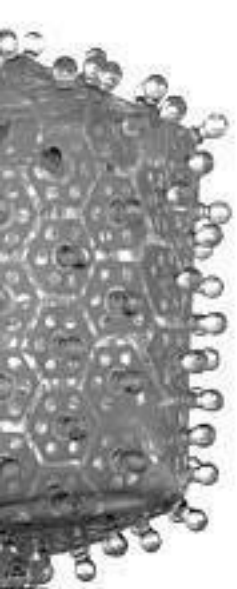
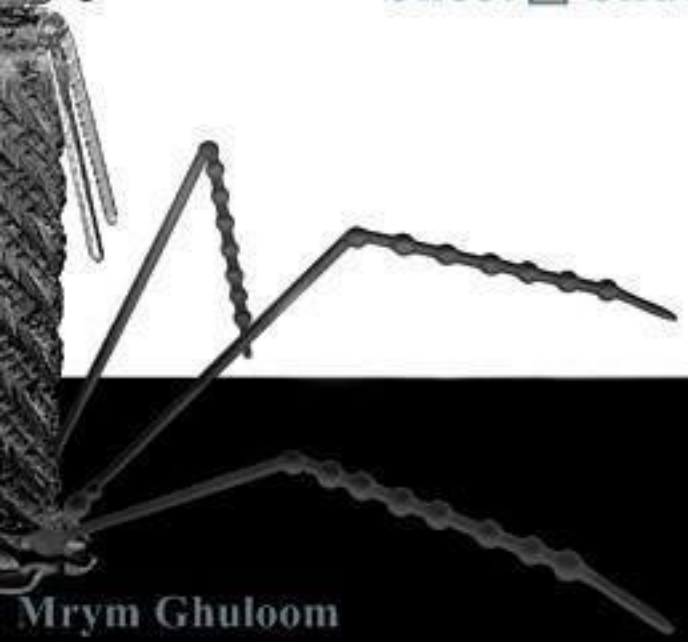


Microbiology

Lecture No: 4 - virology

Dr Name: Hamed Al-zoubi

Sheet ☐ **Slide** ■



Mrym Ghuloom

Virology – replication and genetics
JU- 2nd Year Medical Students

By

Dr Hamed AlZoubi – Microbiology and Immunology Department – Mutah University.

MBBS (J.U.S.T)

MSc, PhD medical microbiology (UK).

FRCPath (associate, medical microbiology).

dr_alzoubi@yahoo.com

Virology – replication and genetics

- Genetics
- Life cycle:

✓ Influenza
✓ Polio
✓ Rabies
✓ Adenovirus
✓ HIV

Viral Genetic Variation

1- Low fidelity of reverse transcriptases and RNA replicases compared to DNA polymerase (proof reading)

- **Mutations**
- **Quasi - species**

Viral Genetic Variation

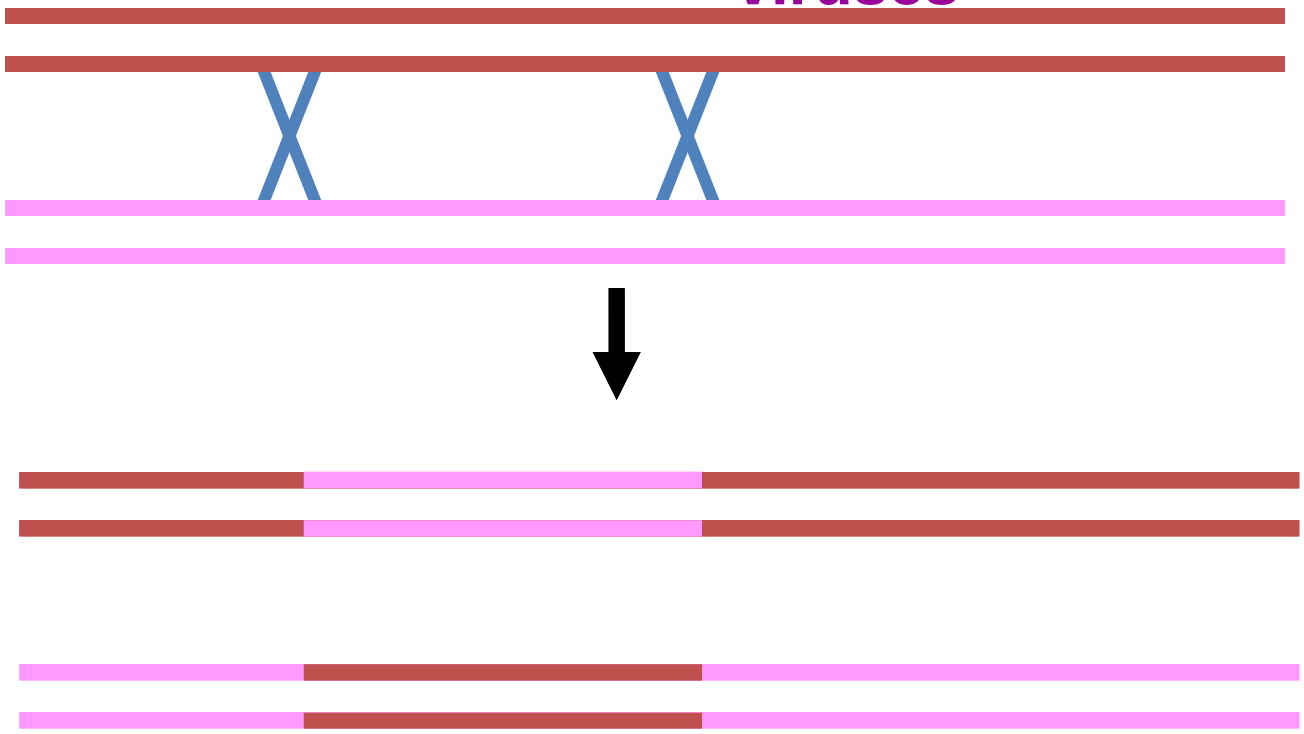
2- Recombination

- ✓ DNA viruses: breakage
- ✓ RNA related viruses:
- when the virus polymerase switches template strands during genome synthesis
- the new recombinant virus has properties incompatible with survival

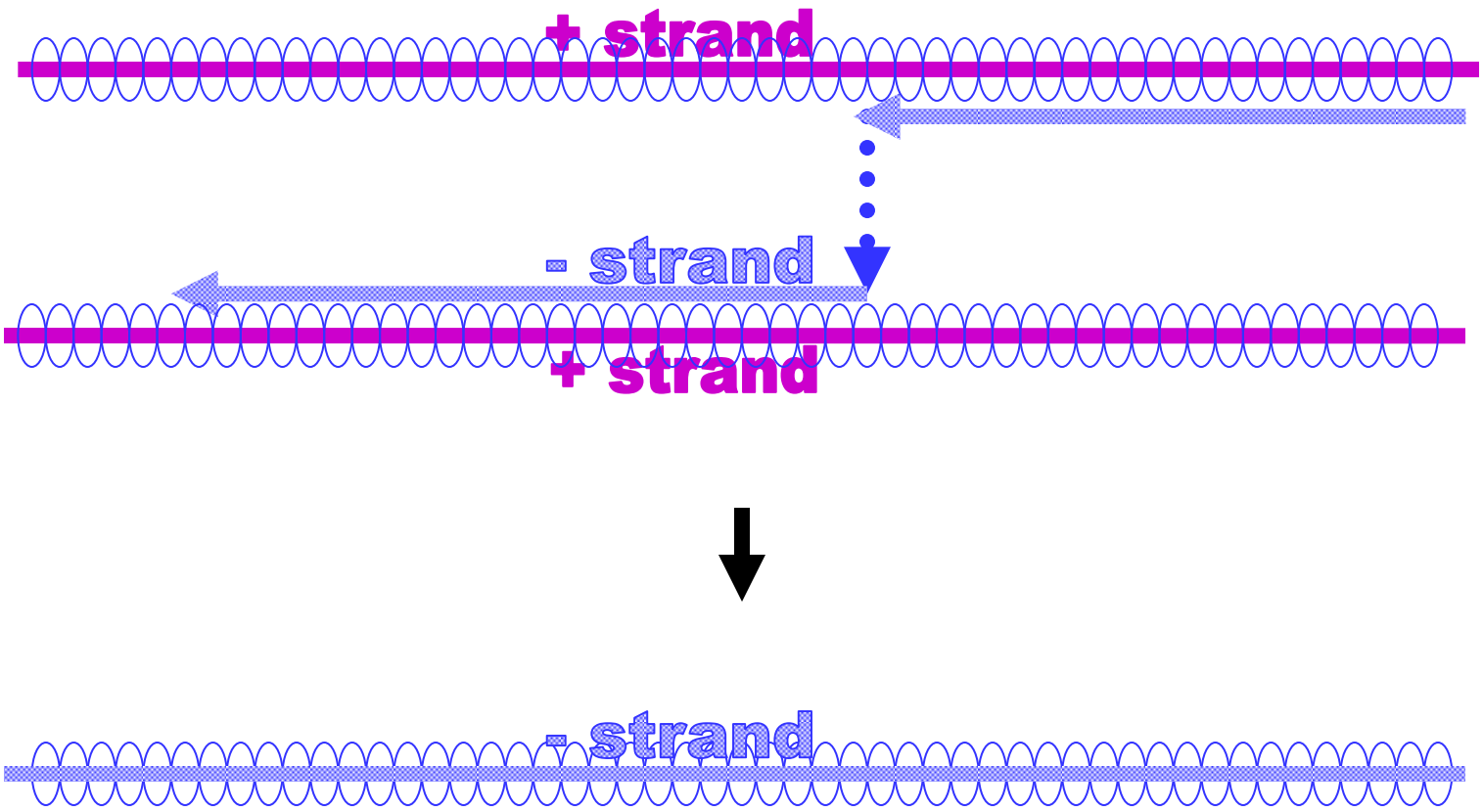
RECOMBINATION-DNA

‘classic’ recombination

common in DNA viruses



RECOMBINATION – RNA viruses



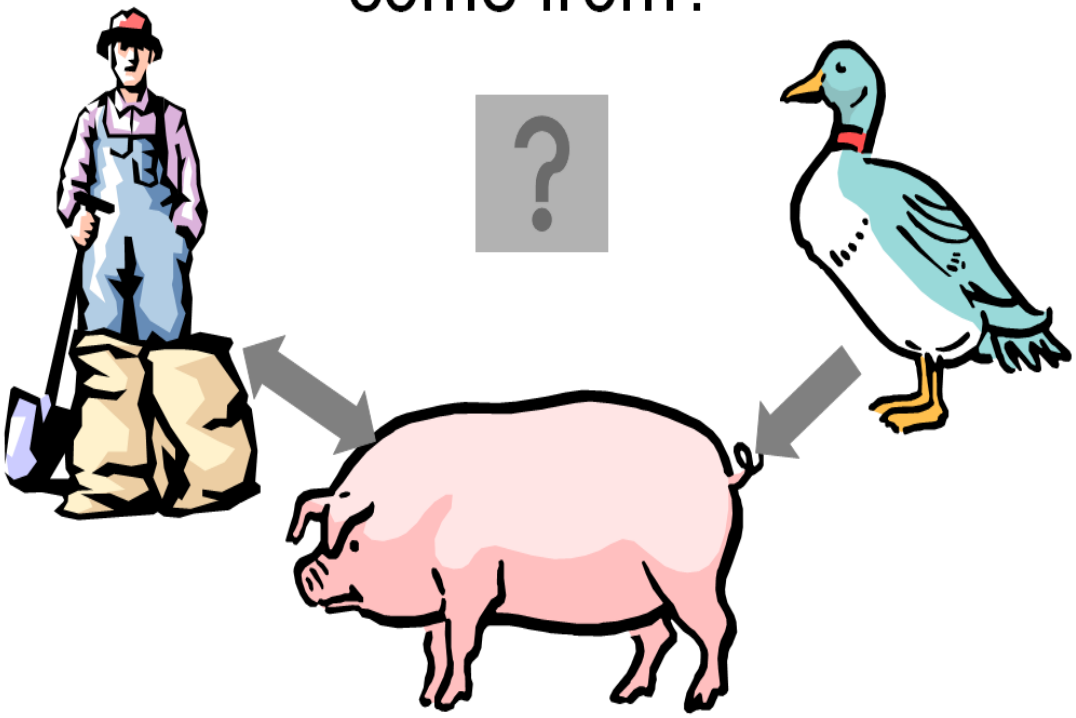
- 3- reassortment:
- In segmeted RNA viruses
- exchange of genes
- More frequent than recombination

- can extend the gene pool of the virus and allow the emergence of new and successful variants.

- An example is the infrequent appearance of pandemics of influenza (in 1918, 1957, and 1968), caused by reassortment of genes between human, avian, and pig influenza A viruses.

- Anovel mutant may be created that can cross the species barrier and infect humans.

where do “new” HA and NA come from?



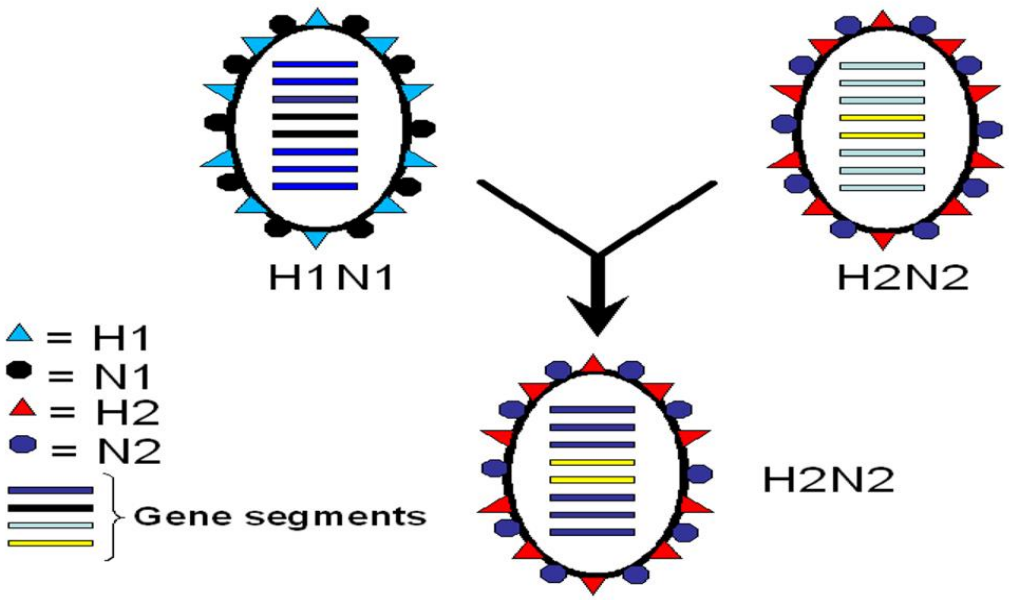


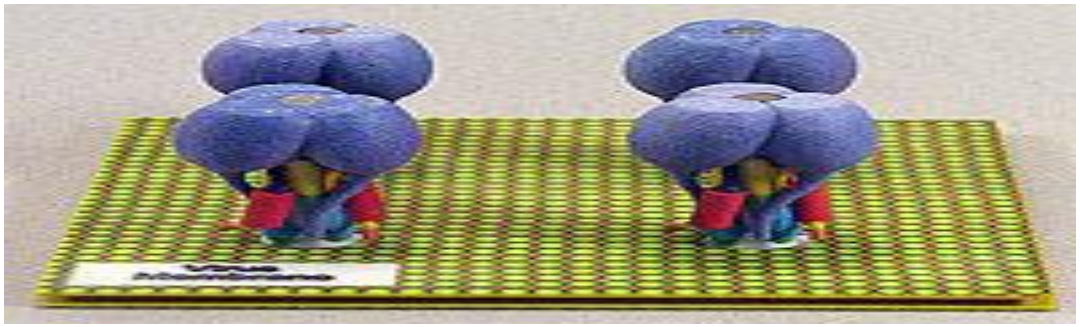
Figure 4. Mechanism of reassortment of RNA segments of influenza virus

Influenza virus (-ssRNA)

- *The influenza HA spike protein is shaped like a 'Toblerone' chocolate bar and protrudes from the virus surface (500 spikes on each virion)*

- *each spike composed of three identical subunits with a bulb-shaped hydrophilic portion furthest from the viral membrane*

- *a narrower hydrophobic stalk attaches the spike to the viral lipid and protrudes through it to anchor the spike to the underlying membrane of matrix protein.*



- *The most exposed and distant region of the HA contains:*

1- The antigenic sites, which often protrude from the HA

2- and a receptor-binding site, a saucer-shaped depression near the HA tip.

- *complete HA molecule, which during synthesis in the cell is cleaved into the two pieces by a protease*

- *Two polypeptides (HA1 and HA2), joined together by disulphide bonds, constitute the HA.*
- *An influenza virion with a cleaved HA is more infectious than one in which the HA remains as a single protein.*

- *The low pH environment of cytoplasmic endosomes triggers a massive movement of the chain of amino acids in HA1/HA2 and the whole HA molecule becomes contorted.*

↓

The central junction of HA1 and HA2 comes into contact with the lipid membrane of the endosome

The fusion motif of HA2 triggers the fusion of viral and endosomal lipids → *RNA release into nucleus* → *replication*

Neuraminaidase:

- *Antigenic sites were identified*
- *the enzyme active site on the mushroom head,*

- *anti-influenza drugs, designed to sit precisely in the NA active site and so block enzyme action.*

- *NA is essential for release of virus from infected cells so that these NA inhibitors should inhibit viral release e.g Oseltamivir and Zanamivir*

Influenza virus

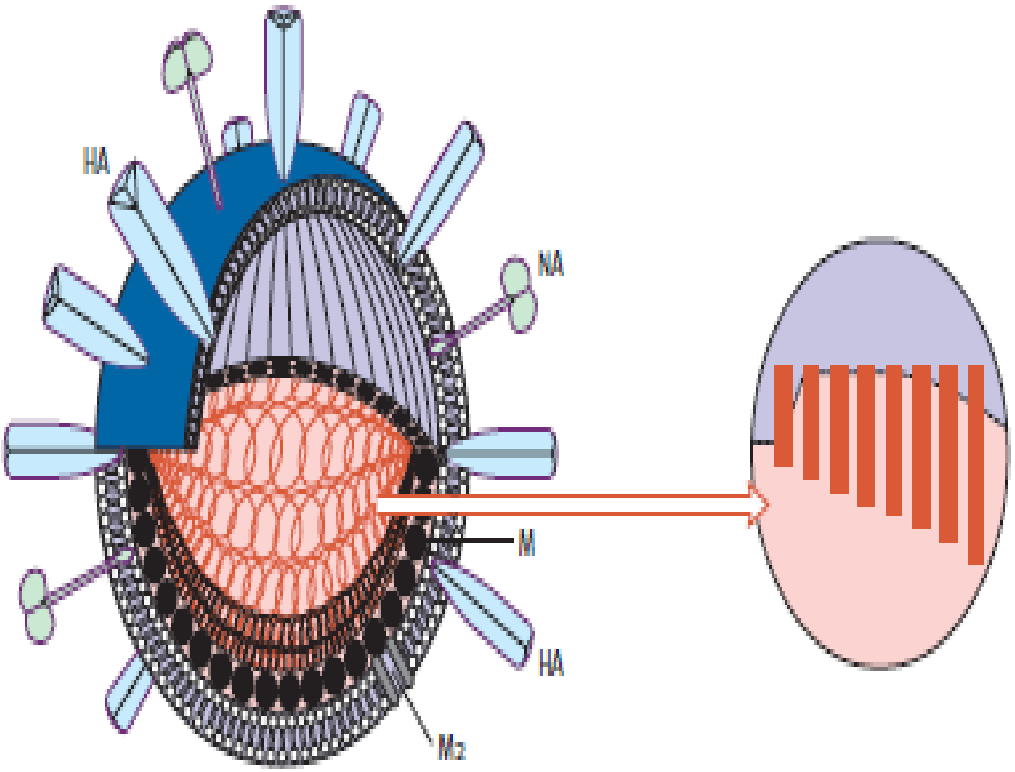
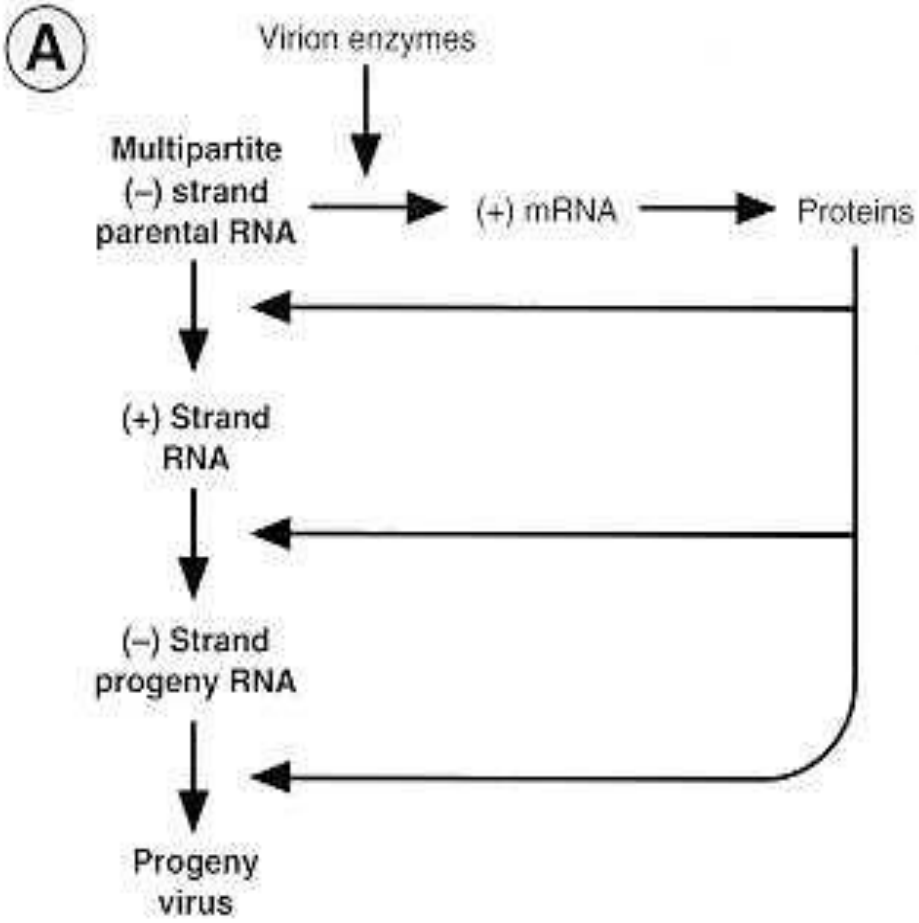


Fig. 2.2 Structural features of influenza virus. The spikes of HA and NA protrude from the lipid bilayer; beneath this is a layer of M protein, which in turn encloses the segmented RNA genome, each segment of which is covered with the nucleocapsid protein and has attached additional structural proteins PB1, PB2, and PA, which are involved in genome replication. 'Pores' of M2 penetrate through the lipid and function as ion channels.

Flow of events during the replication of Orthomyxoviruses.



Influenza virus

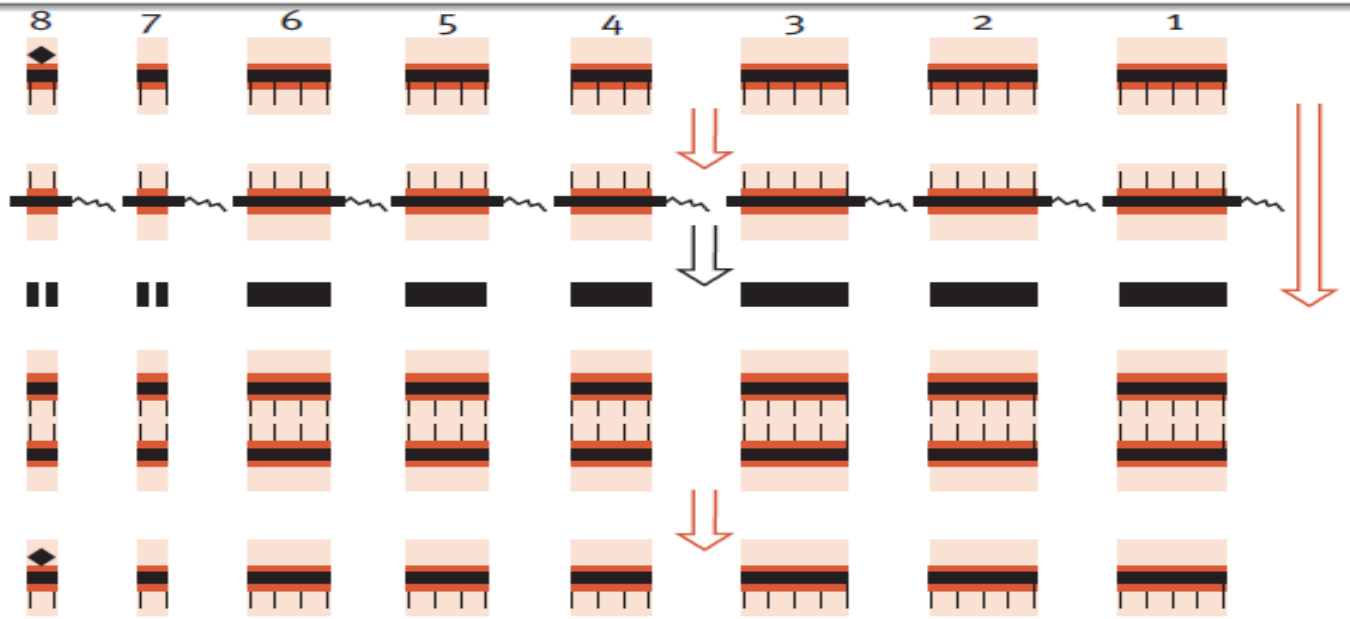
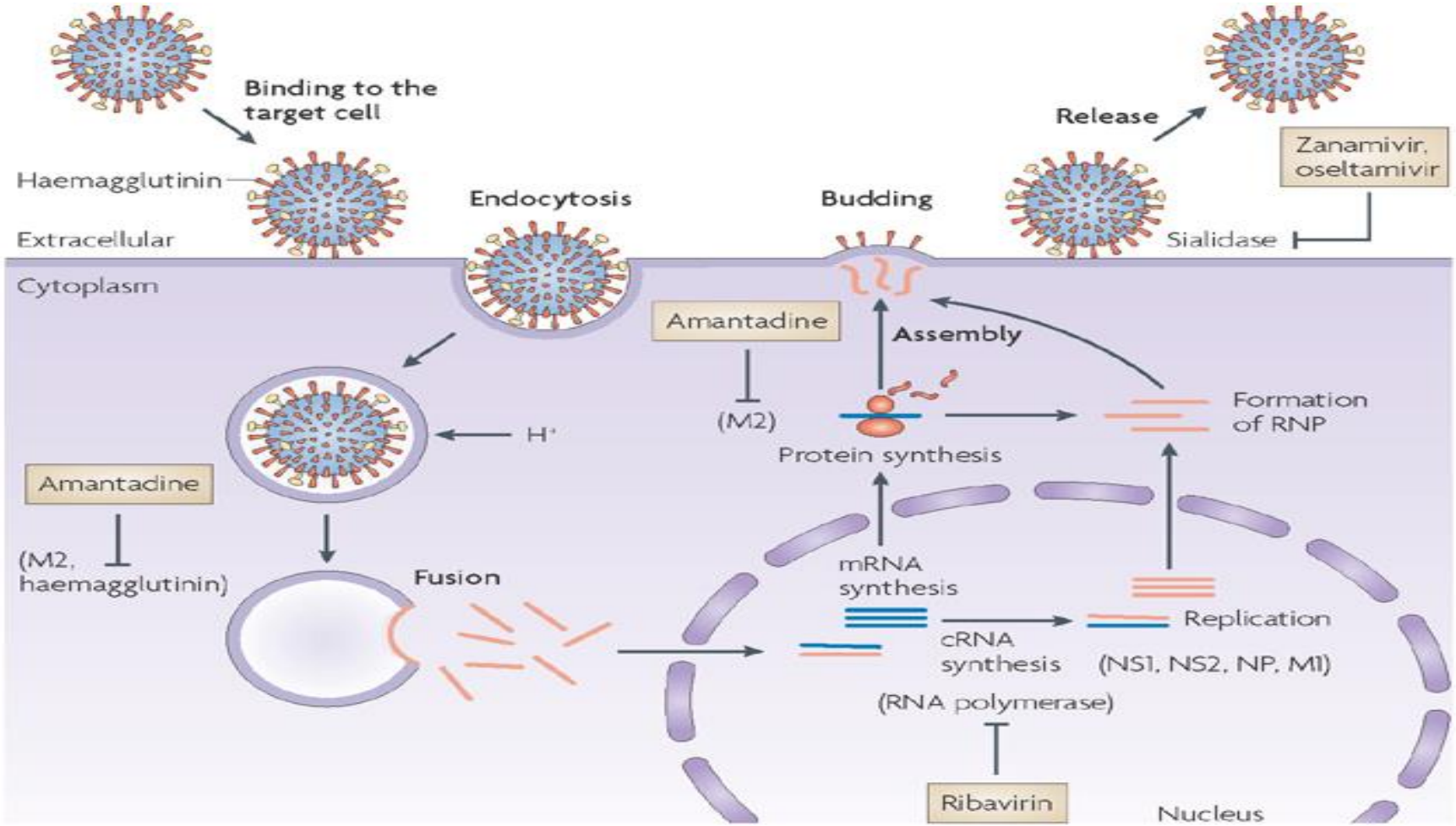


Fig. 3.6 Replication strategy of influenza, a negative-stranded RNA virus. The viral genome is in the form of eight loosely linked single-stranded RNA segments. Most transcribed mRNAs are monocistronic, i.e. they code for a single protein. However, the mRNAs of genes 7 and 8 have undergone splicing and each now codes for two viral proteins. The mode of transcription and replication of influenza virus is unique as it requires co-operation with cellular RNA polymerase II ('cap snatching'). ~, poly(A) tail; ◆, RNA-dependent RNA polymerase.

Influenza



Nature Reviews | Drug Discovery

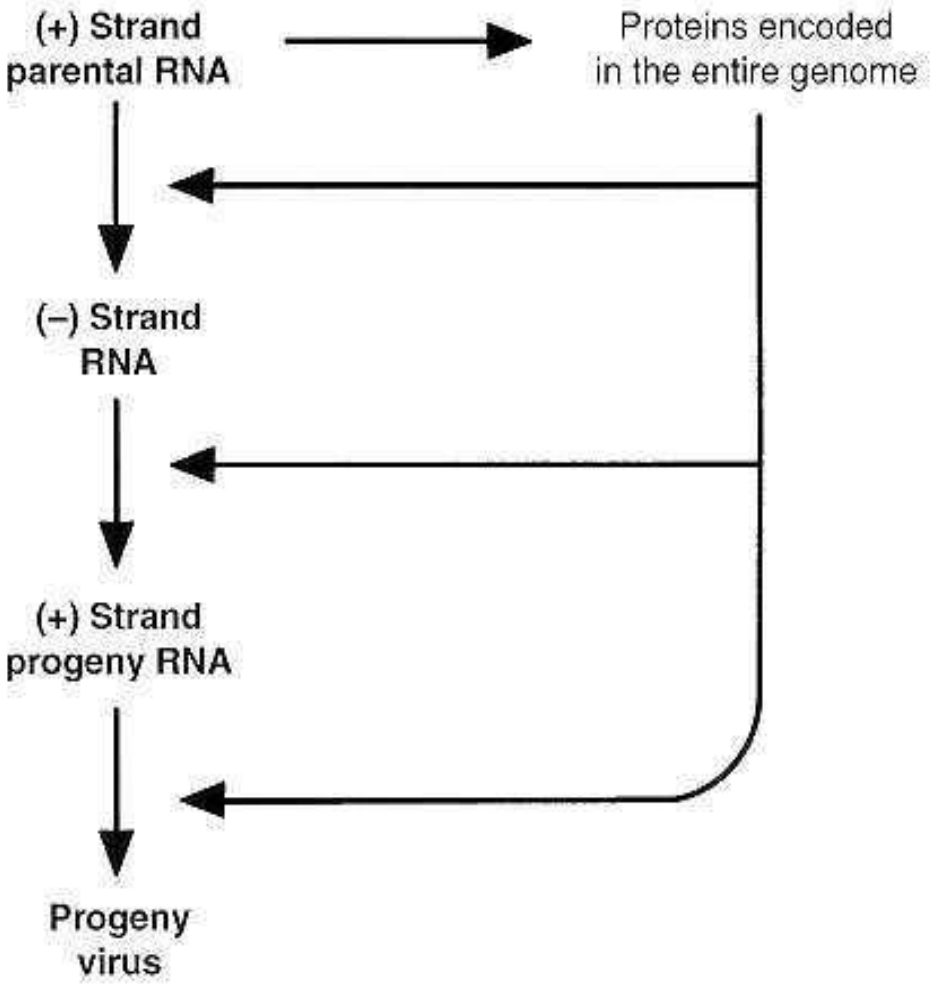
Poliovirus (+ssRNA)

- The virion RNA molecule is polyadenylated at the 3 end and a small virus-coded protein (VPg) is present at the 5 end.
- The genome has a single open reading frame whose primary translation product is a polyprotein, which is cleaved to produce viral capsid proteins:

✓ (VP1,VP2, VP3, and VP4), the RNA polymerase, two proteases, and some minor products

- Non-translated regions (ntr) at each end of the genome are not translated into proteins.
- The 5 end noncoding region has - initiation of protein synthesis.

Flow of events during the replication of Picornaviruses



Poliovirus

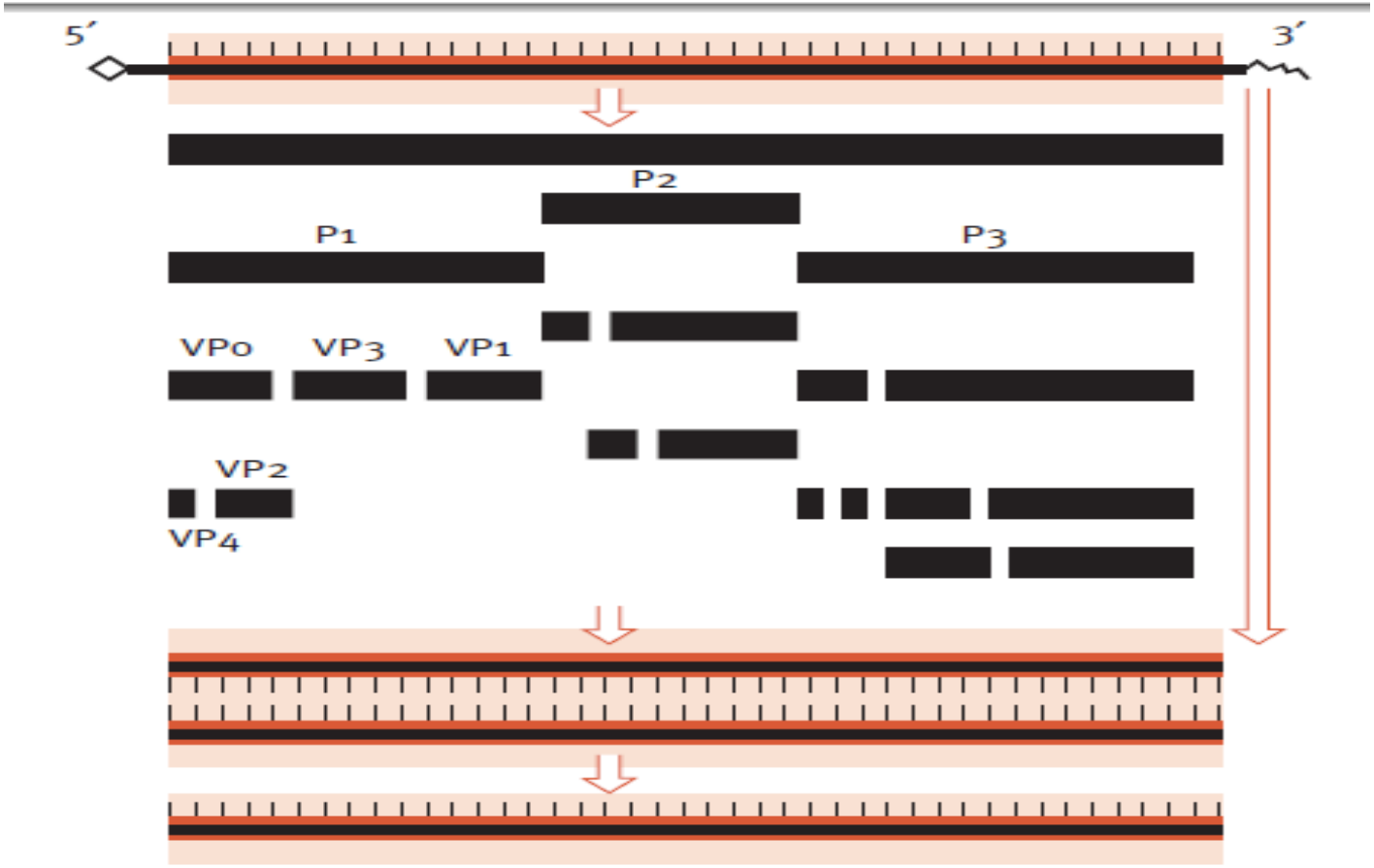


Fig. 3.5 Replication strategy of polio, a positive-stranded RNA virus. The genomic RNA acts directly as mRNA and is translated to give a polyprotein, which is rapidly cleaved by virus-coded proteases into 12 or more smaller proteins (not all illustrated). At a later stage during replication the number of positive RNA strands increases and these are used either as mRNAs or are packaged into virions. ~ poly(A) tail; ◇ 5′ cap.

Rabies: a negative-strand RNA virus

- A virion - associated polymerase transcribes the five genes, which are arranged sequentially on the ssRNA genome into 5 capped, methylated, and polyadenylated mRNAs

- These are translated into the nucleocapsid (N), core phosphoprotein (P), matrix (M), glycosylated membrane spike (G), and polymerase (L)polypeptides.

Rabies

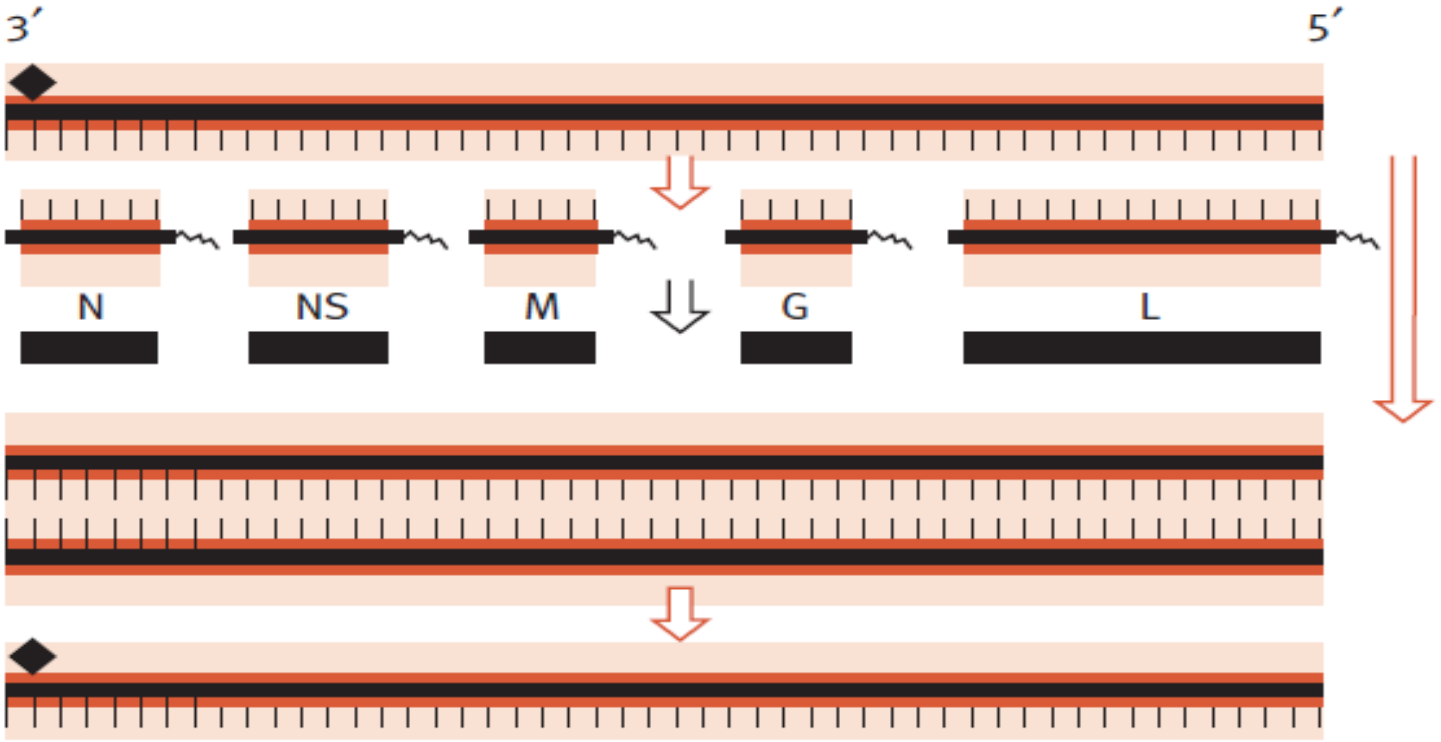


Fig. 3.7 Replication strategy of rabies, a negative-stranded RNA virus. The viral genome is in the form of a single complete strand of RNA. The five genes are positioned in a linear manner. There is an intergenic sequence, a translation start signal and a poly(A) signal at the end of each gene. Five mRNAs are transcribed by a start-and-stop mechanism and each is translated into a viral protein. ∿, poly(A) tail; ◆, RNA-dependent RNA polymerase.

Adenovius: Linear ds DNA

- 36 kbp, containing 30 genes
- There is a terminal protein attached to the 5 end of each DNA strand, which acts as a primer during genome replication and initiates synthesis of new DNA strands.

- Groups of genes are expressed from a limited number of shared promoters; to extend the genetic information

- viral-spliced mRNAs are utilized to produce a variety of polypeptides from each promoter.

Adenovirus

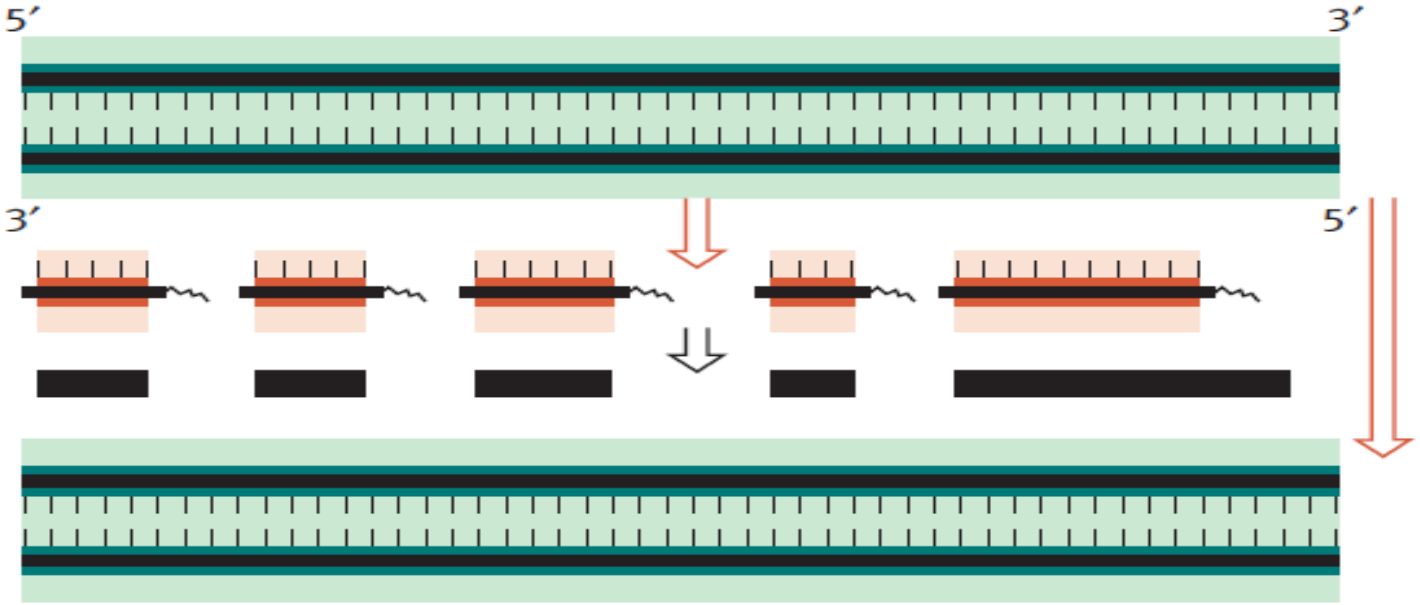
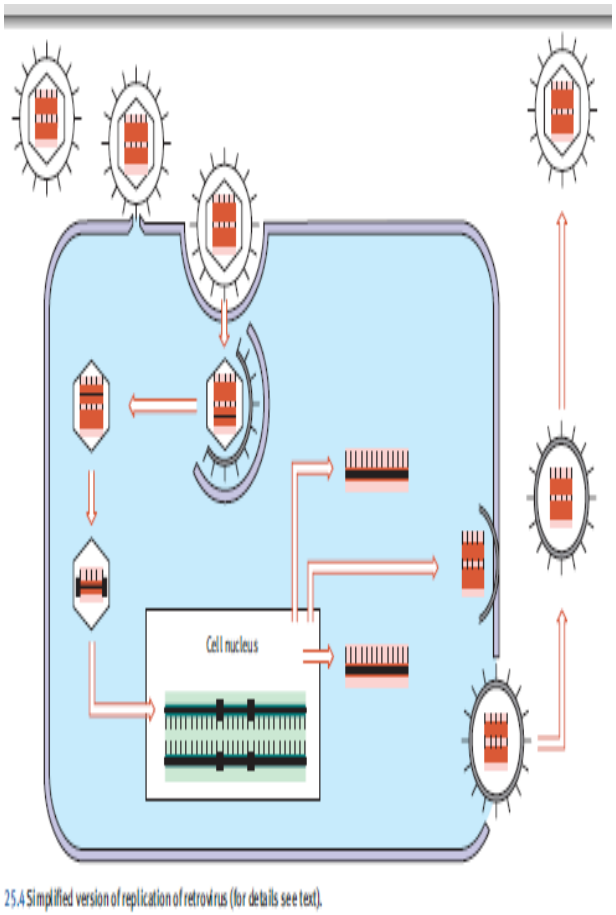
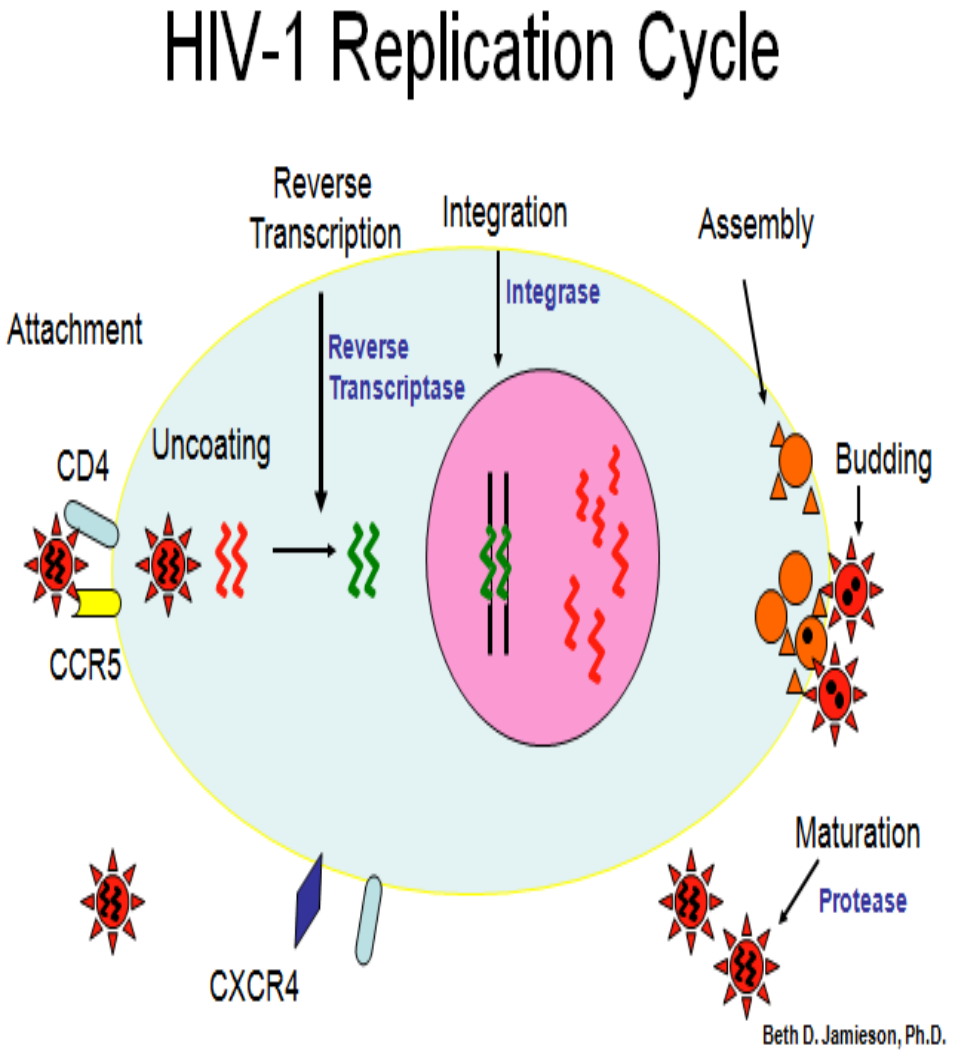


Fig. 3.8 Replication strategy of adenovirus, a DNA virus. The adenovirus genome is transcribed and replicated in the cell nucleus. Replication is mediated by a protein (P) at the 5′ end of each DNA strand. Multiple mRNAs (not all shown) are transcribed from both DNA strands. Early mRNAs are encoded by input parental DNA. Later mRNAs are encoded on both DNA strands. Splicing is extensively utilized and can provide control of different regions of the genome, as well as a means of changing the reading frame. ~~, poly(A) tail.

Retrovirus (+RNA)



25.4 Simplified version of replication of retrovirus (for details see text).



The End