Introduction to Complex Biological Systems
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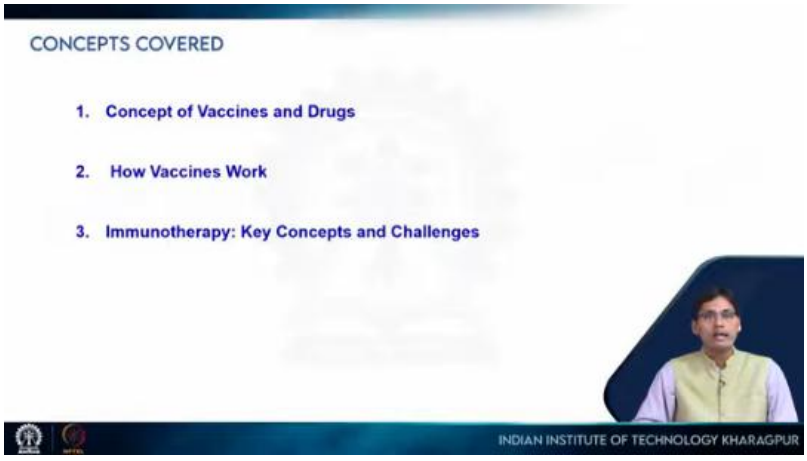
Lecture 50
Vaccines and immunotherapy

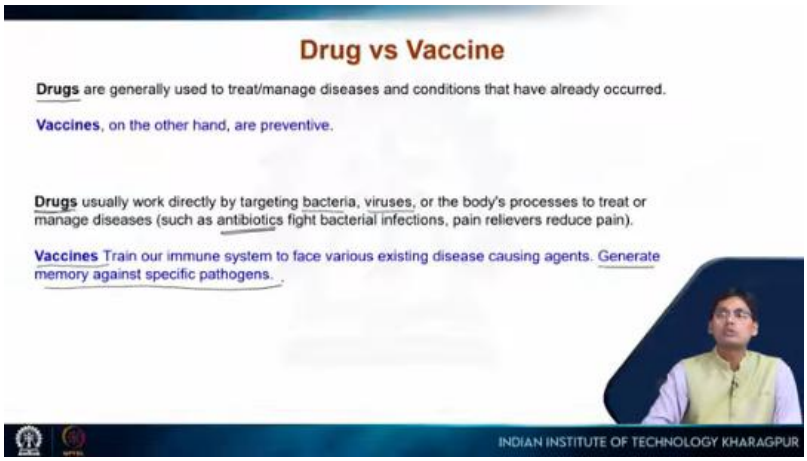
Hello everyone, today I am going to discuss vaccines and immunotherapy. This is the last lecture of Module 10. In this lecture, I will mostly cover vaccines and how vaccines work, particularly different types of immunotherapeutic strategies, mostly key concepts and the challenges. So, here, first, I would like to introduce vaccines, particularly the difference between vaccines and drugs.



So, if you see whatever drugs or medicine we generally take. Those are used to treat or manage some kind of disease, but the disease has already happened. So, to take care of that particular disease, we take drugs. But on the other hand, vaccines are preventive, and more importantly, drugs usually work directly by targeting some kind of pathogen. It can be bacteria, it can be a virus, or they can target some kind of body process to treat or manage diseases.

So, if I give you some examples of antibiotics, for example, tetracycline, streptomycin, doxycycline. Those antibiotics are used against bacterial infections. Similarly, different types of pain relievers can be used to reduce pain. So, all these are examples of drugs. On the other hand, vaccines train our immune system to face various existing disease-causing events. So, sometimes I discuss the huge success of vaccination against the smallpox

virus. So, in this case, what happened in the case of vaccination is that it generates memory against a specific pathogen. So that if the same pathogen again infects us, we will not have the disease. We will not show the symptoms because our immune system already knows how to tackle that particular type of pathogen. So this is a vaccine. So now I will go into how this vaccine works. So, before that let me explain in case of natural infection what happens.

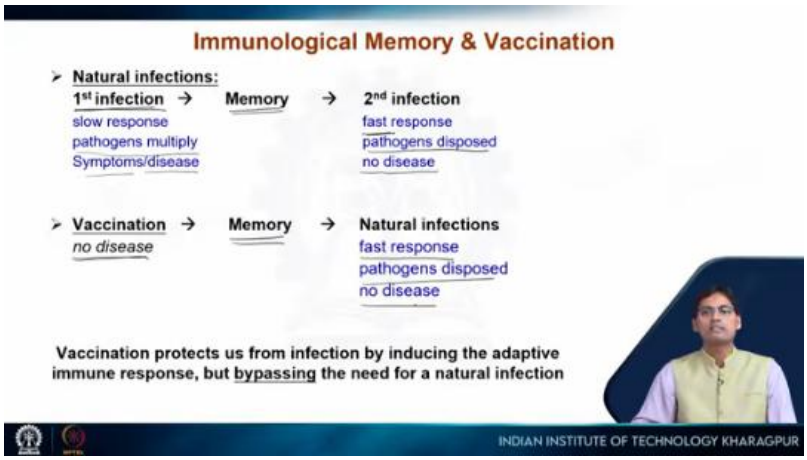


So, in the case of natural infection, the first time we are exposed to some kind of pathogen, some kind of virus or bacteria. What will happen? Our immune system will try to protect us, but that immune response will be slow and pathogens will multiply. Finally, we will get some kind of symptoms, and we said this is the disease. Some kind of deviation from the normal physiology and how many symptoms will be there, how much disease will be there that is depending on the pathogen itself and also our host physiology.

But, the thing is, everything is slow during the first infection, and finally, after some suffering and after some symptoms, we will have the memory, memory means here immunological memory. Again, if the same pathogen infects us, say 6 months later, or sometime it could be 6 years later, but during that time our immune response will be very fast. Pathogens will be disposed of soon, and there will be no symptoms or disease. That is why we will not be able to understand that we are infected by that pathogen. So this is how our immune system always tries to protect us from the same infection. But the same

thing can be happening here by vaccination. But in case of vaccination, we use some kind of weak or attenuated strain of that particular pathogen.

It can be a very weak pathogen or it can be some parts of the pathogen. So, as a result of that, it will not multiply inside the host. So, there will be no disease. But whatever we are administering in the host body, it will teach the immune system to make memory. So, as a result of that, immunological memory will be there, and in the future, when that particular person is naturally infected by the same pathogen, during that time, our immune system will work very fast. The pathogen will be disposed of, and no disease will occur just like whatever I discussed in the top panel. But here, what is happening is that when we are providing a vaccine, the disease is not appearing at all. So, as a result of that, I would like to summarize that vaccination protects us from infection by inducing the adaptive immune response but bypassing the need for a natural infection. So, in the top panel, I mentioned that because of natural infection, our immune system learns how to tackle this pathogen and keeps some memory, particularly memory B cells. But in the case of vaccination, all those things are happening, but we are not getting the disease. Now, vaccination, I would say, is a huge success if you think about it that way.

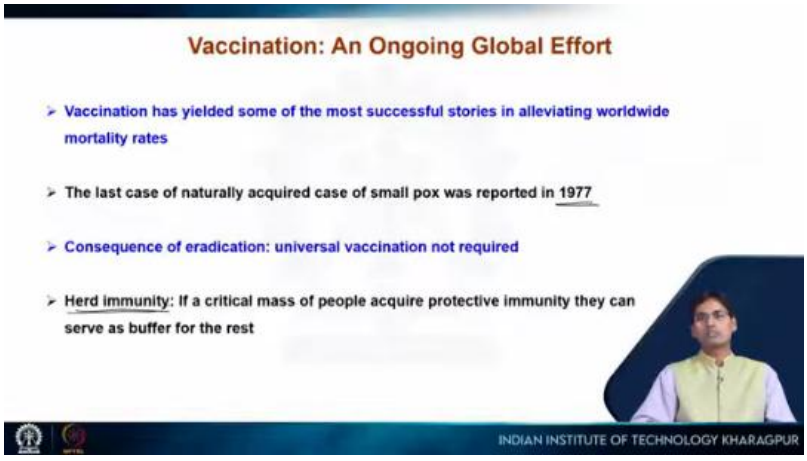


So, it has yielded some of the most successful stories in taking care of worldwide mortality rates. As I already mentioned, in the case of smallpox, vaccination has already eradicated it. Similarly, if you see vaccinations against polio throughout the world, particularly in India, it is a huge success. Now, there is no polio. So, polio vaccination is also a huge success.

So, as I just mentioned, smallpox, the natural occurrence, was last reported in 1977. So, it has completely been eradicated. So what I would like to tell you is that during the last module, when I was discussing microbes, I mentioned that antibiotics are the greatest invention in terms of medicine, as they protect us from different types of pathogens. Similarly, vaccination is another one. So, I would say antibiotics and vaccines are kind of major pillars in biomedical science and we get a lot of benefits out of them.

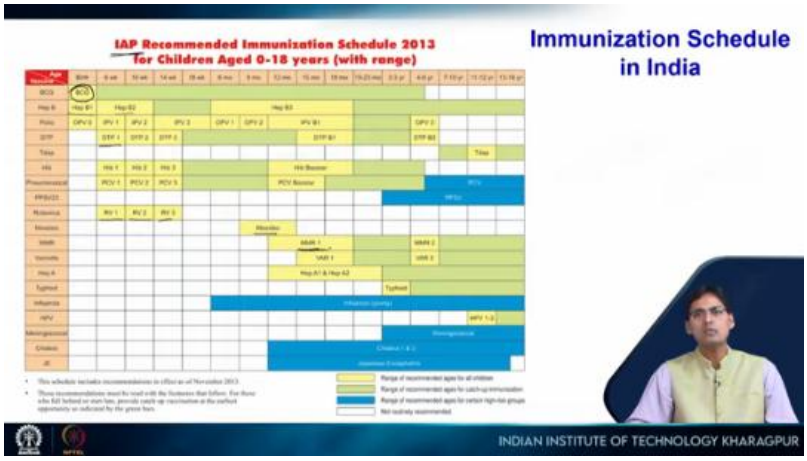
Now, what is the consequence of eradication of that particular pathogen? I would say then that universal vaccination is not required because it has already been eradicated. So, there is no requirement of vaccination at all. For example, for smallpox, no vaccination is required, and this is the last point of this slide: the herd immunity. What is herd immunity?

So, the idea here is that if in a population the majority of the members are protected from a particular pathogen, then it is very unlikely that other people will also get that infectious disease. That infectious agent cannot be spread because most of the people are already protected from that particular pathogen. So this is called herd immunity, which means if a critical mass of people acquires protective immunity, they can serve as a buffer for the rest. So, as a result, most of the time the other will not get the disease. So, this is called herd immunity. You do not need to go through the details of this slide; I am just trying to show you that different countries have different types of recommendations for immunization.

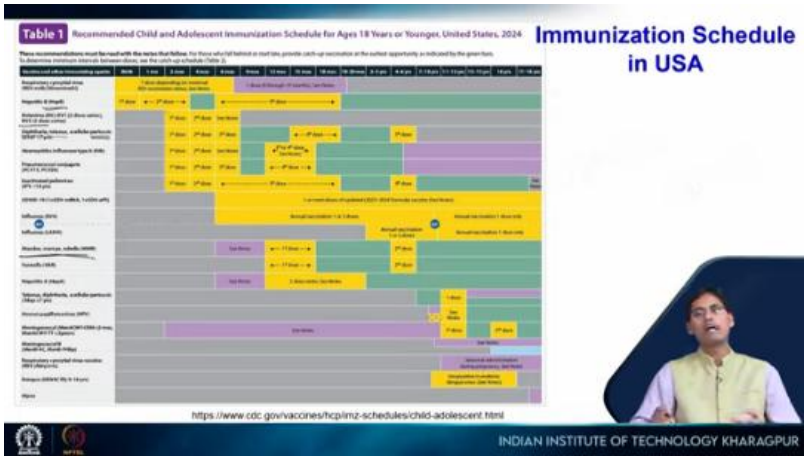


This is particularly, I would say, the child to adolescent stage, 0 to 18 years, and this is, I would say, recommended by the Indian Academy of Pediatrics. So, they have recommended this immunization schedule. And if you see, some of them, I would like to mention, for example, in India, BCG is a must, so BCG is provided at the very beginning, just after birth, and then you can see the hepatitis vaccine, and then DTaP, which is also very common, diphtheria, tetanus, pertussis, and rotavirus, for example. Then, measles virus; mumps, measles, and rubella. So, these are some kind of vaccines against viruses. We have some recommendations in our country, and all kids, after birth, take all those vaccinations on a routine basis.

So, this is a great benefit for their health also. Similarly, I am just showing here in the United States what their recommendation is based on the CDC. So, they also have their own recommendations, but if you see, most of them are very similar, almost overlapping with each other in the vaccines they are recommending. So, for example, here, the BCG is not mandatory, as I mentioned in the previous slide in the context of India. But here, if you see, the Hep B, hepatitis B virus vaccine; then rotavirus and DTaP, diphtheria, tetanus, and pertussis. Those are common, the same thing.



Similarly, in this list, I am sure you will find mumps, measles, rubella, that should also be there, like the MMR vaccine. So, as a result, now we have a very robust schedule for immunization, particularly for kids. Now, I will start discussing immunotherapy. This is one of the important aspects of biomedical science that is coming up and a lot of research is going on in immunotherapy. So, what is immunotherapy?



So, this is a treatment strategy that uses the body's immune system So, I would say in the case of vaccination, they are trying to generate some memory in our immune system. But the difference is the vaccine is preventive, but here, because of the disease, we are trying to manage that disease by this immunotherapeutic approach. So as a result of that, a treatment strategy that uses the body's immune system to fight diseases and it can be different types of diseases, but especially against cancer.

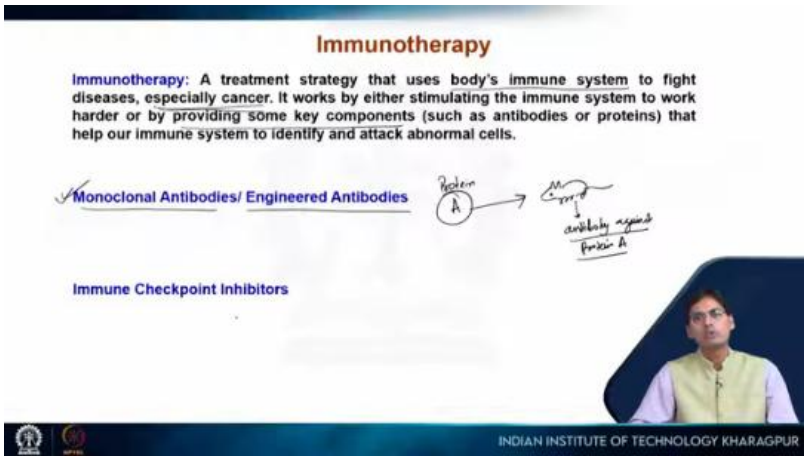
It works by stimulating the immune system to work harder. It can also dampen the immune response. Sometimes it is also required. This is not always true that it stimulates the immune system to work harder. What kind of immunotherapeutic reagent we are talking about and what kind of disease we are managing. Depending on that, we have to think. I will explain both of those things. Either by this way or by providing some key components, such as antibodies or some proteins, that help our immune system to identify and attack abnormal cells. So, therefore, we can try to manage that particular type of disease. There are different approaches and strategies. I will mostly discuss two important strategies since I discussed antibodies during the last lecture.

So, it will be very easy for me to explain this. So, one important thing is monoclonal antibodies and engineered antibodies. So, what are monoclonal antibodies? See for therapeutic purpose Generally, when we try to raise antibody, we try to get antibody. How do we do that? Generally, we use some animal models. It can be a mouse; it can be a rabbit or some other animal model. So I want to raise an antibody against Protein A,

this is my protein Protein A, so what I need to do is I have to inject this protein to some mouse, for example.

So, now there is a detailed protocol on how to do this and after some time you can get some antibodies against Protein A from this mouse blood. But the thing is this antibody is not a monoclonal antibody. Because, when you are actually injecting Protein A into this mouse, the mouse immune system will work on that, and this Protein A might be a very big protein, and several peptide antigens can be presented, and as a result of that, it will be a mixed population of antibody. So we will not say that this is a monoclonal antibody. Monoclonal means it is exactly the same antibody that we are getting, and as a result, their specificity will be exactly the same.

So, that is called monoclonal antibody and also engineered antibody. I will discuss. Monoclonal antibodies can also be engineered and I will explain soon. Immune checkpoint inhibitor. This is again another very important strategy that we have different checkpoints when our immune system is getting activated. So, we can target those checkpoints also to modulate our immune response. Here, particularly I would like to focus on monoclonal engineered antibodies to decrease immunogenicity but maintain their antigenic specificity.



So, now the monoclonal antibody I already mentioned. So, for example, if you see this is fully mouse antibody that means that antibody raised in the mouse. The antibody can be against some human protein, but the antibody we are saying is fully mouse, meaning it is completely raised in the mouse, and I am saying that it is a fully mouse antibody. There

are some conventions that if it is a mouse antibody, the antibody name ends with this letter, and if it is chimeric, I will explain what chimeric is, and it will end up with these letters, and similarly, humanized with this letter is not that relevant in our discussion, but some conventions in nomenclature. Now I will discuss, and give an example of each of these.

Now the thing is why we are trying to reduce the immunogenicity, because when we are getting the antibody from a mouse, then if this antibody is injected to humans, it might show some immunogenic reaction against that antibody because it is after all a foreign protein. So, that is why we are trying to make their immunogenicity as low as we are trying to reduce. So, the first one is mouse antibody. One example is Muromomab. So, as I mentioned, this part you can see is the same as this.

So what does it do? So, this is an Anti-CD3 antibody. It is a monoclonal antibody, but an Anti-CD3 antibody. So, CD3 is a marker of T cells. So, it is present on the surface of the T cell.

So this antibody inhibits the T cell activation. So, if it inhibits T cell activation, then it will dampen the immune response. Somehow, it will be a little bit immune suppressive. So, that is why this kind of antibody can be used during organ transplantation, particularly before it was used for kidney transplantation so that during transplantation organ rejection will not happen if the immune system is a little bit in its suppressive condition.

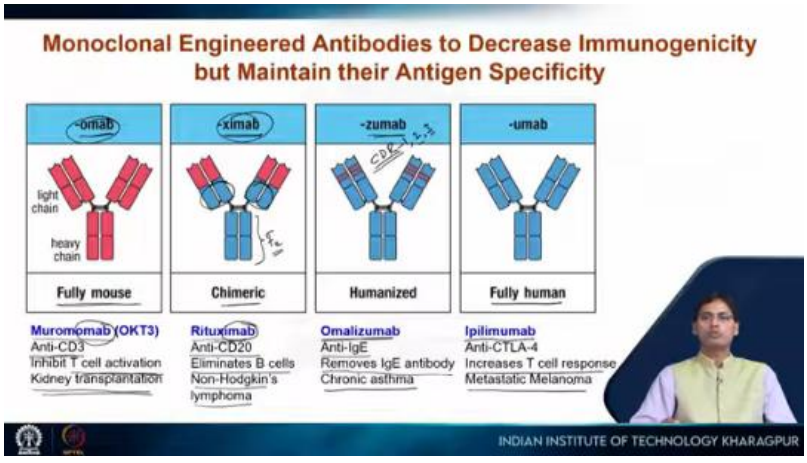
The next one is a chimeric antibody; if you see here, we change some portion of this antibody by a completely human sequence. For example, this whole Fc region, fragment crystallizable region and also, the constant region here of this variable heavy chain as well as light chain; all these portions are from a human sequence. So, only the variable domains of heavy chain and light chain are containing mouse sequences. As a result, it will show less; I would say that immunogenicity will be decreased.

So, it will be a much better reagent compared to the fully mouse antibody. So, now, if I give you one example, this is rituximab. So, again if you see this is matching here and one example already I mentioned. So, this is against Anti-CD20 and it eliminates B cells

and this was used in Non-Hodgkin's lymphoma to treat this kind of disease. Then scientists are trying to make it even better so that it is called a humanized monoclonal antibody where you can see everything replaced by human sequence only that CDR region, the complementarity-determining region. I told you about CDR 1, 2 and 3 here. So, just keeping those things then other portions of human sequence, one example here this one. This is against Anti-IgE.

So, IgE is related to some allergic reactions also. It has some correlation and this antibody removes IgE antibody and as a result this is used against chronic asthma and the last one. This is very important for a fully human antibody, the monoclonal antibody. One example is ipilimumab. It is kind of a tongue twister.

This is against CTLA-4. CTLA-4 is an inhibitory receptor present on T cells. I will be discussing a little bit in detail soon. It increases the T cell response and it is being used now against metastatic melanoma. So, this is some approach, some monoclonal antibodies and particularly engineered antibodies for the therapeutic purpose, either we used before or currently in use. Now I would like to discuss a little bit more engineered version of antibody and one of that example is bi-specific antibody. What is bi-specific antibody?



I mentioned before that, in the case of an antibody, if I say this is one heavy chain and the next heavy chain, two heavy chains and two light chains, this is some antibody. Here I mentioned that it will capture one antigen similarly in this region, but for an antibody, this antigen will be the same because these two arms are identical, so the same antigen

should be there. Now the bispecific antibody means it will recognize two different antigens. This is some kind of engineered antibody. Let us see how we can make that.

So, in order to do that, I am just giving you one example: catumaxomab. This is some kind of bi-specific antibody. It was used particularly in some European countries against some cancers. So, I would like to discuss what is happening here. So, this antibody is made up in this way. So, if you see, as I already mentioned about the antibody. So, this is one antibody, and this antibody recognizes some antigen which is coming from EpCAM. The full form is an epithelial cell adhesion molecule. So, this is some protein, and this antibody is against that. So, now if you see, another antibody I am talking about here.

So, this antibody is against CD3. As I already mentioned it is present on T cells. So, it recognizes CD3. So, now we designed a single antibody where one arm is something like this, that is holding EpCAM and the other half is coming from here. So, one half from here and the other half from here. As a result of this one I am changing the shape of this antigen so that it will be easy to understand. So, this is the case here.

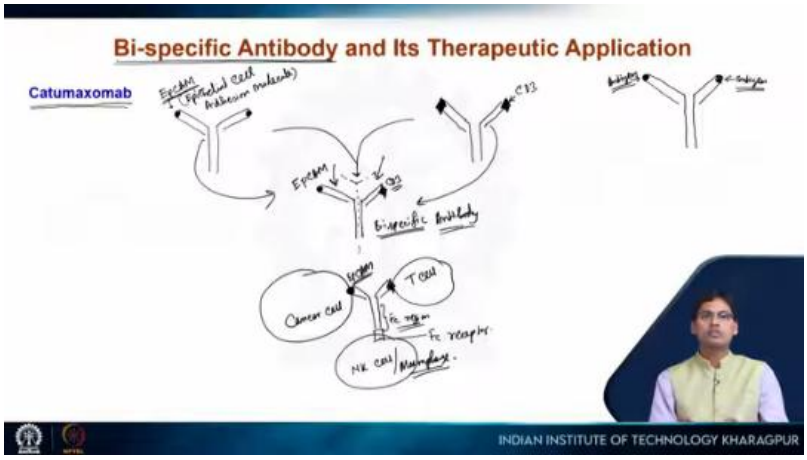
So this is coming from here. So, this is holding this is against CD3. So, this antibody I would say this is bi-specific antibody. So, what are the applications of bi-specific antibody? So, it will be very useful in such a way that I would like to explain.

For example, if you use this antibody. So, first I would like to mention here: if you see this left part and right part are different here. This is against CD3 and this part is against this epithelial cell adhesion molecule. So, now if you apply this antibody what will happen? So, I would say if you have this. This is cancer cell cancer cell why? Because this epithelial cell erosion molecule is generally overexpressed and expressed in some cancer conditions but not in normal cells. So, we have to design in that way. So, this is present in some cancer cells. So, I would say if this is the EpCAM protein. So epithelial cell adhesion molecule here.

Similarly, what will happen? The antibody will bind here and the same antibody since it is bi-specific antibody Here it will bind to CD3 and CD3 is present where? It is present in the T cell. So, therefore, this antibody is driving the T cell to come close to this cancer cell.

So, these two cells are very close to each other. Then what will happen? The T cell will exert its effects. So, it will try to kill the cancer cell. So, this is the strategy.

On top of that, what is going to happen, as you know, is that in the Fc region, the Fc region will bind to some Fc receptor. As a result, if this is an Fc receptor, it could be an NK cell or a macrophage. So now, I hope that you understand what I am trying to say is that this antibody is driving this T cell and NK cell or macrophage very close to this cancer cell, and therefore, it can exert its effect. So, this is how a specific antibody works. Now, I will explain another example. This is called an antibody-drug conjugate.



So, there are many antibody-drug conjugates already in use as therapeutic reagents. Here, particularly, I am mentioning this one: enfortumab vedotin. So, this is again a monoclonal antibody against this protein, Nectin-4. So, the antibody is against Nectin-4.

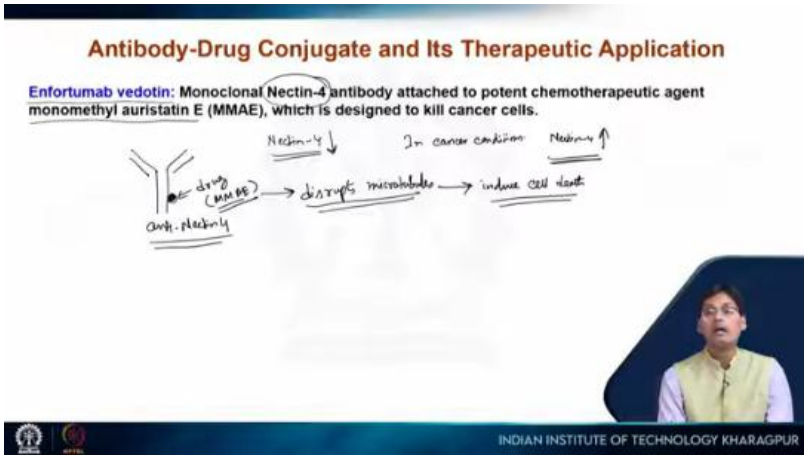
So, Nectin-4 protein is generally present during the embryonic stage, not in healthy humans I am talking about. In healthy adults, the expression of Nectin-4 is very low. In most tissues, they do not even express Nectin-4 in healthy adults. But in cancer conditions the expression of Nectin-4 increases in many different types of cancer. For example, renal cancer and different types of lung cancer, Nectin-4 expression reappears.

So what did scientists do? They developed some kind of monoclonal antibody against Nectin-4. So, I would say if this is the monoclonal antibody against Nectin-4. Now, this antibody is again attached with some drug, and the name of this drug is some kind of chemotherapeutic agent. So, particularly this is monomethyl orystatin.

So, this is MMAE in short form. Now, what is its function? So, the thing is that this antibody will bind to Nectin-4, which is expressed by some cancer cells. Now some of this antibody will go inside the cell because the cell will endocytose some of this Nectin-4 antibody. I already mentioned these antibodies attached to this drug.

So, now this drug will exert its effect. So, this drug particularly MMAE. So, it disrupts microtubules that are present in the cell, particularly in cancer cells, because they only go into cancer cells as cancer cells express Nectin-4. So, now as a result of that it will induce cell death.

So, this is some kind of strategy of conjugating a drug with a specific antibody to target cancer cells, and this is FDA approved for the treatment of locally advanced or metastatic urothelial carcinoma, the bladder cancer. I will discuss immune checkpoint inhibitors. This is the last discussion of this immunotherapeutic part. So, now here if you see, I particularly mentioned antigen presenting cell or APC present antigen through MHC molecule. So, this is our peptide antigen and TCR or T cell receptor recognizes the peptide loaded MHC molecule and then the T cell activation and all those downstream things happen.



So, I just mentioned that I did not discuss in detail what other things are happening, but now I would like to mention; otherwise, I cannot explain this. The thing is, this interaction is very much, if I give you some kind of analogy, like trying to drive a car. So, you need a specific key. So, this key is required for the ignition of that car. So, this

peptide MHC and TCR interaction is so specific that it is just like a particular key is required for that particular car.

Now, after ignition, you are just sitting there inside the car; then what will happen? The car will not move. So, in order to move the car, we have to press the gas accelerator. So, as a result, that car will move on. So, similarly here just this recognition is not enough, you need some additional signaling mechanism.

So, the first thing is that you have a receptor here, which is called CD28. Very important receptors present on T cells, particularly those like T cells and naive T cells, are just saying antigen now. So, CD28 interacts with some ligands. It is called B7-1 and B7-2. So, B7-1 and B7-2 present on the antigen presenting cell and CD28 is present on the T cell when it interacts, then it will provide some kind of signal which is called a stimulatory signal.

So, just as I mentioned the analogy that you are putting pressure on the accelerator, so that the car will move, so that the T cell will function now. Now if you are just moving your car going on and on, but sometimes after seeing a stop signal or something, you have to stop also, so you have to put the brake. So in the T cell also, that kind of mechanism is there. That is kind of homeostasis is required because if our immune system is too active that is not also good. So, we need some optimal function of the T cell. So, it cannot be like that; it is always giving the stimulated signal, and it is always in an activated state. That is not good.

So, as a result of that, at the beginning stimulation is required, and then after some time, eventually another receptor will be expressed on this T cell. So, this is called CTLA-4. It was not there at the beginning, but after some level of activation, CTLA-4 gets expressed and as a result it will also bind to the same ligand B7-1 and B7-2 just like CD28 interacts with B7-1 and B7-2. But this interaction CTLA-4 and B7 interaction is much stronger, almost 10-fold, 20-fold higher in terms of affinity compared to CD28 and B7. So, as a result, although CD28 and CTLA-4 are competing for the same ligand. but, since the affinity is more with CTLA-4, it will bind to B7 molecules and then it will provide an inhibitory signal to that T cell.

So, the T cell homeostasis will be maintained. So, it will not be overactivated. So, this is some kind of balance, and this is called T cell co-stimulation, which is very important for proper immune function. Now, after understanding that, in the biomedical field, scientists develop therapeutic reagents, particularly different types of antibodies and protein-based reagents, to tackle this system and modulate this kind of co-stimulation to address different types of diseases. I am giving you just one example.

For example, this is the FDA approved drug I mentioned during the discussion of monoclonal antibodies, this is ipilimumab. So, this is also called Yervoy, the commercial name, produced by Bristol-Myers, and it is currently being used against melanoma. Now, what is this? So, this is a monoclonal antibody that binds to CTLA-4. So, it binds to CTLA-4 and blocks this inhibitory pathway.

So, then why are we using this? That means during different types of cancer, particularly in melanoma, what we need is more activation of T cells so that they can clear the cancer cell. So we are trying to prevent this inhibitory pathway. So, we want the T cells to remain more active.

So, as I mentioned, it is already in use. So, there are many other examples, but now I will explain a little bit about some other reagents which can be immunosuppressive. Here another FDA approved drug is called Orencia. What is this?

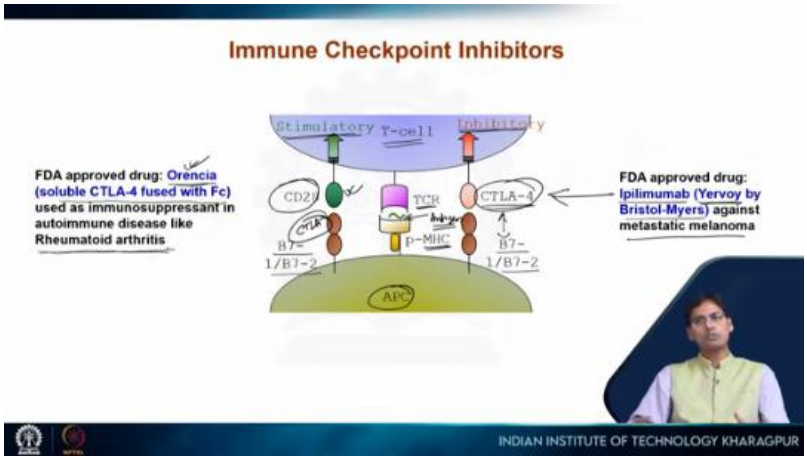
This is a soluble analog, a soluble form of CTLA-4 with Fc fusion. Fc means that part of the constant domain of the antibody. That Fc region, I already discussed that. So, CTLA-4 is fused with Fc. Now, what will happen?

This can be used as an immunosuppressant, and it can be used against some autoimmune diseases, for example, rheumatoid arthritis. How? Because this CTLA-4 is not membrane-bound. It is soluble CTLA-4 I am talking about. So, now if you are adding CTLA-4, what will happen?

This CTLA-4 will bind to B7-1 and B7-2 because I already mentioned the affinity between CTLA-4 and B7 molecules is much higher compared to CD28. So if you administer this soluble form of CTLA-4 fused with Fc, which is much more stable and a

better reagent, that's why we are adding the Fc region to that particular soluble CTLA-4. Now they will bind to B7 molecules, but they are not CTLA-4; they are not membrane-bound and they are not on the T cell. They cannot provide the inhibitory signal to the T cell, then what will happen?

As a result, the CD28 cannot be engaged with B7 here because B7s are already saturated by CTLA-4. So, therefore, our immune system, like the T cell, will not be that much active because the CD28 is not getting B7 on the antigen presenting cell. Because of that, this kind of strategy can be used against autoimmune disease, for example, as I already mentioned that Orencia is being used against rheumatoid arthritis. But there are some pros and cons about this immunotherapy also. So, we have to critically evaluate those things and accordingly need to use all this kind of immunotherapeutic reagent.



So, this is just a touch I started a little bit of this discussion, but if you are interested, you can follow any good textbook and recent literature will get you huge information because particularly immunotherapeutic reagents for recent cancer therapy are coming up. So that's all and thank you very much.

REFERENCES

1. Janeway's Immunology (9th Edition)