- **RNA virus cycles**

Including viruses with:

single-stranded (+)RNA genomes,

single-stranded (-)RNA genomes (segmented and non-segmented),

dsRNA genomes

Viruses with reverse transcriptase in the cycle will be discussed separately.

Viruses with ss(+)RNA genomes

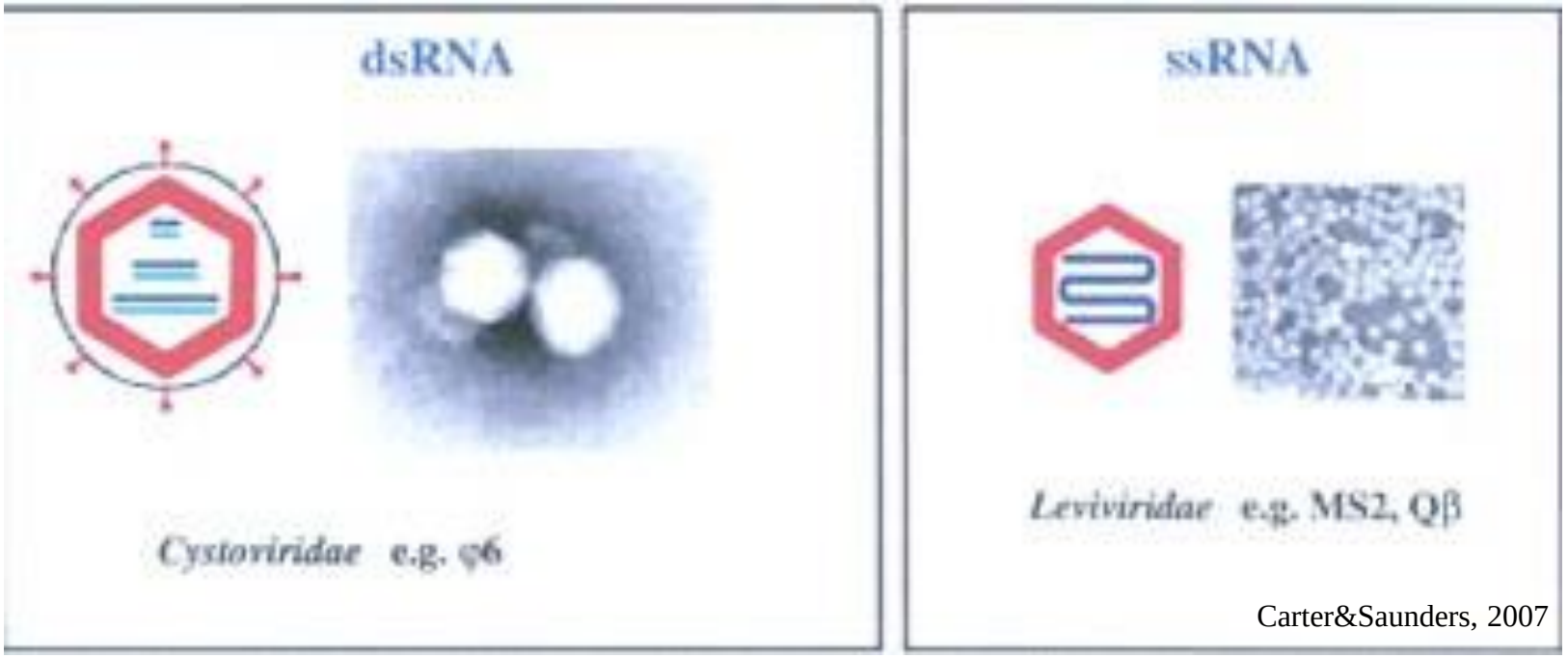
"Size limit" at about 32 kb (*Coronaviridae* – animal viruses) or is it?

Closteroviridae (*Citrus tristeza virus* about 20 kb) largest plant RNA viruses.

Planarian Secretory Cell Nidovirus, PSCNV – 41 103 nts

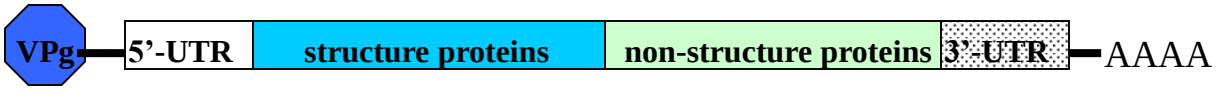
Mollusc - *Aplysia californica nido-like virus, AcNV – 35 906 nts*

Phages (MS2 ~ 3.5 kb) amongst the smallest viral genomes.



All (+)RNA genomes are "infectious", other common features of viruses in this group.

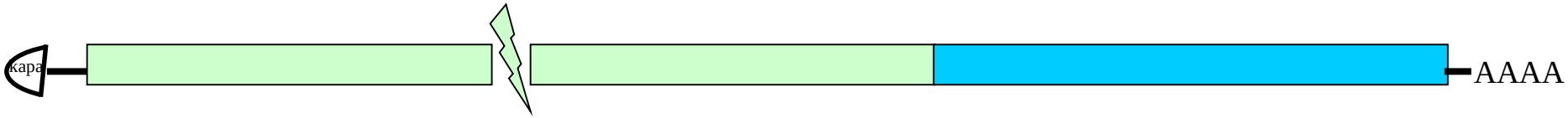
(+)RNA genome organization



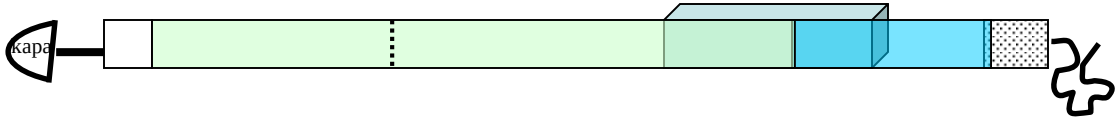
Picornaviridae (PV, HAV)

Togaviridae (*rubella*, W,V, EEE, Sindbis virus – genus *Alphavirus*)

Flaviviridae (*Denge*, HCV, YFV)



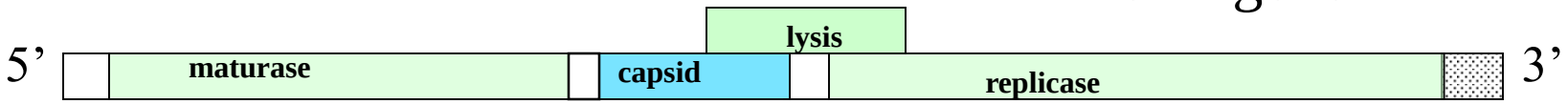
Coronaviridae - 25% of common cold, bronchitis, gastroenteritis, serious respiratory and systemic diseases (SARS, MERS, COVID-19)

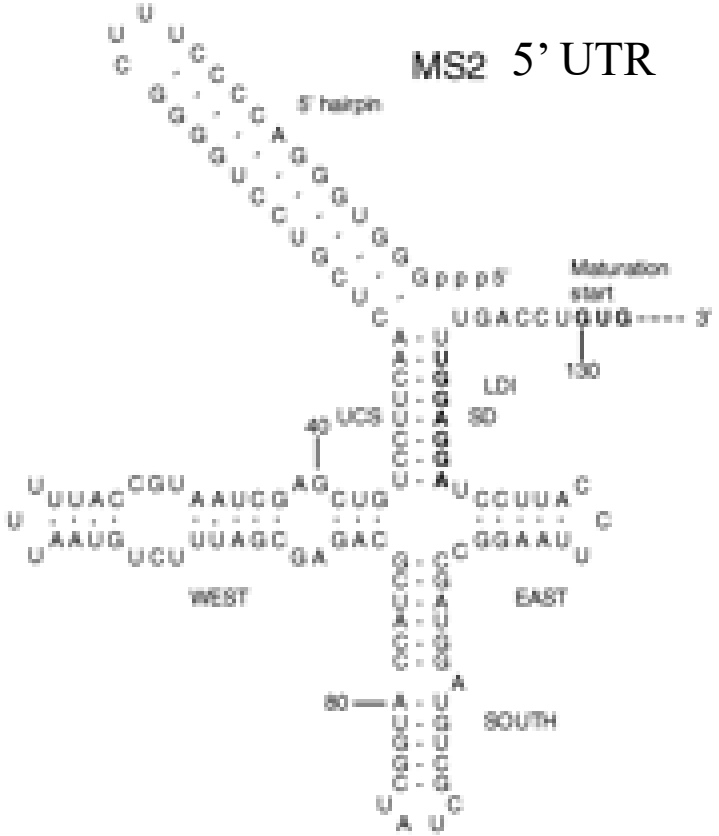


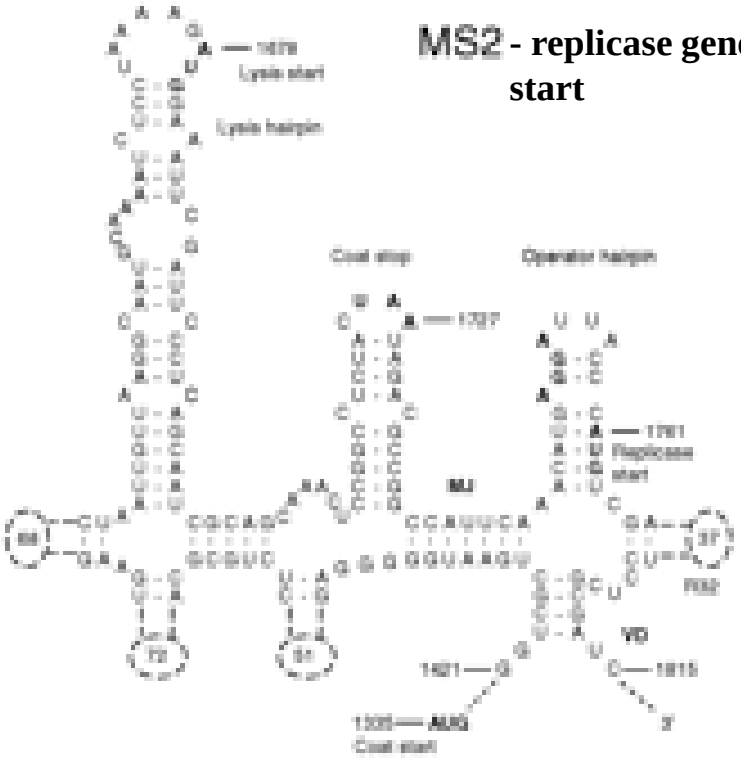
The majority of plant viruses (e.g. TMV) has a genome like this one. There are those with segmented or multipartite genomes. Terminal structures can vary.

Phages MS2, f2, Qβ – different terminal structures than plant viruses, they have introns.

MS2 - genom





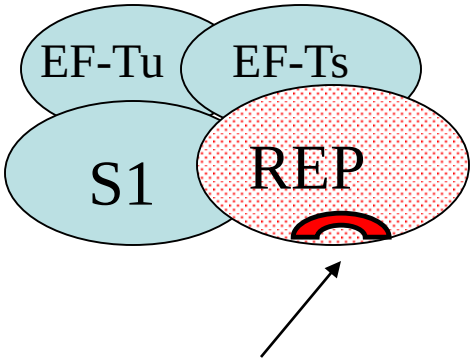


Ruokoranta *et al*. 2006. J. Virol. 80: 9326.

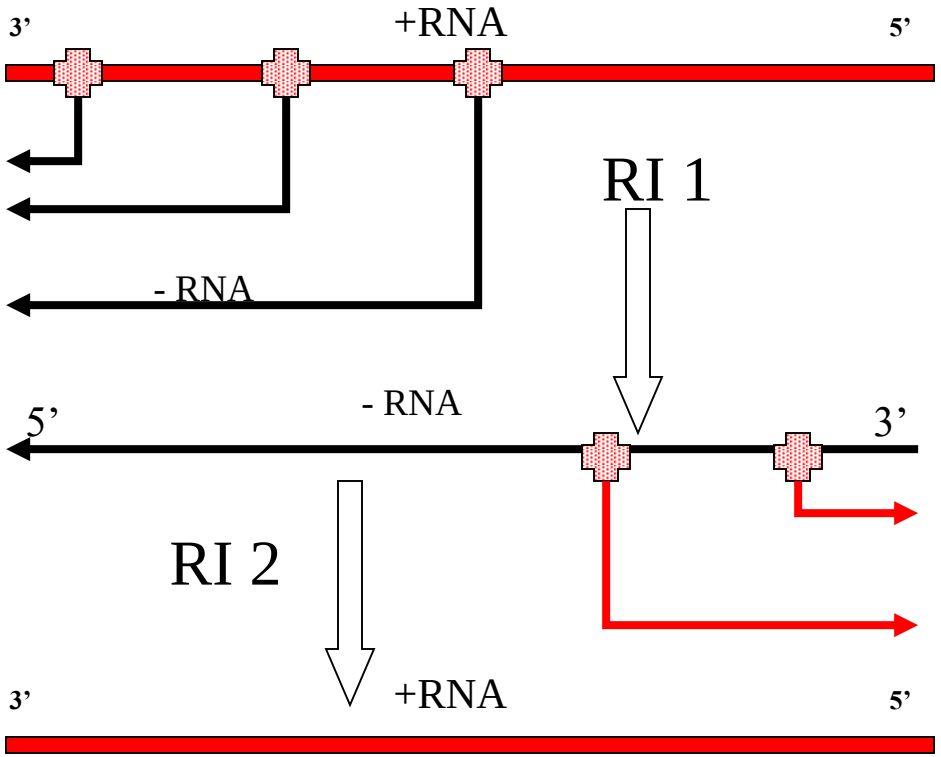
The cell has not enough enzymes for RNA virus genome replication (ssRNA-dependant RNA-polymerases – plants, some eukaryotes).

A virus has to code for at least RNA-dependent RNA-polymerase (RdRp) which can bind cell another virus protein and form RNA replicase.

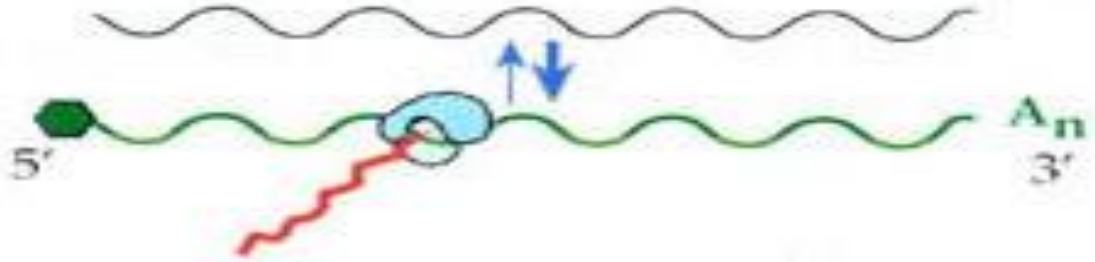
Active ssRNA-phage replicase complex



MS2 phage active site

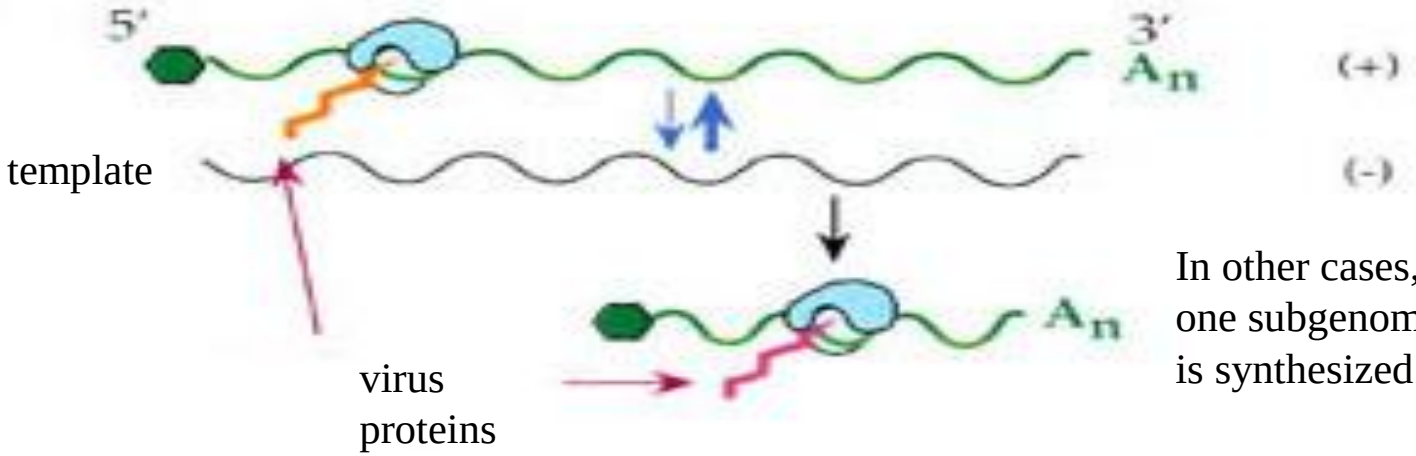


(+)RNA virus transcription and translation strategies



(+) genome is the replication template and mRNA

virus polyprotein



In other cases, at least one subgenomic mRNA is synthesized.

replication ↓ subgenomic mRNA synthesis ↓ cap or VPg ribosome

Prerađeno iz Strauss&Strauss, 2002

Roles of the cap as 5’-terminal structure:

- facilitates the transport of mRNA from nucleus to cytoplasm
- exonuclease protection

- Roles of the cap in translation:

Cell mRNAs need a cap at the 5’-end to initiate translation, cap-binding proteins are a part of the translation initiation complex.

It contains:

binding sites for eIFs, aminoacylated Met-tRNA, and 40S ribosome subunit (eukaryotes).

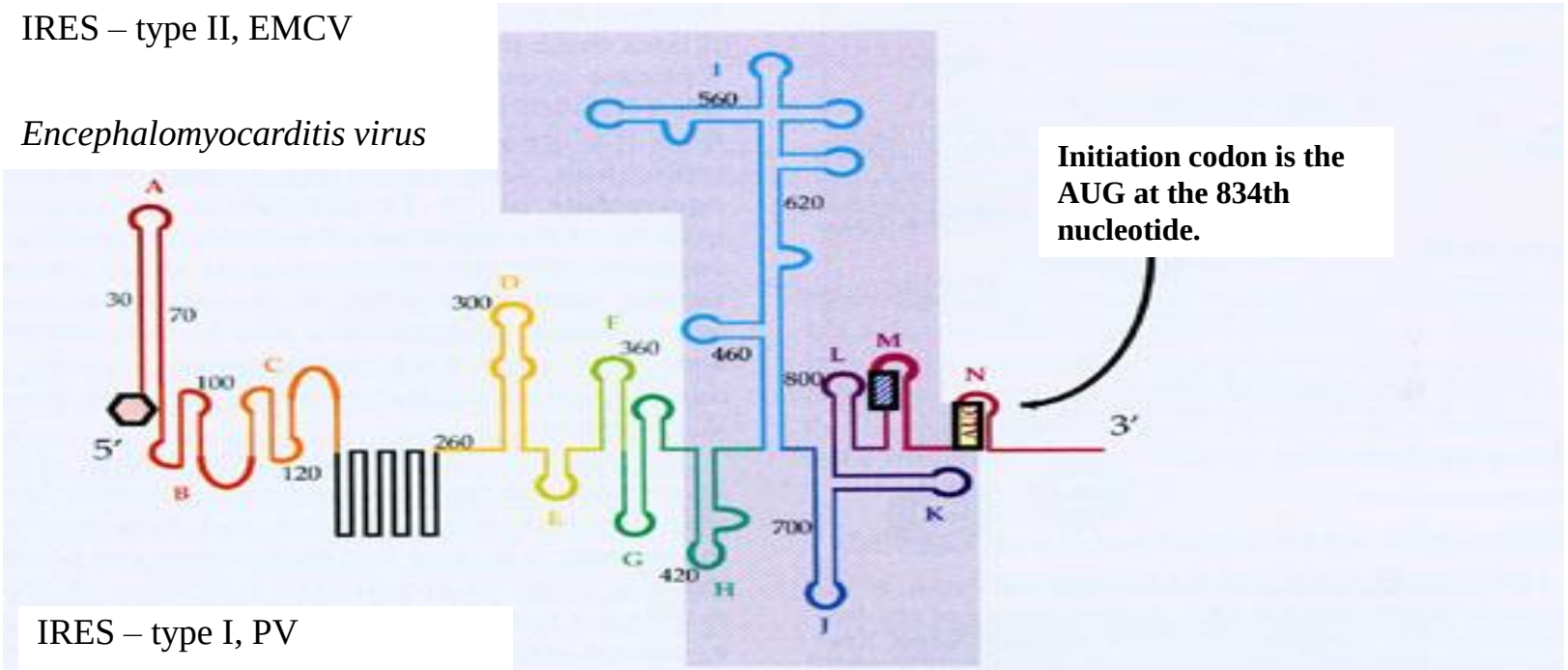
Signal for screening the first suitable AUG codon.

PolyA at the 3’-end and proteins that bind it are also important – translation enhancement probably through interactions with 5’-end (mRNA circularization).

Cap-independent translation (*Picornaviridae* examples)

IRES – type II, EMCV

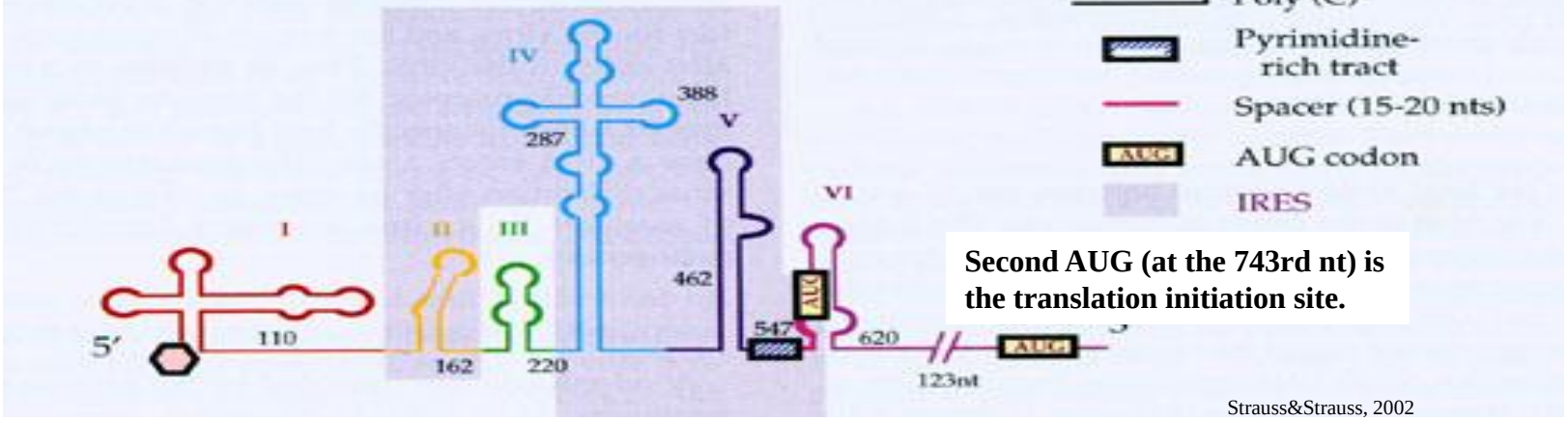
Encephalomyocarditis virus



Initiation codon is the AUG at the 834th nucleotide.

IRES – type I, PV

Poliovirus

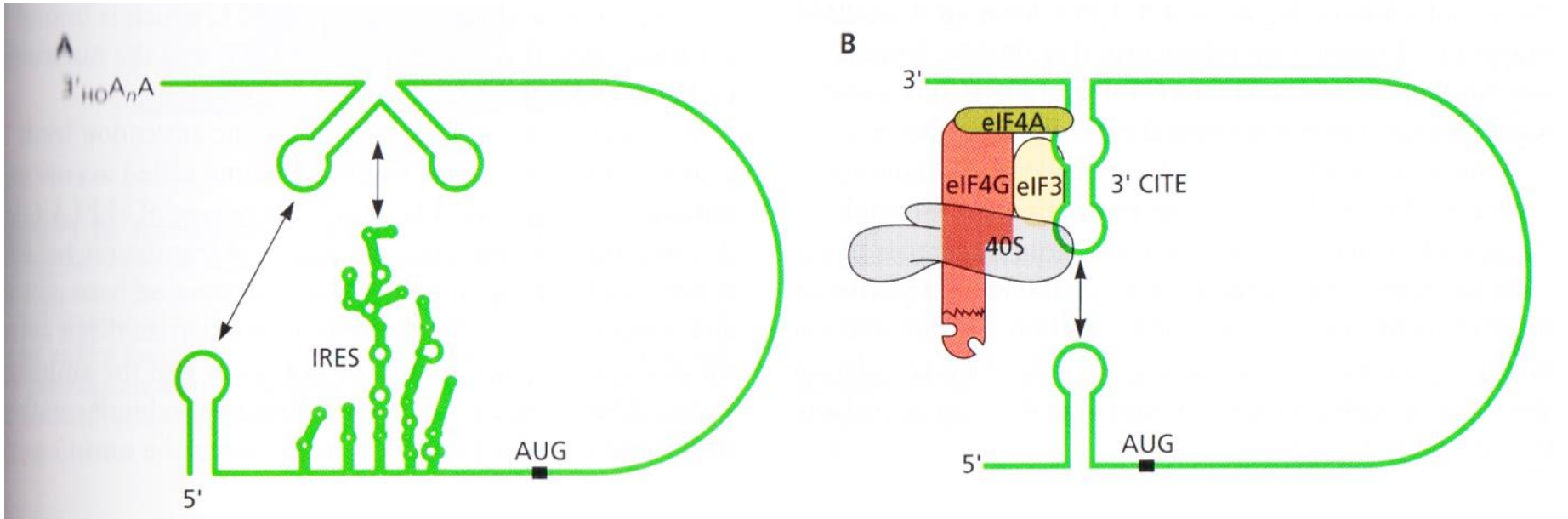


Second AUG (at the 743rd nt) is the translation initiation site.

Strauss&Strauss, 2002

Five IRES-a (I-V) types – classified according to conserved primary and secondary RNA structures , start codon location and activity in certain cell types.
IRES is found in some cellular mRNAs. Used in expression vectors or in gene therapy vectors (between two transgenes).

RNA-RNA-interactions help in translation (A – FMDV- specific sequence at the 3'-end of the genome, B – plant viruses have CITE (3'-cap-independent translational enhancer)

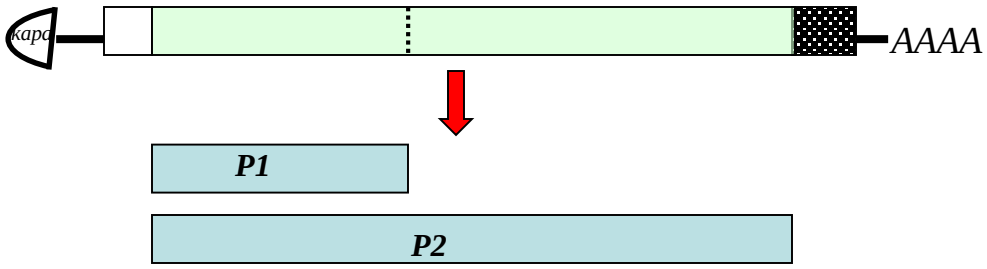


Flint et al. (2015) Principles of Virology

Proteome is bigger than expected from the genome size – condensation of genetic information.

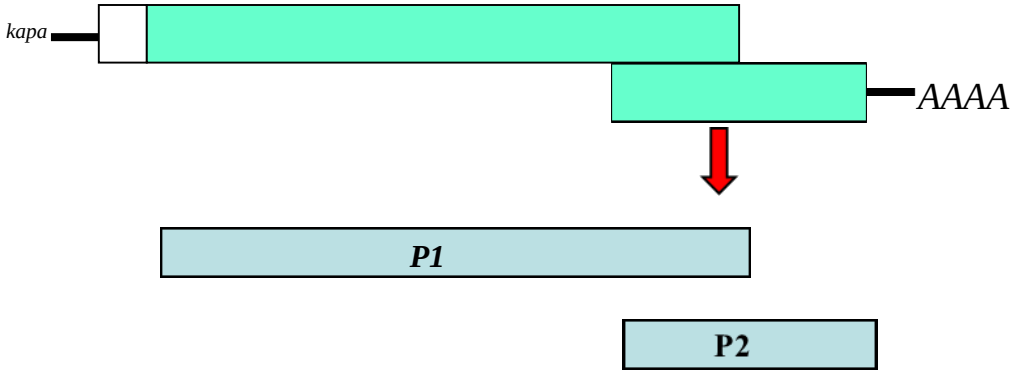
a) leaky stop codon - stop is read like information for an aa with efficiency of 5-20% –amber (UAG), opal (UGA) are usually leaky

– shorter and longer proteins are made sequentially, the bigger is 5'-coterminal with the smaller one.

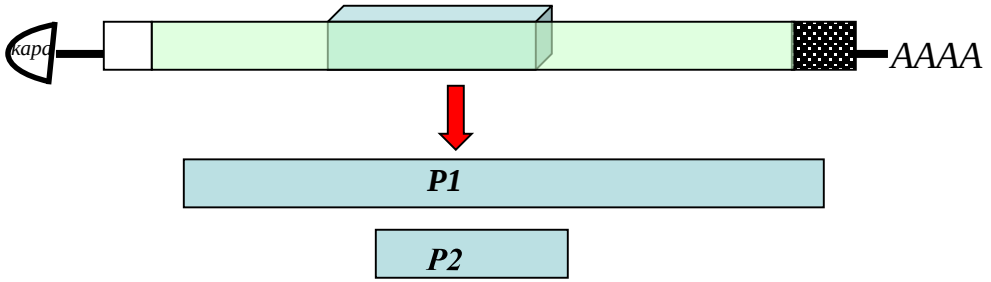


b) leaky scanning – internal start codone and the firts protein stop codone is disregarded (read through) in

overlapping genes



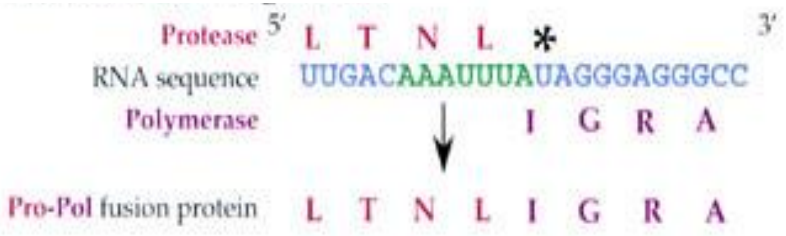
nested genes



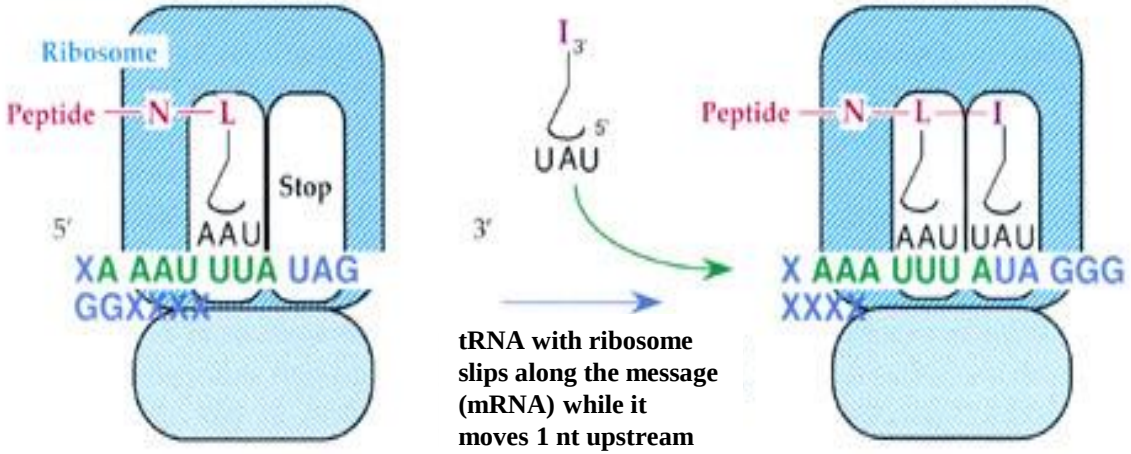
c) ribosome frameshifting – ribosome slips on the “slippery sequence” of a mRNA to position +1 or -1 u relative to stop-codon. Proteins are synthesized “in frame” or “out of frame”.

Frameshifting mechanism (Retroviruses shown but similar in normal RNA-viruses

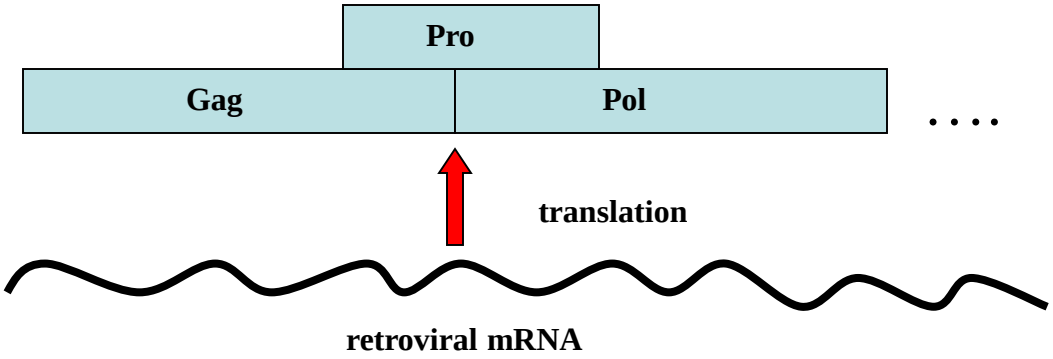
***Avian leucosis virus*, ALV**

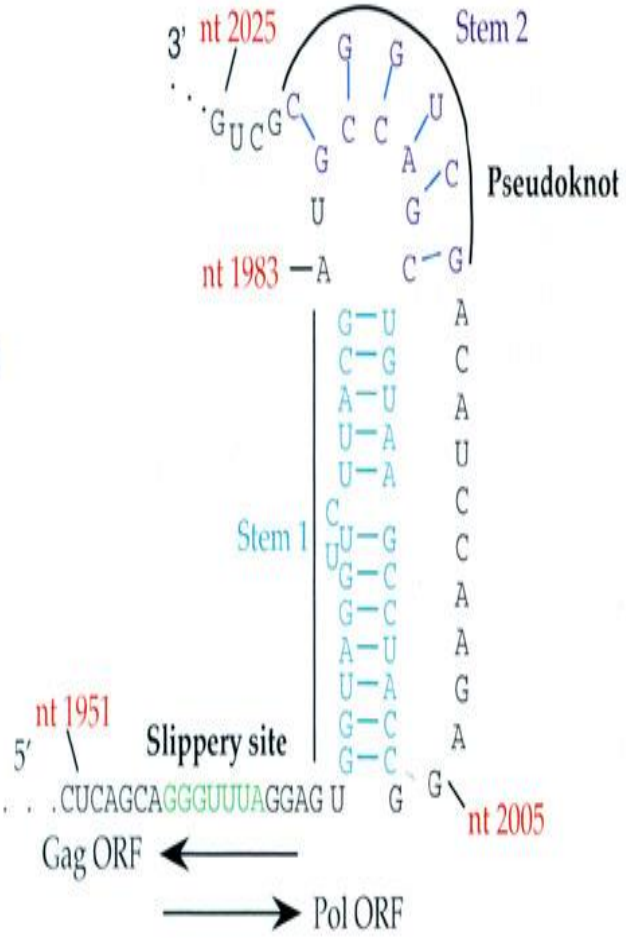


-1 frameshift

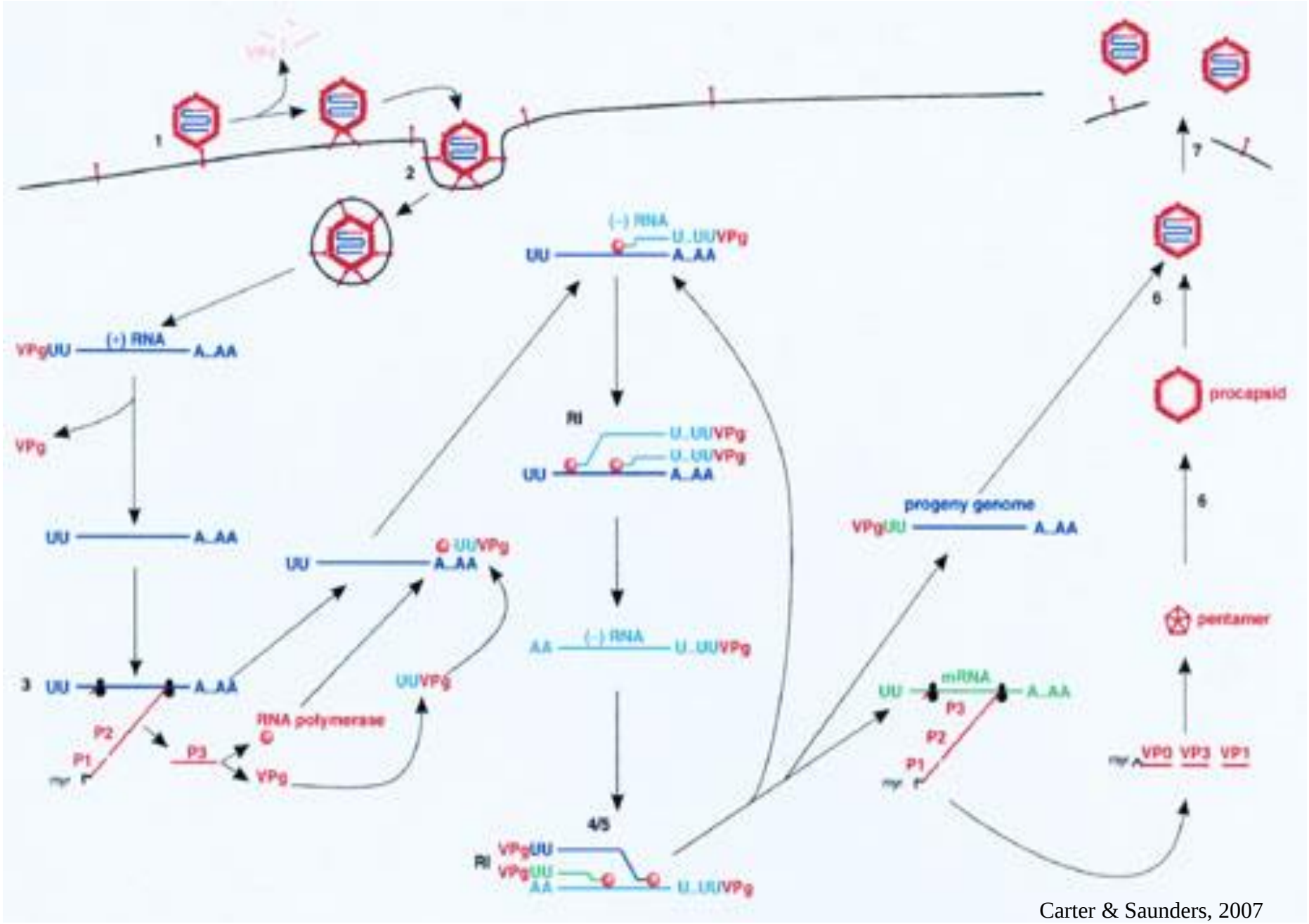


tRNA with ribosome slips along the message (mRNA) while it moves 1 nt upstream



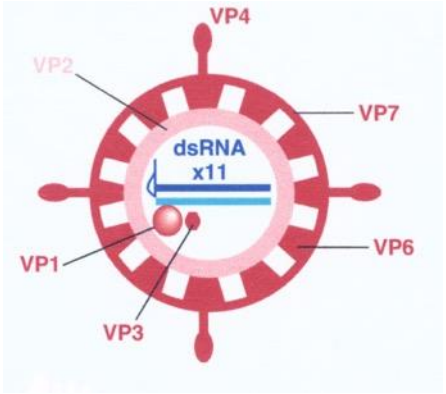


Poliovirus cycle



Carter & Saunders, 2007

dsRNA viruses:



Reoviridae – segmented genome, totally 16-27 kbp, virusi of mammals (rota virion), arthropods, plants,

Birnaviridae – 2 segments 7 kbp, fish, birds, arthropods

Totiviridae – 1 dsRNA, fungi (*S. cerevisiae virus L-A*, *Aspergillus*, *Ustilago*), protozoa (*Giardia*, *Leishmania*)

Partitiviridae– 2 segments, 3-10 kbp, plants, fungi

Endornaviridae – 1 big linear dsRNA , no capsid (protozoa, fungi, plants, asymptomatic)

Cystoviridae – *Pseudomonas syringae* pv. *phaselicola*, lytic, enveloped, L, M, S-segments, (+)RNA is packaged, (–)RNA "disappears" in the capsid