Environmental Chemistry and Microbiology

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Module - 11
Lecture - 55
Pathogens and Diseases - II

Welcome everyone. This is the second part of Pathogens and Diseases. This is lecture 55 of module 11.

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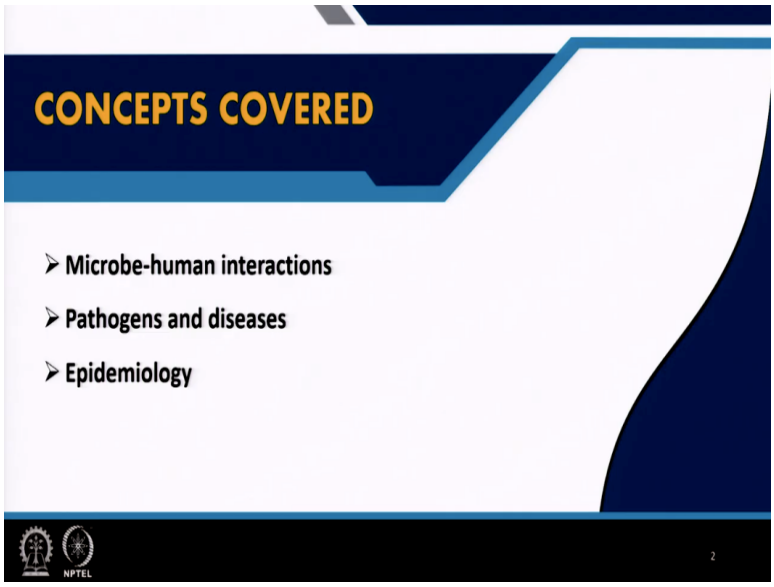

So, like I said in the previous part, we have already covered the nature of microbe and human interactions, what are the different types of interactions, both adverse as well as beneficial. We have also looked at some pathogens and diseases. We are going to continue with that. And towards the end of this topic, we will cover a little bit about epidemiology.

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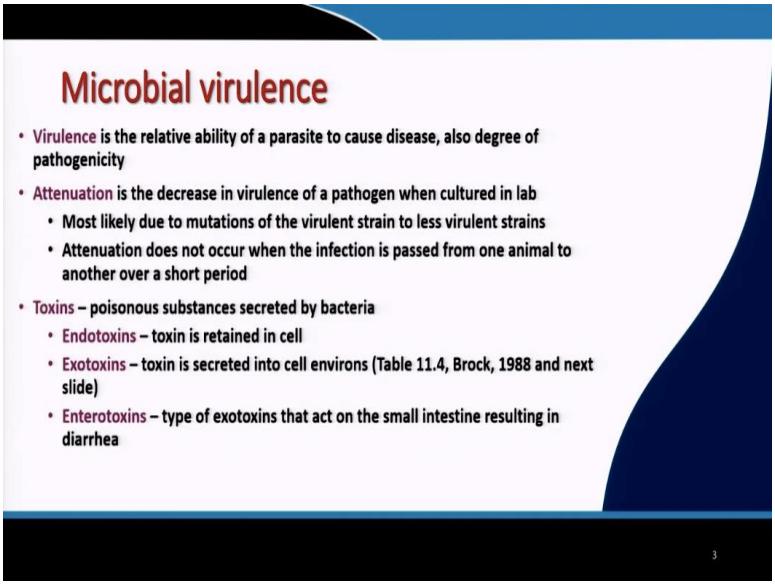

So, let us take a look at microbial virulence. Now, we already know that pathogens can be bacterial, viral, protozoa; all kinds of microbes exist. Do all microbes which cause diseases; let us say that if a particular pathogen or a particular parasite is known to cause a disease, is the relative ability to cause the disease going to be the same? And the answer is no; by no means is it going to be the same.

And this also has to do with the degree of pathogenicity of that organism or the parasite. And I will show you some examples a little bit later. Then second is the attenuation or the decrease in the virulence of the pathogen when it is cultured in the laboratory. So, for example, let us say you take a sample from an infected person. And then you culture it in the lab, and perhaps reinfect another person. There is some likelihood that there will be decreased virulence of that pathogen that has been cultured in the lab. So, I am not going to go into too much detail. Like I said in the last lecture, this remains the domain of medical experts, and we are not trying to compete with medical experts over here. So, we just want to be aware of some of the fundamental principles that are part of microbe-human interactions, diseases and their control and so on.

So, just some idea is necessary for all of us, but by no means are we going to go into any details. So, attenuation is most likely because of mutations that happen to the virulent strain and make it less virulent. And this attenuation does not occur when the infection is passed from one animal to another over a short period of time. I again come back to the example of COVID-19. We have seen that there is very little attenuation of the virulence of this virus, as it is being passed from one human being to another. So, this is not happening from host to host. When the infection is passed from host to host, it is not getting attenuated.

Let us also take a look at another issue that is related to pathogenic organisms, and these are toxins. So, you have poisonous substances that are secreted. In this case, I have written bacteria, but these toxins can be associated with algae. If you remember, in one of the initial lectures, I mentioned something about red algae. Red algae secrete a particular toxin. It is an exotoxin, which is (in the case of red algae), it is a neurotoxin and has been responsible for the death of many people who go swimming in areas that are affected by red algae.

So, these toxins can be poisonous substances secreted by bacteria, algae and many other microorganisms. There are 2 major groups, endotoxins and exotoxins. So, by definition, endotoxins are when the toxin is retained in the microbial cell. If it is an exotoxin, it means the toxin is secreted into the cell environment. Within exotoxins, we have enterotoxins. Entero- by definition means, within the gastrointestinal tract. So, these types of exotoxins that act in the small intestine, can cause diarrhea, stomach upset, and all these kinds of things. So, very often when we talk about contaminated food and water, and we have a stomach upset or diarrhea, that is, perhaps, because of these types of toxins.

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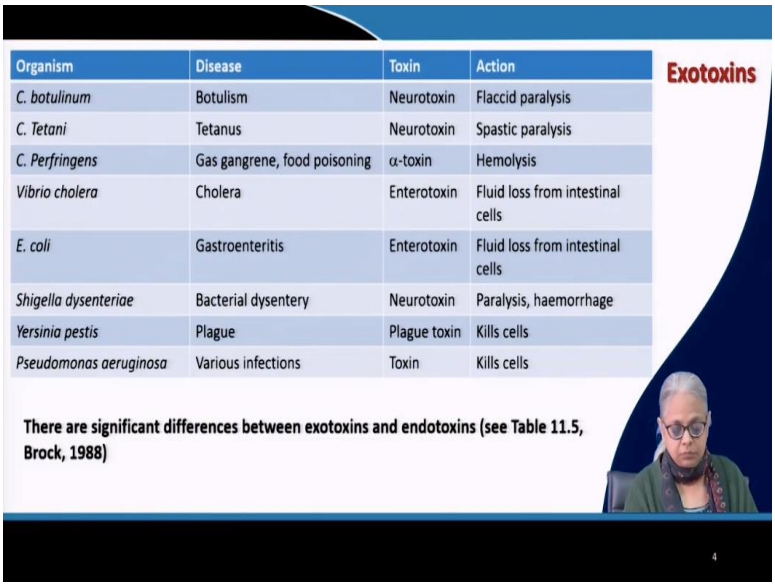

Organism	Disease	Toxin	Action
C. botulinum	Botulism	Neurotoxin	Flaccid paralysis
C. Tetani	Tetanus	Neurotoxin	Spastic paralysis
C. Perfringens	Gas gangrene, food poisoning	α-toxin	Hemolysis
Vibrio cholera	Cholera	Enterotoxin	Fluid loss from intestinal cells
E. coli	Gastroenteritis	Enterotoxin	Fluid loss from intestinal cells
Shigella dysenteriae	Bacterial dysentery	Neurotoxin	Paralysis, haemorrhage
Yersinia pestis	Plague	Plague toxin	Kills cells
Pseudomonas aeruginosa	Various infections	Toxin	Kills cells

Let me show you some examples of these kinds of exotoxins. So, we have here; this is an incomplete list; the actual list is in table 11.5 of the Brock, 1988 edition. So, you have an organism *Clostridium botulinum*. This is responsible for what is called food poisoning or botulism. It is a neurotoxin. It causes flaccid paralysis. Then you have *Clostridium tetani*. And again, same issue. We have a disease called tetanus. And those of you, who are like me, I often fall and injure myself. One of the things that we learned, at least I learned very early in my childhood was that if you have fallen and injured yourself, the first thing you need to do,

especially if you are in contact with dirt on the road and any other place, you need to go to the hospital and get what is called an anti-tetanus injection. So, that is because of this particular organism. This is something that is very common in soil, and it causes a neurotoxic effect and causes spastic paralysis.

Clostridium perfringens causes food poisoning, gas gangrene and the table in the textbook has alpha, beta, gamma, lambda; all these toxins are mentioned and they are all responsible for causing hemolysis. *Vibrio cholerae* is the causative agent of cholera. It produces enterotoxins and results in fluid loss from intestinal cells. *E. coli*, the virulent strain of *E. coli* can cause gastroenteritis. It is an enterotoxin again. Remember, these are both examples of enterotoxins; and the same effect, fluid loss from intestinal cells. *Shigella dysenteriae* causes bacterial dysentery, and it is also a neurotoxin; it causes paralysis, haemorrhage and so on, so many other effects.

Yersinia pestis is the causative agent of the plague. So, the toxin is called the plague toxin and it is responsible for killing cells. *Pseudomonas aeruginosa* is a very common environmental bacterium. You find it anywhere and everywhere. It causes any number of infections including gastroenteritis, stomach upsets all of that, and it secretes toxins; same effect, it kills cells.

So, there is another issue over here and that is the differences between exotoxins and endotoxins. You can refer to the original in the textbook, but I will just explain some of these effects to you right here. So, what are the major differences between exotoxins and endotoxins? The first thing is their chemical properties. The proteins that are excreted in terms of exotoxins, they are actually proteins that are excreted by certain gram-positive or gram-negative bacteria, and they are considered to be heat sensitive. So, if you boil the water, you are likely to be able to get rid of them. And if it is an endotoxin, it is because of the lipopolysaccharides or the lipoprotein complexes that are released, when the cell lysis. And they are often part of the outer membrane of these gram-negative bacteria. They are also considered extremely heat stable. So, I will come back to this point later. Then we think about toxicity.

Exotoxins are considered to be highly toxic; they can often have fatal effects. On the other hand, endotoxins are weakly toxic and they are rarely considered to be fatal. Immunogenicity; highly immunogenic. They stimulate the production of neutralising antibodies. Endotoxins on the other hand are relatively poor immunogens and the immune response is generally not sufficient for neutralising the toxin.

Then we come to the toxoid potential. The treatment of exotoxins with formaldehyde will destroy the toxicity, but the treated toxin remains immunogenic and there is no toxoid potential that is known for endotoxins. In terms of fever, exotoxins do not produce a fever in the host,

while endotoxins are pyrogenic, which means they often produce fever in the host. Let us then come to degree of virulence.

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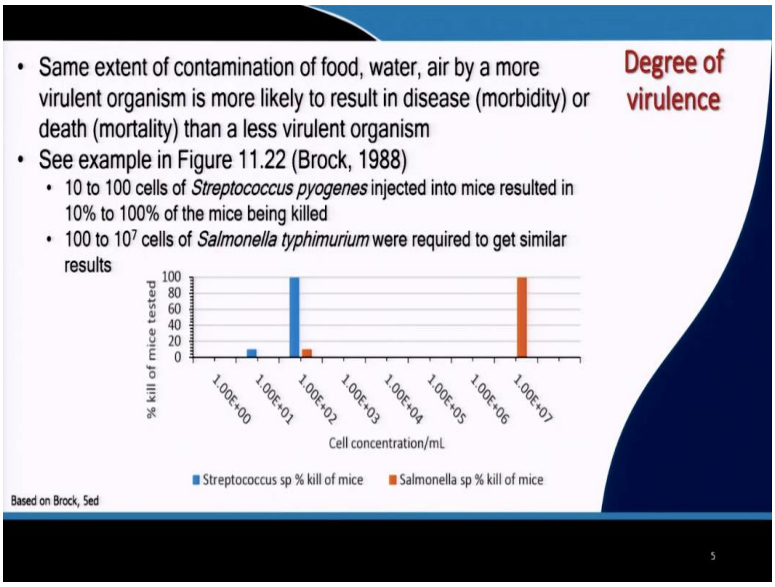

How do we determine the degree of virulence? This is often done in what are called lab or animal assays. And you can do it with bacteria; you can do it with smaller animals like rodents and rats and so on. So, those are standard methods, standard procedures for determining the virulence of different microorganisms. So, the same level of contamination of food, water, air, by a particular virulent organism, when it is going to cause either disease or death, is compared to a less virulent organism.

So, if you are wondering what that means, let us take a look at….so, if we want to look at the degree of virulence in terms of an example, you can refer to figure 11.2 in Brock's textbook. This is an old one. And I have taken some of the data and redrawn the graph over here. So, what you see is, as a function of cell concentration per ml, what was the percent kill of the mice that were tested? Now, its standard procedure to test the virulence or sometimes the degree of fatality associated with either a toxin or a microbial concentration and so on. So, it is standard practice to expose a particular organism. In this case, the mice have been exposed to 2 different species, *Streptococcus* and *Salmonella* species. And these data are simply there to tell you which one is more virulent.

So, let me just explain something. So, we have *Streptococcus pyogenes* that was injected into mice in a bioassay, and 10 cells of *Streptococcus* were related to 10% kill of the mice tested. When 100 cells of *Streptococcus pyogenes* were tested; these 100 cells were capable of killing all of the mice that were tested. Then we come to *Salmonella typhimurium*. Now, 100 cells of

Salmonella were injected into mice and 10% kill was obtained with these 100 cells. To get 100% kill of the mice tested, 10^7 cells of *Salmonella* were required to get that result. So, *Streptococcus* is definitely far more virulent compared to *Salmonella*.

So, the next thing that we want to know is, where are the infections most likely to come from?

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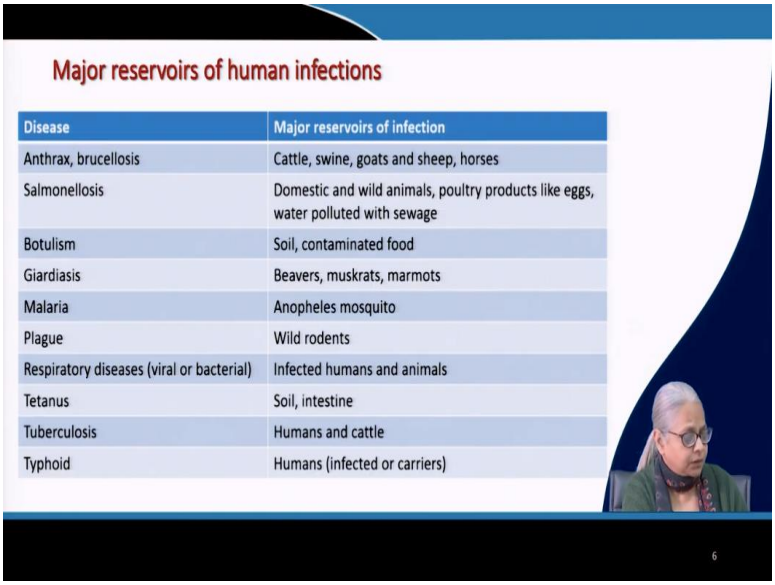

So, what are the major reservoirs of human infections? We have diseases like anthrax, brucellosis. The biggest reservoirs of infection are cattle, swine, goats and sheep, horses. These are the reservoirs where, if these animals are infected, they are also likely to pass it on to any human being that is in contact with them.

Salmonellosis: The biggest reservoirs are domestic as well as wild animals, poultry products like eggs, and water that is polluted with sewage.

Botulism comes from soil as well as from contaminated food. Giardiasis comes from beavers, muskrats, marmots and so on. Malaria is from the Anopheles mosquito. Plague is from wild rodents. Respiratory diseases, viral or bacterial, can be from infected humans as well as animals. Tetanus, I already mentioned it. It is mostly from soil. It can be from the intestines of different animals where there is faecal contamination in the soil.

So, very often, what happens is that faeces from animals and birds, etcetera falls on the soil, and therefore the soil contains these organisms. So, that is the normal route of transmission. Then we have tuberculosis. We have humans and cattle that can be reservoirs. For typhoid, same thing. Infected humans or people who are carriers; they may not be sick, but they are carrying the microorganism in their blood and so on. So, they are also potential reservoirs.

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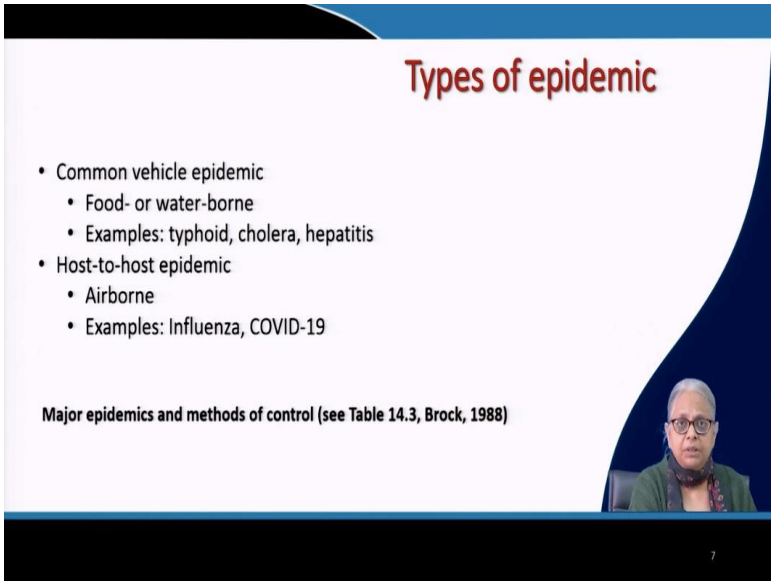

Let us take a look at some types of epidemics. We are living, like I said, we are living right through the COVID-19 pandemic and most of us have become quite familiar with the word epidemic. And let us take a look at some of the methods of control and the types of epidemics. So, there are 2 major categories. One is called the common vehicle epidemic and the second is called host-to-host epidemic. Let us take a look at the first one.

In our country, especially in India, in the monsoon period, you will find that the incidence of food and waterborne diseases generally goes up. And this is because you have waterlogging and because of waterlogging, the water supply lines are often contaminated with sewage. And sewage contains faecal matter. So, faecal matter from healthy as well as infected people is now contaminating the water supplies. So, the incidence of these food and waterborne diseases tends to be higher during the monsoon season, especially in areas where waterlogging happens.

So, the simplest examples in our country are typhoid, cholera, hepatitis. These are normal; widespread occurrences of these diseases (that happen frequently during the monsoon). When you have host-to-host epidemics, just like the one that we are living through, the COVID-19. That is a host-to-host epidemic where the route of (transmission), direct contact between an infected person and a not infected person is likely to transmit the virus. So, that is an airborne virus.

We are calling it an airborne virus. Some of us are very particular and call it an aerosol-associated virus. Regardless, it is coming into the air either through nasal droplets or through droplets from the mouth and so on. Whatever the carrier is, whatever it is present in and with the air, it may be direct contact or contact with these droplets, either way, this is a host-to-host epidemic. And influenza also fits into the same scheme of things.

Let me also share some more examples of epidemics and methods of controlling them. This is from table 14.3 from the text. So, examples of common vehicle epidemics, I have already mentioned that typhoid, paratyphoid, bacillary dysentery, brucellosis; these are all examples of common vehicle epidemics. We have already seen the infective organism and decontamination methods.

For typhoid fever, the best thing to do is decontamination of the food and water that are suspected to be contaminated. That is one thing. And the second thing is the pasteurisation of milk. And the third thing is vaccination. For bacillary dysentery, it again has its roots in contaminated food and water. Detection and control of carriers, inspection of food handlers and decontamination of water supplies are the methods of control.

And that from our point of view is what we need to focus on. Then we come to host-to-host epidemics. So, we have respiratory diseases, let us say like diphtheria. Diphtheria is caused by *Corynebacterium diphtheriae*. The sources of infection are infected people and infected food and fomites. Fomites are basically when you have surfaces that are contaminated by the infective agent, by the bacteria. If someone else comes and touches it, they are likely to carry the infected organism into their system and therefore get infected.

So, immunization with the diphtheria toxoid and quarantine of the infected individuals is the best way to control it. I will come to the next one that is listed in the table.

That is tuberculosis. It is fairly common in India; not very common, but frequently, it has been detected. *Mycobacterium tuberculosis* comes from the sputum of infected human beings and from contaminated milk as well. So, early and extensive treatment of all cases is necessary and pasteurisation of the milk is also necessary.

Influenza which comes from the flu virus; infected human beings are the sources of infection. And the only thing that can be done to control the spread of influenza is vaccination. So, these are some of the examples of major epidemics and methods of control.

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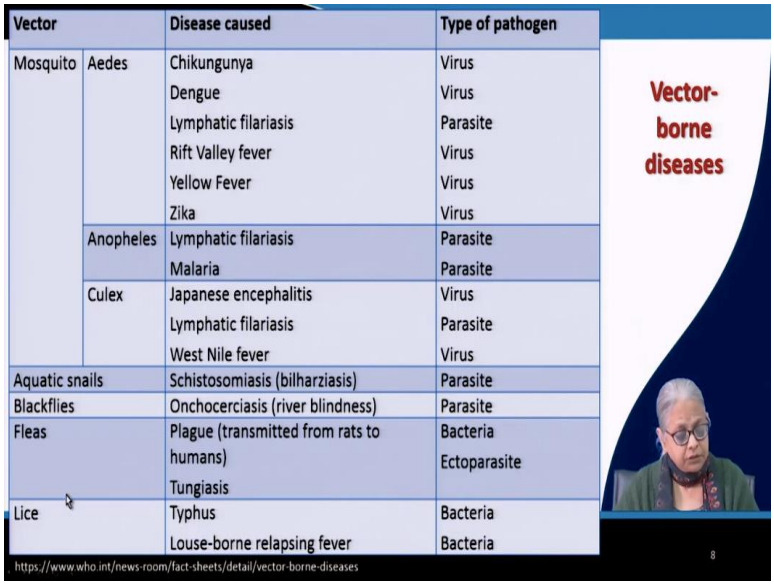

We can now also look at vector-borne diseases. So, having seen airborne, foodborne, waterborne, let us take a look at the next one, which is vector-borne diseases. The biggest one, the biggest vector, if you might say, is the mosquito. These mosquitoes of different species, *Aedes aegypti, Anopheles, Culex*; these are some of the common mosquito species which are carriers of other pathogens. And these other pathogens can be either viruses or protozoa; they are all parasites. And with the *Aedes* species of mosquitoes, the diseases that are caused are chikungunya; dengue, which are viral diseases. You have *lymphatic filariasis*, which is due to a parasite or a protozoa. Then you have rift valley fever, yellow fever, zika fever; all of them are viral fevers.

Then you have *Anopheles*. The most common disease that is associated in India with mosquitoes is malaria. And that is also caused by; the actual causative agent is Plasmodium. And there are several species of Plasmodium, *Plasmodium falciparum, Plasmodium vivax*. All these are common malaria causing agents that are carried by the mosquito and injected, literally injected with a mosquito bite into a human being causing this infection.

Then we come to Culex. Culex is the carrier for Japanese encephalitis and West Nile fever which are both virus-related; and *lymphatic filariasis* which is protozoan or parasitic. There are several other vectors that are mentioned over here, aquatic snails, blackflies and I will mention plague. Plague is carried by fleas from rats, rodents and so on into human beings. So, the fleas that bite the rats which may be carrying this bacterium called *Yersinia pestis*; when it bites a human being, it is going to transmit the bacteria from the rat to the human being. So, that was the causative agent of the plague and so many other problems associated with that. I will stop at this point.

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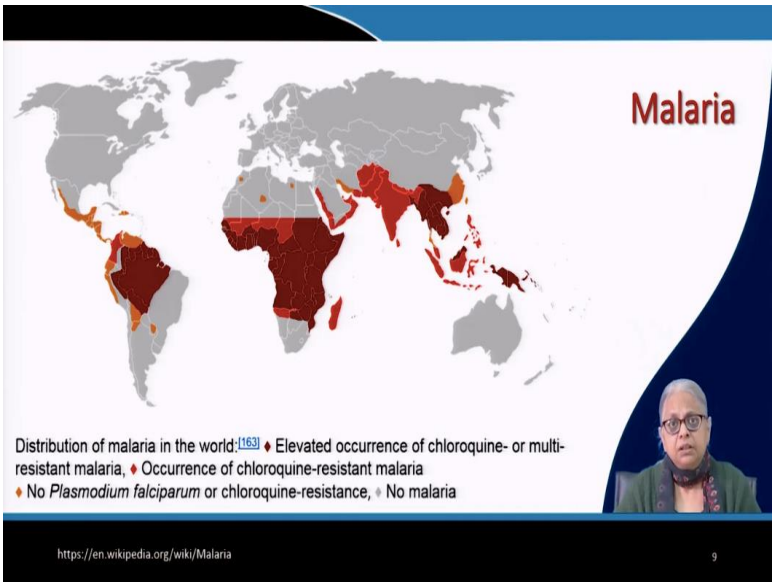

And let us take a look at malaria. Our country is one of the countries that are fairly severely affected by malaria for, it has been around for, God knows how long. You can see here in this map, the distribution of malaria all around the world. Now, all areas of the world are not equally infected or affected by this particular disease. You have elevated occurrence. Now, there is something that is mentioned in the WHO documents as well as other documents that the *Plasmodium* species that is the causative agent of malaria, yes, has become resistant to chloroquine and to other drugs perhaps.

So, what you see in this map is a dark brown set of geographic areas where the elevated occurrence of chloroquine and multi-resistant malaria is already known. In our country, we do have chloroquine-resistant malaria. So, it is no longer effective. And there are other parts of the world, much smaller areas where there is no resistance to either *Plasmodium falciparum* or to chloroquine. And then there are much larger areas where there is no malaria at all. So, you can see that malaria is very frequent in certain parts of the world. And that is clear over here.

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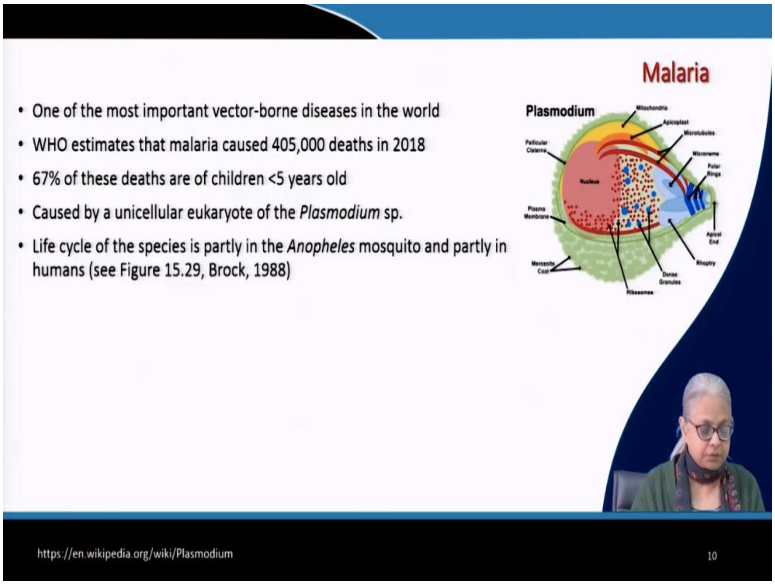

Like I said, it (malaria) is one of the most important vector-borne diseases that is known even to this day and WHO, the World Health Organisation estimates that malaria caused at least 405,000 deaths in 2018. 67%, the bulk of these deaths; two thirds of these deaths were of children below 5 years of age and the causative agent is the *Plasmodium species*. And this *Plasmodium* species is shown over here.

And you can see, it is quite different from other types of microorganisms that we are familiar with. You have a nucleus. You have many of the general organelles that we associate with microorganisms are there. There are mitochondria, there are ribosomes, plasma membrane and so on. But then, there are several other organelles which we are not going to go into, that are also present and particularly important in terms of the virulence of this particular microorganism.

Let me come to the lifecycle of this *Plasmodium* species. So, you can refer to the figure mentioned over here, 15.29 in the text. And I will just briefly describe to you how malaria is transmitted. So, let us say you have an infected human being and you have the infected merozoites that are present in the red blood cells of the infected person. They will actually be expelled into the blood. And they will be producing haploid cells. And these haploid cells are the gametes of the actual diploid cell, which will then further reproduce. So, when a mosquito has these haploid cells in its gut, in its blood, these gametes, both the male as well as the female will be fertilised. They will produce the zygote; the zygote is diploid; and this diploid zygote will grow and result in what are called sporozoites and these sporozoites will be released. When the mosquito bites another human being, these sporozoites will be injected into the blood of that person, from where they will be transferred to the liver, and they will enter into what is called the exoerythrocytic stage. And it will form many other types of cells and cause infection

of the red blood cells. So, that is the life cycle of malaria disease. You can see, part of the life cycle is completed in the gut of the *Anopheles* mosquito and part of it in the human, infected person.

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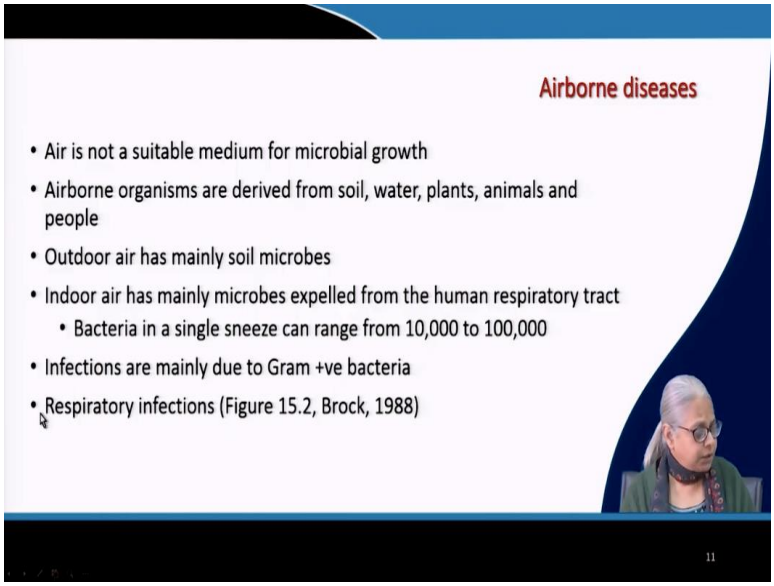

Let us now come to airborne diseases. Air, like we all know is not very suitable for microbial growth, but that does not mean that microorganisms do not exist in air. They are constantly being expelled into the environment by infected human beings, animals and so many other organisms. So, airborne organisms are present, but they do not grow in air. They are there because of other sources. So, airborne organisms are derived from soil, water, plants, animals, people and so on. If you were to take a simple test, you can wipe any surface or you can take your Petri dish with agar in it and just go around the room and let it be exposed to air. Take a sterile Petri dish with agar in it and expose it to air for less than a minute and incubate it after that, you will find it is teeming with all kinds of organisms. So, you will have bacteria, you will have fungus, you will have protozoa; all kinds of organisms will be found on the agar media after incubation. So, that is sort of evidence of the fact that they exist in the air, but they do not grow in the air. So, outdoor air has mainly soil microbes. Indoor air has mainly microbes expelled from the human respiratory tract. So, a bacteria in a single sneeze, the number of bacterial cells can range from 10,000 to 100,000 cells.

Most infections, most airborne infections are due to gram-positive bacteria. And I will also mention another schematic, that is there in the textbook, of respiratory infections. I will just mention some of the common respiratory infections that are mentioned in this figure. So, the

nasal cavity is often colonized by *Staphylococcus aureus*. This is a pathogenic species. The oral cavity, the mouth is colonized by *Streptococcus pyogenes*.

You have the pharynx, the larynx. The larynx is the site of the influenza bacteria; so, *Haemophilus influenzae*. Then you have the primary bronchi or the bronchus which is the (site of the) influenza virus. The secondary bronchus has another species, which I am not familiar with. And then you have alveolar ducts, the final endpoints of the lungs. So, you have the alveolar ducts which are colonised by *Coxiella*, and you have alveolar sacs which are colonised by *Chlamydia*. So, these are some of the respiratory infections which can go from the upper respiratory tract all the way to the ends of the lungs.

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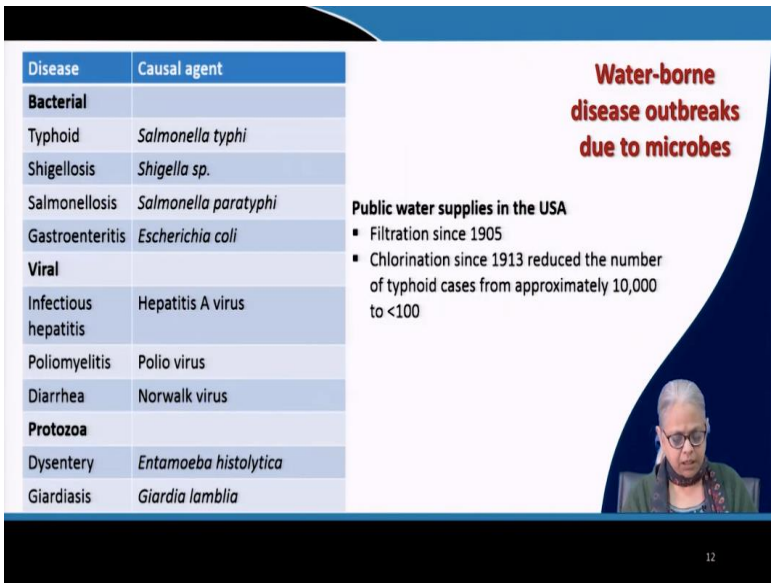

We then come to waterborne disease outbreaks. Most of you are aware, like I said, the monsoon season has always been a time when waterborne outbreaks are very common. And often you hear your friends, family or even the government sending out messages saying that: do not eat food or drink water from sources that are not 100% uncontaminated or safe. So, vendors; you do not want to go to vendors that are selling food and water or beverages for that matter.

So, these kinds of things are very common, especially in our country, in the monsoon season. These are all examples of waterborne disease outbreaks due to microorganisms. And you can have all 3 categories of microorganisms. You can have bacterial, viral, as well as protozoa outbreaks, or rather disease outbreaks due to these kinds of microorganisms. I have already mentioned that typhoid is caused by *Salmonella typhi*.

Shigellosis is caused by various *Shigella species*. Salmonellosis is caused by various *Salmonella species*. Gastroenteritis; many organisms are involved, *E. coli* being one of them. And like I said, *E.coli* is an opportunistic pathogen in many cases. And then you have viral

diseases like infectious hepatitis, which is caused by Hepatitis A virus; poliomyelitis, which is caused by the Poliovirus; diarrhoea, which is caused by the Norwalk virus. Then you have dysentery, which is caused by *Entamoeba histolytica*. And then you have Giardiasis by *Giardia lamblia*. Now, these are all examples of various types of microorganisms that may be present in contaminated water and perhaps even in food.

One of the things from the environmental engineering and science point of view is that for us, control of these outbreaks is our…..; it is within our ability to control these outbreaks; and that is why we are studying microbiology and all of that.

So, one of the biggest interventions, you might call them treatment technologies that are used and that have been successful in reducing the incidence of these waterborne disease outbreaks, is basically due to water treatment. And I will just give you an example. So, public water supplies, especially in the U.S. have been using filtration. So, we have rapid sand filters, which have been in use, perhaps as far back as 1905; and chlorination which has been literally the backbone of water supply systems, public water supply systems since 1913. And that has been credited; these 2 processes together have been credited for reducing the number of typhoid cases in the U.S. from approximately 10,000 to less than 100. So, these are all technological interventions that have helped to bring down the incidence of both morbidity and mortality due to waterborne diseases.

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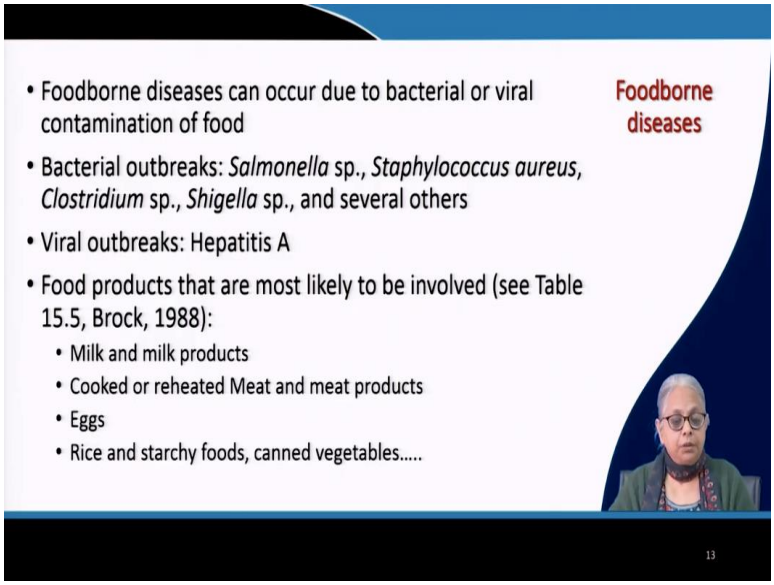

Foodborne diseases can occur, like I said, due to bacterial or viral contamination of food. We have bacterial outbreaks. So, these bacterial outbreaks; I have already mentioned most of the species. And what are the food products that are most likely to be contaminated by these pathogenic organisms? We have milk and milk products; we have cooked or reheated meat and

meat products; we have eggs, rice and starchy foods, canned vegetables. These are all highly vulnerable to contamination by these pathogenic species.

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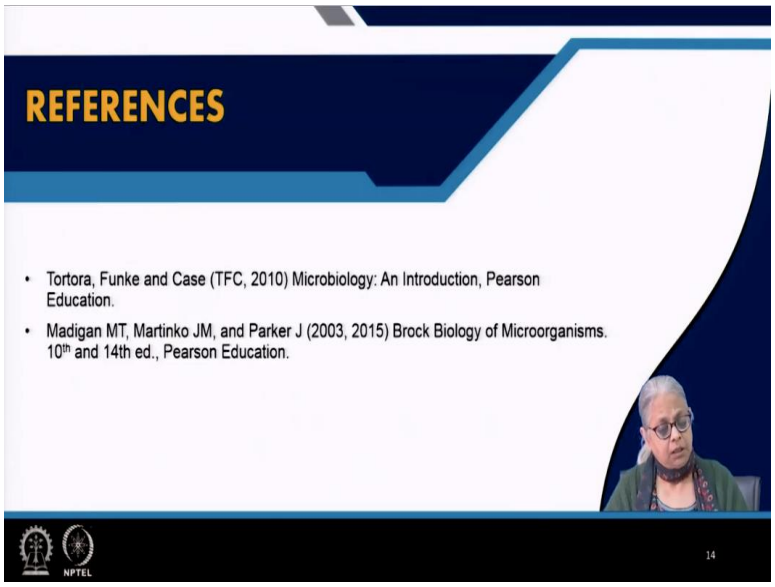

I will stop at this point. Thank you. And this brings me to the end of this topic.