Virology

Lec (2)
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Teaching Objectives:
1. To classify types of replication of viruses.
2. To describe stepwise.
3. To recognize the mechanism of transcription and translation

Replication of viruses

Viruses are obligate intracellular pathogens and require cellular enzymes to help them replicate. Unlike bacteria, which replicate by binary fission, viruses have to 'disassemble' their structure before they can replicate. The steps of viral replication can be broadly studied under two headings

A-The growth curve: Shows the amount of viruses produced at different time during infection. No typical virus is seen in the culture after 3-4 hours of onset. (Eclipse period). After that there will be accumulation of nucleic acid and then the virus appears inside the cell (Rise period). Further ahead happens the killing of the cell and the viruses, burst out and are released from this, it is clear that growth curve of the virus is different from that of the bacterium figure (1).

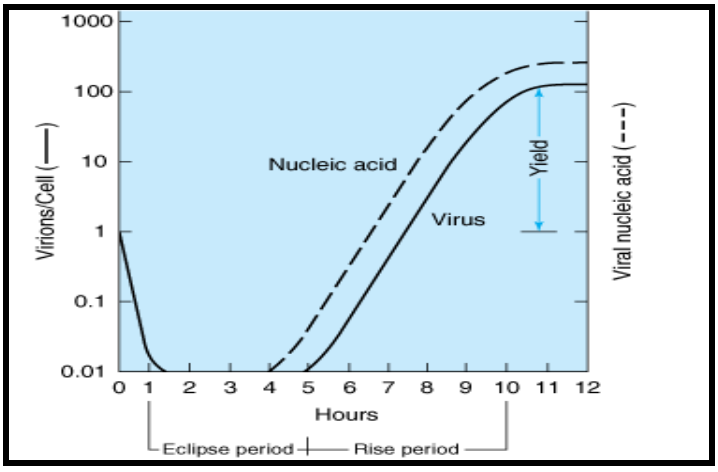


Figure (1): Viral growth curve

B-Stepwise description

1. Adsorption or attachment.

The first step in the replication cycle is the attachment of the virus particle to the cell Surface via ionic interactions which are temperature-independent. The viral attachment protein recognizes

specific receptors, which may be protein, carbohydrate or lipid, on the outside of the cell. Cells without the appropriate receptors are not susceptible to the virus.

Therefore are very specific in the cell type that they can infect this gives them the 'cell tropism' and is important in disease pathogenesis (i.e. why some viruses affect certain organs only). Influenza viruses use the haemagglutinin (HA) protein to attach to the sialic acid-containing oligosaccharides on the cell surface. HSV-1 attaches to the fibroblast growth factor receptor, rabies virus to the acetylcholine receptor EBV (Epstein -Barr virus) to the CD21 (complement protein). Viruses may use more than one cell receptor, for example HIV uses the CD4 receptor to attach to the CD4 T-helper cells, but it also uses a chemokine receptor CCR5 as a co-receptor. It is now believed that most viruses use more than one receptor on the cell surface in a sequential binding process.

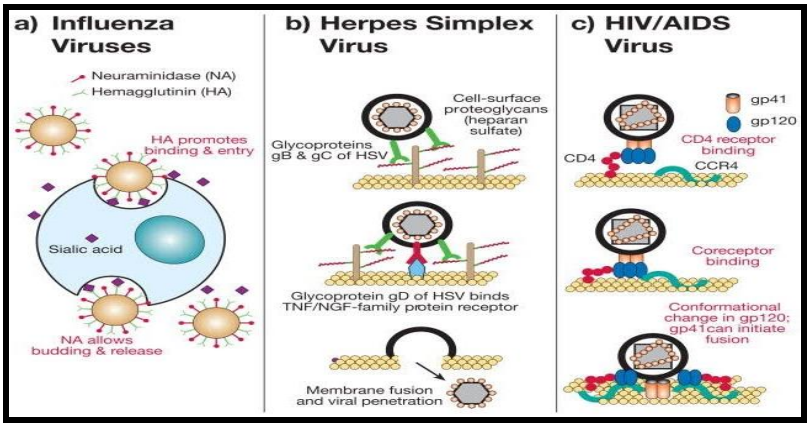


Figure (2): Attachment step in different viruses

2. Penetration or engulfment.

The virus enters the cell in a variety of ways according to the nature of the virus.

- **Enveloped viruses:**

A- Entry by fusing with the plasma membrane. Some enveloped viruses fuse directly with the plasma membrane. Thus, the internal components of the virion are immediately delivered to the cytoplasm of the cell (figure 3).

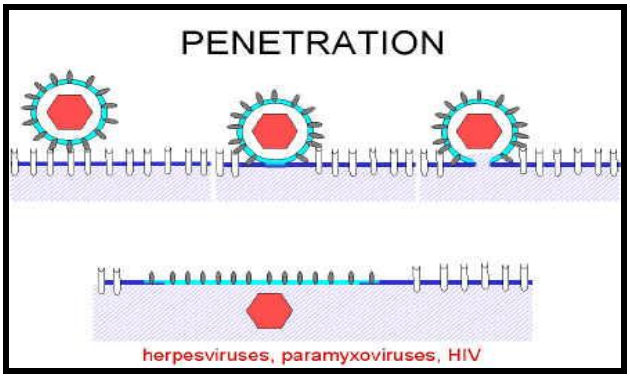


Figure (3): Fusion of a virus with the plasma membrane after attachment to a cell surface receptor

B- Entry via endosomes at the cell surface. Some enveloped viruses require an acid pH for fusion to occur and are unable to fuse directly with the plasma membrane. These viruses are taken up by invagination of the membrane into endosomes (figure 4)

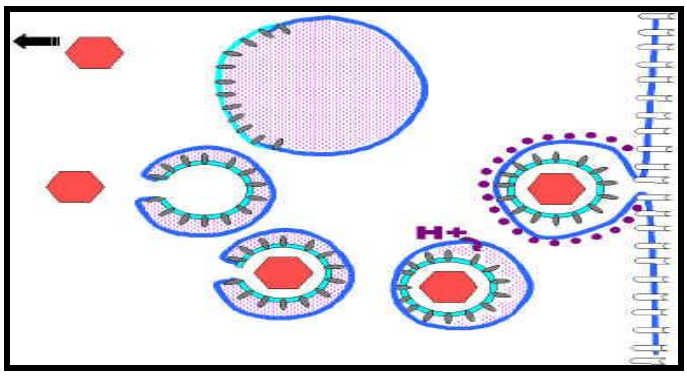
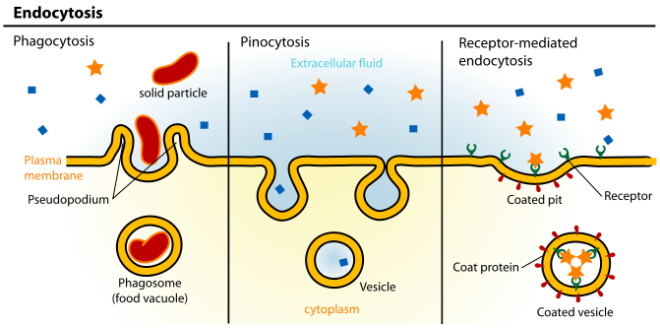


Figure (4): Fusion of a virus with the membrane of an endosome

- **Non-enveloped viruses**.

Non-enveloped viruses may cross the plasma membrane directly or may be taken up into endosomes. They then cross (or destroy) the endosomal membrane.



- **Certain bacterial viruses (bacteriophages)**

This type of viruses have especial mechanism for entering bacteria, T group of bacteriophages infected E. coli by attaching several tail fibers to the cell surface and then using lysozyme from the tail to degrade a protein of the cell wall.

3. Uncoating.

Physical separation of the viral nucleic acid from the outer structural component of the viron.

4. Synthesis of viral nucleic acid and protein. Many strategies are used, some will be discussed.

First step (Transcription)

A- DNA viruses replicate in the nucleus using host cell DNA-dependent RNA polymerase to synthesize their mRNA. Except **Poxviruses** which replicate in cytoplasm, they carry their own polymerase within virus particle.

- DNA virus mRNA is transcribed from the DS DNA viruses in a similar fashion to cellular DNA replication. These viruses can therefore completely depend upon the cellular process to replicate. The genome of these viruses (e.g. cytomegalovirus (CMV), Epstein-Barr virus (EBV)) needs to carry information to code for the virus specific proteins only. Regulatory proteins and those required for viral DNA synthesis are coded early on and the later proteins are generally structural proteins.
- Parvovirus Single stranded DNA viruses are first converted into double stranded, and then mRNA is transcribed as for the DS DNA viruses.

B- RNA viruses divided into four groups.

All RNA viruses replicate in the cytoplasm except **HIV** and **Influenza** viruses (in both cytoplasm and nucleus). And hepatitis dalta virus replicate in the nucleus (is a defective RNA virus which requires the help of a hepadnavirus like Hepatitis B Virus.

- Viruses containing **positive** polarity single strand RNA such as Poliovirus, use their RNA as mRNA and utilize the cell's ribosomes and enzymes to translate the information contained in this RNA to produce viral proteins. One of the first proteins to be produced is a RNA-dependent RNA polymerase, which then transcribes viral RNA into further RNA genomes. These viruses, because they can subvert the cellular system for their own replication, do not need to carry the information for the initial replication enzymes within their genome.

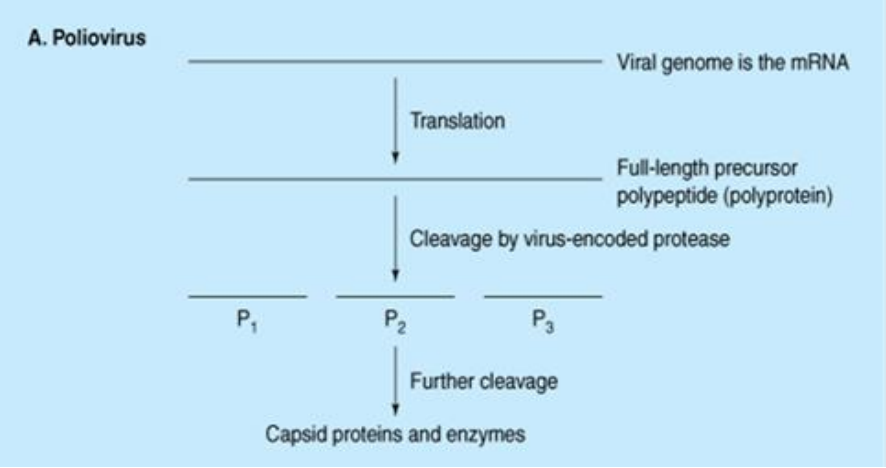


Figure (5): Synthesis of viral nucleic acid and protein in Poliovirus

- Viruses containing **negative** polarity single strand RNA need to convert it first to a RNA strand, which is then used as an mRNA template for translation or direct transcription to the genomic negative RNA. They therefore need to carry a viral-specific RNA-dependent RNA polymerase.
- Single strand RNA of **positive** polarity with reverse transcriptase. The virus in this case carries reverse transcriptase, which changes RNA to single strand DNA and the latter acts as template to produce double strand DNA that is integrated in host cell chromosome and remains asymptomatic, if anything transforms it form latent to active, then host polymerase transcripts virus DNA to mRNA.

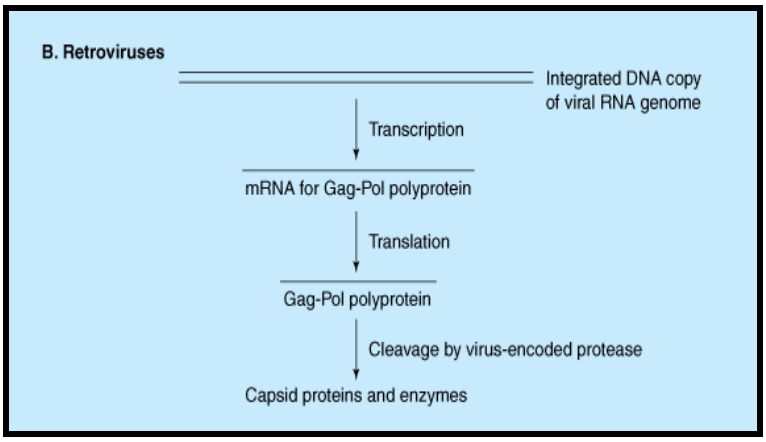


Figure (6): Synthesis of viral nucleic acid and protein in retroviruses

- Ds RNA viruses have to first convert the negative RNA strand of the Ds RNA into a complementary RNA to be used as mRNA. The RNA strand of the DS RNA acts as a template for viral genome replication. These viruses also need to carry the RNA-dependent RNA polymerase to initiate the first steps of viral replication.

Second step (Translation)

Once the viral mRNA of either DNA or RNA viruses is synthesized it is translated by host cell ribosome into viral protein, some of which are early protein, enzyme required for replication of the viral genome and others of which are late protein structural proteins of the progeny viruses. The term early is defined as occurring before the replication of the genome and late is defined as occurring after genome replication. The most important of the early protein for many RNA viruses is polymerase that will synthesize many copies of viral genetic material for make its mRNA.

5. Assembly/maturation: New virus particles are assembled. There may be a maturation step that follows the initial assembly process.

6. Release: Virus may be released due to cell lysis or, if enveloped, may bud from the cell. Budding viruses do not necessarily kill the cell. Thus, some budding viruses may be able to set up persistent infections. Not all released viral particles are infectious. The ratio of non-infectious to infectious particles varies with the virus and the growth conditions.

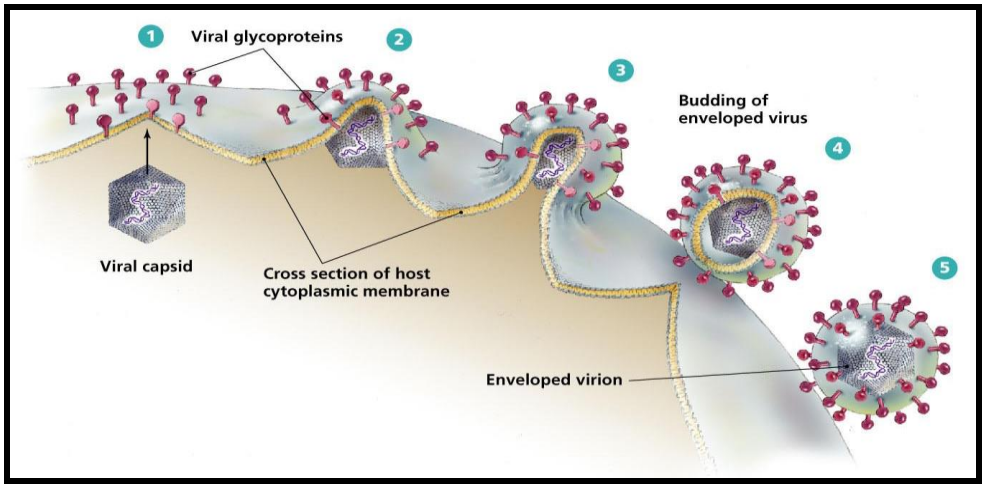


Figure (7): Envelop virus release from cell by Budding

Structure versus non-structural proteins: All proteins in a mature virus particle are said to be structural proteins - even if they make no contribution to the morphology or rigidity of the virion non-structural proteins are those viral proteins found in the cell but not packaged into the virion.

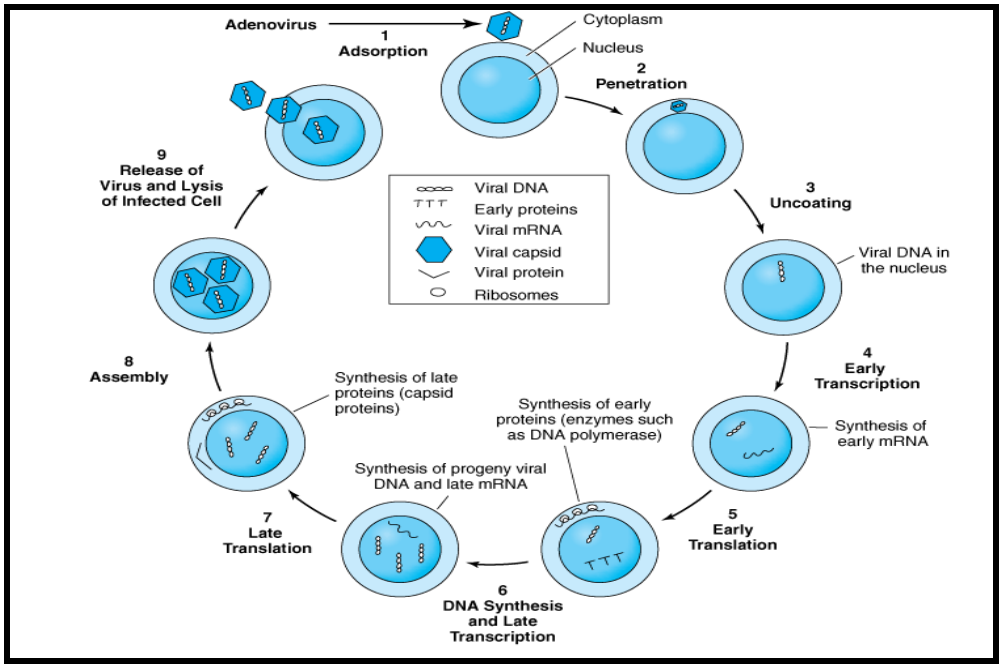


Figure (8): Stepwise of Adenovirus replication