Course Name: I Think Biology

Professor Name: Dr. Sravanti Uppaluri

Department Name: Biology

Institute Name: Azim Premji University

Week: 10

Lecture: 52

W10L52_Public Health-Rotavirus (Case study)

Hello, as part of the unit on public health in the iThink Biology NPTEL course, in this lecture we will be discussing the case study of rotavirus, which is a major public health issue in India today. So viruses come in many different shapes and sizes and they also manifest as disease in with various, really a very wide range of symptoms. So the tobacco mosaic virus that you see on the bottom right has a long elongated shape. There are really geometrical shapes that you see in viruses like the icosahedral viruses. The one that we may must be familiar with is coronavirus because it caused the global pandemic just a few years ago. In general, the mechanism through which viruses attack a host cell is illustrated here.

So a virus initially attaches to a cell, enters the cell. Once a virus enters the cell, it releases its genome into the host cell. And this genome then is replicated using the host machinery. And once that's done, the entire virus is assembled.

And as the virus, viral particles replicate, the viral particles are released and the host cell rises. This is a very general schematic to show you how viruses work. But viruses, as I said earlier, come in different shapes and forms. And the mechanisms through which they enter the cell, whether they have a genome that is made up of RNA or DNA, also depends widely on the evolutionary trajectory of a virus.

So rotavirus in particular is named after its weed-like structure. That's what you see on the right-hand side. And a schematic of the rotavirus shows you that the genome of the rotavirus is made up of double-stranded RNA molecules. And it has viral proteins 1 through 7 that are specifically located at different positions on the viral particle. Okay. So a more more detailed picture and a more specific one for rotavirus is shown here.

It shows you that the rotavirus binds the surface of a cell. And this binding occurs through one of the viral proteins that I talked about in the previous slide. These viral proteins bind to the

surface of the cell, allowing the viral particle to be engulfed and placed inside an endosome within the cell. So you should be asking yourself, why does a host cell actually engulf an a foreign particle, right? Is there no no way for it to recognize that there is a foreign particle that has entered the cell or that is attempting to try and enter the cell? And these are really interesting, actually, evolutionary questions that you might ask. As the question, as the viral particle proceeds through its lifecycle, it releases its RNA into the cytoplasm.

This RNA is then transcribed and translated into viral proteins. And together, these viral proteins and the RNA, the double-stranded RNA of the, of the virus that has been replicated within the host cell are then assembled. And these, this assembled, you know the assembly sort of process or the assembly line of of the virus begins as it goes through an organelle quantum pyroplasm and then travels through the endoplasmic reticulum. And when the viral particle is released, it causes cellular damage. And of course, since this is a schematic, we're just showing you one virus viral particle, right? But in fact, many viruses are being replicated and formed and assembled at the same time.

So this usually or often ends up in the host cell actually rising altogether. So how does, where does, where is the virus formed in found in the first place? So it turns out that rotavirus is, of course, transmitted from human to human through the fecal oral route. So that means if there is, in our drinking water, if there is any kind of contamination, this can lead to rotavirus infection. But this also happens through a process called zoonosis. So zoonosis is where basically a pathogen is transferred from an animal to a human.

And in the case of rotavirus, it has been seen, actually zoonotic transmission has been seen from horses, pigs, cows, dogs, cats, goats, and sheep. So really all kinds of animals. So whether you have pets or you live on a farm with animals, it's not unlikely that members of your family or yourself would have been infected by rotavirus. So I should mention that rotavirus as a public health issue is quite important because nearly 90% of the global human population. So that's not just in India. Over 90% of people have been infected with rotavirus at some point or the other.

So if you think about this, what, what does it mean that rotavirus is a public health issue? So how dangerous is it? What is, what is really the problem and how big is it? And what do we mean by how big is it? How many people does it infect? What are the treatments available? How much does it cost? Who are the people involved who try to solve these kinds of public health issues? And what are the kinds of skills and capacities people need in the professions that address public health issues? Right? So I should tell you, so rotavirus primarily causes diarrhea. And by itself, it is it is a problem that nowadays can be treated. Unfortunately, however, if it's not recognized early enough, rotavirus can cause severe dehydration and can actually be fatal in children. So in the next slide, what I will show you is a video, a video interview of Professor Gagandeep Kang, who is an Indian world renowned expert physician scientist who has dedicated a large part of her life to the study of rotavirus in India.

And in this video, this video interview, you will see and learn about the various facets of rotavirus that she has studied. And this will really give you an insight into how wide ranging public health is as a discipline. A rotavirus has been a big part of your life for so many years, and you've engaged with the rotavirus in several ways as well. Could you tell us a little bit about all the work that you've done with rotavirus and your association with it? Yeah, sure. So I started with rotavirus.

In fact, what is my office now was originally a lab and was the first place that I ran a gel where I could see the, you know, did silver staining so that you could see the genome of the rotavirus. This was at a time when electro-fiber typing was the way to be able to look at the relationships between rotaviruses in the mid 1990s. After that, we started to use more molecular projects. But I've looked at rotaviruses every which way that it's possible to have looked at them in the community and looked at them in people in hospitals and adults and children, in animals also. And there are plenty of rotaviruses by the way. When people think about zoonotic infections, actually zoonosis go in both directions. So if you have pet animals or you have cows and goats, people who look after those animals can transmit their viruses to those animals and can acquire the animals viruses. We've studied it for rotavirus. We've studied it for parasites and bacteria. But whenever you're in proximity with people or animals, there is transmission going on.

So we've looked at rotavirus vaccines. We've looked at clinical trials for rotavirus. What kinds of drugs can be used to treat it? We've looked at does rotavirus change the microbiome? We've looked at what rotavirus does to families in terms of costs of treatment as outpatients, in terms of hospitalizations, interactions of rotavirus with other birds. You name it, we've done it. Alright, so Professor Kang really gave us a wide range of topics that she has studied in relation to rotavirus.

So let's do a brief review of that to reinforce really what the discipline of public health can be, right? So she started off telling us about how she looked at rotavirus in the lab, right? So she was studying genomes. So she did lab work. And also tried to identify different kinds of strains. She also mentioned zoonosis. So studying the transmission between humans and animals in both directions, right? She mentioned that she has looked at rotavirus in individuals, in hospitals, in families, in communities, right? So she's looked at how rotavirus affects basically individuals at different sort of scales, right? Both at a small individual scale, but where you are looking at communities, so many individuals together. She also said how it, she also said that they've looked at the economics. In other words, what are the costs related to rotavirus, right? In terms of hospitalization or treatment.

So I'll put this down as economics of the disease, right? She talked about how rotavirus affects the microbiome. So the microbiome is essentially the set of microbial organisms that are part of your body, right? So being infected with rotavirus, how does it change or influence that microbiome? So you would have to be a microbiologist, at least have the skills of some of, some

skills of a microbiologist to do this study. She then mentioned vaccines and clinical trials. We'll talk about the vaccine, the vaccine development in the next few slides.

And clinical trials basically involve the testing of a vaccine or any kind of treatment. And this usually is runs in several phases. That the clinical trials themselves run in three phases. One where you start off testing on a very small population, you grow the population. And then a third clinical trial where you look at the effect of the treatment, say in a larger set of population over a longer period of time, look for side effects and see whether the vaccine or the treatment actually addresses the problem that you're trying to solve.

So together what you can see is whether you're looking at the economics and the cost related to the disease, or you're looking at how a virus or a health issue affects individuals, families or communities. And if you are doing studies in the lab, you might be a microbiologist or you might be a biochemist, molecular biologist to be studying, you know different aspects of a virus. You might be interested in veterinary science when you're studying zoonosis, or you might be a pathologist or a physician to be studying how zoonosis or zoonotic transmission influences the presence of a pathogen in the, in a population. You might be an immunologist trying to develop a vaccine, or you might be, say, somebody who's working in a pharmaceutical industry looking at clinical trials. So together you can see that there's a lot of different kinds of skill sets and different kinds of professions that are actually involved in looking at an issue that is part of public health, right?

So you really need strong teamwork. In this lecture we'll address two of the things that we talked about. The first is the lab. How do we study a virus in the lab? The other is how is a vaccine produced for rotavirus, right? So in the lab, how do we experiment? In other words, how do we study how a virus works? In general, how do we study how any pathogen works? How do we study how cells divide, right? So for obvious ethical reasons, we can't really experiment directly on humans. And so in general, most of fundamental biological work in the lab has been done using model systems.

So these are drawings of very commonly used model organisms whose sort of biology has been well understood. The most sort of commonly used model organism is E. coli. And E. coli has been used to study very many different aspects of cell biology, everything from cell motility to cell division, cell size, etc etc.

Budding yeast, that's what's shown here. Budding yeast is used or has been used to study the cell cycle. Another example is the fruit fly, Drosophila melanogaster. The fruit fly has been an extremely important model organism for us to understand how genetics works, patterns of inheritance. It has also been important for us to study the process of development, right?

So how do you go from a single celled zygote to an organism that has specialized tissues and organs? Another one is zebrafish. That's what's mentioned here. A zebrafish has been an important model organism, as well as planaria, to study regeneration. Mice, of course, are mammals, so they are much closer to us as humans, have been used for behavioral studies. And, you know, a lot of work has been done in cancer biology as well.

So this is not an exhaustive list by any means, but this gives you an idea of different kinds of systems that are used in the lab. So suppose I wanted to study rotavirus. One of the things that you can also do in the lab is to grow cells on their own in the lab, okay. And these cells can be used to specifically create what we call an organoid, right.

So that was what this picture here. And this organoid is a collection of cells that starts to divide and grow within the lab. And when you provide it the right sort of precursors and mediator molecules, you can actually get it to form a sort of, say, a a mini organ. So say a mini brain. We don't call it an organ. We call it an organoid because it really mimics the organ. It's not exactly the same, right. And the idea is that when you have an organoid, you are able to test, you know, targeted, say, therapies, or you can study in a very targeted manner a specific kind of behavior of a tissue.

So let's think about it now. Depending on the problem that you have that you want to study, model organisms are really important. And it makes sense that you would want to choose the right model organism. Right? So suppose you want to study sleep. Right. So that's what's listed here. I want to study how sleep works. So would I use a bacterial cell, E. coli, to study sleep? Or would it make more sense maybe to use a primate? So maybe study sleep in monkeys. Right. And of course, I would think your answer would be primates because we know that primates sleep. Whereas E. coli, our definition of sleep, will not certainly not match what E.coli do, right?

Likewise for schizophrenic behavior, if you wanted to study behavior, you might study a primate. On the other hand, if you wanted to understand, let's say, a specific gene that you know has been associated with schizophrenic behavior, you might look for that same gene or a homologous gene in another organism. So you might see that even mice have the same mutation that humans have that is correlated to schizophrenic through schizophrenia. So you might see that in mice.

You might also see it in C. elegans because C. elegans also have a very, very well characterized neural network. And because we have, it has such a well characterized neural network, it also allows us to manipulate it easily to see what the effects are on, say, the biology of schizophrenia or schizophrenic behavior, right. On the other hand, if you want to study something at a smaller scale, if you want to understand cell division, right, what would you use to study?

It might be much easier to understand how cells divide just by looking at bacterial cells. Right. Because you can isolate cells easily. You can grow them quickly and you can watch them divide quickly, right. You might also use cell lines. So cell lines are essentially basically cells that have

been taken from an organism. So there may be human cell lines, but you can also get, you know, cell lines from dogs or cats or other kinds of multicellular organisms. And and with the right culture conditions in the lab, they actually grow and proliferate. Often these cell lines originate from some sort of a cancer or tumor tissue because they're able to grow easily on their own.

And so now we are using systems that have the same or very similar sort of genomic content as humans do. Right. So human cell lines are interesting for that reason. So you could study cell division in human cell lines. And likewise, you might use cell lines to study brain cancer. Right. So you might study brain cancer looking at cell division or looking at the effects of some kinds of therapeutic drugs, first by testing on cell lines. And then maybe you would test on a mouse model that has cell lines.

Okay, so for example, it would not make sense then to study brain cancer with in using E. coli. Because E. coli don't have a brain. So you have to really choose the model organism depending on the problem that you're looking at. That's why choice of model organism is really essential for the problem that you're studying.

So I just want to illustrate now an example of how enteroids, so as I said, enteroids are organoids. And these are organized derived from human intestinal cells, right. So they are mini organs, organoids, because they're derived from intestinal cells, they're called enteroids. And the idea of this study by Saxena, K et al, the reference is given at the bottom right, was to test whether a human enteroid that was derived from a a tissue, from a human patient, whether this would be a good model system to study human rotavirus, right.

So what they did was they extracted intestinal cells from a human patient and then tried to grow these intestinal cells and develop them as enteroids in the lab. Then what they did was to try to infect these cells or rather infect these this enteroid with different strains of rotavirus, right. And they wanted to see if the human rotavirus was specific to this enteroid. And if so, that would indicate that the human enteroid is in fact a good model system to specifically study strains of the rotavirus that infect humans.

And what you see on the left hand side are images of the enteroid, right. At the very bottom is the human enteroid infected by the human rotavirus. The second one is the it’s the same similar enteroid infected by the animal rotavirus. And this the top, very top panel is the mark for the control. And what you see at the very bottom is that the enteroid that is infected with the human rotavirus has the most cytopathic effect, right? The most cytopathic effect.

In other words, the cells are affected the most. And you can see this sort of lysed and and dying. And this graph quantitatively describes what they've what they've seen on the left hand side, which is that in the mock. So in the control where there was no rotavirus that was placed on the on the enteroid, nearly none of the of the enteroids actually were destroyed. Whereas with the animal rotavirus, 13 percent were infected.

13 percent of the enteroids were infected. With the human rotavirus, 45 percent were infected. And so what Saxena, et al have done is shown that this human intestinal enteroid could be a good model system to try to study how human rotavirus infects human cells, right. OK, so let's shift gears a little bit to think about how. How the vaccine was developed. So this is a really interesting and heartwarming story because the the rotavirus vaccine is the first indigenously developed vaccine.

So this vaccine was developed in India by Indians for Indians, right. So often a problem with vaccines that are developed globally is that they are developed against strains that are found in other countries, right? And as we know, through the process of evolution, viruses can evolve very quickly and strains can start to diverge very quickly. So what you will see is that the process of the virus, the vaccine being introduced, took many years. So starting from 1988, where doctors at AIMS, the All India Institute for Medical Sciences, discovered a strain of rotavirus that was infecting newborns, but did not really cause any illness. And they so they thought that this particular strain might be useful to help develop a vaccine.

And over the course of the next 10 to 12 years, there was a collaboration, a global collaboration with the USA India vaccine program, together with DBT, the Department of Biotechnology, and the National Institute of Health in the USA. This program, over the course of 10 years, helped develop the vaccine and initiate and fund phase one clinical trials. In 2000, in 2000 phase one and phase two clinical trials were completed, but not without the help of many NGOs, continued support from AIMS and other organizations. By 2008, the phase three clinical trials were being conducted now across multiple centers in India. And towards the end of that period, Bharat Biotech became a partner to make this a public-private partnership, right.

And by the, by 2016, Rotavac, which is the rotavirus vaccine, was introduced into the Universal Immunization Program. This is a program that requires all, requires a minimal set of immunizations in the entire population of newborns, right. And so in summary, in this lecture, what we've had is a very brief introduction to different kinds of viruses. We've looked at how a public health issue can be or should be addressed and all of the different facets of public health issue, of a public health issue. We've looked at two different areas of that public health issue in a little bit more detail.

The first is model organisms, and we looked at an example of intestinal cells being used to develop enteroids. And we also briefly discussed how vaccine development is an example of really teamwork. And this was the development of the Indian rotavirus vaccine is really an amazing example of teamwork. What you would have seen from more recent experiences is the coronavirus vaccine, right. The Covid vaccine was developed on much shorter timescales. And I

think this is really an example, an amazing example of how systems have evolved and how we are able to address health requirements, public health requirements at much faster scales.