Cell and Molecular Biology
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Week 11
Molecular Immunology
Lecture - 43
Vaccines

Hello, everyone. This is Dr. Vishal Trivedi from the Department of Biosciences and Bioengineering, IIT Guwahati. And in this particular module, we are discussing the different aspects of immunology. So far, what we have discussed includes the basics of immunology, the purpose of the immune system, and how the immune system has different types of systems. So we have cell-mediated immunity, we have antibodies to protect us, and so on, right? So we have a different type of cells: the T cells, B cells, and macrophages, and we will discuss how these cells are integrated with each other and how they are actually causing a very robust immune response so that we can be protected from different types of infectious organisms.

Not only do external infectious agents affect us, but our immune system also protects us from internal antigens as well. For example, it is protecting us from the development of cancers or eliciting different types of allergic reactions. Subsequent to that, in the previous lecture, we also discussed the antibodies. So we have discussed the structure of the antibodies, how the antibody is produced within the host system, and how the diversity of the antibody is governed.

By the clonal propagation and selection by the antigen. And then, subsequent to that, we will also discuss how you can produce the antibody in animals. So we have taken an example of a rabbit, and we have shown you all the steps on how the antibody can be produced in rabbits. Subsequent to that, we have also seen the interaction of the antibody and antigens, and we have studied how you can use this particular phenomenon to develop different types of immune assets and how you can use this for different types of biological applications. Now, in today's lecture, we are going to discuss the vaccinations and how the vaccination is helping the organism acquire resistance against different types of infectious organisms and how that can be used or how that is helping society get protected from different types of diseases.

So as far as the vaccine is concerned, it is also called immunization. So immunization is the process of eliciting a long-term state of protective immunity against disease-causing pathogens. Exposure to the live pathogen, followed by recovery, is one route to immunization. Vaccination, or the intentional exposure to form a pathogen that does not cause disease, is called a vaccine, and is another way of mimicking the natural process.

So in nature, what happens when the pathogen comes is that it actually elicits the immune response, and then our immune system illuminates that pathogen.

And in this process, some amount of B cells or some memory B cells is being formed. So when the same pathogen comes the next time, it actually elicits a more robust immune response so that particular pathogen is eliminated. So the similar natural process can be mimicked if you are actually going to give the infection in the form of a pathogen that does not cause disease, right? So in an ideal world, both engage the antigen-specific lymphocytes and result in the generation of memory cells providing long-term protection. However, vaccination does not ensure immunity, and a state of immune protection can be achieved by means other than infection or vaccination. For example, the transfer of antibodies from the mother to the fetus or the injection of antiserum against a pathogen.

Both provide immune protection, but these are called passive vaccinations or passive immunity. Without the development of memory B cells or T cells specific to the organism, however, this state of immunity is only temporary. Thus, vaccination is an event in which immunization, the development of a protective memory response, is a potential outcome of that particular event. So this can be achieved in multiple ways, right? So you can actually have different types of vaccines. You can have the passive vaccine, can't you? So, Edward Jenner and Louis Pasteur are recognized as the pioneers of vaccinations or the early attempts to induce active immunity.

But similar recognition is due to Emil von Boring and Hinderbo Grasso for their contributions to passive immunity. This investigator was the first to show that the immunity elicited in one animal can be transferred to another by injecting the serum from the first animal. So passive immunization, in which the preformed antibodies are transferred to a recipient, occurs naturally when maternal IgG crosses the placenta to the developing fetus. Similarly, the maternal antibodies to diphtheria, tetanus, streptococci, rubella, mumps, and poliovirus are required for passive protection of the developing fetus. Later, the maternal antibody present in breast milk can also provide passive immunity to the infant in the form of the maternally produced IgA.

Then we have passive immunization, which can also be achieved by injecting a recipient with the preformed antibody, which is called an antiserum from the immune individual. Before vaccines and antibodies became available, passive immunization was the only effective therapy for some otherwise fatal diseases such as diphtheria, providing the much-needed femoral defense. Currently, several conditions still warrant the use of passive immunizations, including the following: for example, if there is immunodeficiency, toxins from venous exposure, or exposure to a pathogen that can cause a disease faster than an effective immune response can develop. Babies born with

congenital immune deficiencies are frequently treated with passive vaccinations, as are children experiencing acute respiratory failure caused by the septic neutral virus. Passive immunity is used in an unvaccinated individual exposed to the organisms that cause botulism, tetanus, diphtheria, hepatitis, measles, and rabies.

to protect their travelers and their health care workers who expect exposure to the potential pathogen due to their lack of protective immunity. Antiserum provides protection against poisonous snake bites and insect bites; in all these instances, it is important to remember that passive immunization does not activate the host immune response. Right? So passive immunization only provides the solution; passive vaccine immunization does not provide protection for subsequent infections, right? Passive immunization is a temporary solution. Therefore, it generates no memory response and protection in is transient. Protection is temporary.

It actually happens at that moment. You cannot produce antibodies. So passive immunization may be effective. It should be used with caution because certain risks are associated with the injection of a preformed antibody. The antibodies were produced in another species, such as a horse, right? The recipient can mount a strong response to the isotopic determinants of the foreign antibody or the parts of the antibody that are unique to the horse species.

This anti-isotype response can cause serious complications. Some individuals will produce the IgE antibody specific for horse-specific determinants. High levels of these IgE horse antibody immune complexes can induce the persistent mast cell degranulation, leading to systemic anaphylaxis. Another individual produces IgG or IgM antibodies specific for the foreign antibody, which forms complement-activating immune complexes. The deposition of these complexes in the tissue can lead to a type 3 hypersensitivity reaction.

Even if human antiserum or gamma globulin is used, the recipient can generate an anti-allotype response to human immunoglobulins and recognize specific antigenic differences, although its insensitivity is usually much less than that of anti-isotype responses. There are common agents, you know, that can be used for passive immunization. For example, you can actually use the horse antivenom for the black widow spider mite. Then, for botulism, you can use the horse antitoxins. Bacteria, you can use horse antitoxins.

Then, for hepatitis A and B, measles, and tetanus, you can use the pulled human immunoglobulins. Then, for rabies, you can use human or horse polyclonal antibodies. Then, for the snake bite, you can use the horse antivenom. And for respiratory diseases,

you can use the monoclonal anti-RSV. Then we have the active vaccinations or the active vaccine.

Whereas the aim of passive vaccination is transient, the protection or elevation of an existing condition, the goal of active immunization is to trigger the adaptive immune response in a way that will elicit protective immunity and immunological memory. When active immunization is successful, a subsequent exposure to the pathogenic agent elicits a secondary immune response that successfully eliminates the pathogen or prevents the disease mediated by its products. Active immunization can be achieved through natural infection with a microorganism or it can be acquired artificially by the administration of vaccines. In active immunization, as the name implies, the immune system plays an active role, doesn't it? Proliferation of the antigen-specific T cells and B cells is induced and results in the formation of protective memory cells, and this is a primary goal of vaccination. So active vaccination is actually going to engage the T cells, B cells, and ultimately it is going to produce the vaccine memory cells.

Right. So, these memory cells are actually going to have the memory for a particular antigen. Right. And then this antigen comes naturally. Right. When you are going to get a disease, these memories are actually going to produce the antibodies.

And that's why these antibodies are actually going to protect you against the particular disease. The active immunization with various types of vaccines has played an important role in the reduction of deaths from infectious diseases, especially among children. The use of vaccines for active immunization has proven essential in lowering the number of infectious disease-related fatalities among children. According to the recommendation established by the American Academy of Pediatrics, pediatric vaccination in the United States starts at birth. So you know that there are so many vaccines that we are going to give to the kids.

And it starts on day one. So in India, when the kid is born, on day one, you actually get the vaccine for the BCG. The BCG vaccine is being used to protect a person from one of the deadly diseases, which is called tuberculosis. So, the hepatitis B vaccine, diphtheria vaccines, pertussis vaccines, hemophilia, influenza-type vaccines, inactivated polio vaccines, measles, mumps, rubella vaccines, Wurzel-Joster vaccines, meningococcal vaccines, etc. are among the vaccines that are advised for children up to the age of six. In addition to girls, boys between the ages of 11 and 12 are advised to get the vaccine against HPV or human papillomavirus, which is actually going to be used to generate the immune response against cervical cancer.

So it is recommended not only for girls but also for boys. Then we have the subunit

vaccine. So many of the risks associated with the attenuated or killed whole organism vaccine can be avoided with a strategy that uses only the specific purified micro molecule derived from the pathogen. So the subunit vaccine is part of the pathogen or part of the pathogen in terms of the protein. So you are not using the complete pathogen, but you are using a protein from the particular pathogenic organism.

The three most common applications of this strategy referring to subunit vaccines are inactivated exotoxins or toxoids, capsular polysaccharides or surface glycoproteins, and key recombinant protein antigens. One limitation of some subunit vaccines, especially the polysaccharide vaccine, is their inability to activate T helper cells. Instead, they activate the B cell in a thymus-independent manner. resulting in an IgM production but little class switching. So there is no affinity maturation and little, if any, development of memory cells.

So this is the limitation of some of the subunit vaccines where you are using the polysaccharides or the low molecular weight antigens: it is not going to activate the T cells, and it is not going to activate the B cells in a thymus-dependent process. And because of that, it is actually going to produce IgM rather than IgG. And it is also not going to give you the memory cells. However, the vaccines that conjugate a polysaccharide antigen to a protein carrier can take care of this and induce the T helper cell response. Some bacterial pathogens, including those that cause diphtheria and tetanus, produce the exotoxins that account for all or most of the symptoms.

Diphtheria and tetanus vaccines have been made by purifying the bacterial exotoxins and then inactivating them with formaldehyde to form a toxoid. Vaccination with the toxoid induces anti-toxoid antibodies that are capable of binding to the toxin and neutralizing its effects. Conditions for the production of toxoid vaccines must be closely controlled and balanced to avoid the excessive modification of the epitope structure while also accomplishing complete detoxification. As discussed previously, passive immunity can also be used to provide temporary protection in unvaccinated individuals exposed to the organisms that produce these exotoxins, although no long-term protection is achieved in this case. The essential rule of some pathogenic bacteria depends primarily on the anti-phagocytic property of their hydrolytic polysaccharide capsules.

Coating the capsule with antibody can greatly increase the ability of macrophages or neutrophils to phagocytize such pathogens. These findings provide the rationale for a vaccine consisting of the proliferated capsular polysaccharides. There is a vaccine for Streptococcus pneumoniae that contains 13 antigenically distinct capsular polysaccharides. The vaccine induced the formation of oxygenating antibodies, and it is now on the list of vaccines recommended for all infants. The vaccine for Neisseria

meningitidis, a common cause of bacterial meningitis, also consists of the purified capsular polysaccharides.

Some viruses carry the surface glycoproteins that have been tested for use in antiviral vaccines with little success. Similarly, we have a glycoprotein D from HSV-2, which has been used to prevent genital herpes in the clinical trials of some vaccines, suggesting that they may have a variable approach to antiviral vaccines. Theoretically, the gene encoding any immunogenic protein can be cloned and expressed in a cultured cell using recombinant DNA technology, and this technique has been employed widely in the design of many types of subunit vaccines as well. For example, the safe way to produce a significant amount of purified toxin to be used in the generation of detoxified vaccines is by cloning the exotoxin gene from the pathogenic organism into an easily cultured host cell. A number of genes encoding the surface antigen from the virus, bacteria, and protein pathogens have been successfully cloned into a cellular expression system for use in vaccine development.

For example, a recombinant vaccine developed against hepatitis B was created by cloning the gene of a major hepatitis B surface antigen and expressing it in yeast cells. The recombinant yeast cells are grown in large permits, allowing the HBSE to accumulate in the cells. The yeast cells are harvested and disrupted, leaving the recombinant HBSE antigen, which is then purified using a conventional biochemical technique. And the recombinant hepatitis B vaccine induced the production of protective antibodies and holds much promise worldwide for protection against the human pathogen. So, this is the production of these subunit vaccines, such as the hepatitis B vaccine.

So, this is a hepatitis virus, right? So from the virus, what you do is take out the protein code, right? And then you code the protein, clone it into a suitable yeast vector, transform that into a yeast, and then it will actually start producing the protein. And then you use the purification strategies, and that's how it is actually going to become a vaccine. Then we have a DNA vaccine, you know, so a more recent vaccination strategy called a DNA vaccine utilizes the plasmid DNA encoding the antigenic protein that is injected directly into the muscles of the recipient. This strategy relies on the host cells to take up the DNA and produce the immunogenic protein in vivo, thus directing the antigen through the endogenous MHC class I presentation pathway that helps to activate better CTL responses. The DNA appears either to integrate into the chromosomal DNA or to be maintained for a long period in an episomal form, and this will be taken up by the dendritic cells or the muscle cells in the injected area.

Since muscle cells express low levels of class 1 MSG and do not express co-stimulatory

molecules, delivery to local dendritic cells may be crucial for the development of antigenic responses to the DNA vaccine. So, so far in the subunit vaccines, what we are doing is cloning that part of that particular pathogen into a recombinant vector, then producing the protein, and then injecting the protein. Right. In a DNA vaccine, instead of doing that, we are actually taking out the gene fragment, cloning it into a suitable vector, and then injecting the vector containing that particular recombinant DNA. And once this particular recombinant clone enters the cell, it starts producing the protein.

And then, as a result of that, it is actually going to start eliciting the immune response, and that's how it is actually going to mimic the natural conditions. So tests in animals have shown that DNA vaccines are able to induce protective immunity against a number of pathogens, including influenza and rabies. The addition of a follow-up booster shot with a protein antigen or the inclusion of a supplementary DNA sequence in the vector may enhance the immune response. One sequence that has been able to, you know, add to some vaccine is the common CPG DNA motif found in the pathogens recalled in this sequence in the ligands for TLR9. DNA vaccines offer some protective advantages over many of the existing vaccine approaches.

Since the encoded protein is expressed in the host in its natural form, there is no denaturation or modification. The immune system is directed to the antigen presented by a pathogen, inducing both humoral and cell-mediated immunity to stimulate both arms of the adaptive immune response. Non-DNA vaccines normally require immunization with live attenuated preparations, which can incur additional risks, so DNA vaccines also induce prolonged immunity. Expression of the antigen enhances the induction of immunological memory; finally, the DNA vaccine presents the important practical advantage that no refrigeration of the plasmid DNA is required, eliminating the long-term storage challenges. Remember that vaccine storage and transport is a very important feature.

Because it actually eats up a lot of money, right? So, when you are supposed to, you know, store the vaccines at a very low temperature, then you are supposed to carry the refrigerators and all that, and that actually increases the price of these vaccines. So, in addition, the same plasmid vaccine can be custom-tailored to insert the DNA encoding a variety of proteins, which allows the simultaneous manufacturing of a variety of DNA vaccines for different pathogens, saving time and money. An improved method for administering the DNA vaccine involves coating gold microbeads with the plasmid DNA and delivering the coated particles into the underlying muscle with an air gun, which is called a gene gun, right? This allows the rapid delivery of a vaccine to a large population without the need for a huge number of needle concerns, improving safety and reducing costs. Human trials are underway with several different DNA vaccines, including those

for malaria, HIV, influenza, Ebola, and herpes. Although there are currently no licensed human DNA vaccines, three such vaccines have been licensed for veterinary use, including the Wesson-Lyle vaccine that has been protecting hosts.

This vaccine has been used and tested in humans. After three doses, most volunteers demonstrated a titer of neutralizing antibodies similar to those seen in heart tests, as well as CD8 positive and CD4 positive T cell responses against the viruses. Since the widespread development of the DNA vaccine for use in humans is still in its early stages, the risks associated with the use of this strategy are still largely unknown. So this is the classical strategy for the DNA vaccine. So first, what you're going to do is you're going to take out the gene.

Right. What you are going to clone and then clone into the vector. Then you are going to insert that vector into a particular cell. Right? Then you inject this, right? And then it is actually going to start producing the different types of vaccines, right? And then you can actually use the DNA vaccine. Similarly, we can have RNA vaccines. So, RNA vaccines prime the body to combat external invaders.

The vaccines aid in the prevention of infections. All vaccines cause an immune response by introducing a harmless fragment of a specific virus or bacterium. The majority of vaccines contain a dead or weakened virus or bacteria. But instead of using a component of real bacteria or viruses, researchers have created a new kind of vaccine that is known as the messenger RNA vaccine. So one kind of RNA that is required for the synthesis of proteins is messenger RNA. Cells strictly degrade the messenger RNA after completing the proteins.

So vaccine-derived messenger RNA does not change DNA or enter the nucleus. So a fragment of RNA that matches a viral protein, typically a small fragment of a protein present on the virus, is introduced as part of messenger RNA. So earlier we were administering the DNA vaccine. So DNA produces the messenger RNA, and then messenger RNA produces the protein. Remember that when we were discussing the central dogma of molecular biology.

Now, with the DNA vaccine, there is a danger that the DNA vaccine could integrate into the human genome or the organism's genome. So, to avoid that, people have started injecting messenger RNA. Now, messenger RNA will not integrate into the genomic DNA or the genomic content. So, messenger RNA will produce the protein, and then the protein is going to be used as the antigen, and it is actually going to elicit the immune response. So, those who receive a messenger RNA vaccine are not exposed to the virus, nor can the vaccine cause the infection.

The viral protein can be produced by simply using messenger RNA. The immune system creates a specialized protein known as an antibody in reaction to its recognition as foreign. By identifying certain viruses or other pathogens, binding to them, and designing the pathogens for elimination, antibodies aid in the body's defense against infection. Once created, the antibody stayed in the body even after the pathogen had been eliminated, allowing the immune system to react promptly in the event of re-exposure. After being vaccinated against the virus using messenger RNA, antibodies may strictly identify the virus, bind to it, and label it for destruction before it can cause major illness. The only recognized or permitted messenger RNA vaccine at this time is for COVID-19, an illness caused by the SARS-CoV-2 coronavirus.

These vaccines employ messenger RNA, which instructs the cells to make copies of the spike protein, a protein found on the surface of the coronavirus. Scientists are looking into the possibility of using messenger RNA to create vaccines for adult illnesses. This is, you know, an example of the COVID virus. So this is the SARS-CoV-2 virus, right, which is responsible for the famous COVID-19. So the SARS-CoV-2 virus, which was initially discovered in China in late 2019, is the cause of the viral disease called COVID-19.

When an infected person coughs, sneezes, talks, or breathes, respiratory droplets are the main way the virus spreads, causing different types of illness and symptoms. And talking about the COVID vaccine, the COVID virus is a primary target for vaccination because of its spike protein, which enables it to bind to and penetrate human cells. The COVID vaccines are being designed to train the immune system to recognize and fight SARS-CoV-2, preventing illness or reducing the severity of the infections. The spike protein or the whole viruses, right, these are the two things we can use for developing the COVID vaccine.

Now, let's talk about the messenger RNA vaccine. So messenger RNA vaccines introduced the cells to how to produce the spike protein that is presented on the COVID-19 virus's surface. So following the immunization, the protein fragments are produced by the body's muscle cells and displayed on their cell surface. The body produces antibodies as a result. These antibodies are then employed to aid in the removal of the COVID-19 virus if they acquire it. Once the protein fragments are formed, the instructions are broken down and eliminated by our cells.

The vaccine's messenger RNA does not penetrate the cell's nucleus, which houses some DNA. Messenger RNA is used in the Moderna and Pfizer's BioNTech COVID-19 vaccines. So in a COVID-19 vaccine, you are actually producing messenger RNA from

the COVID-19, right? Which is responsible for making the spike protein. And then you are putting this messenger RNA into a human cell, correct? And then this human cell started producing the protein, and it is actually expressing that protein, the spike protein, on its surface. And because it is immunogenic, it is actually going to be recognized by the immune system, and it is actually going to start producing antibodies.

And now, when there is a subsequent infection with the real virus, these antibodies are actually going to bind to these viruses, and they will not allow the virus to cause the disease. Then we have the vector vaccines. So, this is a kind of vaccine involving the insertion of a component from the COVID-19 virus into a modified strain of another virus, referred to as a viral vector. The cells are instructed to replicate the COVID-19 S protein by means of a viral vector. The immune system reacts by producing antibodies and protecting white blood cells when the cells exhibit the S protein on their surface.

So, the antibody aids in the removal of COVID-19 viruses. So neither the viral vector nor the COVID-19 virus can interact with or infect a person who has received the viral vector. So, for example, the Johnson & Johnson COVID-19 vaccine. So this is a strategy for developing COVID-19 vaccines. So you actually have the COVID-19 virus.

We just take out the gene for the spike protein. Then you clone it into the viral vectors, and then you are going to insert that into the human cell, and then it is actually going to start producing the spike proteins, and then these spike proteins are going to be recognized by the immune system. And then it is actually going to produce the antibodies, and these antibodies can be used in a subsequent infection to provide the immunization. Then we also have the protein subunit vaccines. So only the portion of a protein that caused the effect effectively activates the immune system and is included in the subunit vaccine. So you're basically going to take the spike protein, produce the spike protein, and then use that.

So examples of the subunit vaccine are Novavax COVID-19 vaccines. And this is the strategy, right, where you are actually going to take out the spike proteins and clone them into a bacterial, yeast, or animal system. Then you produce the protein, purify that protein, and inject that protein for vaccinations. And then the spike proteins will be recognized by the immune system. It will produce antibodies, and then these antibodies are going to give you protection. Then, in some cases, we have used the inactivated COVID vaccines or inactivated COVID virus as a vaccine.

A complete virus that has been killed so that it cannot reproduce or spread illness is used in inactivated vaccines. An example is Covaxin, which has been developed by Bharat Biotech. And this has been mainly used for vaccination in India. So what you have is the

virus that causes a disease. So what you can do is culture this virus, and then inactivate it by chemical treatment or some other method.

Then you take this and inject it into the human, and then the human is going to produce the antibody, and it is actually going to give you the protection. Then we can also have DNA vaccines. So a small fragment of DNA, known as a plasmid, is introduced into the body in a DNA vaccine. And what you can do is just take the gene for the spike protein, and then you are actually going to, you know, encode that into a vector, right? And then you inject that as a DNA vaccine. So the example for DNA vaccine is ZYCOVD, the first authorized DNA-based COVID-19 vaccine, which is produced by the Zydus Cadila in India.

And this is the classical examples, right? So you're going to have a COVID-19. You are going to take out a portion of the chromosome, which is responsible for generating the spike protein. then you clone that into a suitable viral vector and then you inject that into the cell and they will start producing the proteins and these proteins are immunogenic so they are actually going to be recognized by the immune system and that's how they are actually going to produce the antibodies and that's how these antibodies are going to give you the protection when there will be a real infection of the COVID-19. So this is all about the vaccines that we have discussed. We have discussed about the basic phenomena, what is happening when you are getting the vaccinations. So when you are getting a vaccination, the purpose of the vaccine is that it should actually elicit the immune response, but it will not cause a disease.

And as a result of that, it is actually going to train the immune system and it's also going to reduce the memory cells. Right. And these memory cells are actually going to keep a memory. And as a result of that, you are actually going to whenever there will be a subsequent encounter with the real organisms, it is actually going to elicit the immune response.

And that's how it is actually going to clear the infection. As far as the purpose is concerned, you can actually have the different types of vaccines. You can have the passive vaccinations. You can have the active vaccinations. And within the active vaccinations, you can actually use the different strategies.

So in some cases, people are using the inactivated organisms. And in some cases, people are using the product from the pathogens, such as toxides or toxins. which will not cause the disease but it is actually going to train the system so that you can actually be able to elicit the immune response. Then we can also use the subunit vaccines.

You can actually use the DNA vaccines. You can use the mRNA vaccines and so on. And lastly, we have also discussed about the different types of vaccines which are being developed against the COVID-19. And we have also understand the mechanism by which these vaccines are being developed and how they are actually giving the protection against this deadly disease. So with this, I would like to conclude my lecture here. In our subsequent lecture, we are going to discuss some more interesting aspects. Thank you.