MICROBIAL BIOTECHNOLOGY

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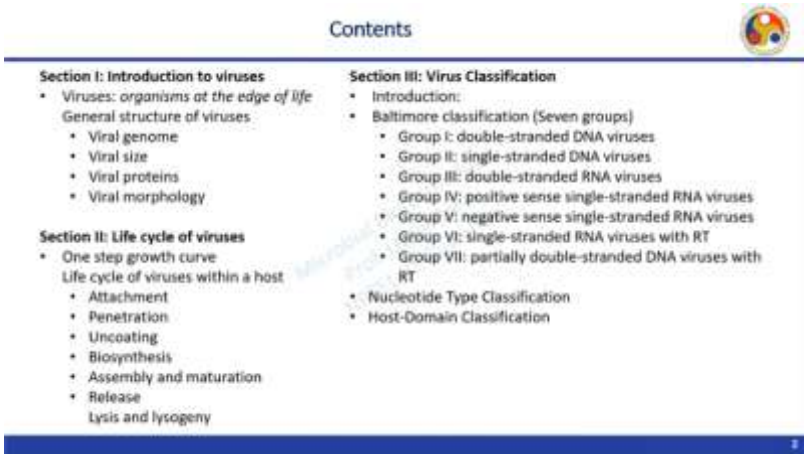
Lecture05
Lec 5: Structure, life cycle and classification of viruses

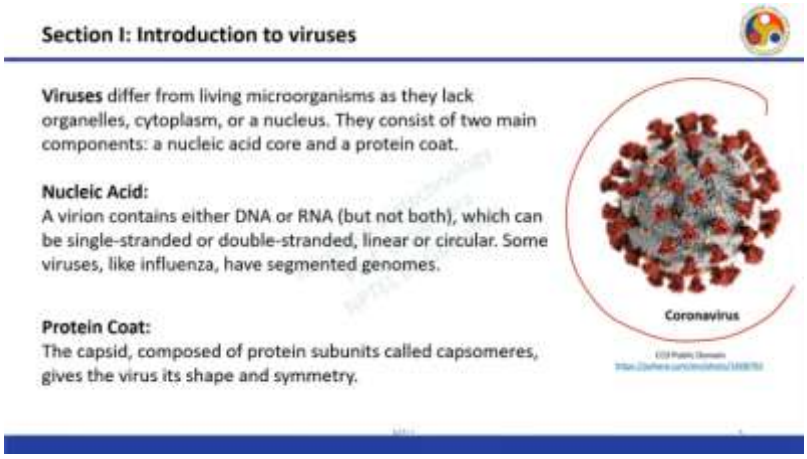
Welcome to my course on microbial biotechnology. Today, we are going to start Module 2, which will deal with the structure and life cycle of representative groups of viruses, prokaryotic, and eukaryotic microorganisms. Let us start with the structure, life cycle, and classification of viruses because viruses are altogether different from other microorganisms. So, this lecture is divided into three sections. We will start with an introduction to viruses, where we will discuss the general structure of viruses regarding their genome size, viral particle size, viral proteins, and morphology.



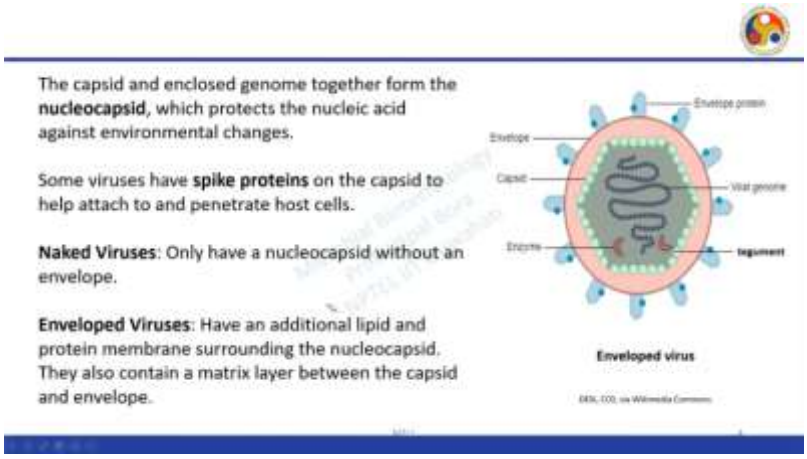
Then, we will go on to discuss the life cycle of viruses and the one-step growth curve. Finally, we will deal with virus classification. We'll have an introduction to various virus classification schemes and then deal at length with the Baltimore classification. So, let us try to understand what a virus is. A virus differs from living organisms as it lacks organelles, cytoplasm, or a nucleus.

It consists of two main components: a nucleic acid core and a protein coat. What is this nucleic acid core? A virus contains either DNA or RNA but not both at the same time, and these can be single-stranded or double-stranded, linear or circular. Some viruses, like influenza, have segmented genomes.

Now, what is the protein coat? The protein coat, basically referred to as the capsid, is composed of protein subunits called capsomers, and these give the virus its shape and symmetry. In this picture, you can see the structure of a coronavirus with some very interesting features on its surface. We will discuss these in length today.



The capsid and enclosed genome together form the nucleocapsid, which protects the nucleic acid against environmental changes. But there are cases where virions do not contain any capsid, and the nucleic acid stays naked. Many viruses have spike proteins, as you can see in the case of coronavirus in the earlier picture, on the capsid to help attach to and penetrate host cells. As I already told you, there are certain naked viruses; they have only nucleic acid without a capsid or envelope. Enveloped viruses have an additional lipid and protein membrane surrounding the nucleocapsid.



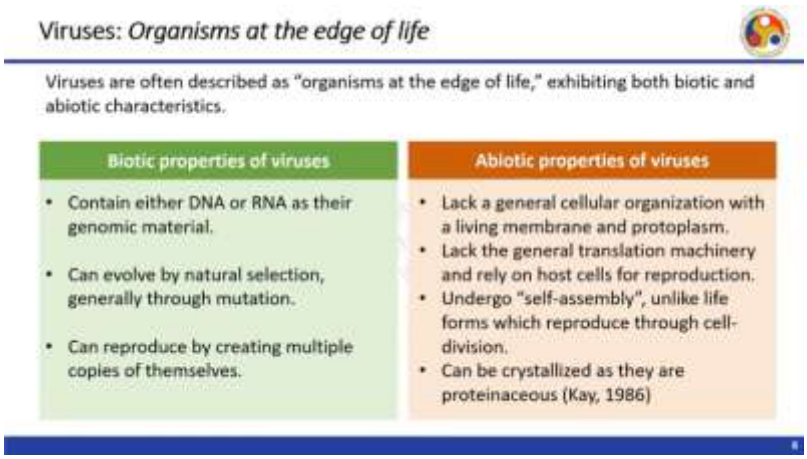
They also contain a matrix layer between the capsid and envelope. In brief, viruses are obligate intracellular parasites. They cannot replicate without a host, making them intermediates between biotic and abiotic forms, or what we generally call the living and

the dead. They infect organisms of all three domains: Eukarya, Bacteria, and Archaea. Since the discovery of the first virus, the tobacco mosaic virus, in the mid-1800s, over 11,000 virus species have been detailed. However, this represents only a small fraction or minuscule, as millions of virus species most likely remain undiscovered.

So, this is the atomic model of a SARS-CoV-2 virus in A, which we have already shown you in an earlier slide. And in B, you can see a tomato leaf infected with tomato TMV, the tomato mosaic virus. And in C, a tomato leaf infected with TMV, another picture. And in D, you can see the atomic model of T4 bacteriophages. What are bacteriophages?

We will discuss this soon. So, viruses are often described as organisms at the edge of life. We refer to them as in between biotic and abiotic forms, and therefore they exhibit both biotic and abiotic characteristics. What are the biotic and abiotic characteristics exhibited by viruses? Let's start with the biotic properties of viruses.

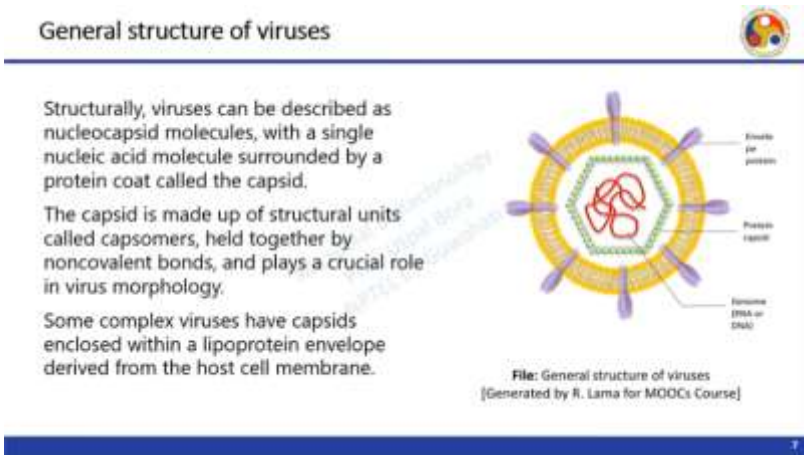
We already told you that they contain either DNA or RNA as their genomic material. Like other living organisms that have nucleic acid, mostly DNA. Viruses can evolve by natural selection, generally through mutation. In fact, the mutation rate in viruses is very, very fast. That is why the influenza virus



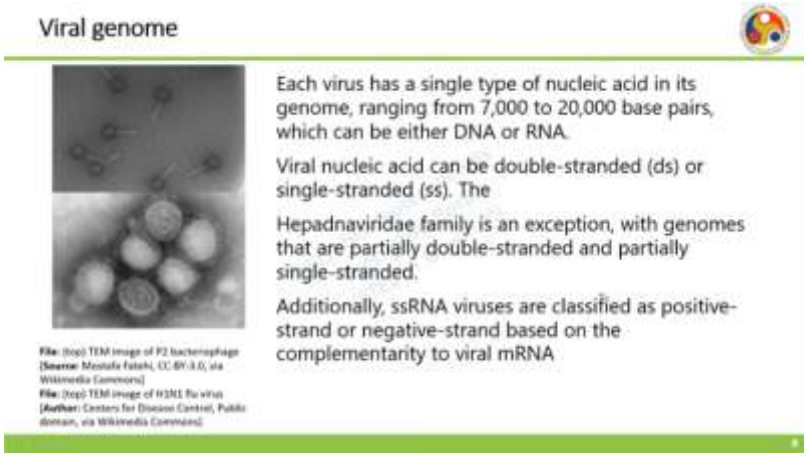
which mutates very fast or several other viruses which mutate very fast are challenges to therapeutic interventions because we cannot develop stable vaccines against them. Viruses can reproduce by creating multiple copies of themselves, but abiotic forms or components cannot do this. So, in this way, they are similar to the living world. What are the abiotic properties of viruses?

Viruses lack a general cellular organization with a living membrane and protoplasm. They lack general translation machinery and therefore rely on host cells for reproduction. Viruses undergo self-assembly, unlike life forms which reproduce through cell division. Viruses can be crystallized as they are proteinaceous. So, these are

some of the abiotic properties of viruses. Now, if you look into the general structure of viruses, we have already described that it can be described as a nucleocapsid molecule with a single nucleic acid molecule surrounded by a protein coat, which we now call the capsid. This capsid is made up of structural units called capsomers, held together by non-covalent bonds, and plays a crucial role in virus morphology. Some complex viruses have capsids enclosed within a lipoprotein envelope derived from the host cell membrane.



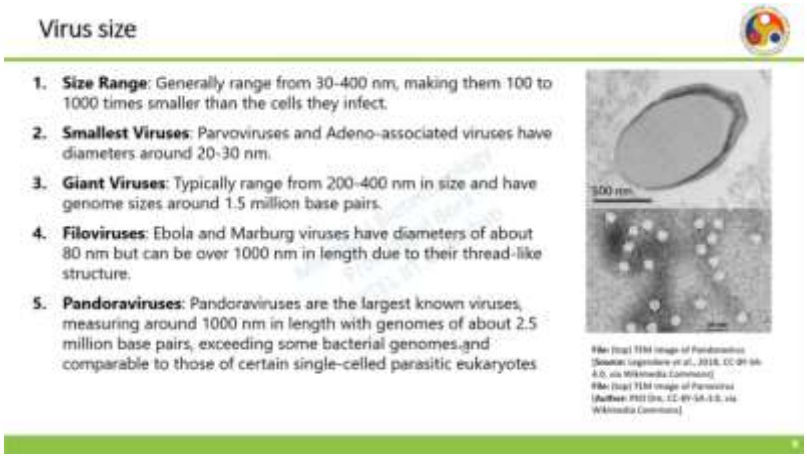
Each virus has a single type of nucleic acid in its genome, ranging from 7,000 to 20,000 base pairs, which can be either DNA or RNA. Viral nucleic acids can be double-stranded or single-stranded. The Hepantiviridae family is an exception with genomes. They are partially double-stranded and partially single-stranded. Additionally, single-stranded RNA viruses are classified as positive-strand or negative-strand based on their complementarity to viral mRNA.



Now, how big or small is a virus particle? Generally, a virus's size could range from 30 to 400 nanometers, making them 100 to 1,000 times smaller than the cells they infect. Parvovirus and adeno-associated viruses have diameters around 20 to 30 nanometers. There are some giant viruses, which typically range from around 200 to 400 nanometers in size and have genome sizes around 1.5 million base pairs.

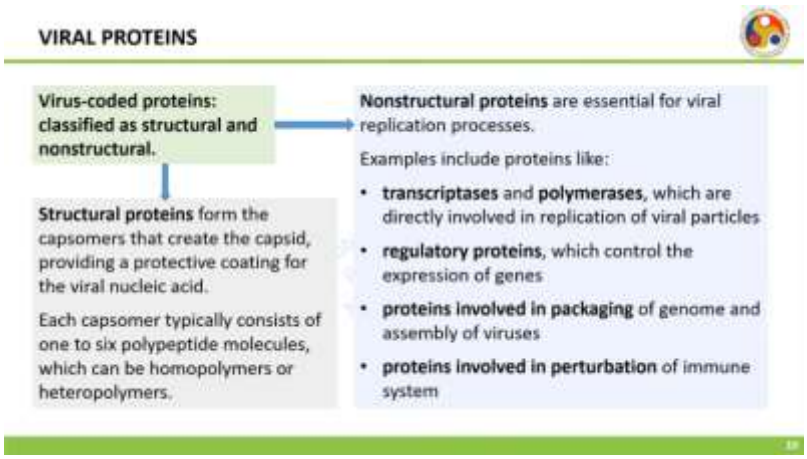
Filoviruses like Ebola and Marburg have diameters of about 80 nanometers but can be over 1,000 nanometers in length due to their thread-like structure. Pandoraviruses are the largest known viruses, measuring around 1,000 nanometers in length and with genomes of about

2.5 million base pairs, exceeding some bacterial genomes and comparable to those of certain single-celled parasitic eukaryotes. So here you can see the same image of Pandoravirus, and this is something around a 500-nanometer scale, and you can very well make an assessment of the giant size of this particular virus.



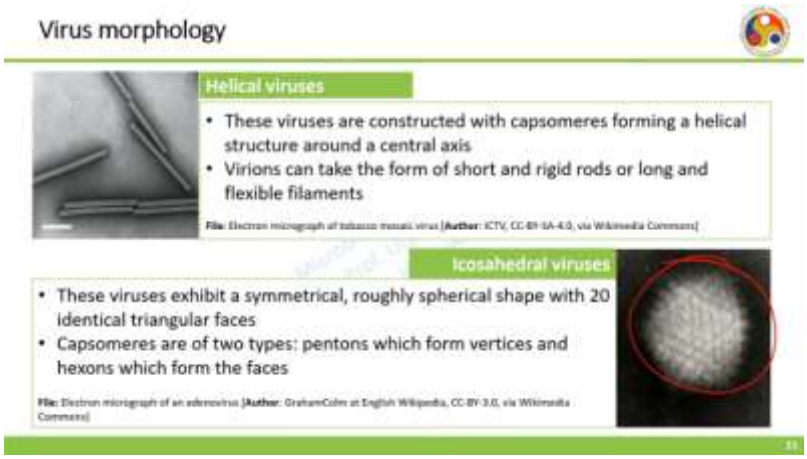
Now let us discuss the viral proteins. Viruses contain two types of proteins. The first type is the structural proteins, and the second type is the non-structural proteins. These structural proteins form the capsomers that create the capsid, providing a protective coating for the viral nucleic acid. Each capsomer typically consists of 1 to 6 polypeptide molecules, which can be homopolymers or heteropolymers.

The non-structural proteins coded by the virus are essential for viral replication processes. Some examples of these proteins are transcriptases and polymerases, which are directly involved in the replication of viral particles. Then there are regulatory proteins, which control the expression of genes. Then there are proteins involved in the packaging of the genome, assembly of viruses, and proteins involved in the perturbation of the immune system. What does a virus look like, or what is the physical appearance of a virus or viral morphology?

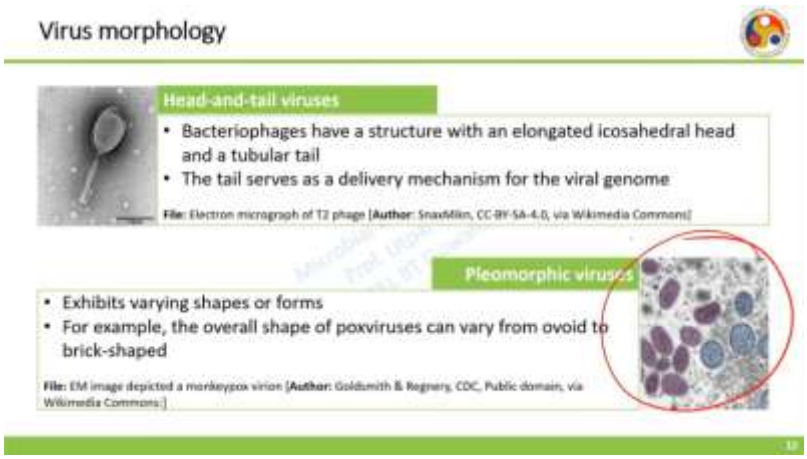


They may be helical in appearance. These viruses are constructed with capsomers forming a helical structure around the central axis. Variants can take the form of short and rigid rods or long and flexible filaments. In this electron micrograph of the tobacco mosaic virus, you can see these particular shapes. Then there are viruses that are icosahedral.

These viruses exhibit a symmetrical, roughly spherical shape with 20 identical triangular faces. Capsomeres are of two types: pentons, which form vertices, and hexons, which form faces. Here, you can see the electron micrograph of an adenovirus. Then, there are certain viruses called head-and-tail viruses. Bacteriophages have a structure with an elongated icosahedral head and a tubular tail. The tail serves as a delivery mechanism for the viral genome.

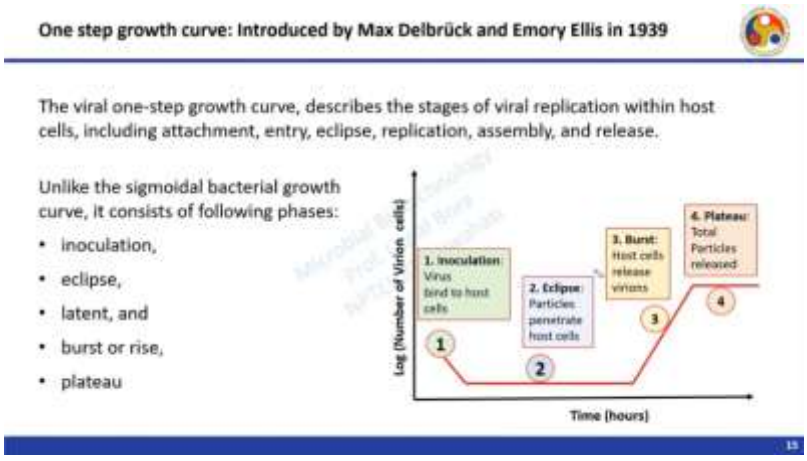


Another type of morphology is pleomorphic viruses. These exhibit varying shapes or forms, as you can see in this picture. The overall shape of poxviruses can vary from ovoid to brick-shaped. Then, we have enveloped viruses. These are viruses that contain a viral envelope obtained from the host cell membrane.



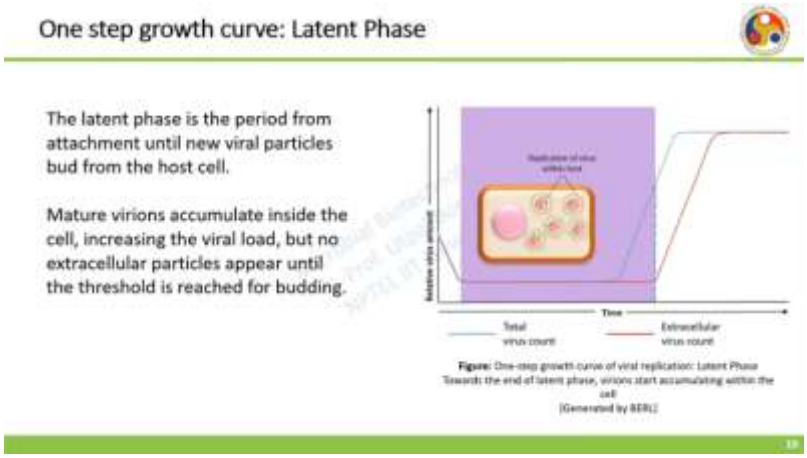
This envelope consists of a lipid bilayer with proteins encoded by both viral and host genomes. In the picture, you can see the electron micrograph of the SARS-CoV-2 virus. Let us now move on to the second section of this lecture, which deals with the life cycle of viruses. The life cycle of viruses can be easily explained with a one-step growth curve, introduced by Max Delbrück and Amory Ellis in 1939. The viral one-step growth curve describes the stages of viral replication within host cells, including attachment, entry, eclipse, replication, assembly, and release.

Unlike the sigmoidal bacterial growth curve, it consists of the following phases. The first phase is inoculation, where the virus binds to the host cell. The second step is the eclipse phase, where particles penetrate the host cell. The third step is the burst phase, where the host cells release variants, and the fourth is the plateau, where virus particles are released. This diagram shows a detailed view of the viral one-step growth curve, including the latent phase, the eclipse phase, and the burst size.



Let us examine the various phases one by one. The inoculation phase. When a host is inoculated with a virus, the initial viral count is high but gradually decreases as viruses attach to host cells via specific receptors. This decline occurs due to saturation and can also be manipulated by adding antibodies to the medium. The second step is the eclipse phase.

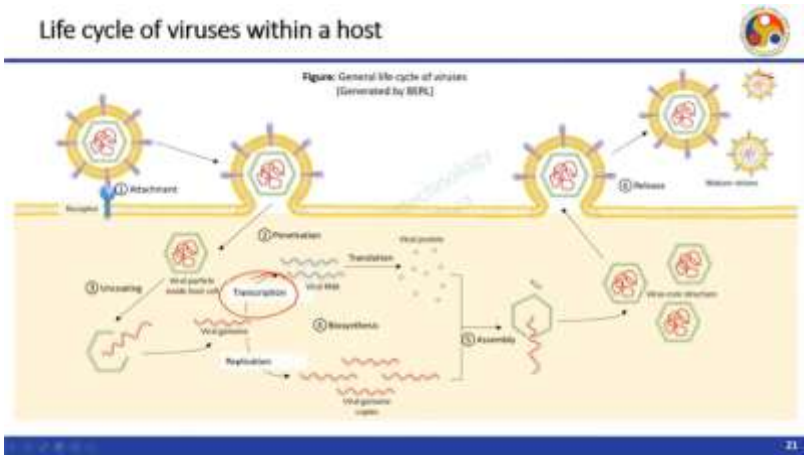
After attachment, viruses enter the host cell, where biosynthesis of components for new variants occurs. In the later stage, variant assembly begins, but the total number of viruses inside and outside the cell remains unchanged during this phase. In the latent phase, we consider the period from attachment until new viral particles bud or emerge from the host cell. Mature variants accumulate inside the cell, increasing the viral load, but no extracellular particles appear until the threshold for budding is reached. The next phase is the burst phase.



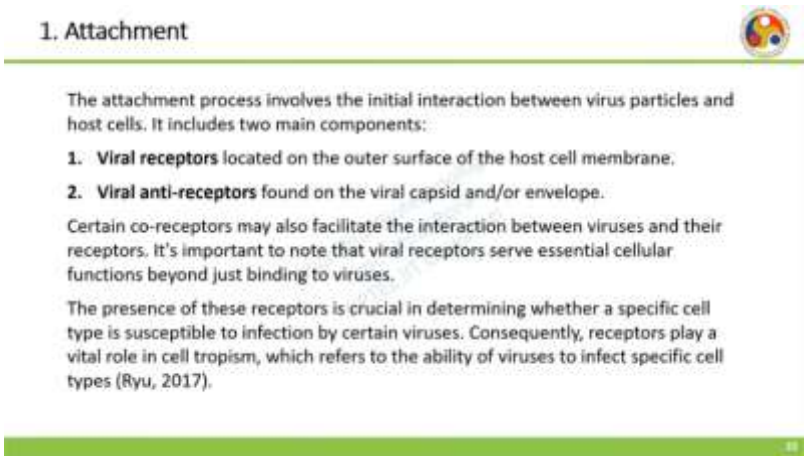
Once the retention threshold is reached, cells either lyse that is breakup or release variants by budding causing a sharp rise in extracellular variants and you can see the virus particles coming out here. Eventually no more viruses are released and we can estimate the number of variants released per infected cell called as the worst size. Overall this is the life cycle of viruses within a host. The first is the attachment.

The virus is getting attached through a receptor here as you can see. Then the next step is the penetration. The virus will penetrate the host cell and it will put the viral particle inside the host cell. here the uncoating will happen so the nucleic acid is released or the viral genome is released and then these viral genome will undergo various molecular events like transcription, translation or replication. So, this is the step of biosynthesis and where both the viral genome is copied as well as the viral proteins are produced.

These two together will then go for self-assembly as you can see here and the viral core structures are formed and finally released and we obtain the mature variants. So, from attachment to penetration to uncoating to biosynthesis to assembly to release, we consider as a life cycle of a virus overall. So, a little bit of detail regarding these various stages of the life cycle. First, the attestment process involves the initial interaction between biased particles and the host cells.



It includes two main components: viral receptors located on the outer surface of the host cell membrane, as we have already discussed or mentioned in the earlier slide. Then, viral anti-receptors are found on the viral capsid and/or envelope. Certain co-receptors may also facilitate the interaction between viruses and their receptors. It is important to note that viral receptors serve essential cellular functions beyond just binding to viruses. The presence of these



receptors are crucial in determining whether a specific cell type is susceptible to infection by certain viruses. If the receptor is absent, the cell will not allow the virus to attach and

therefore will become immune to it. Consequently, receptors play a vital role in cell tropism, which refers to the ability of viruses to infect specific cell types. So, here is a summary of the viral receptors and tropism for human viruses. For ease of discussion, we have divided these viruses into DNA viruses, RNA viruses, and reverse transcriptase viruses (or retroviruses).

There are various examples, such as parvovirus, polyomavirus, adenovirus, herpesvirus, picornavirus, flavivirus, coronavirus, orthomyxovirus, rhabdovirus (which causes rabies), and retrovirus (which causes HIV and hepatitis B virus). These remain very challenging problems that still haunt the world. You can see the corresponding receptors in this column. For example, parvovirus's receptor is HSPG, and for polyomavirus, it is GM1, and so on. For coronavirus, you have the ACE2 receptor, CD4 for HIV-1, and NTCP for the hepatitis B virus.

Table: Viral Receptors & Tropism for Major Human Viruses

Family: Prototype virus		Receptor (Coreceptor)	Viral Antireceptor	Tropism
DNA viruses	Parvovirus: AAV	HSPG (FGFR, integrin)	CAP	variable
	Polyoma: SV40	GM1 gangliosides	VP1	neurotropic
	Adenovirus: Ad5	Integrin	Fiber protein	variable
	Herpesvirus: HSV-1	Nectin-1/HVEM, PILRα	gD, gB	oral epithelium
	Herpesvirus: EBV	CD21	gp350	B cells and epithelial cells
RNA viruses	Picornavirus: Poliovirus	PVR/CD155	VP1, VP2, VP3	neurotropic
	Flavivirus: Hepatitis C virus	CD81, Claudin-1, Occludin	E2	hepatotropic
	Coronavirus: SARS virus	ACE2	Spike	variable
	Orthomyxovirus: Influenza virus	Sialic acid	HA	respiratory tract
	Rhabdovirus: Rabies	NCAM-1/CD56	G protein	neurotropic
RT viruses	Retrovirus: HIV-1	CD4 (CXCR4 or CCR5)	gp120	Macrophages (M-tropic) T-cells (T-tropic)
	Hepatitis B virus	NTCP	Pre-S1	hepatotropic

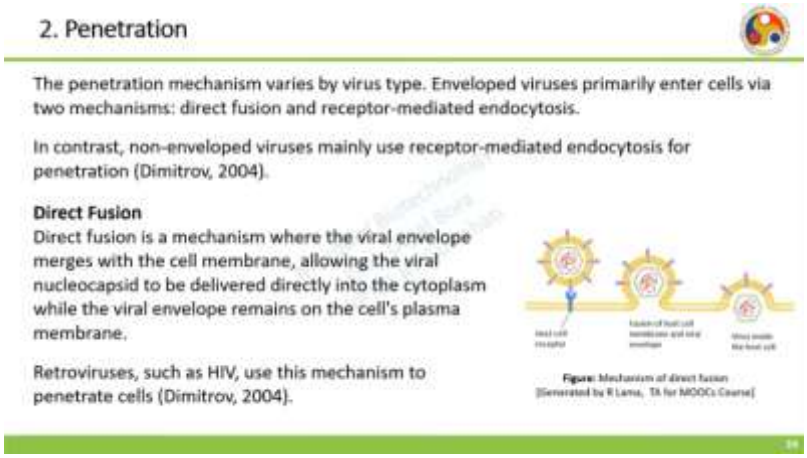
ACE, angiotensin-converting enzyme
FGFR, fibroblast growth factor receptor
PILR, Paired immunoglobin like type 2 receptor
NCAM, neural cell adhesion molecule
HSPG, heparin sulfate proteoglycan
HVEM, herpes virus entry mediator
ICAM, intercellular adhesion molecule
NTCP, sodium taurocholate cotransporting polypeptide
PVR, poliovirus receptor

Adapted from: Ryu, 2017

And then there are certain viral anti-receptors. For example, you have these CAPs in the case of parvovirus and VP1 in the case of polioma, and so on. And in the case of coronavirus, you have this spike, and then you have these GP120 in the case of HIV and pre-S1 in the case of hepatitis B virus. So, based on these tropisms, they can be classified as, for example, neurotrophic or they may be oral epithelium, beta cell, and epithelial cell tropic. Then they may be hepatotrophic or they may have tropism toward the respiratory tract.

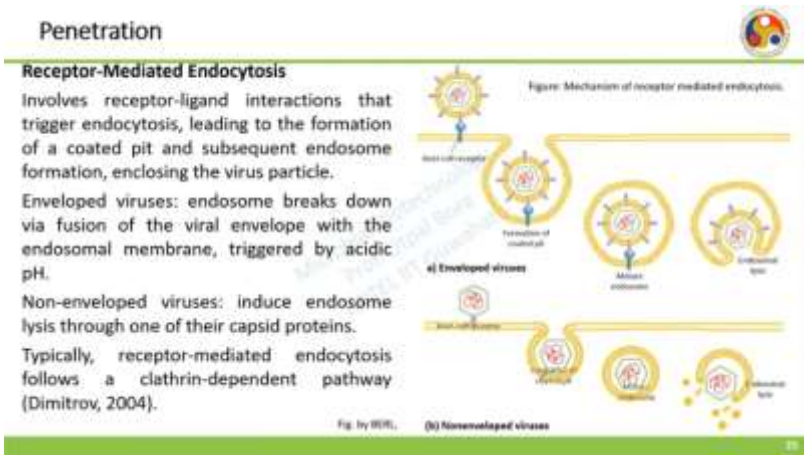
And then they may have tropism for macrophages and so on. Next comes penetration. The penetration mechanism varies by virus type. Enveloped viruses primarily enter cells via two mechanisms. Number one is direct fusion and receptor-mediated endocytosis.

In contrast, non-enveloped viruses mainly use receptor-mediated endocytosis for penetration. Direct fusion. Direct fusion is a mechanism where the viral envelope merges with the cell membrane, allowing the viral nucleocapsid to be delivered directly into the cytoplasm while the viral envelope remains on the cell's plasma membrane. So here you can see the mechanism of direct fusion, where there is a host cell receptor and the fusion of the host cell membrane and viral envelope taking place soon after these attachments through the receptor, and the virus is thereby internalized subsequently. So retroviruses such as HIV use this mechanism to penetrate the host cells.



Receptor-mediated endocytosis. This involves receptor-ligand interactions that trigger endocytosis, leading to the formation of a coated pit and subsequent endosome formation enclosing the virus particles. So, you can see here the formation of the coated pit. Enveloped viruses, the endosomes,

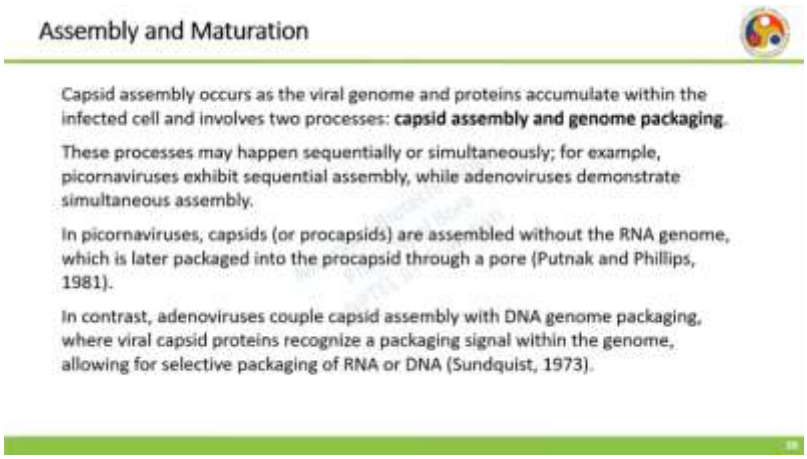
break down by fusion of the viral envelope with the endosomal membrane, triggered by acidic pH. In the case of non-enveloped viruses, they induce endosomal lysis through one of their capsid proteins. Typically, receptor-mediated endocytosis follows a clathrin-dependent pathway in this case. The third step is uncoating during viral replication, as virus particles move from the cell's periphery to the perinuclear space or are internalized. The viral genome is exposed to the cellular machinery for gene expression.



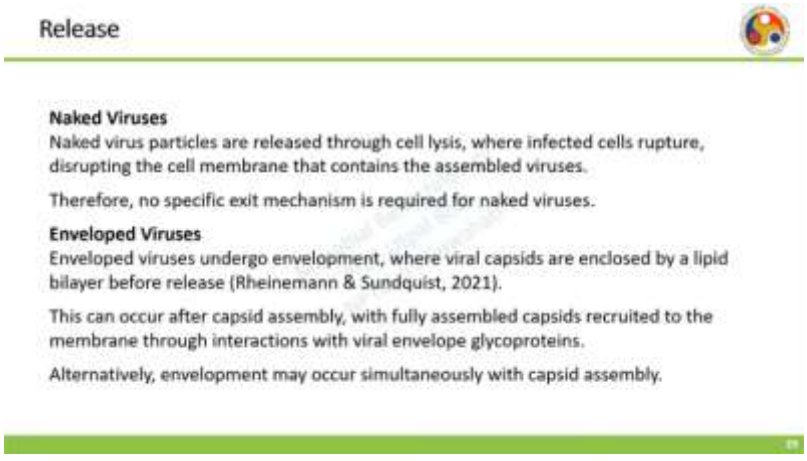
This process, known as uncoating, is often associated with the endocytic pathway or cytoplasmic trafficking. For viruses that replicate in the nucleus, the viral genome must enter through nuclear pores. Similar viruses, like polyomaviruses, allow the viral capsid to enter the nucleus. In contrast, larger viruses dock their nucleocapsids to nuclear pore complexes, leading to partial capsid disruption that enables the DNA genome to enter the nucleus.

In the biosynthesis stage, viral genome replication strategies vary significantly among virus families, distinguishing each family. We will discuss this soon in this lecture. The extent to which virus families rely on their host cellular machinery also varies, from complete dependence to relative or partial independence. The same is true for viral replication. However, all viruses rely entirely on the host translation machinery—whether complete or relative—specifically ribosomes, to synthesize their proteins.

In the assembly and maturation stage, capsid assembly occurs as the viral genome and proteins accumulate within the infected cells and involve two processes. Number one, the capsid assembly and genome packaging. These procedures may happen sequentially or simultaneously. For example, picornaviruses exhibit sequential assembly, while adenoviruses demonstrate simultaneous assembly. In picornaviruses, capsids or procapsids are assembled



without the RNA genome, which is later packaged into the procapsid through a pore. So the procapsid will be formed, and through this pore, the genetic material will be internalized. In contrast, adenoviruses couple capsid assembly with DNA genome packaging, so it goes side by side, where viral capsid proteins recognize a packaging signal within the genome, allowing for selective packaging of RNA or DNA. Finally comes the release phase, where the viruses are released. In the case of naked viruses, virus particles are released through cell lysis, where infected cells rupture, disrupting the cell membrane that contains the assembled viruses.

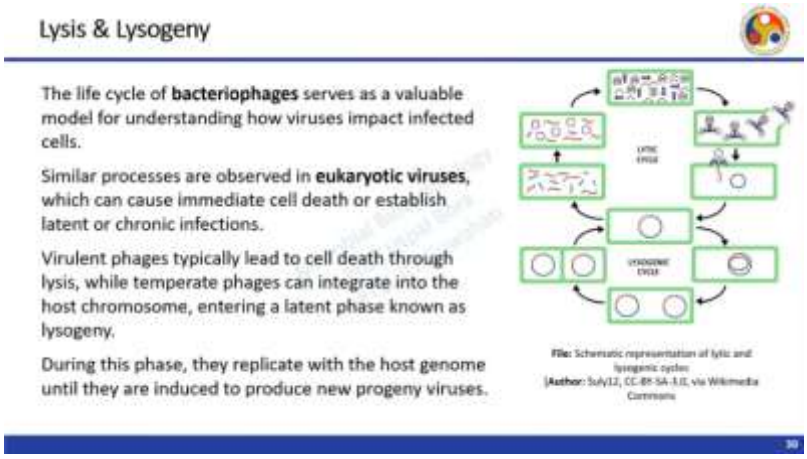


Therefore, no specific exit mechanism is required in the case of naked viruses. In the case of enveloped viruses, they undergo envelopment, where viral capsids are enclosed by a lipid bilayer before release. If you compare these to sending letters in older times, here you write a letter and then put it inside an envelope. And then seal it.

But in the case of naked viruses, you do not use any envelope. You directly use a postcard to write your message and send it. So, these are the two different release mechanisms. So,

this can occur after the case of capsid assembly, with fully assembled capsids recruited to the membrane through interactions with viral envelope glycoproteins. Alternatively, envelopment may occur simultaneously with capsid assembly.

Let us now discuss two important topics in the virus life cycle, which are lysis and lysogeny. The life cycle of bacteriophages serves as a valuable model for understanding how viruses impacts infected cells. Similar processes are observed in eukaryotic viruses, which can cause immediate cell death or establish latent or chronic infections. Virulent phages typically lead to cell death through lysis, while temperate phages can integrate into the host chromosome, entering a latent phase known as lysogeny. During this phase, they replicate.



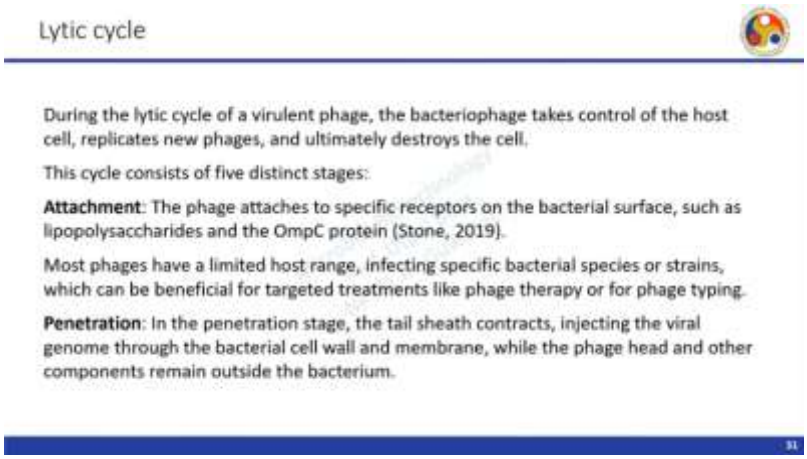
Along with the host genome until they are induced to produce new progeny viruses. So here we can see the two different life cycles: the lytic cycle on the top and the lysogenic cycle below, and also how these two life cycles are interconnected. So here we can see that after the attachment and internalization of the virus into the host cell, multiplication of proteins and DNA occurs, and they are self-assembled into viral particles. The cell is broken open here to release the mature virus particles. This is known as lysis, which kills the host cell.

In the case of lysogeny, once the viral genome is internalized, it gets integrated with the host genome. So it becomes a part of the host genome. Genome. Now, when the host genome replicates, as you can see here, the integrated viral genome will also be replicated. So, from one copy, it becomes two copies here along with the two

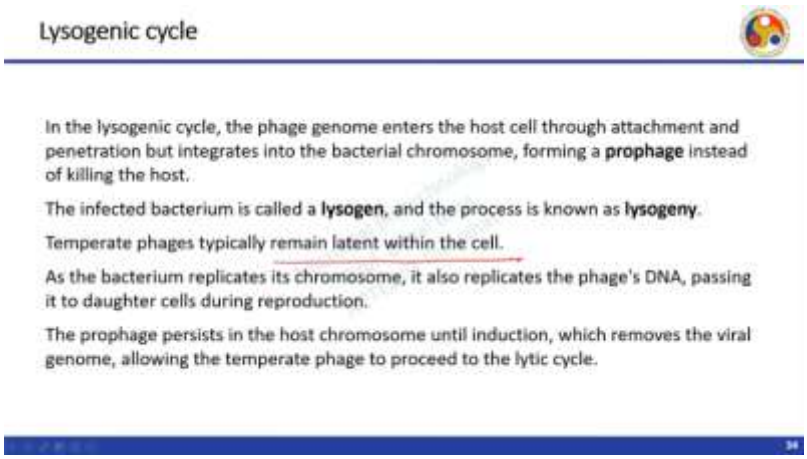
host daughter molecules. Then, as the cell divides to distribute the genetic material, both daughter cells will have a copy each of the viral genome along with their own host genome.

So, this cycle can go on for quite a long time. But finally, the virus will undergo certain molecular events, due to which these integrated viral genomes will, at a particular point in time, get released through various mechanisms. They will also produce the proteins required for their packaging, and then they will enter the lytic cycle. So a virus that was in the lysogenic cycle, in order to be released as mature viruses,

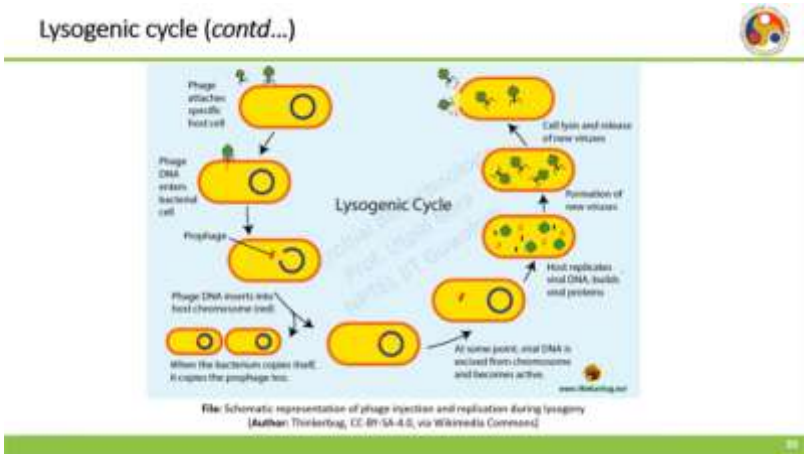
It has to undergo a transformation in its life cycle; not necessarily does it go into the lytic cycle. It can also be released without lysing the host cell by other mechanisms. So, during the lytic cycle of a virulent phage, the bacterial phage takes control of the host cell, replicates new phages, and ultimately destroys the cell, which we have already discussed in the earlier diagram. This cycle consists of five distinct phases, which we have discussed at length earlier, including the attachment phase, the penetration phase, biosynthesis, and finally assembly and release. So overall, starting from the attachment, we proceed to the penetration phase, then the biosynthesis phase, followed by assembly, and finally release. In the lysogenic cycle, the phage genome enters the host cell through attachment and penetration but integrates into the bacterial chromosome, forming a prophage instead of killing the host.



The infected bacterium is called a lysogen, and the process is known as lysogeny. Temperate phages typically remain latent within the cell. As the bacterium replicates its chromosome, it also replicates the phage's DNA, passing it to daughter cells during reproduction, which we have discussed with the help of the diagram earlier. The process persists in the host chromosome until induction, which removes the viral genome, allowing the temperate phage to proceed to the lytic cycle.

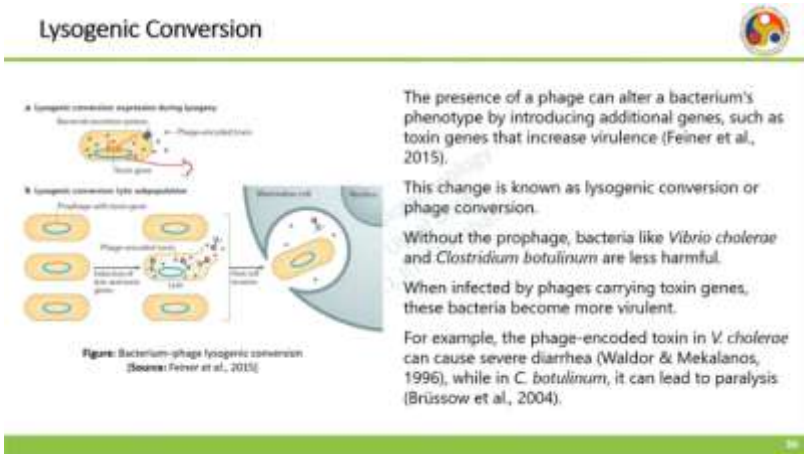


So here is the lysogenic cycle, which was already discussed, starting from the attachment, then the internalization of the DNA, followed by the formation of the prophage and distribution, replication, and distribution into the two daughter cells. And then, as already discussed at some point, the viral DNA is excised from the chromosome and becomes active. Then the host replicates viral DNA, builds viral proteins, and then forms new viruses, which go on to cause cell lysis and release of new viruses. An important concept is lysogenic conversion. The presence of a phage can alter a bacterium's phenotype by introducing additional genes, such as toxin genes, that increase virulence.



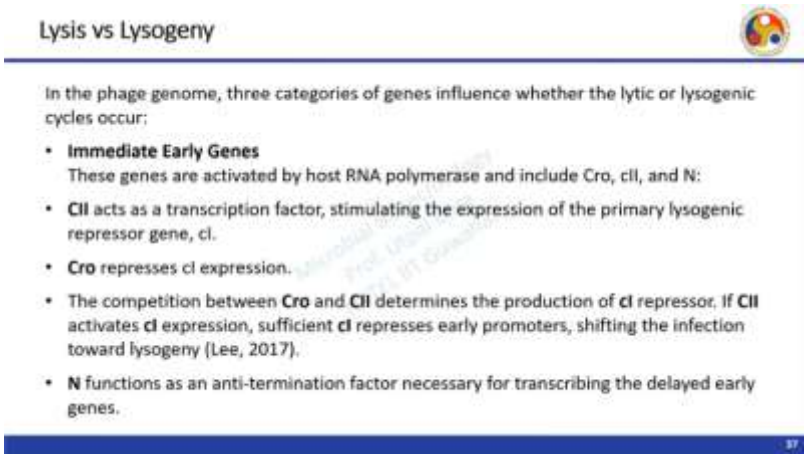
This change is known as lysogenic conversion or FAS conversion. Without the prophage, bacteria like Vibrio cholerae and Clostridium botulinum are less harmful. When infected by phages carrying toxin genes, these bacteria become more virulent. For example, the phage-encoded toxin in Vibrio cholerae can cause severe diarrhea, while in Clostridium botulinum it can lead to paralysis.

So here you can see the lysogenic conversion expressed during lysogeny. So this is a FAS-encoded toxin which comes from the viral genome. So this is the bacterial genome into which this viral genome has integrated along with the toxin gene. So here is the case of lysogenic conversion where the lytic subpopulation phenomenon can be observed.



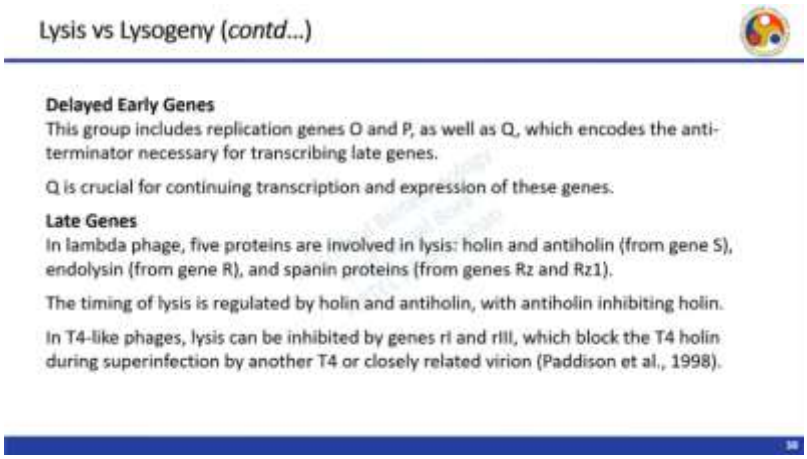
Where is the prophage? with toxin, and then here it produces FAS-encoded toxin. Induction of the lytic and toxin genes, which have been integrated here, results in this FAS-encoded toxin, which leads to diarrhea. Then here you can see the host cell invasion. Let us compare lysis and lysogeny. In the first genome, three categories of genes influence whether the lytic or lysogenic cycle occurs. Number one is the immediate early genes.

These genes are activated by host RNA polymerases and include Cro-CIINN. CIINN acts as a transcription factor. Stimulating the expression of the primary lysogenic repressors in CI. Cro represses the CII expression. The competition between Cro and CII determines the production of CII in the process.

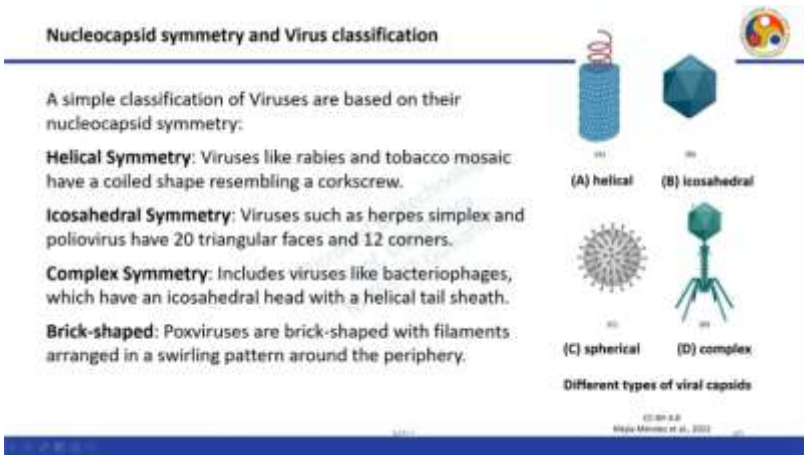


If CII activates CI expression, sufficient CI represses early promoters, shifting the infection towards lysogeny. N functions as an anti-termination factor necessary for transcribing the delayed early genes. The delayed early genes include replication genes O, P, and Q, which include the anti-terminator necessary for transcribing late genes. Q is crucial for continuing transcription and expression of these genes. Late genes in lambda phage include five proteins involved in lysis: holin and antiholin from the S gene, lysin from the R gene, and spanin proteins from genes RZ and RZ1.

The timing of lysis is regulated by holin and antiholin, with antiholin inhibiting holin. In T4-like phages, lysis can be inhibited by genes R1 and R3, which block the T4-holin during superinfection by another T4 or closely related variant. Let us now discuss the classification of viruses. So, in this section, we will discuss at length the Baltimore classification of viruses. Before that, let us look into other kinds of classification, particularly based on nucleocapsid symmetry.

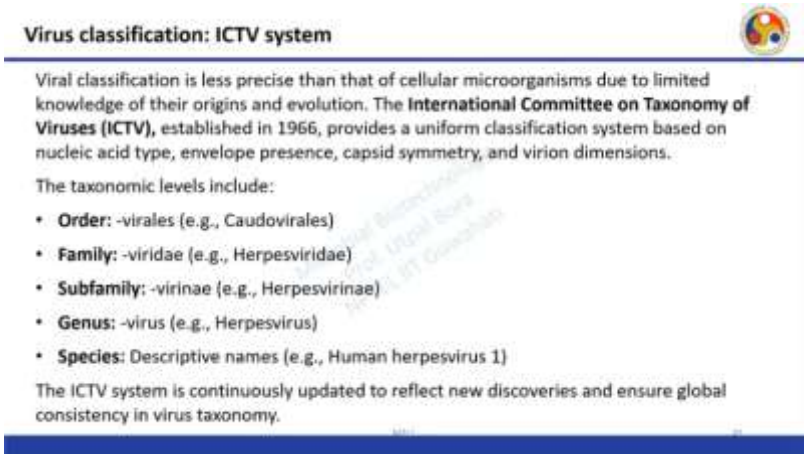


So here you can see a simple classification of viruses which are based on their nucleocapsid symmetry. For example, viruses can be described as helical. Like rabies and tobacco mosaic. They have a coiled shape resembling a corkscrew, as you can see here. Then viruses can have icosahedral symmetry.



These viruses, such as herpes simplex and poliovirus, have 20 triangular faces and 12 corners. And you can see the beautiful geometric shape here. Then we may have certain complex symmetry. Like these in D. So, these include viruses like bacteriophages, which have an icosahedral head, as you can see on the top, and a helical tail sheath. Then we may also have brick-shaped viruses.

For example, pox viruses are brick-shaped with filaments arranged in a swirling pattern around the periphery. Virus classification is based on the ICTV system, or the International Committee on Taxonomy of Viruses, which was established in 1966. This is less precise than that of cellular microorganisms due to limited knowledge of their structure and evolution. The ICTV provides a uniform classification system based on nucleic acid types, envelope presence, capsid symmetry, and virion dimensions. The taxonomic levels include order virales, for example, caudovirales.

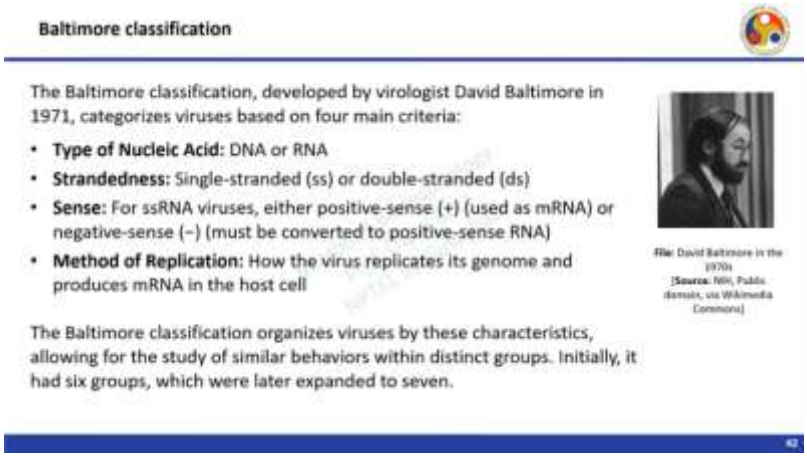


Then family viridi, example herpesviridi. Then subfamily virini, example herpesvirini. Then genus odovirus, example herpesvirus. Then species, descriptive names like human

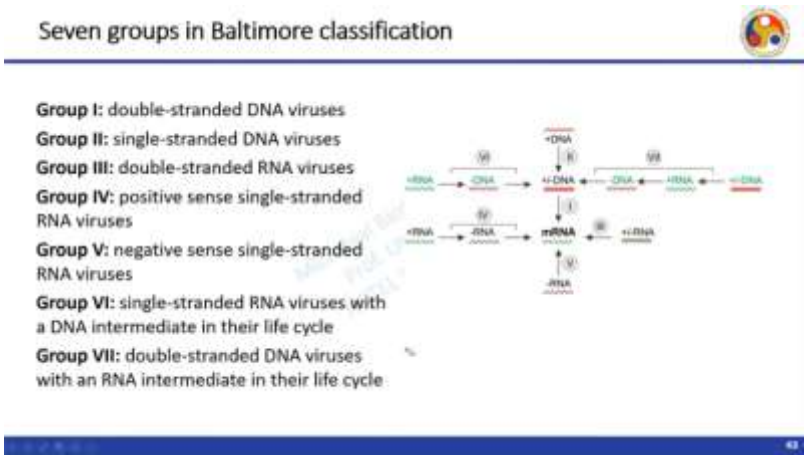
herpesvirus 1. The ICTV system is continuously updated to reflect new discoveries and ensure global consistency in virus taxonomy.

Let us have a detailed look of the Baltimore classification. So this was developed by virologist David Baltimore in 1970s and it categorizes viruses based on four main criteria. When we tell type of nucleic acid, what type of nucleic acid this particular virus has DNA or RNA. When we tell strandedness, whether it is single stranded or double stranded and for single stranded RNA viruses either positive sense plus or negative sense minus must be converted to positive sense rRNA in this case.

And in the method of replication, how the virus replicates its genome and produces mRNA in the host cell are the important criteria in this classification. The Baltimore classification organizes viruses by these characteristics, allowing for the study of similar behaviors within distinct groups. Initially, it had six groups, which are later expanded to seven. We will look into these seven Baltimore classes or BC. So these are divided into group 1 to group 7.

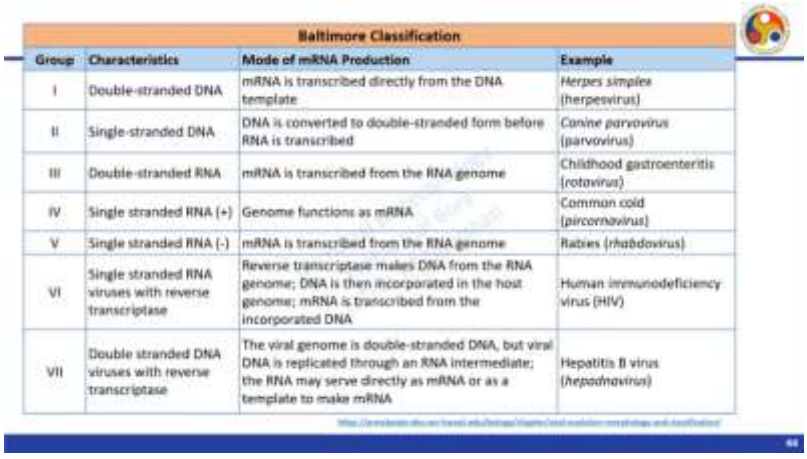


For example, you have Group 1 double-stranded DNA viruses in this case. Then you have Group 1, which is single-stranded DNA viruses. Then Group 3, double-stranded RNA viruses. Then Group 4, positive-sense single-stranded RNA viruses. And then Group 5, which is negative-sense RNA viruses.



Then Group 6 is single-stranded RNA viruses with a DNA intermediate in their life cycle. Double-stranded DNA viruses with an RNA intermediate in their life cycle. So, this particular diagram comprehensively describes the seven groups in the Baltimore classification. So, we have seen the seven groups based on the nucleic acid they include or the strandedness and the intermediates in their life cycle, RNA or DNA in between. And if you look into the mode of mRNA production from 1 to 7, in the first case, mRNA is transcribed directly from the DNA, for example, in herpes simplex virus.

In Group 2, like those in canine parvoviruses, DNA is converted to double-stranded form before RNA is transcribed. In Group 3, mRNA is transcribed from the RNA genome, for example, in the case of rotavirus. Then in the fourth case, the genome functions as the mRNA because this has a single-stranded positive RNA. Picornaviruses, which cause the common cold, are classic examples of this. Then in the case of single-stranded negative RNA, mRNA is transcribed from the RNA genome, such as in the case of rabies.



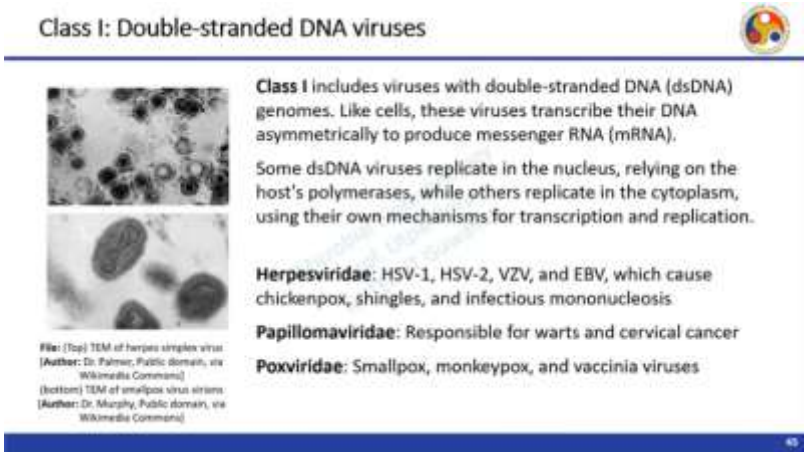
Baltimore Classification

Group	Characteristics	Mode of mRNA Production	Example
I	Double-stranded DNA	mRNA is transcribed directly from the DNA template	*Herpes simplex* (herpesvirus)
II	Single-stranded DNA	DNA is converted to double-stranded form before RNA is transcribed	*Canine parvovirus* (parvovirus)
III	Double-stranded RNA	mRNA is transcribed from the RNA genome	Childhood gastroenteritis (*rotavirus*)
IV	Single stranded RNA (+)	Genome functions as mRNA	Common cold (*picornavirus*)
V	Single stranded RNA (-)	mRNA is transcribed from the RNA genome	Rabies (*rhabdovirus*)
VI	Single stranded RNA viruses with reverse transcriptase	Reverse transcriptase makes DNA from the RNA genome; DNA is then incorporated in the host genome; mRNA is transcribed from the incorporated DNA	Human immunodeficiency virus (HIV)
VII	Double stranded DNA viruses with reverse transcriptase	The viral genome is double-stranded DNA, but viral DNA is replicated through an RNA intermediate; the RNA may serve directly as mRNA or as a template to make mRNA	Hepatitis B virus (*hepadnavirus*)

Then in sixth group, reverse transcriptase makes DNA from the RNA. DNA is then incorporated in the host genome. mRNA is transcribed from the incorporated DNA. So, the best example of this is the human immunodeficiency viruses. In the seventh group, the viral genome is double-stranded DNA.

But viral DNA is replicated through an RNA intermediate. The RNA may serve directly as mRNA or as a template to make mRNA. For example, the hepatitis B virus. So, let's look into a little bit of details of the class 1 viruses. The class 1 includes viruses with double-stranded DNA, as already mentioned.

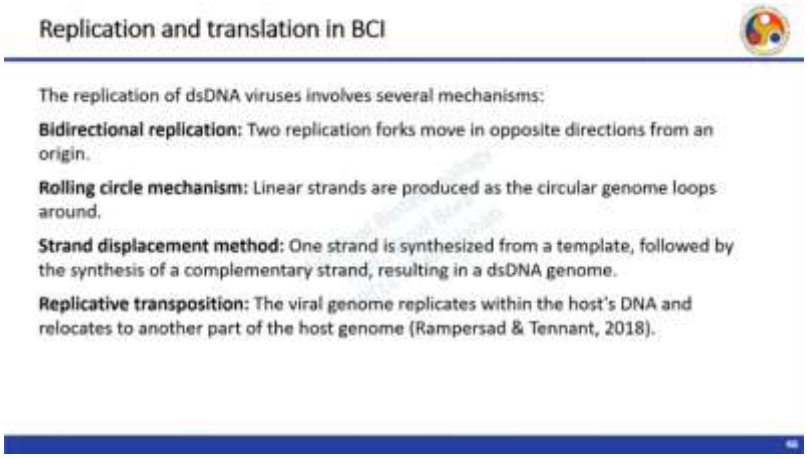
Like cells, these viruses transcribe their DNA asymmetrically to produce messenger RNA. Some double-stranded DNA viruses replicate in the nucleus, relying on the host polymerases, while others replicate in the cytoplasm using their own mechanisms for transcription and replication. Examples include herpes viridi 1, 2 and EBV which causes sick and pox, singles and infectious mononucleosis. Then we have papilloma viridi, these are responsible for warts and cervical cancer and pox viridi, this causes smallpox, monkeypox and vaccinia viruses.



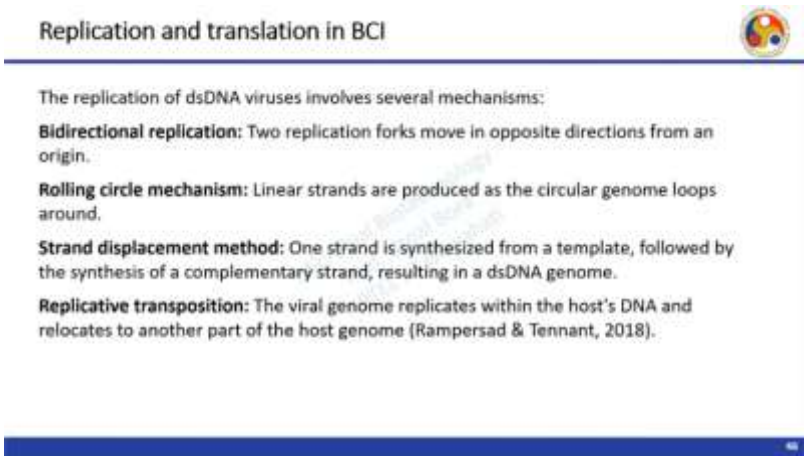
In this picture we can see the electron micrograph image of a herpes simpleus virus on the top and in the bottom, we can see the smallpox virus. Let us now look into the replication and translation of Baltimore class 1 type of viruses. The replication of double-stranded DNA viruses involves several mechanisms. The first one is the bi-directional replication. where two replication forks move in opposite direction from an origin.

But this model has now been revised where in fact the moment is in the same direction due to the involvement of one single polymerase and there is a looping which occurs to accommodate the static hindrances resulting out of these kind of complex structures. We

will not discuss about that in length. Those who are interested can refer to the replication fork model, revised replication fork model. The second one is the rolling cycle mechanism where linear strands are produced as a circular genome loops around.

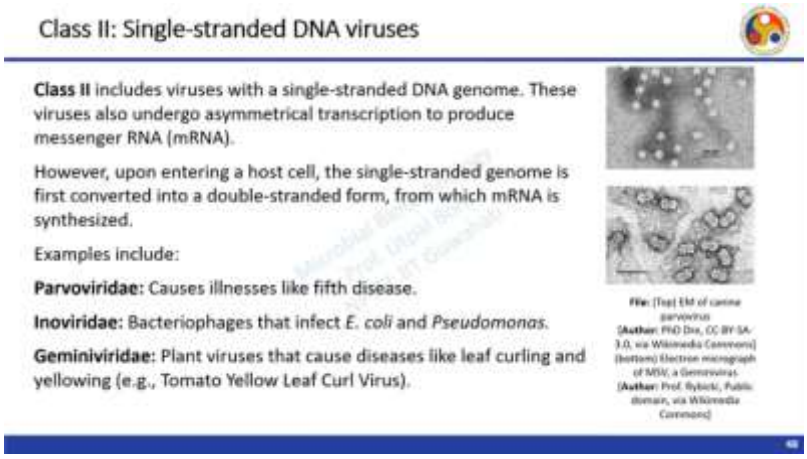


And the third one is a strand displacement method. One strand is synthesized from a template followed by the synthesis of a complementary strand resulting in double-stranded genome. The fourth one is a replicative transposition. The viral genome replicates within the host's DNA and relocates to another part of the host genome. mRNA synthesis occurs in three steps.



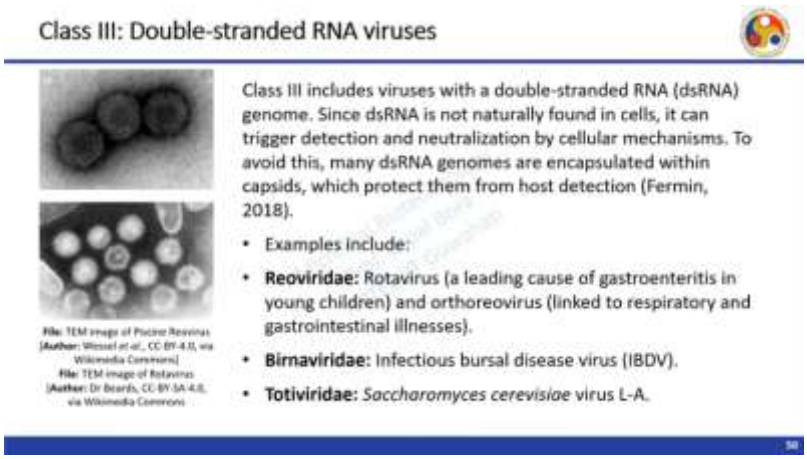
A transcription pre-initiation complex binds to the DNA upstream of the start site, recruiting the host RNA polymerases. RNA polymerase uses the negative strand as a template to generate mRNA. The transcription ends when RNA polymerase reaches specific signals such as a polyadenylation site. In the class 2 viruses, which are single-stranded DNA viruses, they include a single-stranded DNA genome. These viruses also undergo asymmetrical transcription to produce messenger RNA.

However, upon entering a host cell, the single-stranded genome is first converted into a double-stranded form from which mRNA is synthesized. Some of the examples include Parvoviridae, which causes inlaces like 5th disease and then Inoviridae, includes Dactyophages, then infect E. coli and Pseudomonas, then Gemini Viridae, plant viruses that causes diseases like leaf curling and yellowing. So here is the electron micrograph of Canine Parvovirus on top. And then we have these on the bottom, the Gemini virus. And you can see these particles always occurring in pairs as the name Gemini suggests.



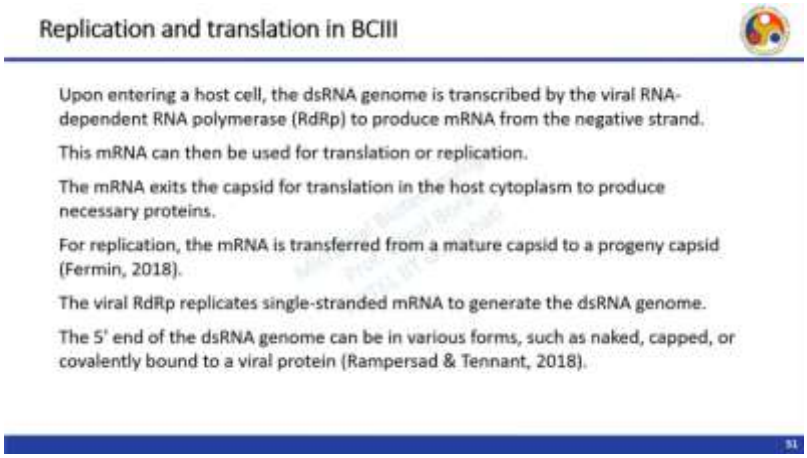
Replication and translation in these Baltimore class 2 type of viruses. Eukaryotic single-stranded DNA viruses primarily replicate in the nucleus. Most have circular genomes that utilize rolling circle replication. In contrast, parvoviruses have linear single-stranded DNA genomes that replicate via rolling happen replication, which is similar to rolling circle replication. Parvoviruses can package either the positive or negative sense strand into capsids depending on the virus.

The newly formed single-stranded DNA may be packaged into variants or replicated by a DNA polymerase to create a double-stranded form for transcription or further replication. The class III double-stranded RNA viruses, since double-stranded RNA is not naturally found in cells, can trigger detection and neutralization by cellular mechanisms. To avoid this, many double-stranded RNA genomes are encapsulated within capsids, which protects them from host detection—a very clever way for the virus to save itself. Some examples include Rio Viridi rotavirus, which causes gastroenteritis in young children, and orthoreoviruses, linked to respiratory and gastrointestinal illnesses. Then Birnaviridae, Infectious Bursal Disease Virus.



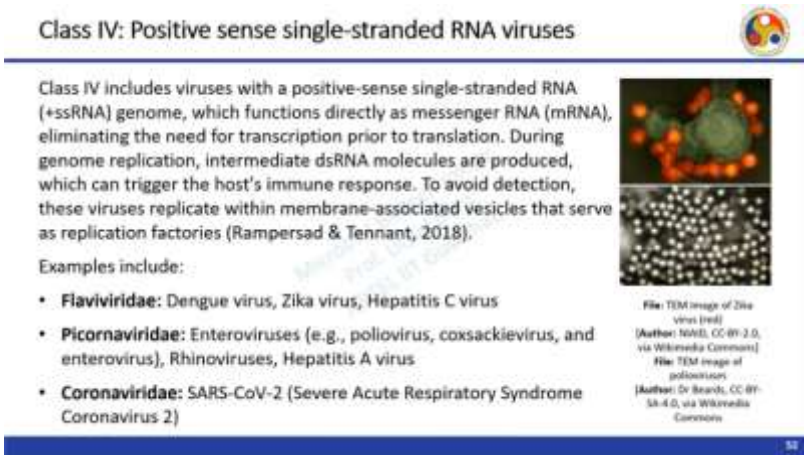
Then Totiviridae, for example Saccharomyces cerevisiae, are some of the examples. Here you can see the TEM image of Piscian reovirus on the top and then the TEM image of rotavirus below. How does replication and translation happen in Baltimore Class III viruses? Upon entering a host cell, the double-stranded RNA genome is transcribed by the viral RNA-dependent RNA polymerase to produce mRNA from the negative strand. This mRNA can then be used for translation or replication.

The mRNA exits the capsid for translation in the host cytoplasm to produce necessary proteins. For replication, the mRNA is transferred from a mature capsid to a progeny capsid. The viral RdRP replicates single-stranded mRNA to generate the double-stranded RNA genome. The 5' end of the double-stranded RNA genome can be in various forms, such as naked, capped, or covalently bound to a viral protein. Let us now look into the BC4 type of viruses, which are positive-sense single-stranded RNA viruses.

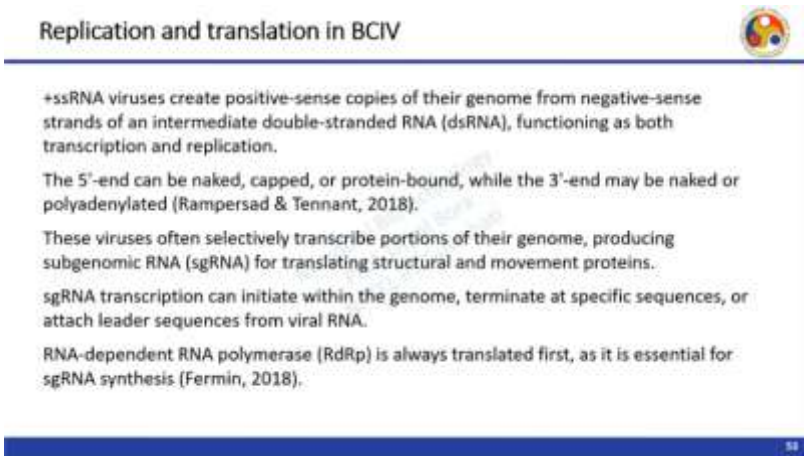


They function directly as messenger RNA, eliminating the need for transcription prior to translation. During genome replication, intermediate double-stranded RNA molecules are

produced, which can trigger the host's immune responses. To avoid detection, these viruses replicate within the membrane-associated vesicles that serve as replication factories. Some examples include Flaviviridae, which causes diseases such as dengue, Zika, and hepatitis C; Picornaviridae, including enteroviruses, rhinoviruses, and hepatitis A virus; and Coronaviridae, which caused the recent pandemic, SARS-CoV-2. Here, you can see the TEM image of the Zika virus at the top and the TEM image of polioviruses at the bottom.

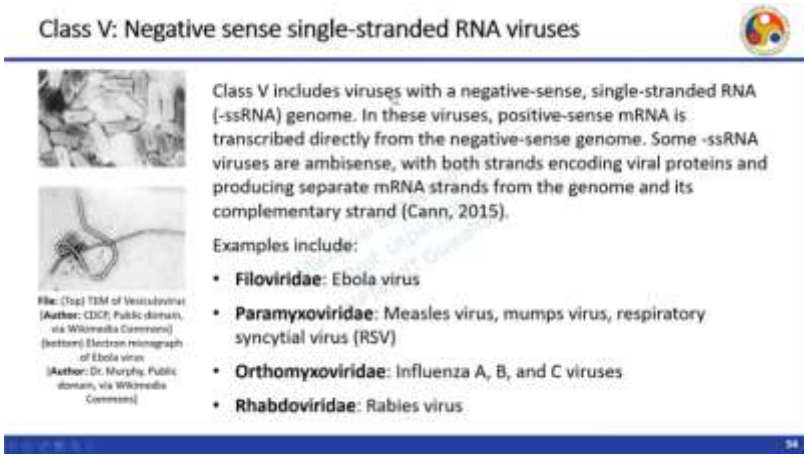


How do replication and translation occur in Baltimore class 4 viruses? These positive single-stranded RNA viruses create positive-sense copies of their genome from negative-sense strands of an intermediate double-stranded RNA, functioning in both transcription and replication. The 5' end can be naked, capped, or protein-bound, while the 3' end may be naked or polyadenylated. These viruses often selectively transcribe portions of their genome, producing subgenomic RNA for translating structural and movement proteins. As RNA transcription can initiate within the genome, terminate at specific sequences, or attach sequences from viral RNA, RNA-dependent RNA polymerases (RdRp) are always translated first, as they are essential for subgenomic RNA synthesis.



Next are the BC class IV viruses. These include viruses with negative-sense single-stranded RNA genomes. In these viruses, positive-sense mRNA is transcribed directly from the negative-sense genome. Some negative-sense single-stranded RNA viruses are ambisense, with both strands encoding viral proteins. They produce separate mRNA strands from the genome and its complementary strand.

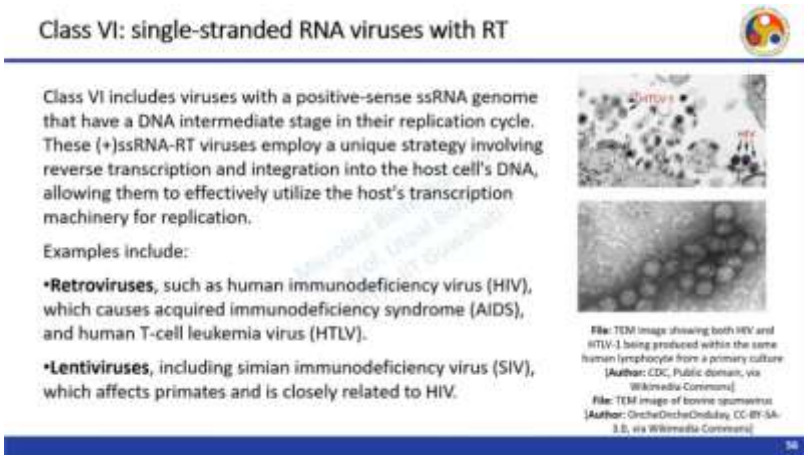
Some of the examples include phyloviridae, the Ebola virus, then measles virus, mumps virus, and then we have influenza A, B, C viruses, and then we have the rabies viruses. On top, you can see the same image of vesiculovirus. Then we see the Ebola virus on the bottom in this illustration by Murphy. How does the replication and translation happen in Baltimore class 5 type of viruses? During transcription, RDRP or RNA-dependent RNA polymerase generates a 5'N triphosphate leader RNA.



When it encounters a leader sequence, the 5'N capping or cap snatching from host mRNA allows viral mRNA recognition and translation by host ribosomes. R-DRP initiates transcription at a gene start signal and terminates at the gene end signal. At the end of transcription, it adds a poly-A tail to the mRNA stream and consisting of hundreds of adenine nucleotides, often through stuttering on uracil sequences. Replication. The genomic negative SSRNA is replicated from the positive sense antigenome similar to transcription but in the reverse way.

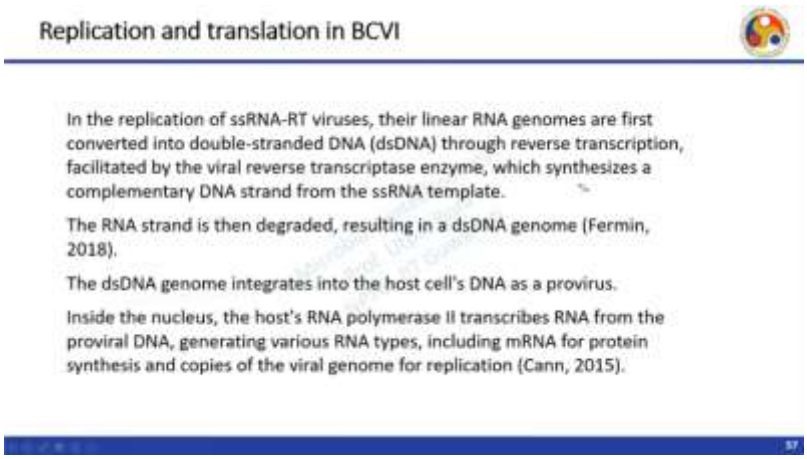
RDRP moves from the 3' end to the 5' end of the antigenome ignoring transcription signals and uses the antigenome as a template for the genome. Class 6 Baltimore viruses. This includes viruses with a positive sense single-stranded RNA genome that have a DNA intermediate stage in their replication cycle. These positive strand Single-stranded rRNA reverse-transcripted viruses employ a unique strategy involving reverse transcription and

integration into the host cell's DNA, allowing them to effectively utilize the host transcription machinery for replication.



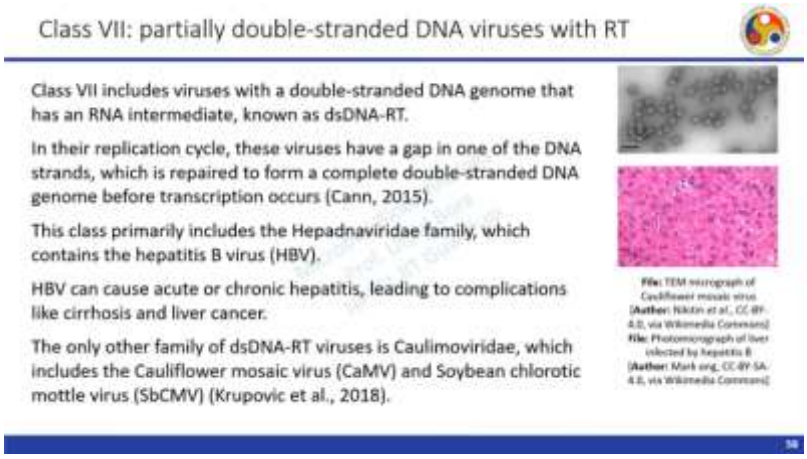
Some of the examples include retroviruses such as HIV, which causes AIDS, and the human T-cell leukemia virus or HTLV. Then we have lentiviruses. These include simian immunodeficiency viruses, which affect primates and are closely related to the human immunodeficiency virus. So here we can see the HIV virus on the top and HTLV in this TEM image. And then we can see here bovine spumavirus in the bottom panel.

The replication and translation in Baltimore class VI viruses, in this replication of single-stranded RNA-RT viruses, their linear RNA genomes are first converted into double-stranded DNA through reverse transcription. This is facilitated by the viral reverse transcriptase enzyme, which synthesizes a complementary DNA strand from the single-stranded RNA template. The RNA strand is then degraded, resulting in a double-stranded genome. The double-stranded genome integrates into the host cell's DNA as a provirus. Inside the nucleus, the host RNA polymerase II transcribes RNA from the proviral DNA, generating various RNA types, including mRNA for protein synthesis and copies of the viral genome for replication.



The last class, or class 7, includes viruses with a double-stranded DNA genome that has an RNA intermediate, known as DS-DNA-RT. In their replication cycle, these viruses have a gap in one of the DNA strands, which is repaired to form a complete double-stranded DNA genome before transcription occurs. This class primarily includes the Hepadnaviridae family, which contains the hepatitis B virus. Hepatitis B virus can cause acute or chronic hepatitis, leading to complications like cirrhosis and liver cancer.

The only other family of DNA-RT viruses is Cholmogoridi, which includes the cauliflower mosaic virus or CaMV and soybean chlorotic mottle virus. The transcription of double-stranded DNA RT viruses resembles that of double-stranded DNA viruses, but these viruses use reverse transcription to replicate their circular genome while still in the viral capsid. In the host cell cytoplasm, the cell's RNA polymerase II transcribes RNA strands from the viral genome. Replication of the double-stranded DNA genome occurs based on these RNA strands and follows a similar mechanism to that in single-stranded RNA RTE viruses. However, in double-stranded DNA RTE viruses, replication occurs in a loop around the circular genome.



After replication, the double-stranded genome can either be packaged into new viral particles or transported to the nucleus for further transcription. So with this, we come to the end of this Baltimore classification. Thank you. Thank you.