

ESALQ

Escola Superior de Agricultura Luiz de Queiroz Universidade de São Paulo

LFT5870 AGENTES CAUSAIS DE DOENÇAS DE PLANTAS: VÍRUS

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PARTE IV

PROCESSOS DE INFECÇÃO E REPLICAÇÃO



1. INTRODUÇÃO

2. PROCESSO INICIAL DE INFECÇÃO

A. Vírus de animal

- a) Adsorção: contacto com a célula
 Proteínas ou glicoproteínas da partícula.
 Receptores na superfície da célula.
 (glicoproteínas ou lipoproteínas)
- b) Penetração: entrada no citoplasma
 - Vírus sem envelope: translocação por endocitose
 - Vírus com envelope:

Endocitose: Partícula é presa numa invaginação da célula, e liberada dentro desta na forma de vesícula.

Fusão: envelope viral funde-se com a membrana e o virion passa diretamente para dentro.

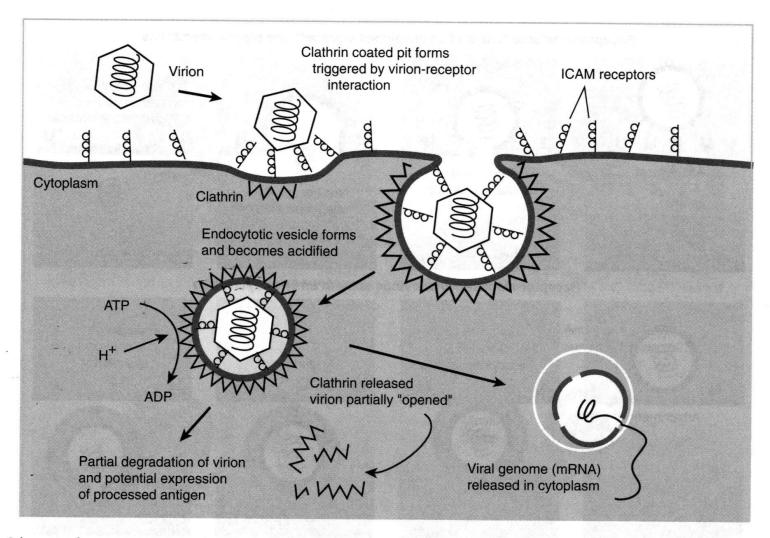


Fig. 6.2 Schematic of receptor-mediated endocytosis utilized by poliovirus for entry into the host cell. The endocytotic vesicle forms as a consequence of close association between the poliovirus-receptor complex and the plasma membrane.

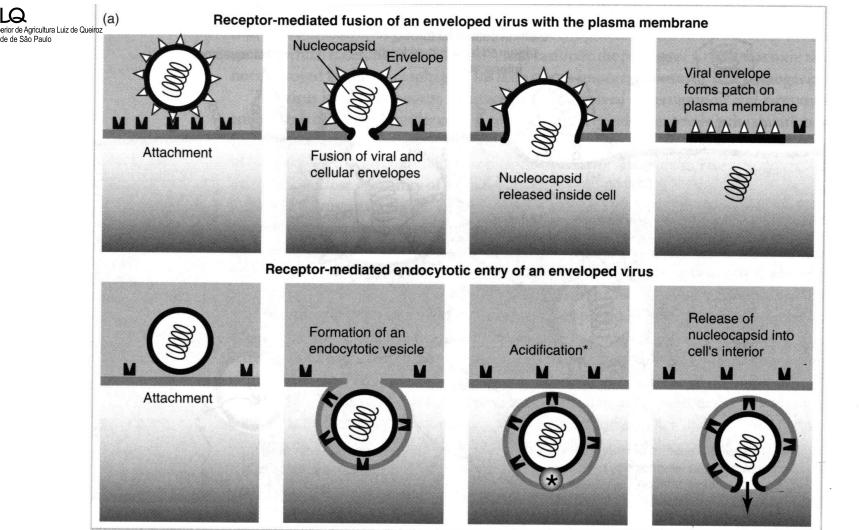


Fig. 6.3 a. The two basic modes of entry of an enveloped animal virus into the host cell. Membrane-associated viral glycoproteins either can interact with cellular receptors to initiate a fusion between the viral membrane and the cell plasma membrane, or can induce endocytosis. The fate of the input virus membrane differs in the two processes. b. The fusion of pseudorabies virus with the plasma membrane of an infected cultured cell is shown in this series of electron micrographs (the bars represent 150 nm). Although each electron micrograph represents a single event "frozen in time," a logical progression from the initial association between viral envelope glycoproteins and the cellular receptor on the plasma membrane through the fusion event is shown. The final micrograph contains colloidal gold particles bound to antibodies against the viral envelope glycoproteins (dense dots). With them, the envelope can be seen clearly to remain at the surface of the infected cell. (Micrographs reprinted with the kind permission of the American Society for Microbiology from Granzow, H., Weiland, F., Jöns, A., Klupp, B., Karger, A., and Mettenleiter, T. Ultrastructural analysis of the replication cycle of pseudorabies virus in cell culture: a reassessment. Journal of Virology 1997;71:2072–82.)

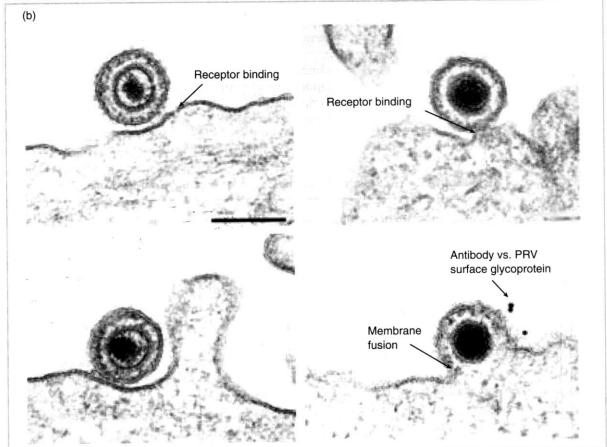


Fig. 6.3 Continued

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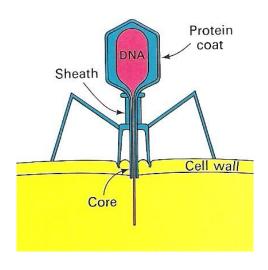
B. Vírus de bactéria (bacteriófagos)

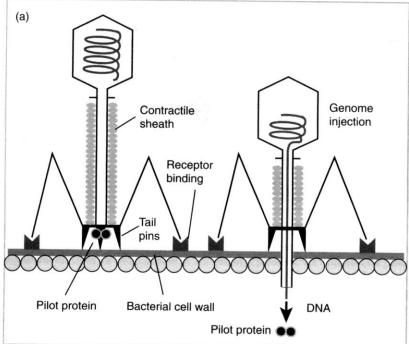
a) Adsorção

Fibras longas: reconhecimento e aderência na célula. Glicoproteínas, lipopolissacarídeos, "sex pilus": receptores na célula.

b) Penetração

c) Retirada da capa protéica





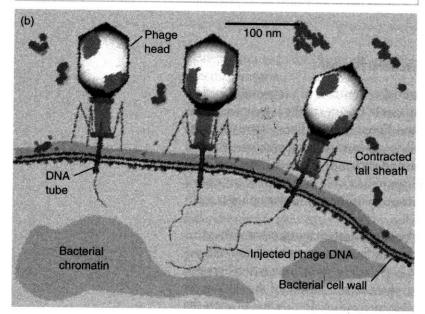


Fig. 6.4 Entry of T4 bacteriophage DNA into an *E. coli* cell. Initial attachment is between the fibers to the ompC lipopolysaccharide receptor on the bacterial cell wall (a). The binding of protein pins on the base plate to the cell wall leads to contraction of the tail fibers and sheath proteins, leading to insertion of the tail tube through the cell wall. As shown in the electron micrograph (b), phage pilot protein (arrow) allows the highly charged viral DNA genome to penetrate the bacterial plasma membrane and enter the cell. Phage DNA can be seen as shadowy lines emanating from the tail tube. (From Dimmock, N.J., and Primrose, S.B. *Introduction to Modern Virology*, 4th edn. Boston: Blackwell Science, 1994.)



C. Vírus de plantas : inoculação por ferimento

a) Adsorção

- Não há evidências de que a capa protéica tem função de reconhecimento ou adsorção.

EX:

RNA Cowpea chlorotic mottle virus (CCMV) em capa protéica de Brome mosaic virus (BMV). Ambos gênero Bromovirus. Infecção em caupi.

CP - BMV

 Não há evidências de que há receptores de vírus nas células da planta.

b) Penetração

- Dano na parede celular: entrada direta.

c) Retirada da capa protéica

- Ácido nucleico é retirado da capa em alguns minutos.



REPLICAÇÃO DE VÍRUS DE PLANTAS

RNA, fita simples (+)	62 (68,8)	76,6%
RNA, fita simples (-)	07 (7,8%	13%
RNA, fita dupla	06 (6,8%)	4,3%
DNA, fita simples	06 (6,8%)	4,1%
DNA, fita dupla	09 (10%)	2%
	Hull, 2014	2020

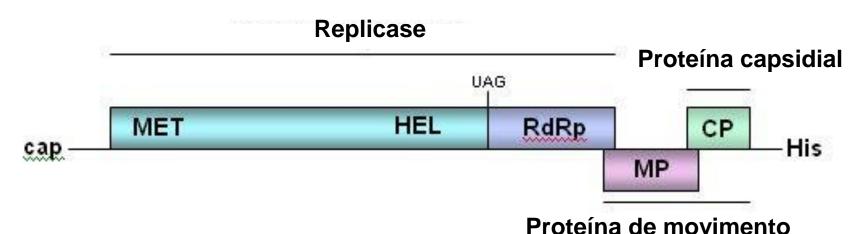


REPLICAÇÃO DE VÍRUS DE PLANTAS: ss+RNA

Organização do genoma viral

Modelo: Tobacco mosaic virus (TMV)

6.395 nt



MET: methyltransferase

HEL: helicase

RdRp: RNA dependent RNA polymerase

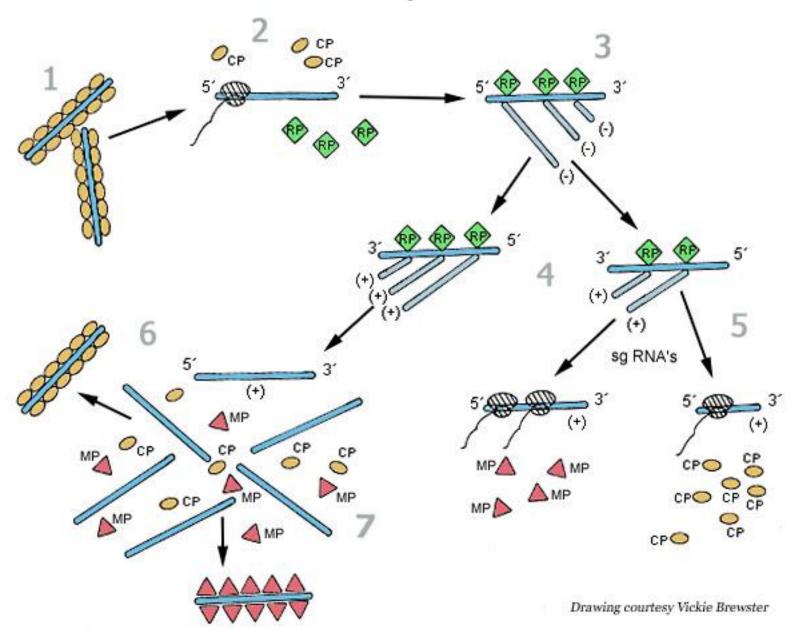


ETAPAS DO CICLO DE REPLICAÇÃO DO TMV

- 1. O TMV entra na célula por ferimentos;
- 2. A capa protéica é retirada e ao mesmo tempo os ribossomos traduzem o RNA para síntese da replicase (RP);
- 3. As replicases transcrevem o RNA viral (+ss) produzindo cópias complementares (-ssRNA);
- 4. O –ssRNA serve de molde para gerar RNA viral (+ss);
- 5. Ao mesmo tempo o –ssRNA serve de molde para gerar RNAs sub-genômicos, que serão usados para sintetizar as proteínas de movimento (MP) e capsidial (CP);
- 6. Parte do RNA viral (+ss) é encapsulado pelas proteínas capsidias;
- 7. Parte do RNA viral é "protegido" pela proteína de movimento para mover-se para a célula vizinha e novo ciclo de replicação.



REPLICAÇÃO DO TMV





REPLICAÇÃO DO TMV

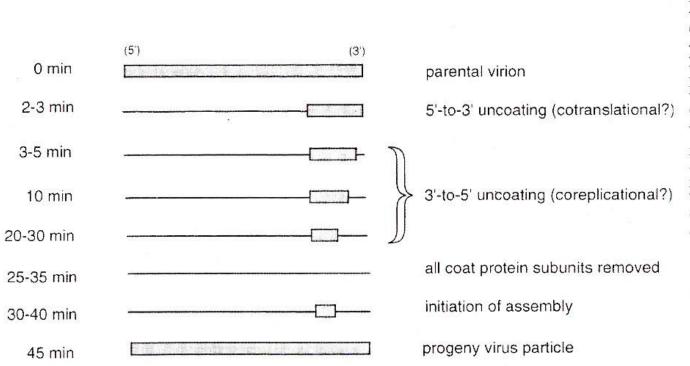


Fig. 7.6 Bidirectional disassembly of TMV particles in vivo. Coat protein subunits are removed in a $5' \rightarrow 3'$ direction from c. 75% of the viral RNA in the first 2-3 minutes after inoculation of protoplasts. Uncoating the 3'-end of the RNA begins shortly thereafter and is completed by removal of subunits in the $3' \rightarrow 5'$ direction. From Wu et al. (1994), with kind permission of the copyright holder, © The National Academy of Sciences, USA.



ROTAS PARA A EXPRESSÃO DE GENOMAS VIRAIS

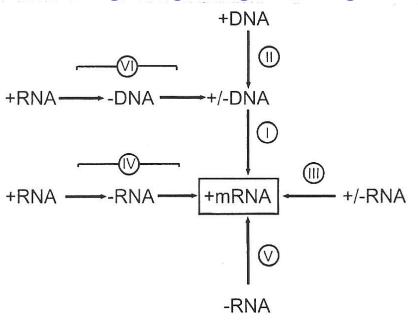
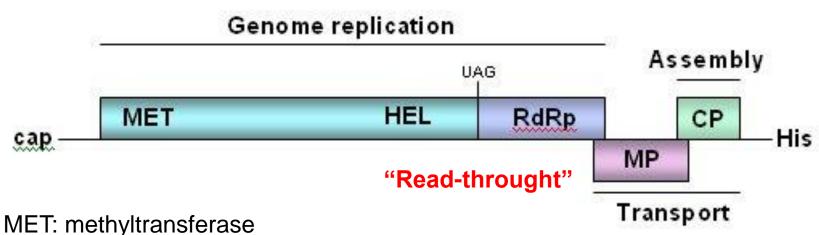


Fig. 7.1 Routing of viral genome expression through mRNA. Route I is transcription of dsDNA usually by host DNA-dependent RNA polymerase. Route II is the transcription of ssDNA to give the dsDNA template for I (e.g. geminiviruses). Route III is transcription of dsRNA, usually by virus-coded RdRp (e.g. reoviruses). Route IV is replication of (+)-strand RNA via a (-)-strand template by virus-coded RdRp—the viral (+) strand is often the template for early translation (the (+)-strand RNA viruses). Route V is transcription of (-)-strand virus genome by virus-coded RdRp (e.g. tospoviruses). Route VI is reverse transcription of RNA stage of retro- and pararetro-viruses leading to the dsDNA template for mRNA transcription. From Baltimore (1971), with permission.



"ESTRATÉGIAS" DO GENOMA PARA A SÍNTESE DE PROTEÍNAS

Tobacco mosaic virus (TMV)



HEL: helicase

RdRp: RNA dependent RNA polymerase

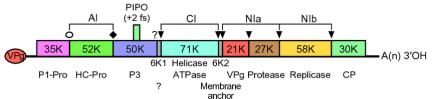
RNA sub-genômicos



"ESTRATÉGIAS" DO GENOMA PARA A SÍNTESE DE PROTEÍNAS

POLIPROTEINA (Potyvirus)

Tobacco etch virus, TEV (9,496 nts)



Poli-proteína: 320 kDa

CP: 20 kDa

2000 capsômeros para uma CP

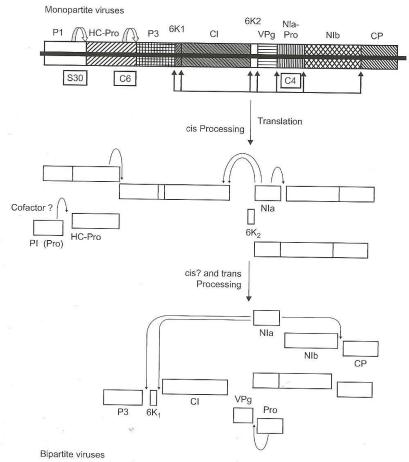


Table 7.1Summary of Genome Strategies Adopted by 18 Single-Stranded Positive Sense RNA Plant Virus Groups

Number	Strateg	y (see Fig. 6.2)	(Gênero) Virus group	Number of ORFs	Number of proteins coded
I	One strategy	Polyprotein	Potyvirus	1	8
II .	One strategy	Subgenomic RNA	Potexvirus Tombusvirus	5 5	4–5 5
III	Two strategies	Subgenomic RNA plus read-through or frameshift protein	Tobamovirus Luteovirus Carmovirus	5 6 5	4-5 6-7 5-7
IV	Two strategies	Subgenomic RNAs and polyprotein	Tymovirus Sobemovirus	3	3-5 4-5
V	Two strategies	Multipartite genome and polyprotein	Comovirus Nepovirus	2 2	≃9 ≃6
VI	Two strategies	Subgenomic RNAs and multipartite genome	Bromovirus Cucumovirus Alfalfa mosaic virus	4 4 4	4 4 4
			Ilarvirus Hordeivirus	4 7	4 7
VII	Three strategies	Subgenomic RNAs, multipartite gen- ome, and read- through protein (or frameshift)	Tobravirus Furovirus Dianthovirus	5 9 4	5 6–9 4