# Viral replication and genetics

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**A virus enters the cell with nothing but its own puny molecule of nucleic acid, which may have only 20 or so genes compared with100 000 genes for a mammalian cell, and sometimes without even a single enzyme to start the process of replication.**

**This is why it must rely so heavily on the host cell for the materials it needs for reproduction and why the replication of viruses is more complicated in some respects than that of other microorganisms.**

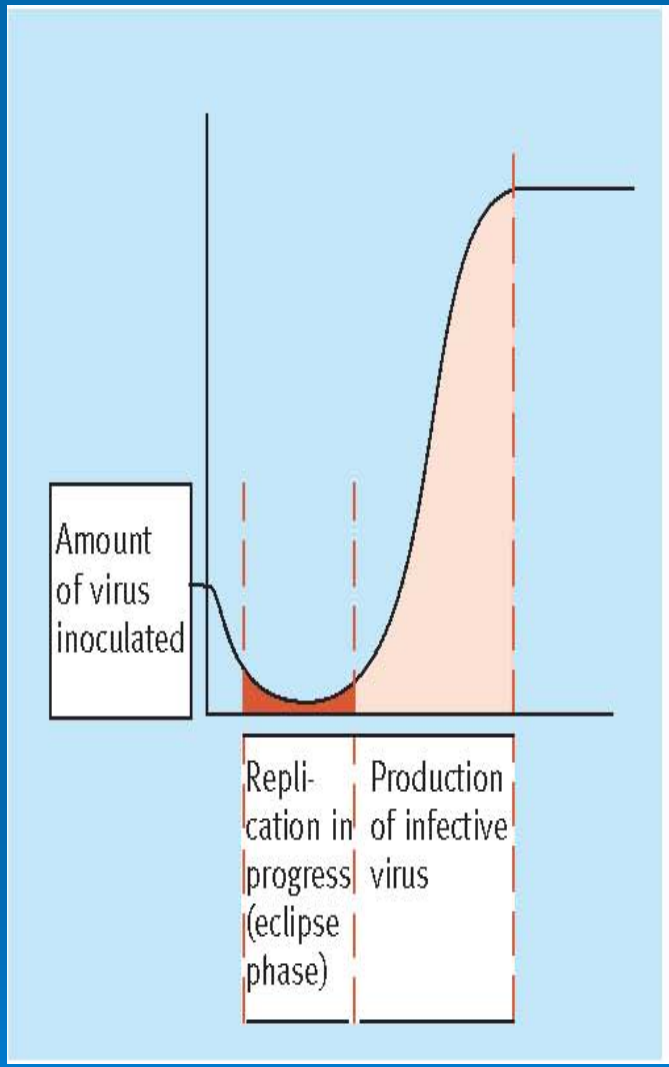
## Virus infection and replication in a host cell

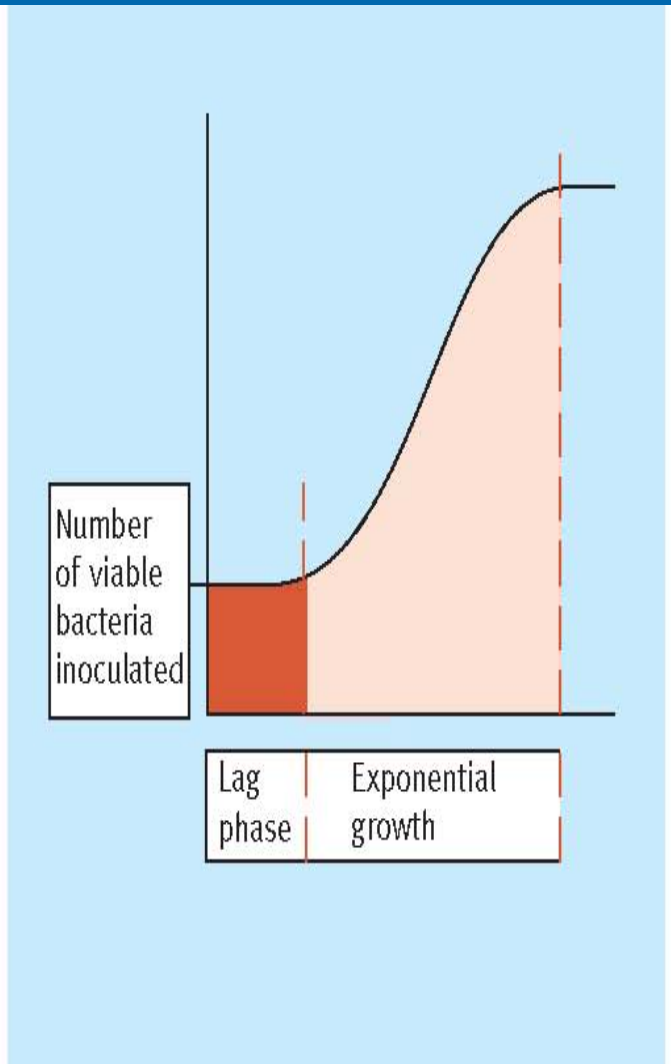
**The initial infection of a cell is a rather hit or miss process, depending upon chance contact, but is greatly helped if a virus enters the body at a suitable site and in large numbers.**

**Often thousands of viruses may enter the body and yet only two or three actually establish an infection. The remainder are destroyed by the general defenses before they have a chance to infect.**

**There follows a period of a few hours during which nothing seems to be happening.**

**This appearance is, however, deceptive because much is going on inside the cell at the molecular level, such as transcription of the 'incoming' viral genes to form viral mRNAs, and their translation to produce early viral proteins, including the enzymes necessary to replicate viral DNA viral RNA.**




Typical growth curve of a virus compared with that of a bacterium.

**A fundamental difference between the replication of viruses and bacteria; the latter retain their structure and infectivity throughout the growth cycle, whereas viruses lose their physical identity and most or all of their infectivity during the initial stage of replication, which for this reason has been termed the eclipse phase.**

The next stage, the **productive phase**, is even more full of action as new virus particles are produced and released from the cell.

## General plan of viral replication

**No single virus is typical of them all.**

**We have chosen a DNA poxvirus because the sequential steps in its replication are comparatively easy to follow. It should, however, be mentioned that this example is not typical of DNA viruses in that it replicates entirely in the cytoplasm, carries many of the enzymes needed for viral transcription and replication and sets up small virus 'factories'.**

**(1) Penetrating any mucus or other physical barriers, the virus adsorbs to a host cell using a specific receptor on the cell membrane.**

**(2) A few minutes later it has entered the cytoplasm of the cell, after which**

**(3) it ‘uncoats’ (i.e. sheds its protective protein shell). In the case of poxvirus this uncoating is only partial. In other viruses the uncoating is more complete and the viral nucleic acid completely frees itself**

**(4) The poxvirus input DNA is transcribed to produce various viral mRNAs, which code for:**

(5) **‘Early’ viral proteins. Viral proteins are produced by the ribosomes of the host cell.**

**(There are as many as 100 early genes distributed throughout the poxvirus genome. Other viruses have only a handful of genes. The early viral proteins may have various functions; for example, some are DNA-dependent DNA polymerases that catalyse and direct the synthesis of new viral DNA molecules).**

**(6) Other early proteins are transcriptional activators that speed up the viral transcription process.**

**In contrast, the late viral mRNAs are transcribed only from newly synthesized viral DNA. The ‘late’ proteins translated from these late viral messages are mostly viral structural polypeptides**

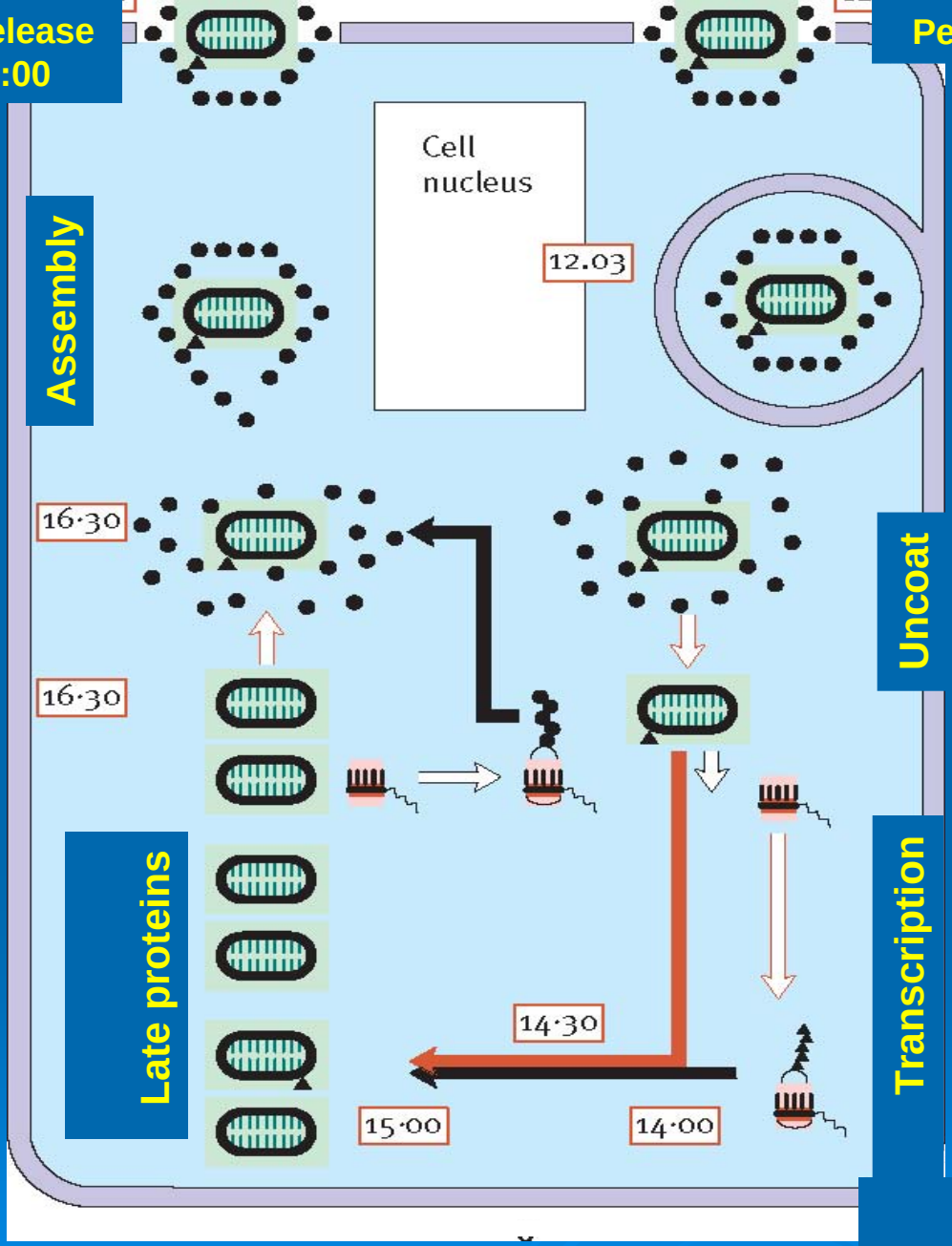
**(7) Assembled with the new DNA to form progeny virions. Assembly occurs in circumscribed areas of the cytoplasm in the case of the poxvirus and immature virions can be seen easily by EM.**

**Other viruses assemble at the plasma membrane itself or in the nucleus.**

**(8) The new virions are then released from the cell by a mixture of budding and cell lysis.**

**- In this example, the whole process takes a minimum of 6–8 hours.**

**- The new infective virions are then free to infect neighbouring cells and start the process over again. As many as 10 000 virions may be released from an infected cell.**




Life cycle of vaccinia, a DNA virus. Adsorption and penetration occur rapidly. Unlike other DNA viruses, vaccinia replicates exclusively in the cytoplasm. Initial transcription takes place in the core of the virion. The genomic DNA strands are covalently linked at their ends. Early mRNAs, coding for enzymes that have an early function, such as replication of input DNA, are transcribed from input DNA. Late mRNAs coding for viral structural proteins are transcribed from newly synthesized DNA.

## Recognition of a ‘target’ host cell

- All viruses have on their outside a receptor-binding protein, which often has a saucer-shaped pocket that reacts specifically with a corresponding receptor on a cell surface.

-These receptors usually have other functions and viruses simply use them for attachment. The virus receptors on cells are often glycoproteins or glycolipids.

- Once attached, which may be a more or less instantaneous process, viruses are almost impossible to dislodge.

-This precise key-and-lock interaction explains why many viruses are restricted to a given host and, within that host, to particular cells and tissues.

## Internalization of the virus

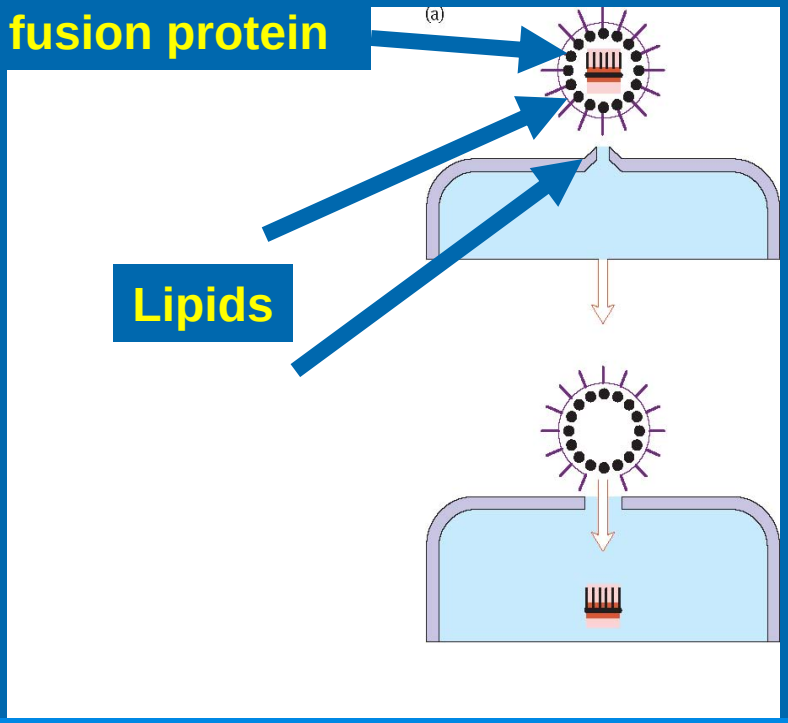
Having attached to the viral host cell, the virus must penetrate the external plasma membrane of the cell rapidly and release its genome into the cellular milieu for subsequent replication. This internalization is accomplished in one of three ways.

## (A) Fusion from without

**Fusion at the cellular external plasma membrane, namely 'fusion from without', is the strategy of entry of paramyxoviruses such as measles and mumps viruses, and also HIV.**

**Such viruses have a 'fusion protein', with a short stretch of catalytic hydrophobic amino acids, which mediates fusion between the lipids of the virus and the lipids of the cell membrane.**

Viral fusion protein




(a) Fusion from without

# (B) Receptor-mediated endocytosis (viropexis)

Viropexis is the most common cellular entry technique for viruses.

Mammalian cells developed methods of attachment and entry of a range of essential molecules, such as nutrients and hormones.

Viruses can exploit these existing avenues of entry.

Viruses attach at special virus receptor areas on the cell membrane.

The cellular protein, clathrin, which underlies the membrane, forms a so-called coated pit and, once the virus has attached, inversion of the cellular membrane and associated virus occurs.

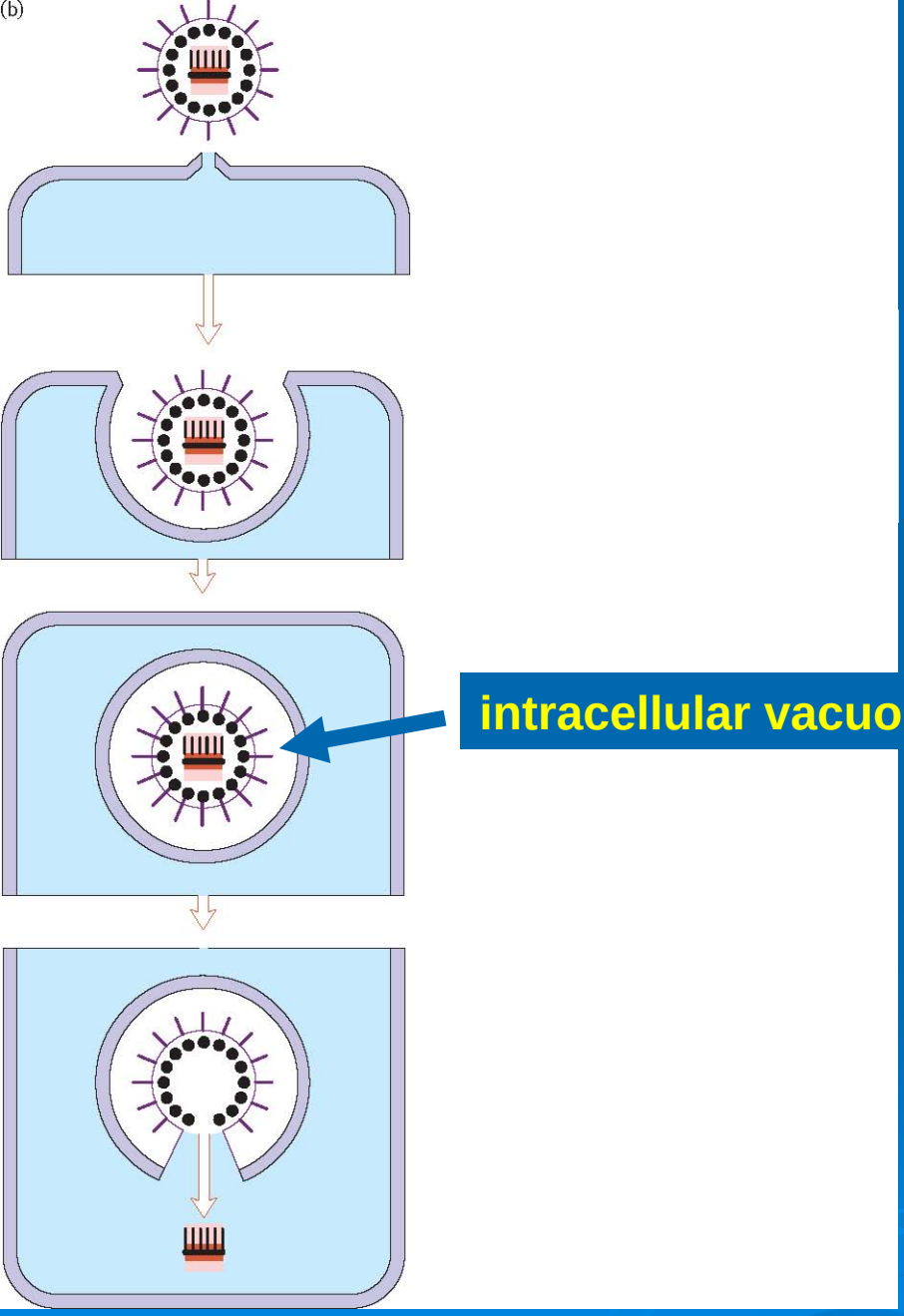
The virus is now in the cytoplasm but is still bounded by the cell membrane through which it has to negotiate a route to the true internal environment and often to the nucleus of the cell.

**It is a mystery how the viral nucleic acid, particularly ssRNA, protects itself from destruction by the many nucleases present in the cytoplasmic vacuole, but presumably the tightly bound viral nucleoproteins provide protection.**

**These endosomes offer a convenient and rapid transit system across the plasma membrane and also through the cytoplasm to the nuclear pore.**

**Some viruses, such as influenza, achieve release from the internal endosomal vacuole by internal fusion ('fusion from within') mediated by the viral haemagglutinin (HA) protein.**

**A further requirement of internal fusion with influenza is a low pH in the cytoplasmic vacuole; this triggers a movement of the HA protein, and the release of nucleic acid to cytoplasm.**




intracellular vacuole.

## (c) Non-clathrin-mediated endocytosis

A few viruses may enter by a third technique known as nonclathrin-mediated endocytosis or via a caveolae assisted entry. (Cavaole= latin for little caves, 50-100 nm invaginations of the P.M.)

In all cases quite extensive internal trafficking occurs before the virus RNA is released from the internalized virus and enters the nucleus via the nuclear pore.

## (D) Viral translocation

**The entire virus particles crosses the P.M. of host , it is rare entry method and is poorly understood.**