, different names say I say this blue stands for silicon, dark green stands for carbon, red stands for say nitrogen, light green this say stands for, so the surface has lithium I am just for your understanding sake I am telling you and lighter pink which may stands for say hydrogen and this one stands oxygen.

now you shine emerging beam out here, do not worry about it what you are shining at this point what this will do is on the surface whatever is present it will eject out the outermost electrons and each one of those outermost electrons which are being ejected out. So, I am representing them by the respective colors of that atom or of that element and now each one of these electrons which are ejected out from the surface are coming with a different kinetic energy, the energy of emitting an electron is different from different atom, where the different atoms are of the different colours.

Now, if you have a detector sitting here which could detect those speeds and it will be giving you say for example, in that case 1 2 3 4 5 6, 6 different signatures and if you have already done your 6 different energy signature and if you have already done your

base calculations right say for example, you already have a library how you create the library. So, you do the same experiment with another situation.

Say for example, you have a, so these are pure substrates what you are talking about and exactly do the same experiment you shine each one of them with particular radiation. So, each one of them will be emitting it with that unique velocity. So, you have already the table at your disposal where you know the basic table is the library of e values energy values for different elements.

If you know that value and what you can do is you can back calculate from this next substrate and you can figure out what all elements are present on that particular substrate right. So, this is how you analyse a substrate that what all different elements which are presenting on a substrate and this is one powerful tool which help chemist, biochemist, material scientist everybody to understand the surface chemistry.

Especially, people who does work on electrochemistry in all other fields analysis of electrodes because, you have to because anything any phenomena which is a surface phenomena. So, you have to realize what I meant by surface phenomena. So, when we talk about a substrate here is a substrate and cells are attaching on the substrate we are talking about some form of surface interaction. So, one needs to analyze this surface interactions very carefully and for surface analysis one of the most powerful tool.

So, what you see is a basically a not a bulk property of the material, but a surface interaction between cell and surface it could be any surface and the interactions are shown here in the red colour . So, now, unless otherwise you know the surface chemistry of any material it is extremely tough for you to quantify that why this interaction is happening.

Who is bonding with what to that level the technique is. So, powerful within this particular say I talk about the red here, talk about nitrogen the speed with which the different electrons from nitrogen are ejected from which server orbital it is coming from most of the outer orbitals. Of course, you can even go down and figure that out it is that powerful a technique, but remember, remember it is a surface phenomenon it is not going to tell you anything about the bulk material and we are not concerned about the bulk and when you talk about the bulk, I talk about suppose this is the material. I am talking about

I am not bothered about the whole bulk property of it this is what I am hatching the lines this is the whole bulk of that material ok.

I am only bothered about just what is happening on the surface the surface reaction and of course, there is always a very interesting interface. So, we are not talking about it. So, any surface phenomena needs to be analyzed very analytically about the presence of different kind of elements which are there because, unless otherwise you know this say for example, thing of this suppose your surface has higher carbon.

So, now having said this I will take you to the next flayer in to the gain using this technique you can even quantify its little tricky, its little tricky to do so, but you can actually quantify the amount of carbon nitrogen and all other xyz elements are there and if you can quantify. So, you can develop a ratio say for example, I say c is to n is to say silicon is to say lithium is just put some number say, say 5 is to 1 is to you know 2 is to 0.5 or one whatever you know. So, in that situation you know that. So, this is the composition I am getting whenever when I am coating the substrate with this is specific material right.

So, that way you not only have a qualitative estimate you also have a quantitative estimate of individual elements present on the substrate or surface. So, this is a powerful tool which generally not used by the biologist very frequently, but those who are intensively into cell culture they may like to look at this technique. Apart from it when we talked about in the analysis we talked about contact angle measurements, this contact angle measurements could be done with the instrument called goniometer and of course, atomic force microscopy you need an afm set up if you do the surface analysis.

So, now if you summarize you have a physical surface analysis in the form of afm you have a contact angle measurements which will tell you the interactive behavior of the material in the presence of water or aqueous system and you have a surface analysis both quantitative and qualitative. So, once you analyse the surface or you know the tools to analyse a surface you know how much power you have now really to approach to a problem.

So, now I give you any material irrespective of whether you know the nature or not I give me give me a random stuff, I say you know this is going to work how you should scientifically proceed in the experimental science wise you first of all do different dose

dependency of dissolving if of course, the assumption here is, if it is a water soluble if it is water insoluble of course, you can do you just have to know how to you know while you have to dissolve an you know put it on the surface. It would if it does not dissolve in a aqueous it will dissolve in a its non aqueous solvence, then you can measure (Refer Time: 16:23) also of course, that is another applications is not to a cell culture application may be something else, but the see the point is we are not bothered about where are you are going to apply because you will be going to going places and doing different kind of scientific endeavor.

What is important is that you should have an idea that what all tools are there in my armory, which I can use to analyze surfaces. So, definitely these 3 are some of the very important tools. So, you code the subsurface see you code at different concentrations, suppose I take concentration 1, concentration 2, concentration 3, concentration 4, in our case say (Refer Time: 17:12) we take 4 different concentration since this is aqueous or dissolved in water it is easy for us to do it.

Now, I take afm. So, I have 4 different afm images, and then on top of that I do a goniometer analysis or contact angle analysis. So, now, I know now what is the difference, if at all so, my hypothesis is that whether with the increase in concentration the hydrophobicity or hydrophobicity will change. The alternate hypothesis could be it will not change right you can test your hypothesis by simple experiment you can say if I deposit say this milligram per ml on the surface this is what I am going to see or if I have y milligram per ml on the surface this is what I am going to see, if I have a z milligram per ml this is what I am going to see. So, with different dosage you can see different kind of interactions which are happening. So, that followed by this you can go for what we call as surface chemical analysis, where you will be knowing that which all atoms are or elements of atom. So, atoms of element are present on the surface. So, that will give you an idea that these are the possible interactions which can happen with the cell.

So, now here after what will be, what all will be discussing about the different kind of extracellular matrix what will be using or what people use, I wish all of you should be or at least I request all of you should be aware about the tools at your disposal what all you can use to analyse surfaces with few slight changes we will continue from here ok.

Thank you.