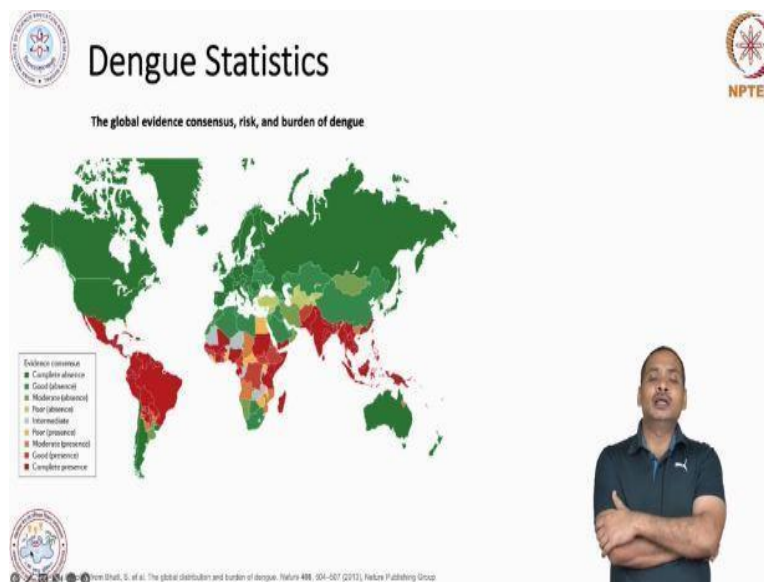


Host-Pathogen Interaction (Immunology)
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Lecture-70
Arbovirus and Dengue Virus Infection

Hi. So, in previous session we have discussed about the arboviruses and now we will take a couple of example about the arboviruses and we will discuss in little more detail about those arbovirus or the disease caused by that arbovirus. So, in this session we will take the dengue virus, which causes dengue.

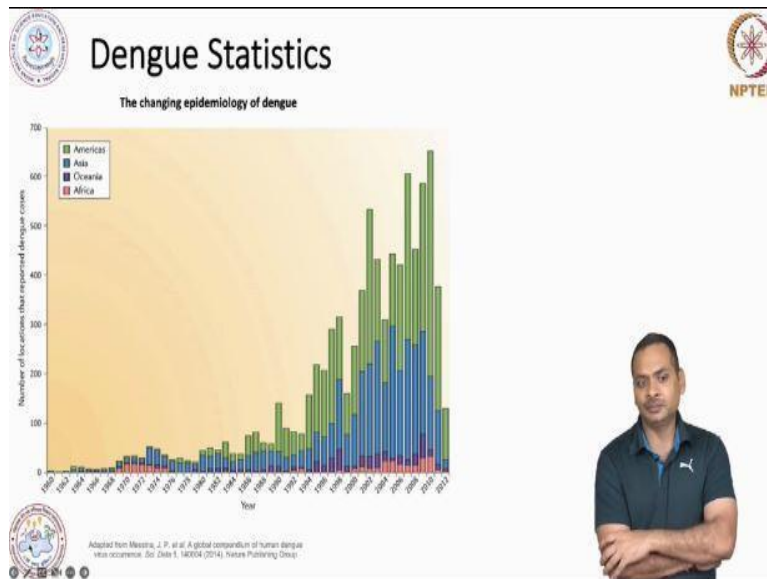
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So, here I would like to show you the statistic of dengue, here you can see that it is severely affecting various parts of the world and particularly our country, here you can see that in India every part is having this dengue virus infection. If you see this image then you can understand that this virus or this viral infection is prominent near the equator region. So, you can understand why this disease is dominant near the equator region.

You can understand that over there is a little higher temperature, which is a very good breeding factory for these arthropods. If you go up then the temperature is very low and there is almost no cases of dengue virus infection.

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And more interestingly this infection these dengue incidences are increasing with the years, here you can see this graph. So, the number of this reported cases are increasing with the year, here I am showing you the statistic up to 2012 and this is increasing in America, in Asia, Oceania and Africa. So, over there you can see in Asia also there is a quite good rising number of dengue infection, dengue, chikungunya and all those things.

So, this you can understand, the season for this disease is when the temperature is not very high, not very low. At that time, these mosquitoes are very active. And generally, this occurs immediately after the monsoon in India is particularly I am talking about. After a monsoon, these mosquitoes breed a lot and then they infect the individual. So, this is important thing to note.

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Dengue

Dengue virus


- The dengue viruses (DENV) are members of the *Flavivirus* genus of the *Flaviviridae* family
- DENV contains a single-stranded, positive-sense RNA genome
- It can cause the full spectrum of clinical manifestations following DENV infection, ranging from asymptomatic infection to dengue fever (DF) and the most severe disease, dengue hemorrhagic fever (DHF) and dengue shock syndrome (DSS)

from Blomender, CDC

So, how this virus looks like and what kind of things it has let us look at. So, the dengue viruses are members of the Flavivirus family Flaviviridae family, basically. And it is a genus is a Flavivirus and the family is Flaviviridae Flaviviridae is the family. The dengue virus contains single-stranded positive-sense RNA genome. It may cause a full spectrum of a clinical manifestation followed by dengue infection, which is ranging from asymptomatic. So, many people are not showing any symptom.

And this is true when there is a first infection. When there is a first infection, then in general the individuals show very mild symptom. And when there is a second or subsequent infection with a little deviated or different strain then that result to the severity. And this severity you can define as a dengue fever, dengue hemorrhagic fever and dengue shock syndrome. And these conditions particularly dengue hemorrhagic fever and dengue shock syndrome can cause the fatality. So, this is quite dangerous.

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The slide is titled "Dengue" and features two photographs of mosquitoes. The left photograph shows a mosquito with a red abdomen, labeled "Aedes aegypti". The right photograph shows a mosquito with a black and white striped abdomen, labeled "Aedes albopictus". Below the photographs is a bulleted list of facts:

- Aedes aegypti is the main vector for dengue transmission to humans
- Aedes albopictus can also sustain transmission
- About one in four people infected with dengue will get sick
- About 1 in 20 people who get sick with dengue will develop severe dengue
- Severe dengue can result in shock, internal bleeding, and even death

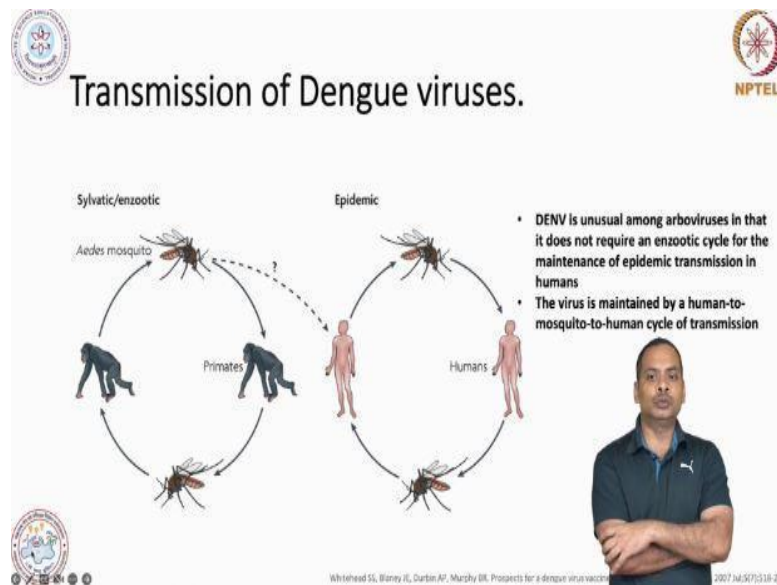
The slide also includes logos for the institution and NPTEL in the top corners, and a small inset image of a man in a blue shirt in the bottom right corner. A small text "from @orender, CDC" is visible at the bottom right of the slide.

So, here you can see that how the mosquito looks like. The dengue is spread by this Aedes aegypti and this is the main vector for dengue virus transmission to humans. There is another species also Aedes albopictus, this can also transmit the virus. About one of four people infected with a dengue will get sick. And about 1 in 20 who gets sick with dengue will develop severe dengue symptoms.

Severe dengue can result in shock, internal bleeding like hemorrhageoids that is why we call it as a dengue hemorrhagic fever. There will be internal bleeding and if it is not managed if the

patient is not receiving appropriate health care support, then that may result to the death of individual, it is a very fatal, very dangerous.


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Transmission of this dengue viruses are basically, in general this dengue virus is a transmitted from human to mosquito and mosquito to human. This cycle is a predominant, people has a little evidences that there is a sylvatic transfer of the virus. Basically, this Aedes mosquito infects a non-human primate. And over there this will be in cycle the number will increase, then mosquito will infect then this primate will again infect. This is one cycle.

Basically, this cycle maintains the viruses. Generally, those primates are not severely affected, but they are the reservoir and they can provide the virus, so for example this mosquito if it will jump or if it will bite to the human, then that will be in human circulation. So, the involvement of this pathway is this sylvatic or enzootic cycle is not very prominent in case of dengue. There is a few evidences, but it is not very prominent for the dengue viruses. Basically, this virus is maintained by human to mosquito to human cycle of transmission.

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Different stages of Dengue Fever


a Dengue fever (DF)

Febrile illness with 2 or more of the following: headache, retro-orbital pain, myalgia, arthralgia, rash, leukopenia, haemorrhagic manifestations, and virus recovery, serological response or temporal occurrence with other cases

b Dengue haemorrhagic fever (DHF)

Dengue haemorrhagic fever (DHF)		Grade
Fever 2-7 days	Petechiae, bruising, or (+) tourniquet test	I
Other haemorrhagic manifestations	Rising haematocrit Hypoproteinaemia Serum effusion	II
	Hypovolemia Weak pulse Hypotension	III
Severe bleeding	Disseminated intravascular coagulopathy	IV

Whithead SJ, Wainy JJ, Durbin AP, Marshy JR, Prospects for a dengue virus vaccine



There are different stages of dengue fever, here I will discuss that. So, there is a dengue fever, which is basically characterized by a febrile illness with two or more of following symptoms that is a some individual show a severe headache, some **individuals** also retro-orbital pain near eye region, some individuals show the myalgia and arthralgia. Myalgia is a basically a muscle pain, we call it as a myalgia as a muscle pain. Arthralgia is the joint pain.

The meaning of dengue itself is a bone breaking fever. So, this is the literal meaning of dengue. There will be a rashes, there will be a leukopenia reduced number of white blood cells; there will be a hemorrhagic symptom; hemorrhagic manifestation and one can recover the virus. So, serological responses or temporal occurrence with other cases. So, these are the characteristic of dengue fever. And it is associated with 2 or more these symptoms.

Here you can see that, there is a different grade of dengue hemorrhagic fever; here you can see that grade 1, 2, 3 and 4. So, basically grade 1 is included that there will be a fever for 2 to 7 days. There will be a petechiae is nothing it is a there will be appearance of red spot or dot over the skin, red or dark brown. Basically, this spot is due to internal bleeding.. There is a bruising and there is some test, tourniquet test.

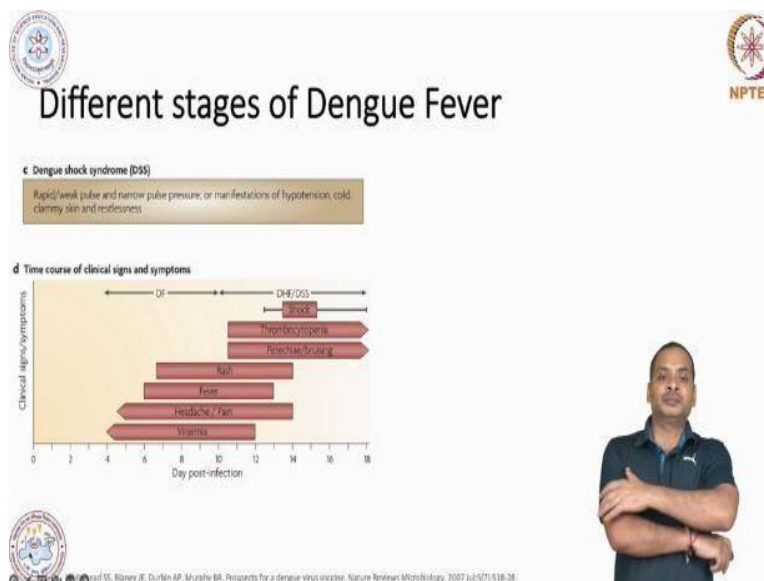
So, this test is basically the doctor perform in order to find out the capillary fragility. So, you can understand these capillaries are quite fragile when there is a dengue virus infection, because this virus infects the endothelium of these blood vessels. So, if it is a severely damaged then these capillaries or blood vessels will be more fragile. So, there is a test, this is a more clinical component.

Grade 1 is also characterized by increased blood vascular permeability, there will be a thrombocytopenia. So, there will be a reduced number of blood platelets. The grade 2 is characterized by this is other hemorrhagic manifestations, rising in haematocrit, hypoproteinemia, there will be a reduced amount of protein in serous effusion. So, this is the characteristic of grade 2.

Grade 3 is a hypovolemia, there is a reduced volume of the blood in the individual, which is associated with weak pulse and hypotension and there could be a possibility of coagulopathy. So, grade 4 which is more risky and dangerous is characterized by severe bleeding. There will be a quite a lot of bleeding, even if individual will pass this stool that will be a very dark colour, because of blood, because there is a lot of internal bleeding and that internal bleeding is resulting to the ~~plaque~~-black stool.

There will be a profound shock, there will be a severe hypotension condition, pulses will be very weak, heart beats will be also slow. And there will be a disseminated intravascular coagulopathy. You can understand there is a reduced number of thrombocyte and all this result to this disseminated intravascular coagulopathy, which is quite a complicated situation. And this situation is basically managed in a critical care wards like ~~intensive care unit~~intrinsic area network.

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Here, I will tell more about the dengue shock syndrome, which is basically rapid and weak pulses and narrow pulse pressure or manifestation of a hypotension ~~cold, altered~~, clammy skin

and restlessness. Clammy skin is nothing it is a there will be a sweat or the skin will be wet that condition we call it as a clammy skin. Restless, you can understand in Hindi we call it as a bechanihona, the individual will be very in this thing restless.

And here you can see the time course of this symptom. Initially, it started with a fever some headache. Initially, the first symptom there will be some headache or pain and then that will be accompanied with a fever and with time there will be a appearance of rashes mainly hand, legs and this trunk in front side or back side. And generally, the things are managed over there.

But if it is not managed then that individual may enter in the dengue hemorrhagic fever condition or dengue shocks syndrome condition, which is consists of this is a petechiae, bruising, thrombocytopenia and the shock. So, shock is a more complicated situation.

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The slide is titled "Different serotypes of Dengue" and features the NPTEL logo in the top right corner. It contains a bulleted list of key points and a diagram illustrating the process of antibody-dependent enhancement (ADE). The list includes: "Based on the neutralization assay, four genetically similar but antigenically distinct serotypes of DENV identified are DENV1, DENV2, DENV3, and DENV4"; "Infection with one serotype of DENV confers lifetime immunity against that serotype but not against heterologous serotypes"; and "Secondary infections with a heterologous serotype can lead to severe complications like DHF and DSS due to the process called Antibody-dependent enhancement". The diagram shows DENV particles being taken up by a monocyte. It depicts the interaction between DENV and antibodies, including "Heterotypic Ab from previous infection" and "FcγR". This leads to "Increased viral load" and "Disease". A presenter is visible in the bottom right corner of the slide.

There are different serotypes of dengue here; there is some very important point, which you need to understand. So, based on neutralization assay there are four genetically similar but antigenically distinct serotypes of dengue is there which we name it as a dengue 1, 2, 3, 4. Infection with 1 serotype of dengue confers basically lifetime immunity against that serotype but not against heterologous or other serotypes.

For example, if individual is infected with dengue 3, so that individual will have a lifetime immunity against the dengue 3, but the if the same individual is infected with say dengue 1, 2 or 4 then that is a more complicated situation. Why because second infection with heterologous serotype can result or lead to severe complication that will cause more severity

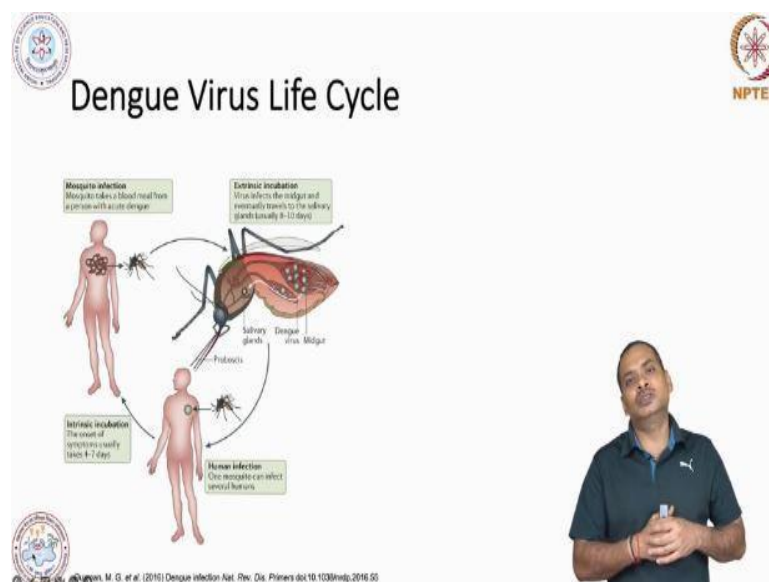
like a dengue hemorrhagic fever, dengue shock syndrome and due one phenomena, which we call it as a antibody-dependent enhancement, which I have mentioned in previous session. So, what is this antibody-dependent enhancement?

Here, for example, individual is infected first time it is infected by dengue 3. And now second time it is infected with dengue 1, 2 or 4. So, the individual will be having the antibodies against dengue 3 and this antibody is weakly binding with the dengue 1, if it individually second time infected with 1 or 2 or 4. So, you know that once this antibody binds with this virus then there will be Fc receptor over the phagocytic cells or immune cells.

And then it will be phagocytosed or it will be taken care by the complements and so on so forth. Here, what it is doing? Once it is attaching with the antibody, it is facilitating its infection to the immune cells like a macrophages, which is a myeloid origin; it can facilitate multiplication in lymphoid origin cells. So, after attaching it is very readily entering in the target cell and over there instead of it is digested and cleared it is basically using this facility to infect the cell more and increase more replication and that result to the very high viral load.

And that result to the dengue hemorrhagic fever and dengue shocks syndrome condition. I hope you understand this thing very well and that result to more complicated disease.

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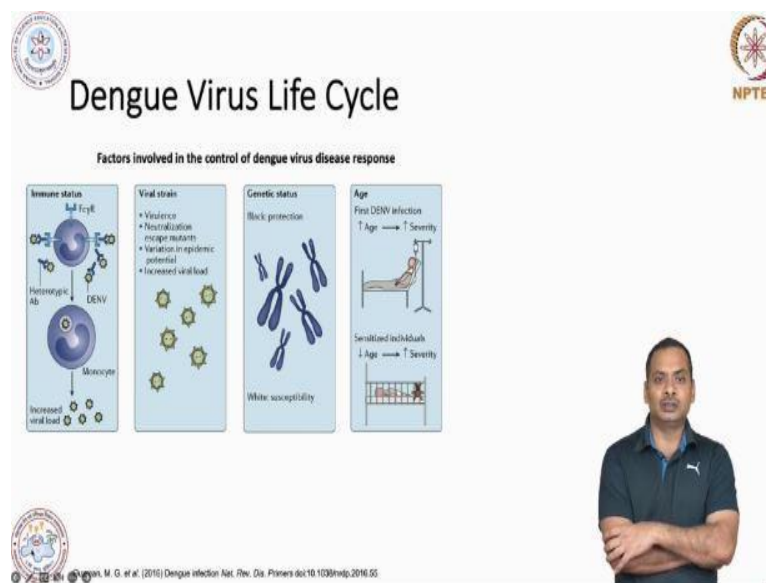


So, dengue virus life cycle; here you can see there is a mosquito and mosquito takes a blood meal from the person with acute dengue. And then there will be extrinsic incubation period, where virus infect the midgut of the mosquito and travels to the salivary gland, over there it

usually it takes about 8 to 10 days. Over there, they will multiply, these viruses will multiply and then when this mosquito will bite another healthy individual.

So, one mosquito can infect the several human; that is very interesting to know. It is not that one mosquito bite will exhaust that mosquito for the viruses and subsequent infection, but if one mosquito is infected if this mosquito bites several human, then this mosquito can infect the dengue virus to all those individuals. Then there will be intrinsic incubation period and the onset of symptom, which usually takes 4 to 7 days. So, then this cycle of this virus is completed.

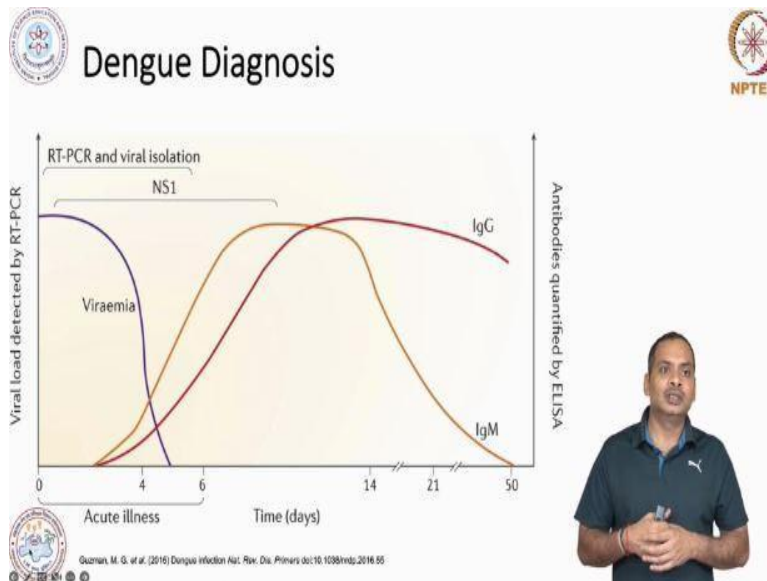
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So, there are factors involved in controlling of dengue virus disease response, one is that immune status as I have explained you. There is a viral strain: virulence, neutralization escape mutants, variation in epidemic potential and increased viral load. Genetic status of course, genetic plays a very important role not only defense in all aspects. So, genetics is also there. There is another factor, which is age.

First dengue virus infection that will not cause so much severity, but in sensitizing individual if there is a heterologous dengue virus infection, then that will cause the severity and that depends on the age. If the individual is very young or if the patient is very elder then that will affect quite severely. The outcomes could be reasonably bad.

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Here, this is a dengue diagnosis. There are of course one can easily detect this virus. At initial phase by simple RT-PCR for viral gene generally people use this NS1 gene. And one can also detect these antibodies, dengue specific antibody that is IgG and IgM and here you can see that duration. And in that way one can diagnose the disease and this is also prognostic marker. When this antibody declines, particularly IgM then one can say that the individual is recovering.

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Dengue Treatment and Prevention

- No specific treatment for dengue is currently available
- Analgesic and antipyretic drugs, such as paracetamol
- Fluid therapy is key to dengue management and is applied based on disease severity
- Prophylactic platelet transfusions are given for dengue fever with thrombocytopenia to prevent hemorrhagic complications
- Bed nets or repellents to avoid mosquito bites

Vaccine type	Developer	Approach	Current status
Live attenuated	Mahidol University	Live attenuated tetravalent virus isolates	Not being tested
	WRAIR and GSK	Live attenuated tetravalent virus isolates	On hold
	US NIAID, US NIH and Instituto Butantan	Recombinant live attenuated, directed mutagenesis and DENV-DENV chimeras	Phase 3
	Azambis and Sanofi Pasteur	Live attenuated tetravalent chimeric YFV-DENV (CYD-TDV) (also known as Chimerivax)	Registered in several countries
Subunit	US CDC, Inovagen and Takeda	Live attenuated chimeric DENV2-DENV (DENVax)	Phase 3
	Hawaii Biotech and Merck	Truncated recombinant envelope protein (DEN-80F)	Phase 3
Inactivated	IPK and CIGB	Tetravalent recombinant subunit vaccine comprising domain III of the envelope protein fused to the capsid protein (DIII-C)	Phase 3
Inactivated	WRAIR and GSK	Purified inactivated virus	Phase 3

Guzman, M. G. et al. (2016) Dengue infection. *Anal. Rev. Dis. Primers* doi:10.1038/nrdp.2016.55

So, dengue treatment and prevention: So, now as I told you for all arboviral infection there is no specific treatment and this is true for dengue. There is no treatment currently available for dengue. Basically, the patient is treated with a paracetamol, which is antipyretic and analgesic but try to understand there is a limit the individual cannot receive more amount of drug even though the symptom is not reducing.

Fluid therapy, since there is a reduced in the volume of blood, so they were continuously given the fluid, but this is also associated with some problems, sometimes the indeed will receive too much fluid and that is also resulting the problem. And when they receive intravenously the fluid then there will be an inflammation of blood vessel due to this catheter. So, all those things need to be managed.

The prophylactic platelet: So, if platelets are reducing, then the individual can receive the platelets and then that can be managed if the thrombocytopenia are quite severely reduced and of course there is a precaution: one should use the net while sleeping, one can use some insect repellent so and so. So, there is a huge effort to make the dengue vaccine, there is a live attenuated; there is a subunit; an inactivated vaccine.

So, here you can see all this thing, but none of them are so far successful few were started in order for trial. However, they showed the negative result. Basically they trigger the antibody-dependent enhancement. So, currently, we do not have a meaningful vaccine against dengue virus. So, this is all about the dengue virus and with this I will stop here. And in next session, we will discuss about the Zika virus. Thank you.