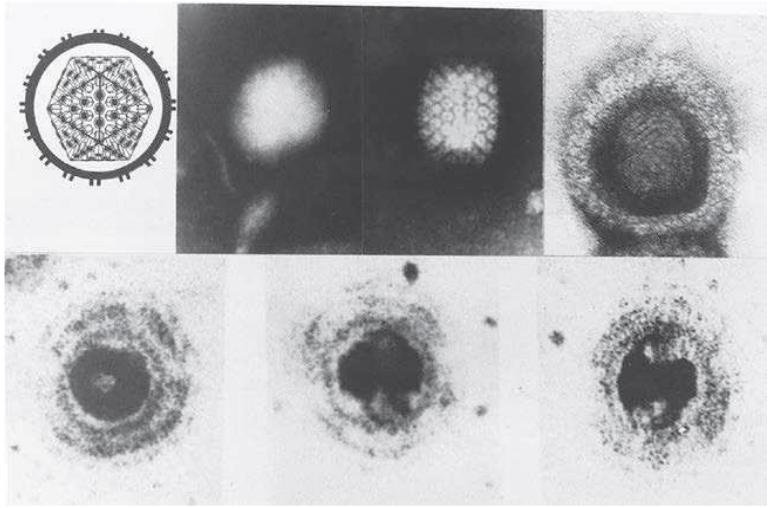
Herpesviridae: Common Biological Properties

Large linear dsDNA genome (125-236 kb; ~70-200 genes)

Herpesvirus DNAs extracted from virions and characterized to date are linear and double stranded, but they circularize immediately on release from nucleocapsids into the nuclei of infected cells



Herpesviridae: Common Biological Properties

Enzymology: Encode enzymes for DNA metabolism, DNA synthesis, protein processing/modification

Nuclear replication: transcription, synthesis of DNA and capsid assembly occur in the nucleus (part of the tegument and envelope derived from the cytosol)

Cytolytic: Production of infectious progeny results in host cell death

Latency: All herpesviruses have the capacity for latency

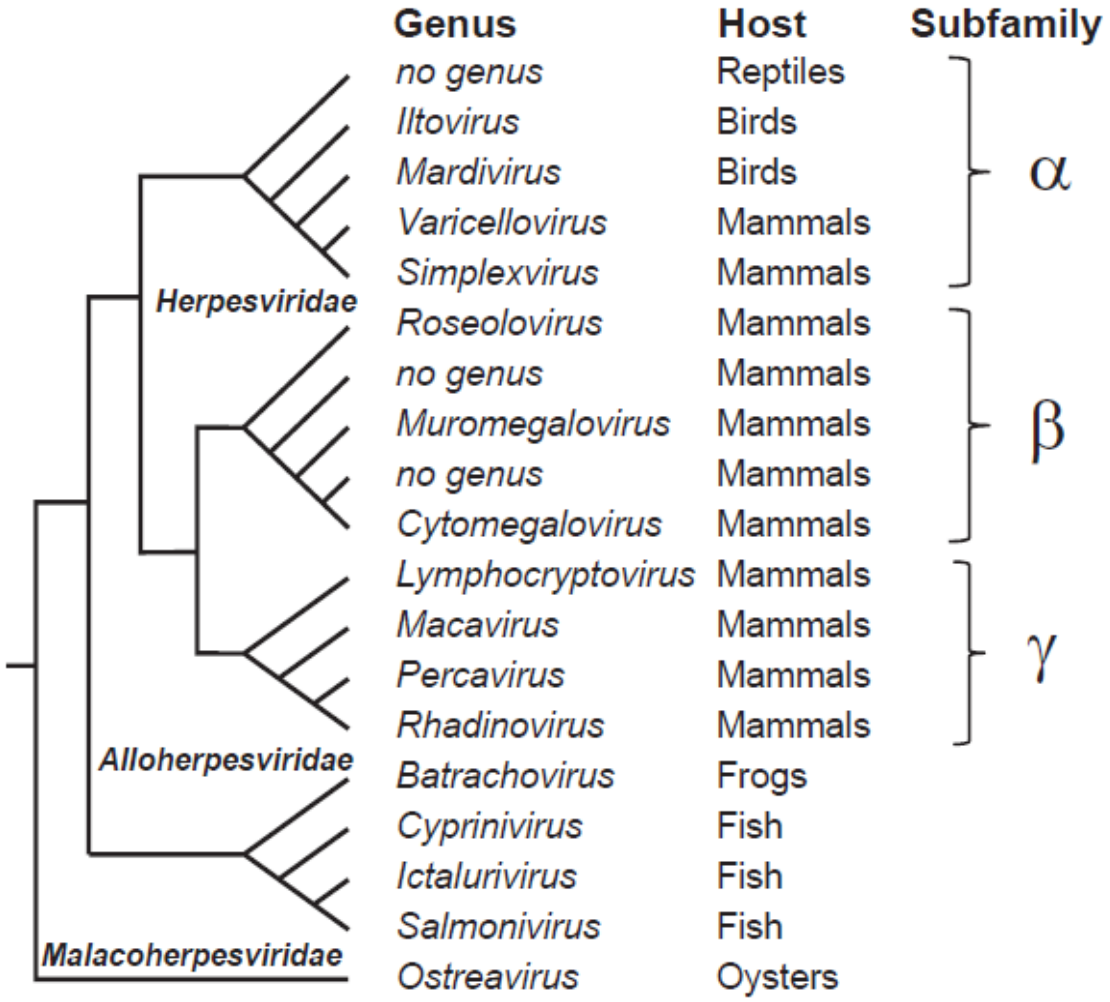


FIGURE 59.2. Major phylogenetic relationships and taxonomic subunits within the order *Herpesvirales*.[102] The schematic shows branching patterns, not evolutionary distances.

Human Herpesviruses:

Alphaherpesvirinae (Neurotropic):

Human herpes simplex virus 1 and 2 (HSV1, HSV2 or HHV-1, HHV-2)
Primary agents of recurrent facial and genital herpetic lesions

Varicella-zoster virus (VZV or HHV-3). Causative agent of cicken pox and shingles

Betaherpesvirinae (Lymphotropic):

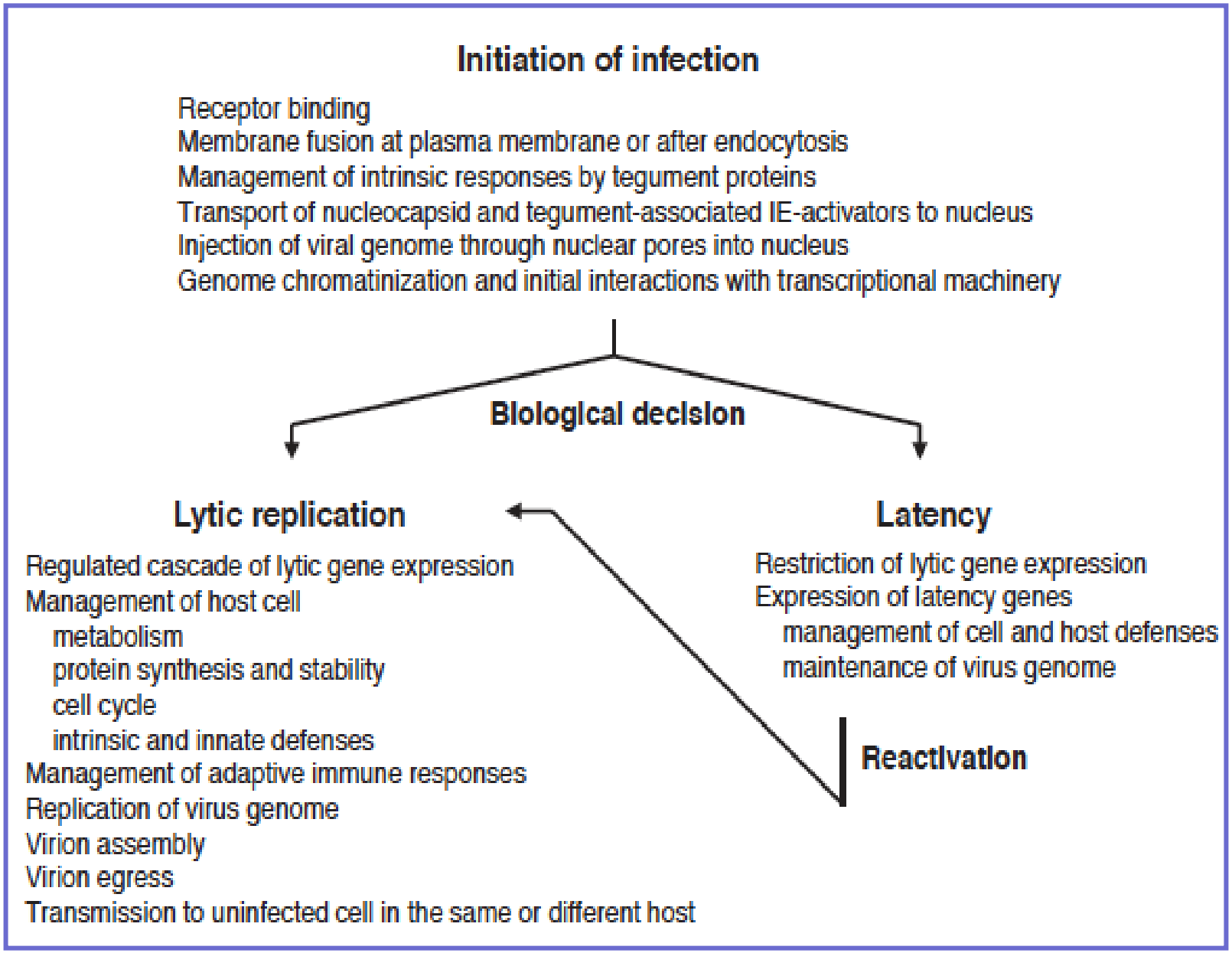
Cytomegalovirus (HCMV or HHV-5). Congenital infections; pneumonia

Human herpesvirus 6A, 6B, 7 (HHV6, HHV7). Some roseola

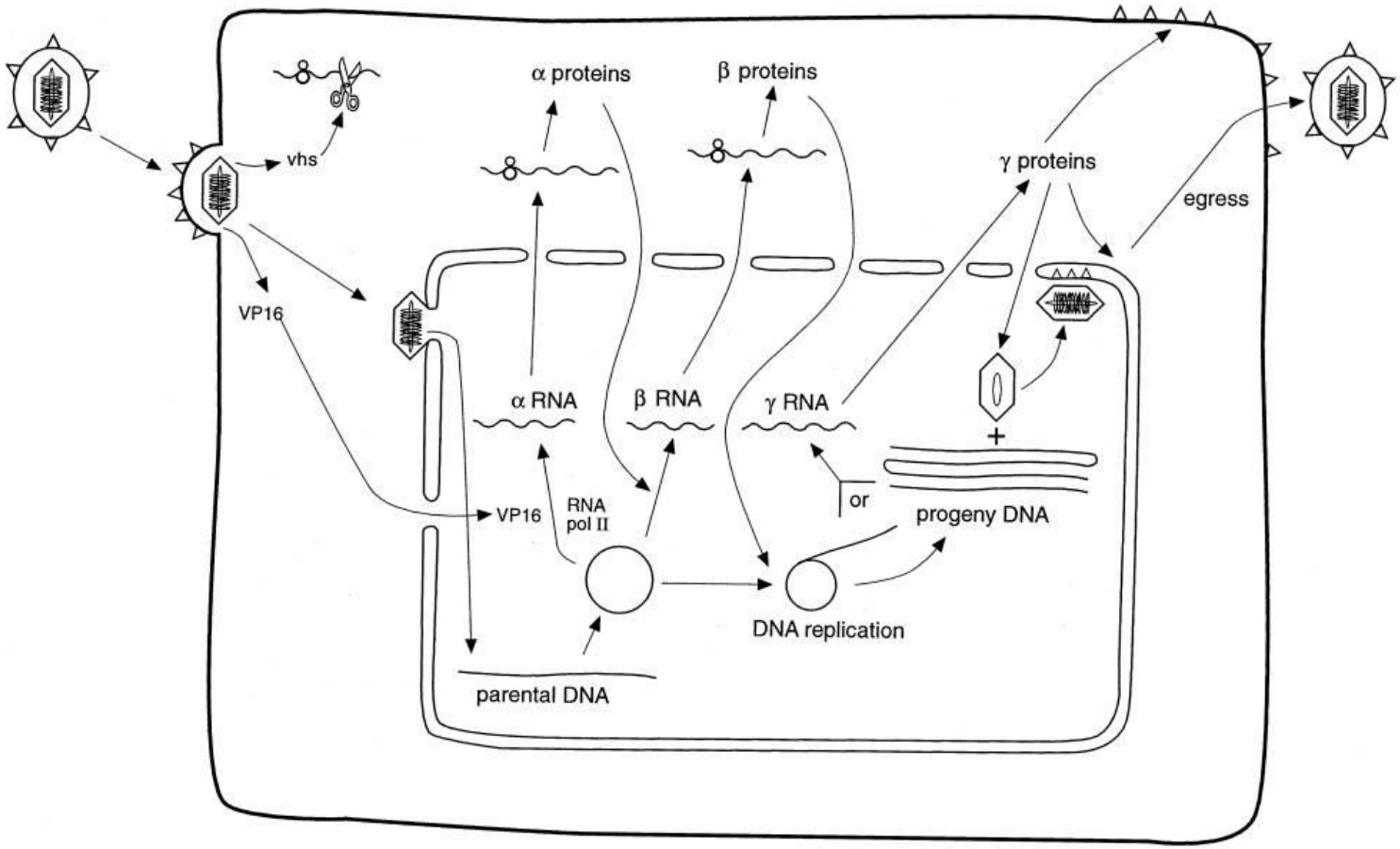
Gammaherpesvirinae (Lymphotropic, tumor-associated):

Epstein-Barr virus (EBV or HHV-4) Tumors (B cells, epithelial cells)

Kaposi's sarcoma herpesvirus (KSHV or HHV-8)


Initiation of infection
Receptor binding
Membrane fusion at plasma membrane or after endocytosis
Management of intrinsic responses by tegument proteins
Transport of nucleocapsid and tegument-associated IE-activators to nucleus
Injection of viral genome through nuclear pores into nucleus
Genome chromatinization and initial interactions with transcriptional machinery
Biological decision
Lytic replication
Regulated cascade of lytic gene expression
Management of host cell
metabolism
protein synthesis and stability
cell cycle
intrinsic and innate defenses
Management of adaptive immune responses
Replication of virus genome
Virion assembly
Virion egress
Transmission to uninfected cell in the same or different host
Latency
Restriction of lytic gene expression
Expression of latency genes
management of cell and host defenses
maintenance of virus genome
Reactivation

Lytic phase replication



<u>Espressione gerarchica in tre fasi temporali:</u>
alpha (immediato precoce);
beta (precoce);
gamma (tardiva)

La replicazione del DNA nel nucleo avviene attraverso la **DNA polimerasi virale**.

Latency

"Reversible non-productive infection of a cell by a replication-competent virus"

Requirements for latency:

1. Evasion of host immunity

e.g., by minimizing gene expression (HSV-1 in neurons) or specific genes designed to suppress immunity (various)

2. Genome persistence

Relatively easy in non-dividing cells (neurotropic herpesviruses)

More complex in dividing cells (lymphotropic herpesviruses)

e.g., EBV *oriP* permits genome replication, retention and segregation.

Host-virus interaction

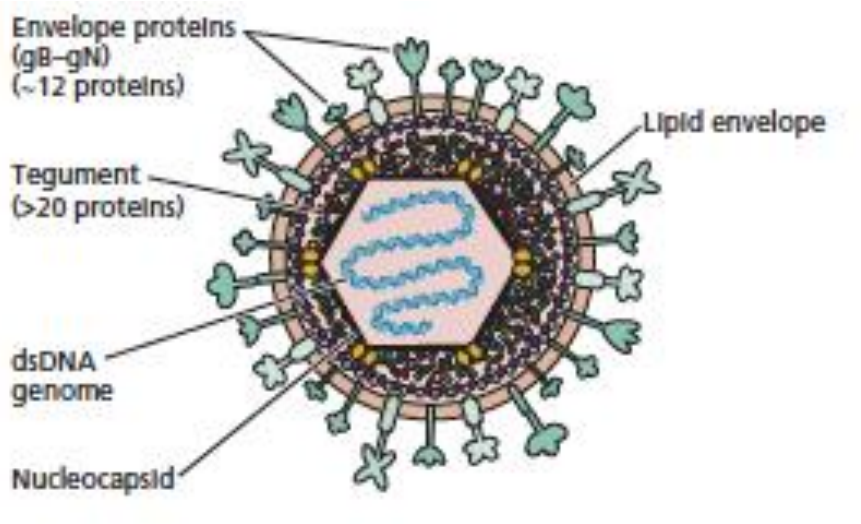
At the cellular level, the virus **blocks the induction of programmed cell death** and the activation of the **interferon pathway**
At the organism level, the response is highly varied and depends in part on the cells in which the virus replicates (from blocking the presentation of antigenic peptides to molecular mimicry).

A common consequence is a **delay in the elimination of the infected cell**—long enough to enable the virus to replicate, colonize the host, and become available for transmission to another nonimmune individual.

The range of genes evolved to this end varies from one genus to another, with limited conservation among members of subfamilies.
Alphaherpesviruses sequesters cellular proteins and modifies them to perform novel functions (e.g., HSV-1 $g_1$34.5 and the cellular protein phosphatase 1$_a$).

Whereas, beta and gamma- herpesviruses encode numerous orthologs of cellular proteins designed to mislead the cell or directly perform the desired function (often with altered regulatory properties).

Herpesvirus Structure



Nucleocapsid icosaedron made up of six different proteins

Matrix called *tegument* lies between the capsid and the envelope and contains at least 15 or 20 proteins (and virion RNAs).

Envelope contains 10 or more glycoproteins

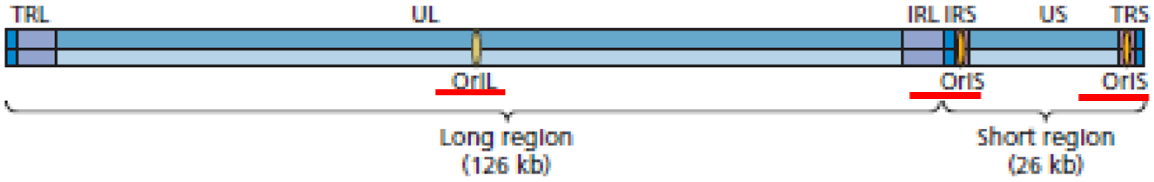
Note: Herpesvirus particles also contain virion RNAs. This may allow for immediate synthesis of new gene products following virus entry, setting up the cell for efficient viral replication. RNA packaging may depend on specific viral RNA-binding proteins (e.g., HSV-1 VP22) and may reflect the concentration of RNAs within producer cells.

From Flint et al. *Principles of Virology* (2000), ASM Press

Herpesviridae

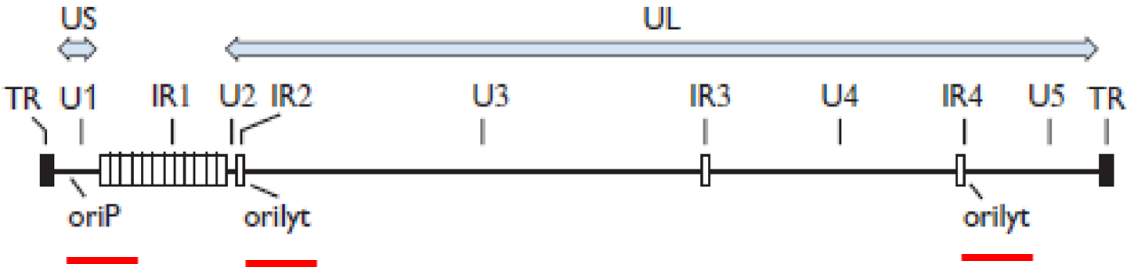
I vari membri sono piuttosto diversi in termini di sequenza genomica, ma condividono una struttura e organizzazione comune: il genoma (lungo da 130 kbp 240 kbp) contiene due regioni uniche lunghe e corte (UL e US) fiancheggiate da ripetizioni invertite (TR e IR)

HSV-1



152 Kbp

EBV



172 Kbp

Although most genes are transcribed by RNA polymerase II; some small transcripts are transcribed by RNA polymerase III, for example, the EBV EBER transcripts. Th e genomes of herpesviruses, such as Epstein-Barr virus and herpes simplex virus type 1, contain three origins of replication.

Genomes of members of the Herpesviridae encode between 70 to 200 protein-coding genes. Herpesviruses also encode noncoding RNAs, some of which are highly abundant, e.g., the OriS RNA and latency-associated transcripts of HSV, the EBERs of EBV, and microRNAs.

Genes of host origin

Herpesviruses generally encode at least one gene of obvious host origin. Examples include thymidylate synthase, *bcl2* or virokines (immunomodulatory function since derived from cytokines)

Some of these genes appear to have been acquired independently by different herpesvirus lineages (again, *Bcl2* and thymidylate synthetase).

Host acquired genes tend to be located toward the viral genomic termini and between core gene blocks. In some cases, the host acquired genes retain a function similar to its cellular counterpart; in other cases, the host acquired gene has been modified to alter its function

The mechanism(s) by which herpesviruses pilfer host genes are still enigmatic. Up to now, the thief has not been caught in the act.

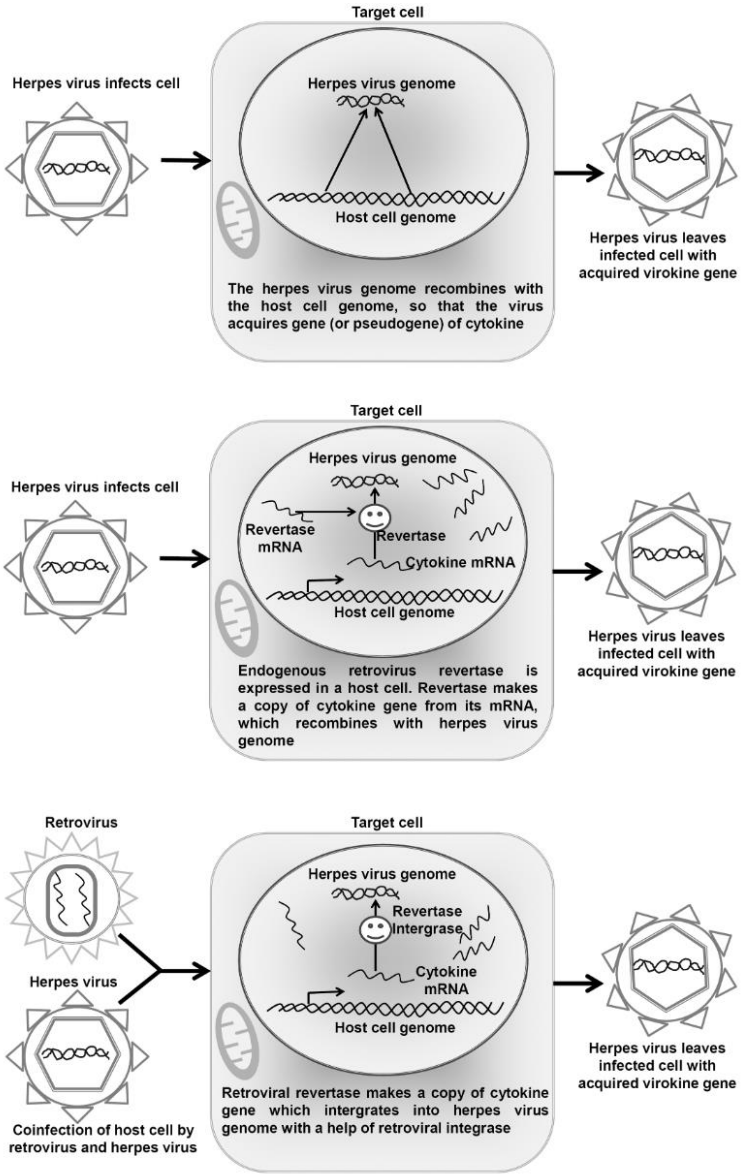
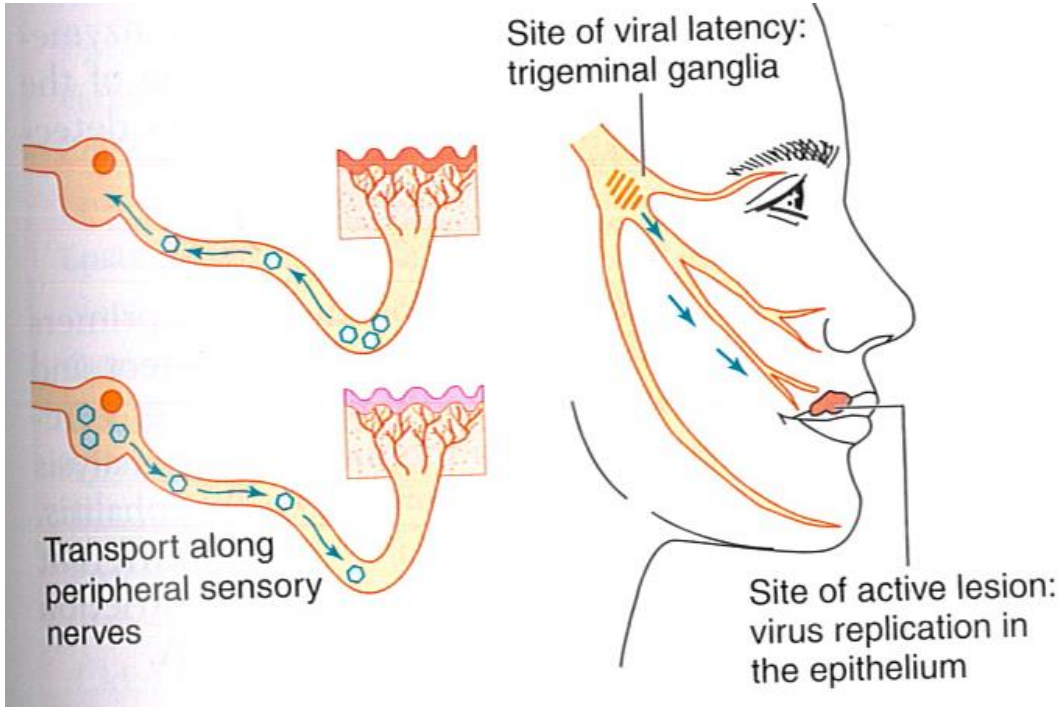
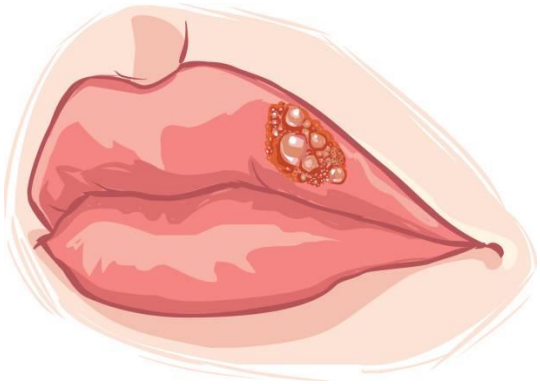


Fig. 1. Three probable scenarios of virokine gene acquisition by herpesvirus.

The mechanism(s) by which herpesviruses pilfer host genes are still enigmatic. Up to now, the thief has not been caught in the act.

Il virus dell'herpes symplex di tipo 1 stabilisce un'infezione latente nei gangli del trigemino.

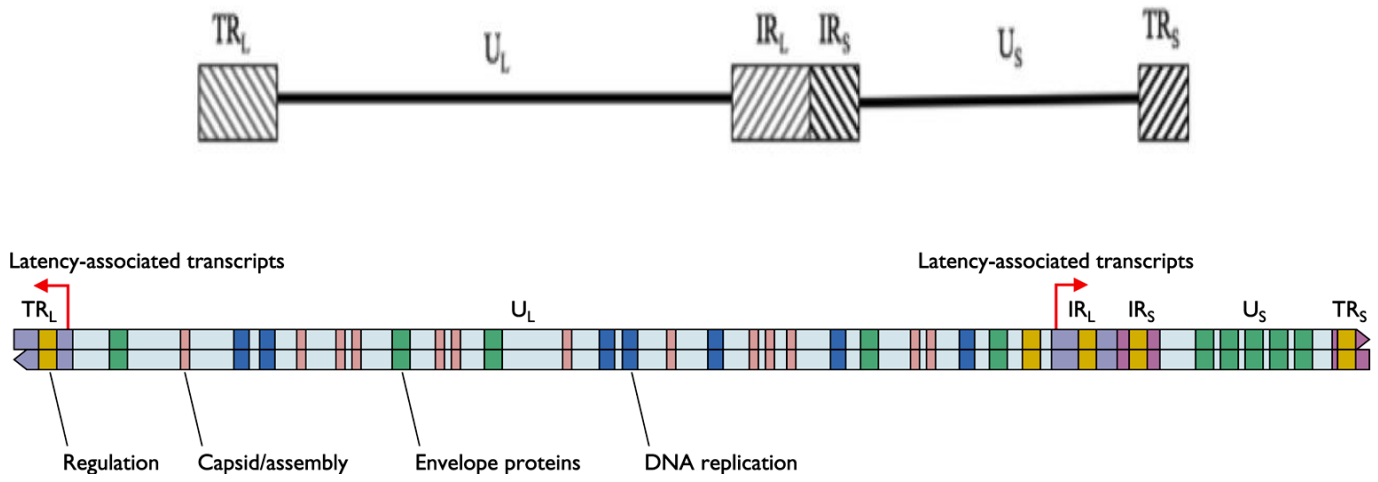




An estimated 3.8 billion people under age 50 (64%) globally have herpes simplex virus type 1 (HSV-1) infection (WHO)

Type 1 (HSV-1) mostly spreads by oral contact and causes infections in or around the mouth (herpes labialis). It can also cause genital herpes. Most adults are infected with HSV-1. Rarely, HSV-1 infection can lead to more severe complications such as encephalitis (brain infection) or keratitis (eye infection). In case of genital infection, it may cause neonatal herpes.

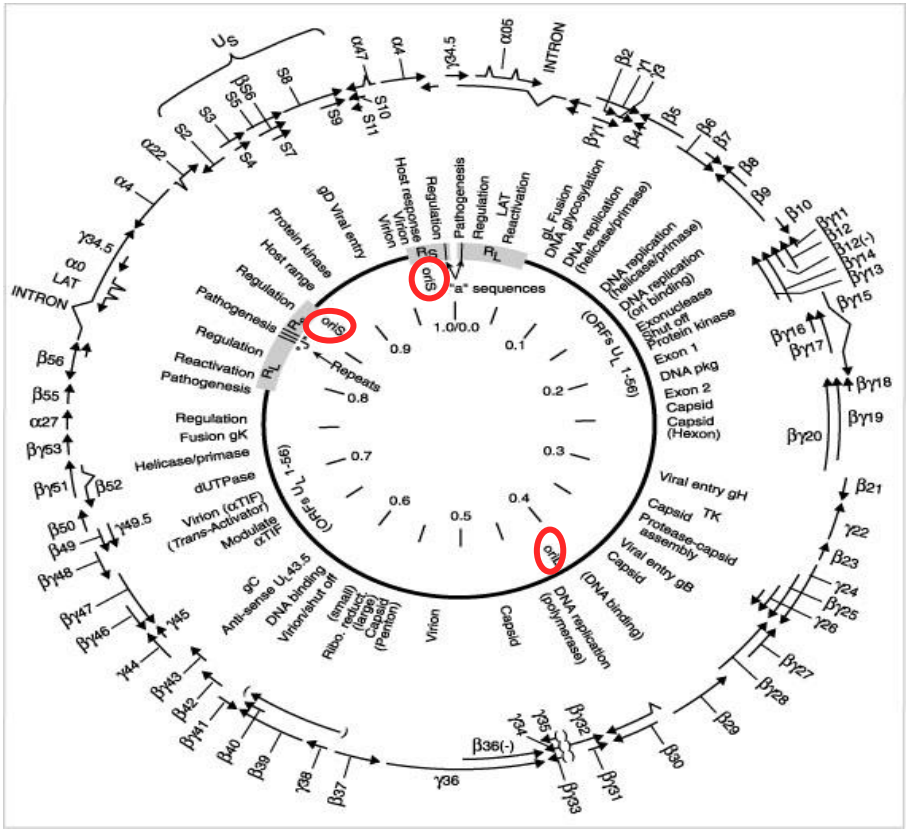
HSV 1 Genome (152 kbp)



HSV 1-around 84 ORF. The position of genes and transcription units associated with the latency is indicated . Genes encoding similar functions are marked with the same color. Genes are dispersed rather than clustered. Few events of splicing.

From Flint et al. *Principles of Virology* (2000), ASM Press

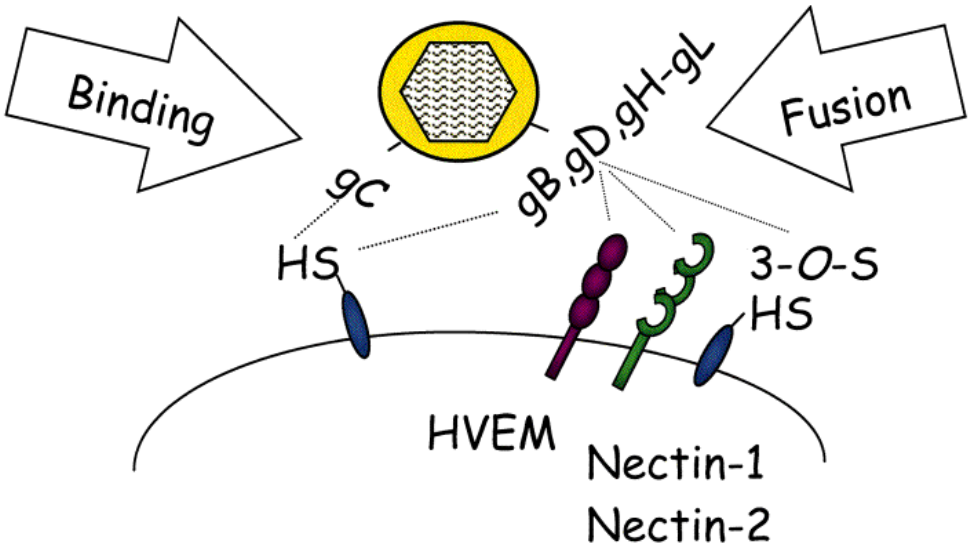
HSV-1 Genome ca 152kbp



The total coding capacity of HSV includes at least 84 transcripts that encode a diversity of proteins, several long noncoding RNAs, and as many as 16 to 17 microRNAs. it has been suggested that replication from OriL may be particularly important during the transition to a productive infection

From Wagner and Hewlett *Basic virology* (2003) Blackwell Science Press

HSV-1 Attachment and Entry



Spear, P.G. Cell. Microbiol. 6:401, 2004.

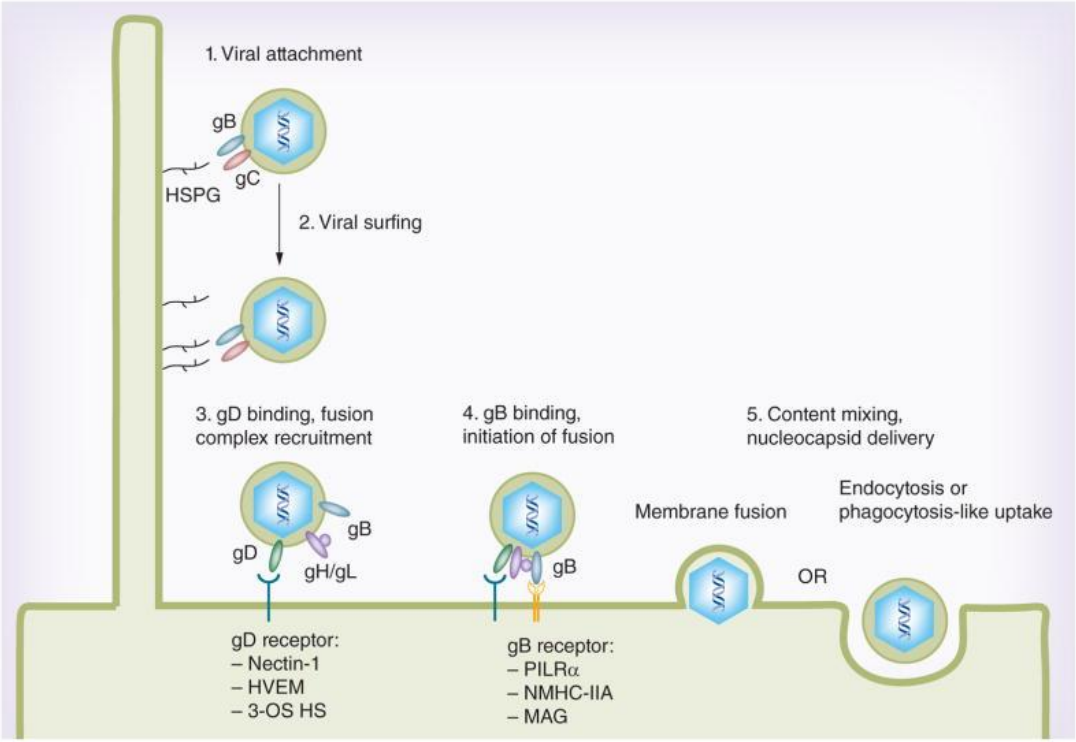
HVEM herpes virus entry mediator

Binding: gC and gB bind Heparan Sulfate (HSPG) on cell surface proteoglycans.

Entry Receptors: 3 receptors are bound by **gD - (1) HVEM**, a TNF receptor superfamily member; **(2) Nectin 1 & 2**, Ig-gene superfamily members & (3) modified HS sites generated by **3-O-sulfotransferases.**

Binding causes a **conformation change** in gD, facilitating fusion. *gB/gH & gL are needed also for fusion as they allows the lipids in both the viral and cell membranes to mix.*

HSV-1 Attachment and Entry



Initial attachment to cells is facilitated by binding of HSPGs by HSV glycoproteins gC and/or gB. Virions are then transported along cellular filopodia toward the cell body in a process termed viral surfing. Recruitment of a fusion complex comprising gB, gD and gH/gL is initiated by gD binding to one of its receptors, nectin-1, herpes virus entry mediator or 3-O-sulfated heparan sulfate. Binding of gB to one of its receptors, PILRα, NMHC-IIA or MAG, is then required for delivery of the viral nucleocapsid to the cytoplasm, accomplished either by membrane fusion or endocytosis/phagocytosis-like uptake. 3-OS HS: 3-O-sulfated heparan sulfate; HVEM: Herpes virus entry mediator.

Future Virol. 2015 Oct 1; 10(10): 1145–1154.

HSV Gene Expression

Often infect post-mitotic cells & consequently encode DNA synthetic machinery of their own.

Viral gene expression is tightly regulated:

1. All viral proteins fall into groups whose synthesis is coordinately regulated (i.e., all genes within a group are turned on at the same time)
2. All viral proteins are expressed in a cascade fashion - from immediate early (IE or α) to early (E or β) to late (L or γ)

The α (immediate-early) genes are expressed first, followed by $\beta 1$ (early-early), β_2 (late-early), γ_1 (leaky-late), and γ_2 (true late)

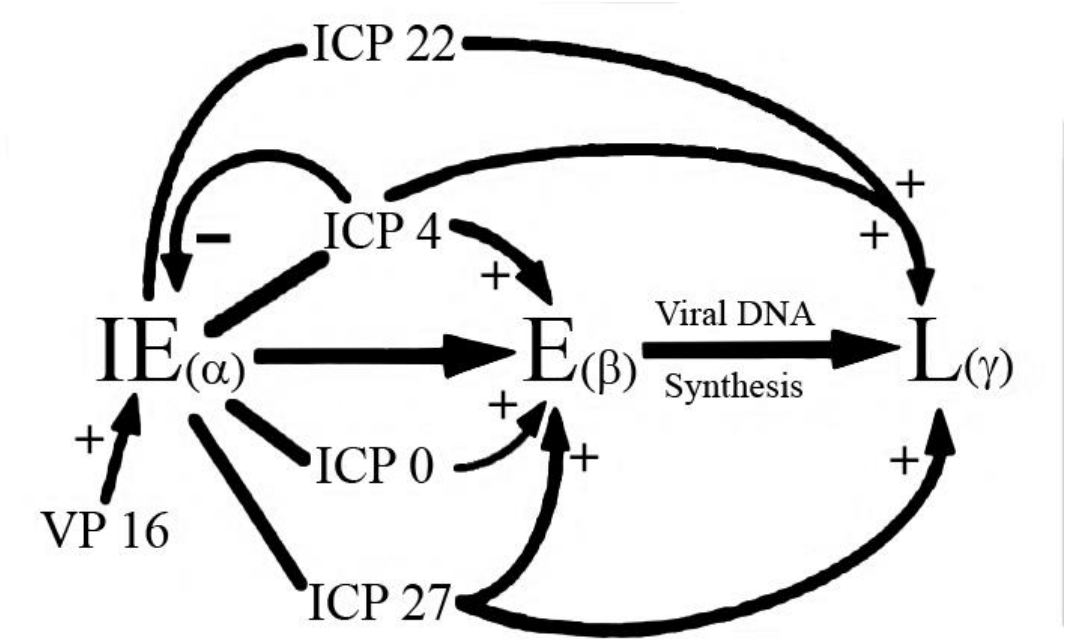
HSV-1 Gene Groups

Immediate-early (IE): 6 genes, which are designated as ICP (infected cell proteins) 0, 4, 22, 27 and 47, plus Us1.5. The RNAs are made even in the presence of protein synthesis inhibitors. All are transactivators of other viral genes. ICP4, ICP0 and ICP27 are essential for virus replication *in vitro*; other proteins are not essential *in vitro.*

Early (E): Made before viral DNA replication. Include enzymes involved in DNA metabolism, including thymidine kinase, ribonucleotide reductase, DNA polymerase.

Late (L): Made after viral DNA replication. Typically, structural proteins, as well as VP16 and vhs.

Herpesvirus Gene Expression



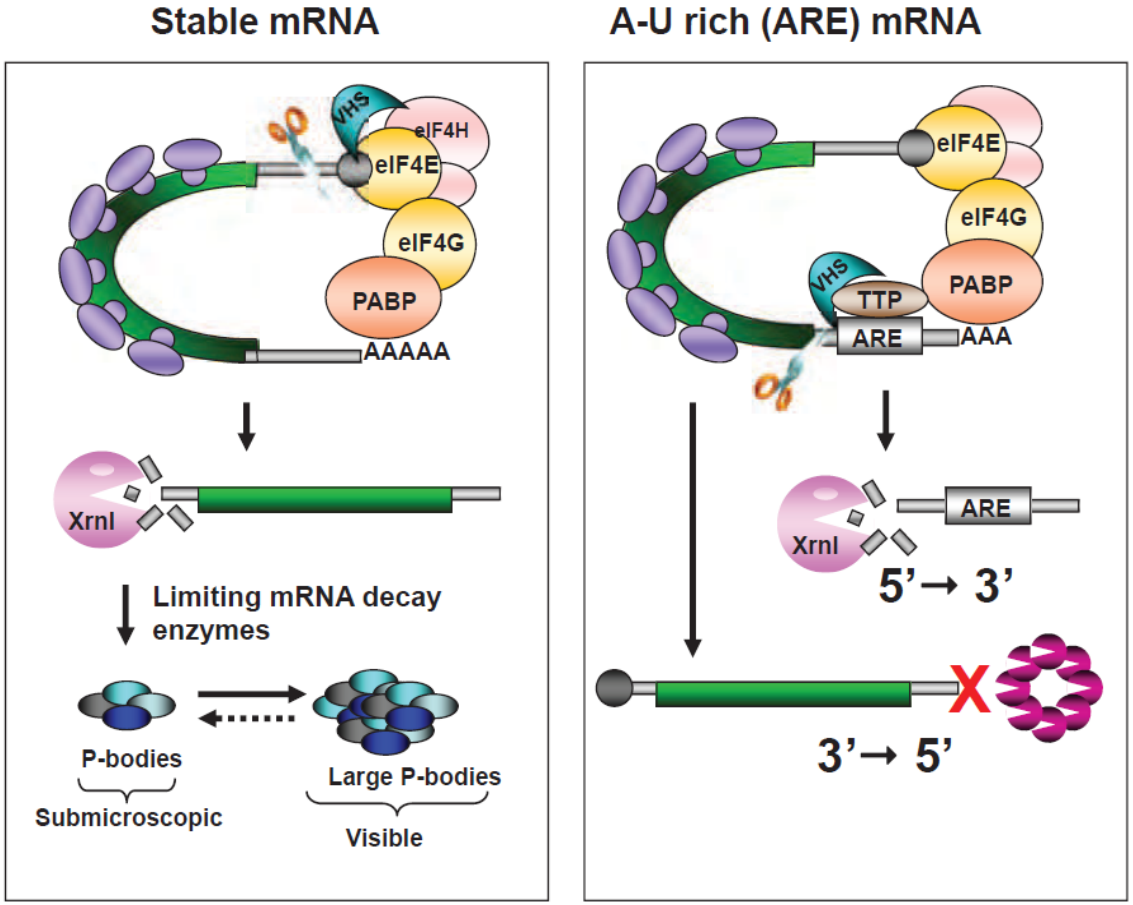
β1 and β2 genes require for their synthesis at least two of the six alpha proteins, ICP4 and ICP0.

The accumulation of the products of γ2 genes, require viral DNA synthesis and the viral proteins ICP4, ICP0, ICP22, ICP27, and US1.5.

Key HSV-1 Virion Proteins

Vhs (virion host shutoff or UL41): Not essential, but enhances replication efficiency. Causes non-specific mRNA degradation. Role of ICP27 in degradation specificity. Late in infection, it is complexed to VP16 and inactivated.

VP16 (α–TIF or UL48): An essential transactivator of viral immediate-early genes. Acts in concert with cellular transcription factors (including Oct-1 & the endogenous regulator of cell proliferation, HCF).



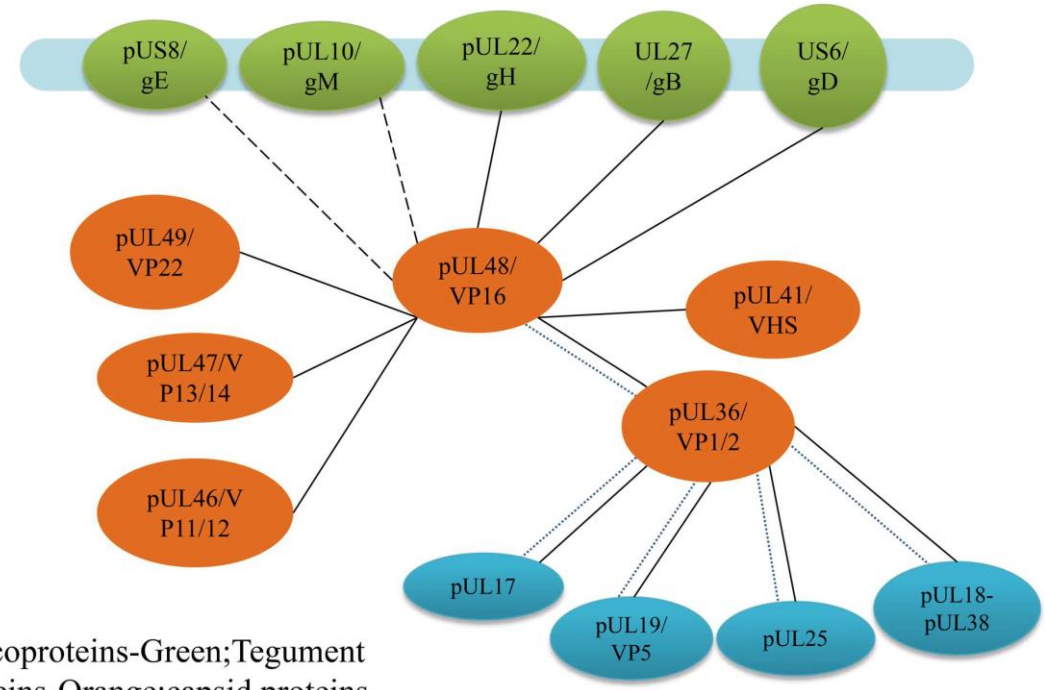
VHS is an endoribonuclease with the specificity of Rnase A. It has been shown to bind to both cap- and poly(A)-binding proteins.

Within a very brief interval after exposure to HSV, the cell responds by activation of NF-kB and the synthesis of a wide array of stress response mRNAs with the ultimate objective to block the synthesis of viral gene products. After a brief burst of activation of NF-kB by gD binding to HVEM and the UL37 tegument protein binding to TRAF6, HSV reduces this response by degradation of the stress response mRNAs by the VHS RNase, a tegument protein encoded by the *UL41* gene

Action of VP16 in activating HSV α genes

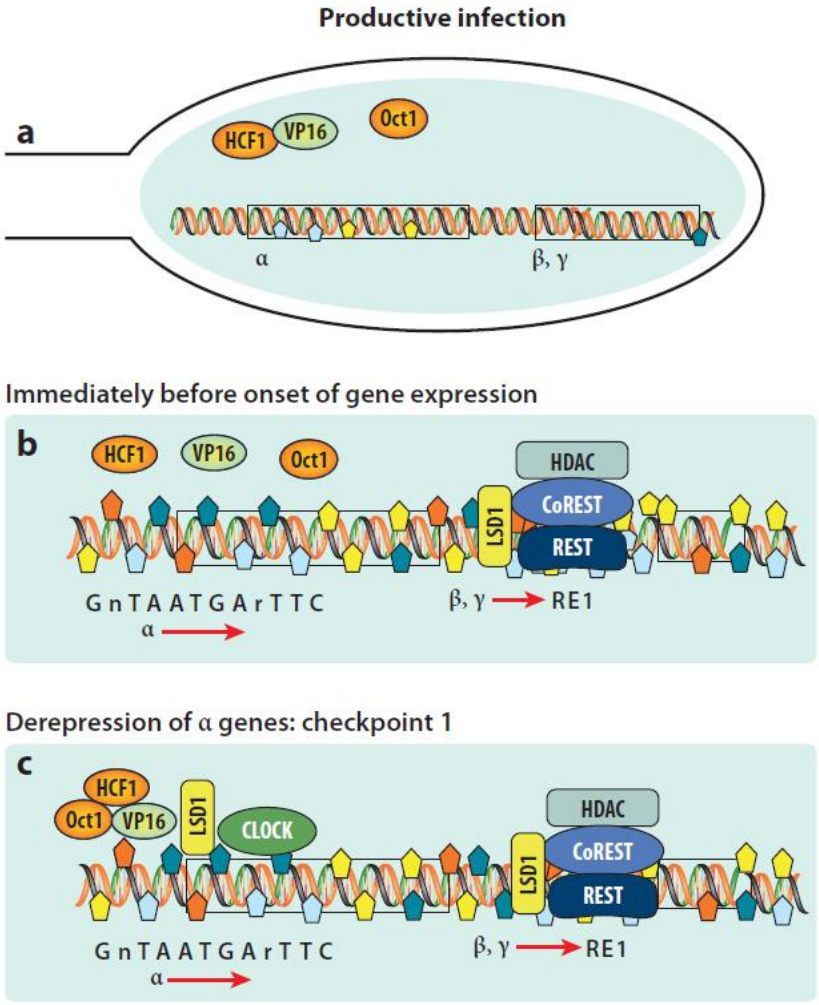
- VP16 has a potent transcriptional activation function when recruited to a promoter.
- Two host proteins, Oct1 and HCF, are required for VP16 to up-regulate α gene expression
- HCF associates with VP16 and mediates its transit into the nucleus
- Oct1 is a DNA binding transcription factor that binds to α gene promoters adjacent to a VP16 binding site. VP16/HCF complex can only bind DNA in the presence of Oct1
- Why the virus has evolved this indirect mechanism for activation by VP16 is uncertain; it may relate to the switch into and out of latency.

VP16 HAS ALSO A ROLE IN VIRAL ASSEMBLY



Glycoproteins-Green;Tegument proteins-Orange;capsid proteins-Blue

Model of regulation of viral gene expression in productively infected cells.

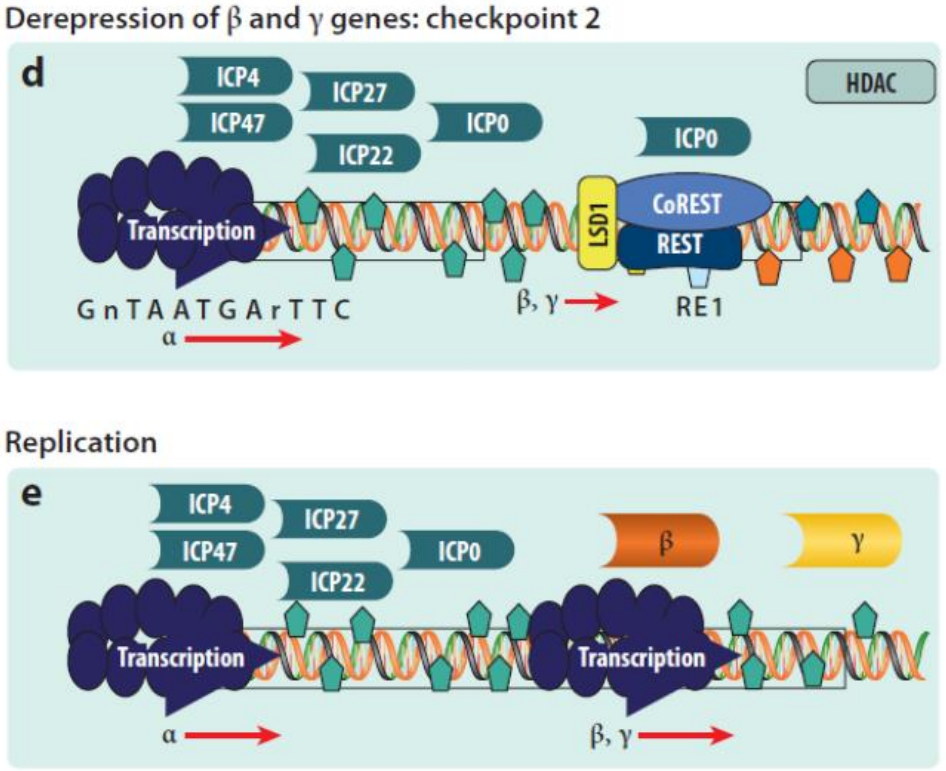


(a, b) Upon entry into cells, viral DNA is immediately bound by histone- and DNA-modifying enzymes, and repressors.

(c) VP16 assembles with HCF1, Oct1, to derepress gene promoters. The complex recruits transcriptional factors to transcribe α genes and overcome checkpoint 1.

Annu. Rev. Microbiol. 2013. 67:355–74

Model of regulation of viral gene expression in productively infected cells.



(*d, e*) ICP0, an α protein, binds to CoREST and dislodges HDAC1 or HDAC2. The complex dissociates from DNA and is translocated to the cytoplasm. Checkpoint 2 is overcome and β and γ genes are expressed. Viral replication ensues

Annu. Rev. Microbiol. 2013. 67:355–74

Replication and γ2 trancription

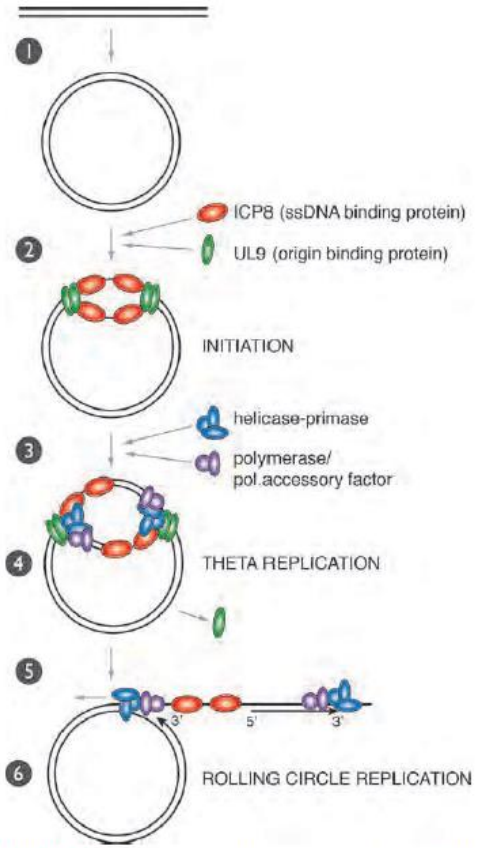
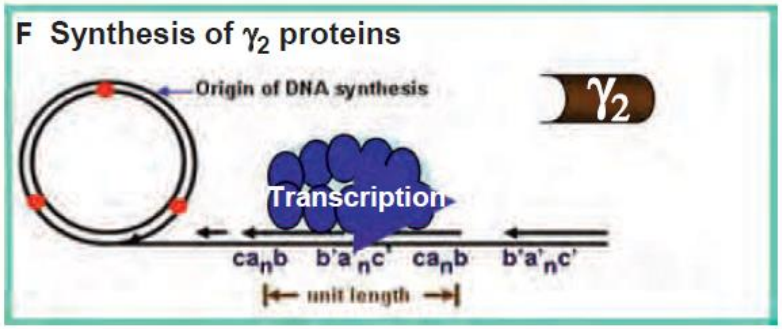


FIGURE 60.10. Diagram of a model of herpes simplex virus DNA replication. 1: Input DNA is circularized upon entry into the nucleus. **2:** U_L9 (the origin binding protein) initially binds to specific elements in the origin (either *ori*L or *ori*S) and begins to unwind the DNA. U_L9 then recruits ICP8 (the single-stranded DNA binding protein) to the unwound single-stranded DNA. **3:** U_L9 and ICP8 recruit the five remaining viral DNA replication proteins to the replication forks. **4:** The helicase-primase proteins and the viral polymerase complex assemble at each replication fork for initial rounds of theta form replication. **5:** Replication switches from theta to rolling circle mode by an unknown mechanism. U_L9 is not necessary for rolling circle replication, as it is not origin dependent. **6:** Rolling circle DNA replication produces long head-to-tail concatamers of viral DNA, which are cleaved into monomeric molecules during packaging. (Copyright Lynne Chang and David Knipe.)



α gene products

ICP0: is believed to enable the expression of beta genes by countering the epigenetic silencing of these genes. ICP0 promotes (E3ubiquitin ligase activity), either directly or indirectly, the degradation or reduction in the levels of several cellular proteins (innate response inhibition, TLR2, IFI6, PML…)

ICP4, fulfills the second function needed for post-a gene transcription: recruitment of RNA polymerase II, TATA-binding protein and the Mediator complex to these gene promoters. It can repress its own expression.

ICP22, It has been detected in complexes containing ICP4, ICP27, TFIID, and CLOCK HAT

ICP27 promotes beta and gamma transcription, stimulates viral DNA replication, inhibits splicesome assembly, it chaperone viral mRNAs virtually from the time of synthesis to their translation (protection from degradation mediated by vhs?)

ICP47 selectively binds to human TAP1/TAP2 and blocks the transport of antigenic peptides into the ER for presentation at the cell surface

Us1.5. functions less defined, contained within the ICP22 Orf

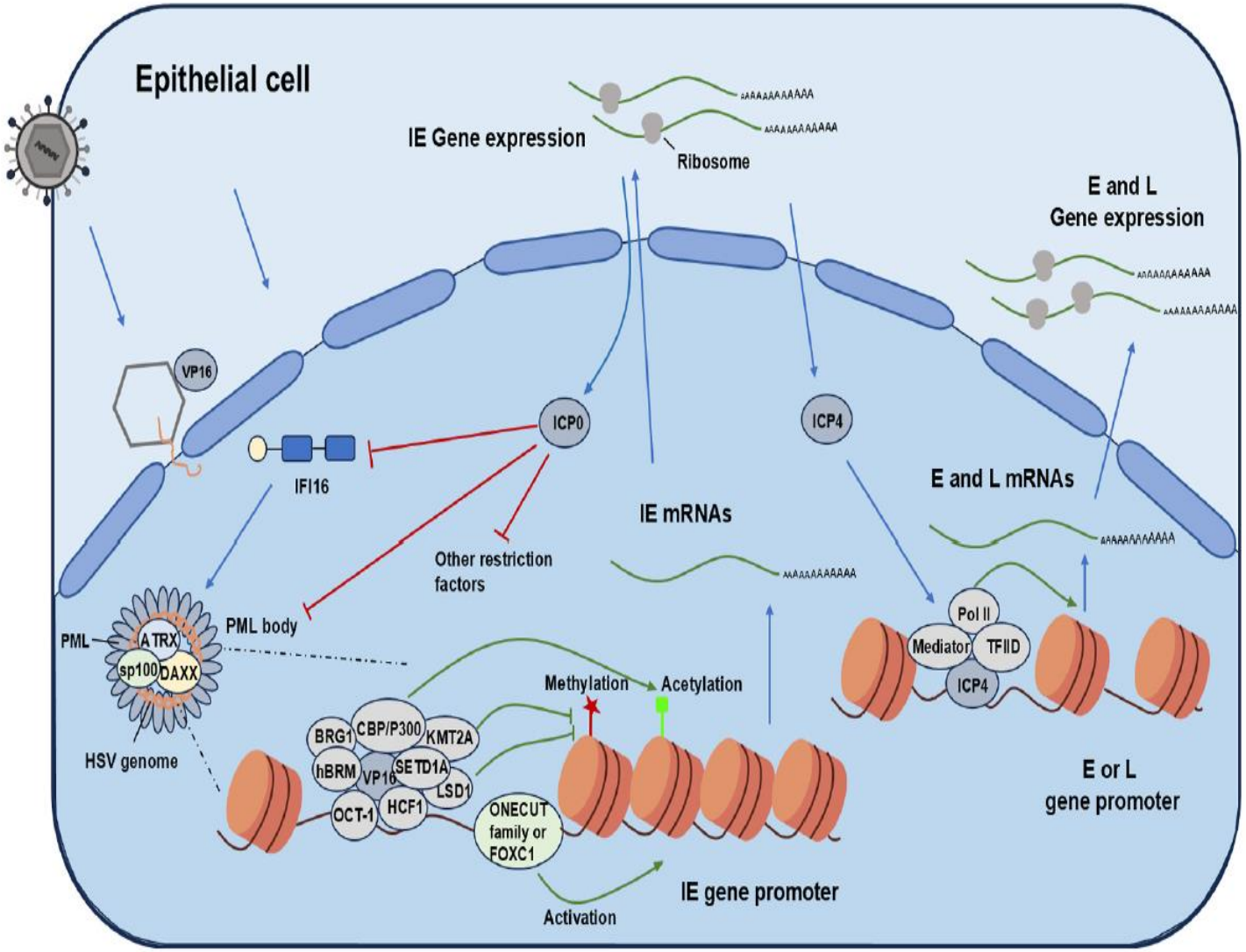


Fig. 2. Mechanisms of viral gene activation during lytic infection in epithelial cells. After nuclear entry, the viral genome froms heterochromatin within PML-NBs containing restriction factors. The VP16-induced complex recruits host chromatin modifying and remodeling proteins to the IE gene promoters to activate IE gene expression. Host ONECUT family and FOXC1 also bind to viral genes to facilitate activation. The IE protein ICP0 counteracts host restrictive mechanisms through the E3 ligase activity. ICP4 binds to the viral genome to recruit cellular general transcription factors to stimulate transcription of E and L genes. Some graphic elements were obtained from Server Medical Art (SMART) (https://smart.servier.com).

Among the β gene products: TK and RR

Function: Thymidine kinase and ribonucleotide reductase are involved in the generation of DNA precursors.

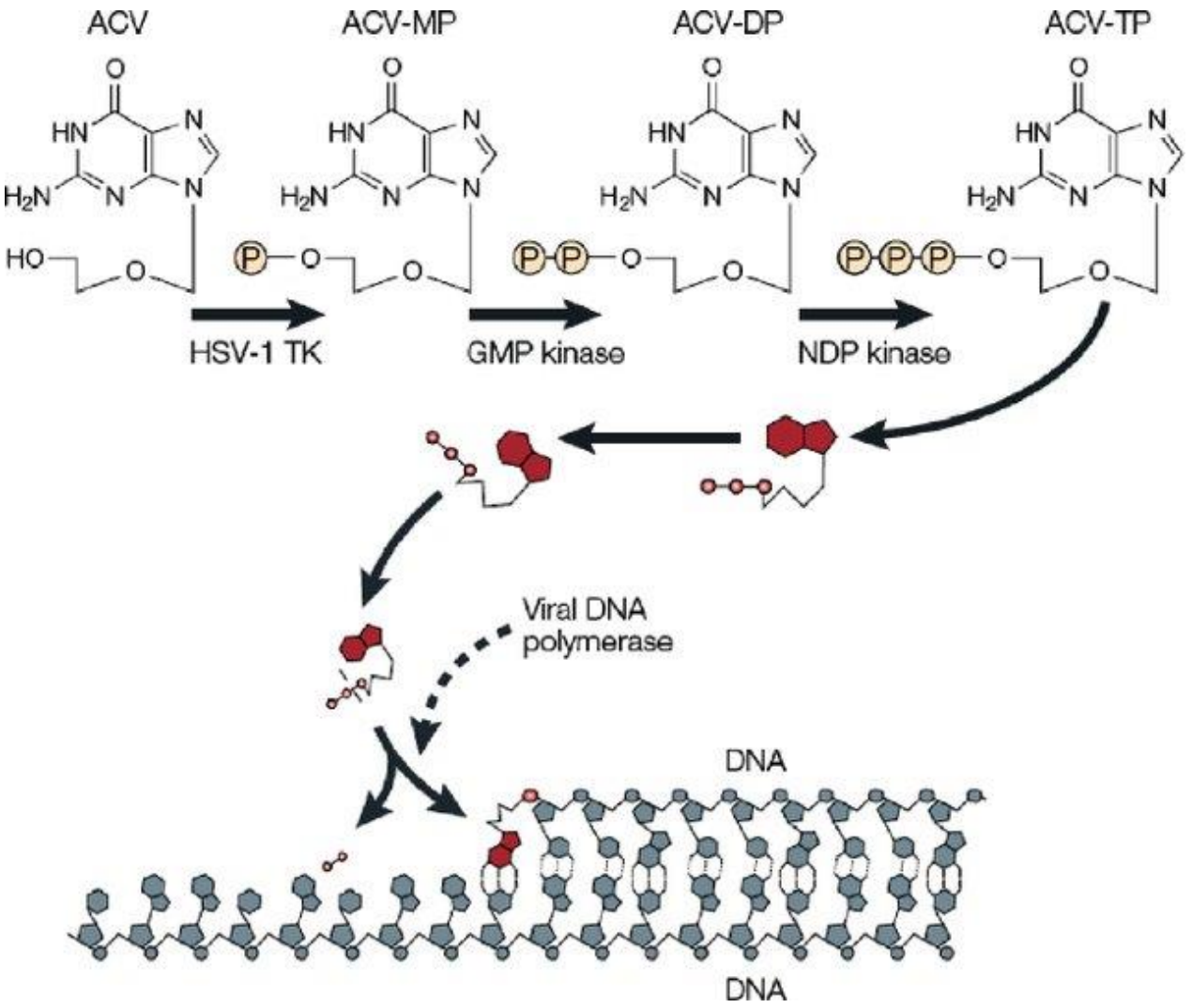
- Thymidine kinase (phosphorylates thymidine and other nucleosides)
- Ribonucleotide reductase (converts ribonucleotides to deoxyribonucleotide)

Phenotype of TK- or RR- HSV-1: Such mutants replicate fine in dividing cells, but fail to replicate in post-mitotic cells like neurons (where the available pool of DNA precursors is small).

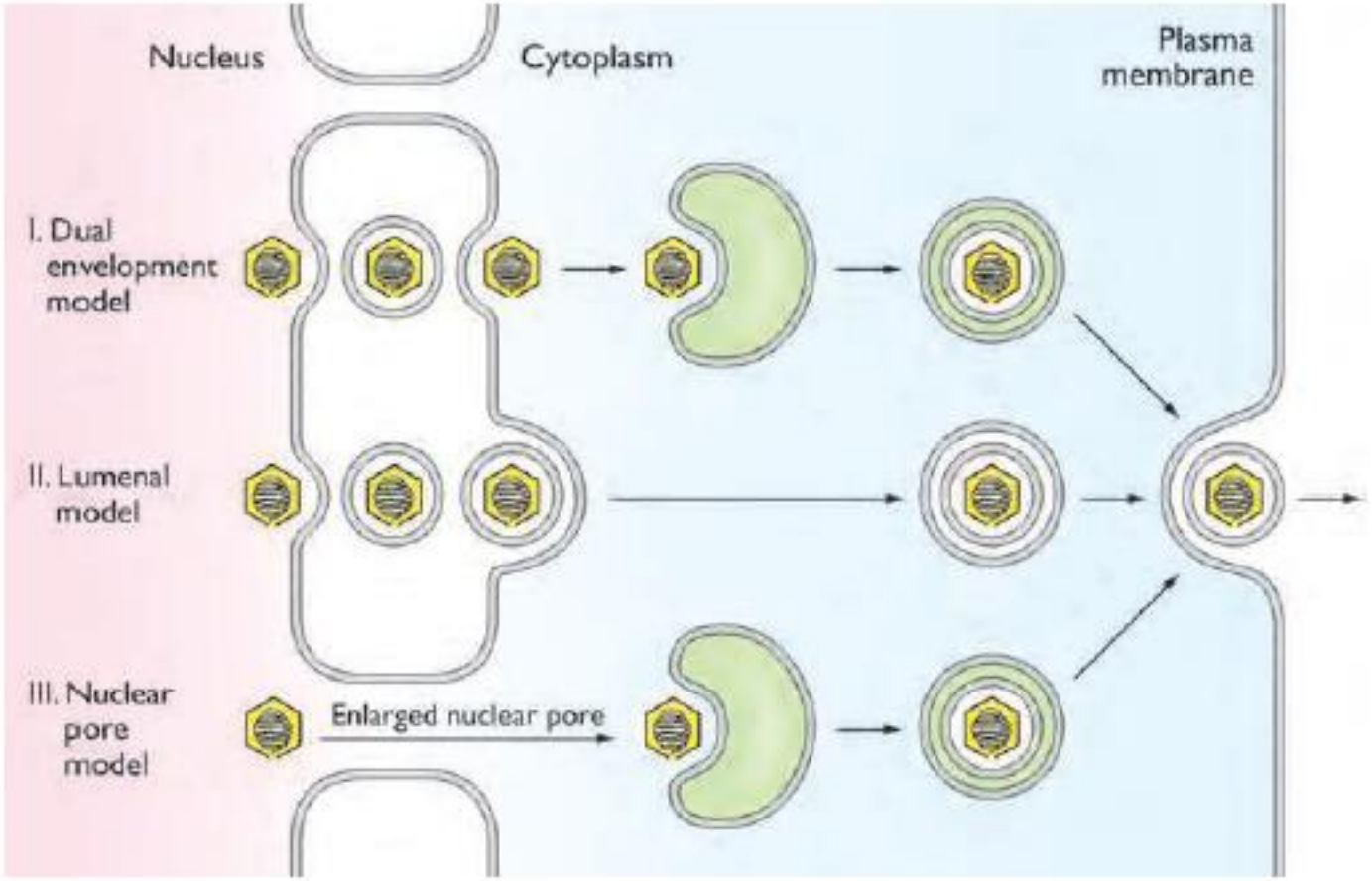
Use of TK and RR deleted viruses: Being used for therapy of brain tumors: CNS cells (including neurons) are all essentially post-mitotic, while tumor cells are not. So, these viruses can replicate only in the tumor cells (killing them).

Moreover…TK activity has been exploited to develop the first antiviral specific drug

Mechanism of antiviral action of acyclovir



Models for egress of herpes simplex virus (HSV) from the host cell



Although this has been a controversial area, the first model—envelopment–de-envelopment–re-envelopment—is supported by substantial data and is considered to be the “most prominent model of virion egress”.

Models for egress of herpes simplex virus (HSV) from the host cell. Shown are the proposed pathways for maturation of filled capsids from the nucleus to the exterior of the infected cell.

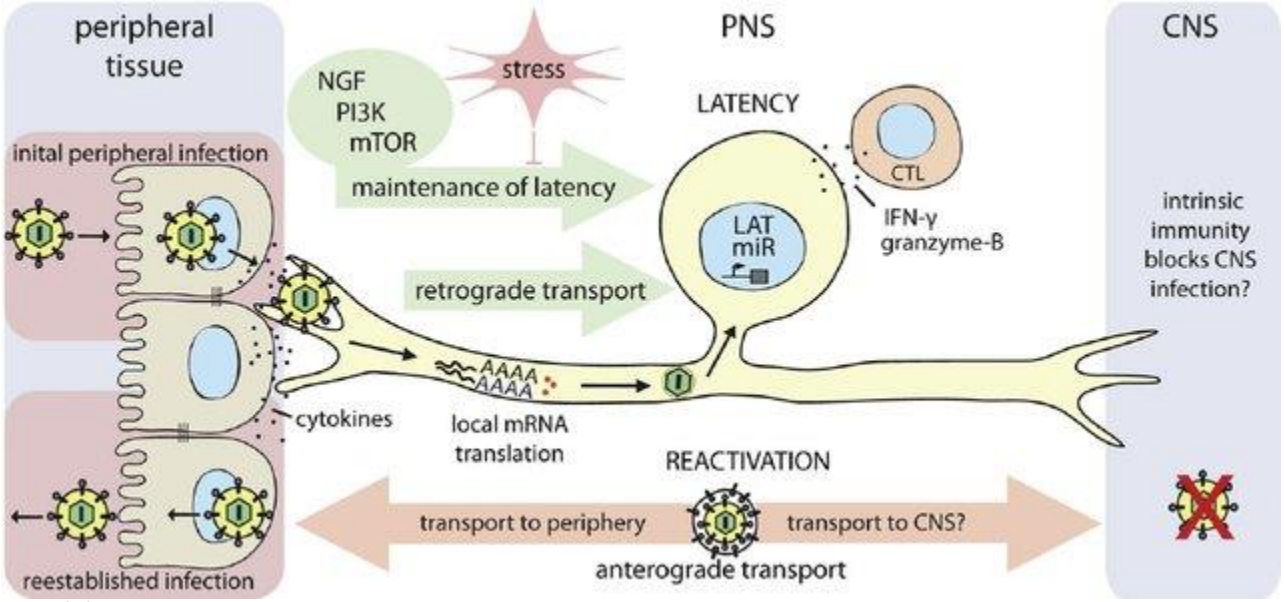
I: In the dual envelopment pathway, nucleocapsids undergo primary envelopment at the inner nuclear membrane, become de-enveloped at the outer nuclear membrane, undergo secondary envelopment at cytoplasmic membranes, and then are transported in vesicles to the plasma membrane where fusion of the vesicle with the plasma membrane releases the virion to the extracellular space.

II: In the lumenal model, the nucleocapsid is enveloped at the inner nuclear membrane, enters a vesicle at the outer nuclear membrane, and is then transported to the plasma membrane where the virion is released.

III: In the nuclear pore model, filled capsids exit the nucleus through enlarged nuclear pores and become enveloped by budding into cytoplasmic vesicles, and the virion is transported to the plasma membrane where the virion is released.

Wild et al have proposed a dual pathway for egress, in which HSV uses the luminal pathway at early times of infection but the bulk of extracellular virus is formed by the nuclear pore pathway at late times of infection.

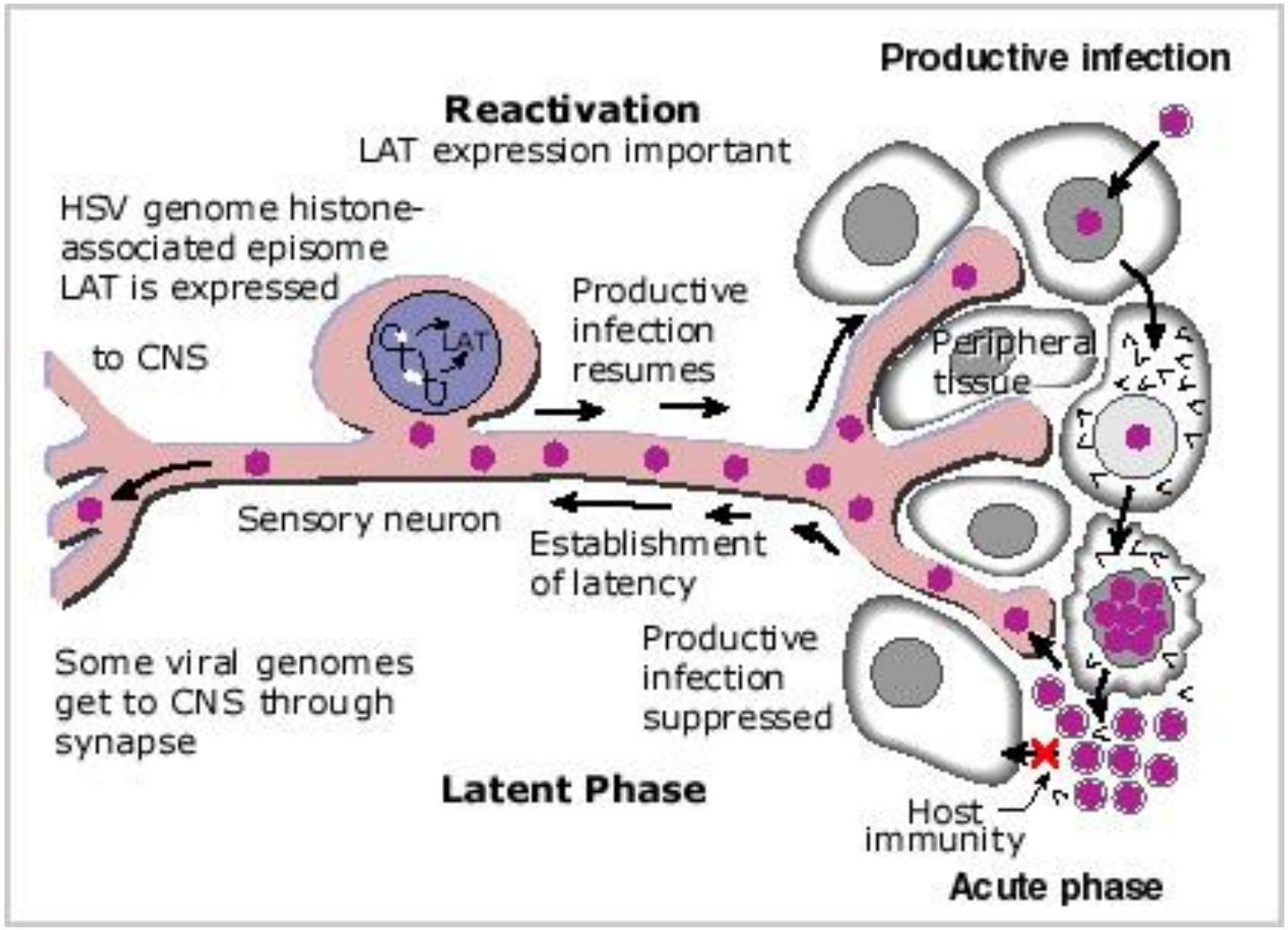
After replicating in surface epithelia, herpes simplex virus type-1 (HSV-1) enters the axonal terminals of peripheral neurons. The viral genome translocates to the nucleus, where it establishes a specialized infection known as latency, re-emerging periodically to seed new infections



2013 Cell Host & Microbe 13(4):379-93

Along the axon, the nucleocapsid is retrograde transported to the nucleus in the neuronal cell body where lifelong latent infection is established. Undetectable production of infectious virus, an episomal viral genome in the nucleus, silencing of virtually all genes required for the lytic cycle but relatively high expression of latency-associated transcripts (LATs) and some LATderived microRNAs.

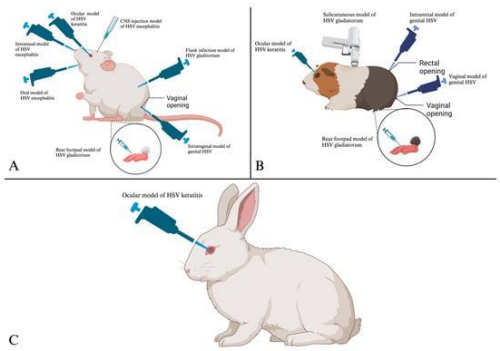
Latent Infections by HSV 1

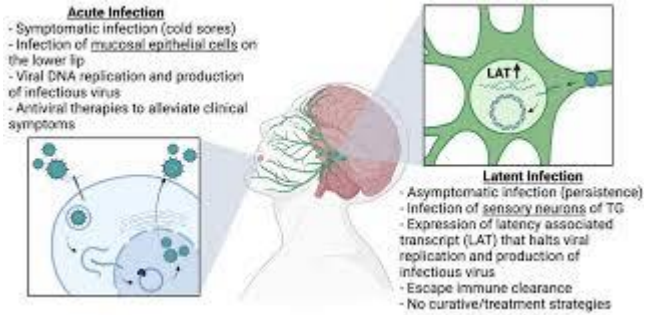


Undetectable production of infectious virus, an episomal viral genome in the nucleus, silencing of virtually all genes required for the lytic cycle but relatively high expression of latency-associated transcripts (LATs) and some LAT-derived microRNAs.

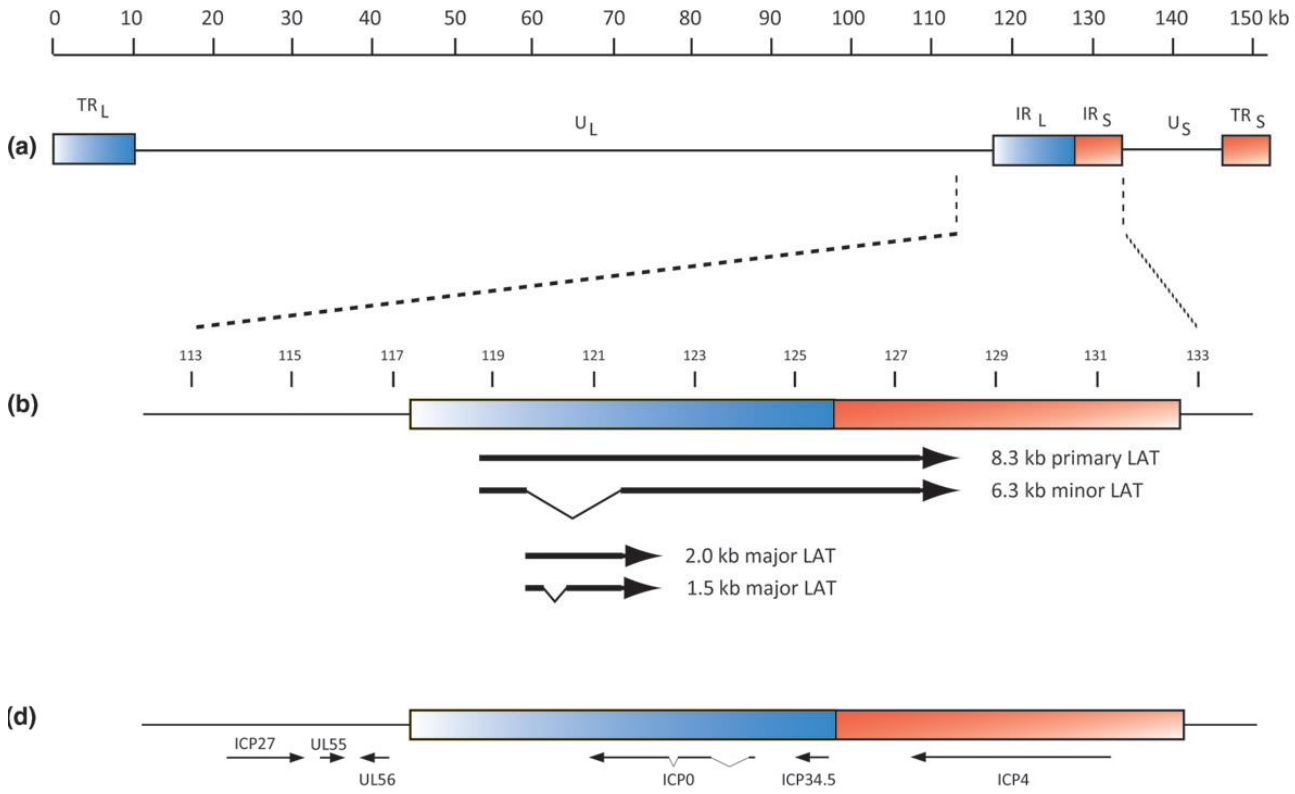
During latent infection, lytic genes are globally silenced whereas the LAT gene locus shows high transcription activity. The silencing is not absolute as lytic transcripts are still detectable at low levels in latently infected ganglia

More than 50% of latently infected neurons express more than one viral lytic gene at any one time





Viral RNA mediated repression of lytic genes during latency:
LAT transcripts



The 8.3 kb polyadenylated primary LAT is spliced into 1.5 and 2 kb stable introns that are abundant during latency, as well as a 6.3 kb exon, which itself is hard to detect but is processed into multiple miRNAs that are easily detectable

M.P. Nicoll et al., FEMS Microbiol Rev 36 (2012) 684–705

Viral RNA mediated repression of lytic genes during latency:
LAT transcripts

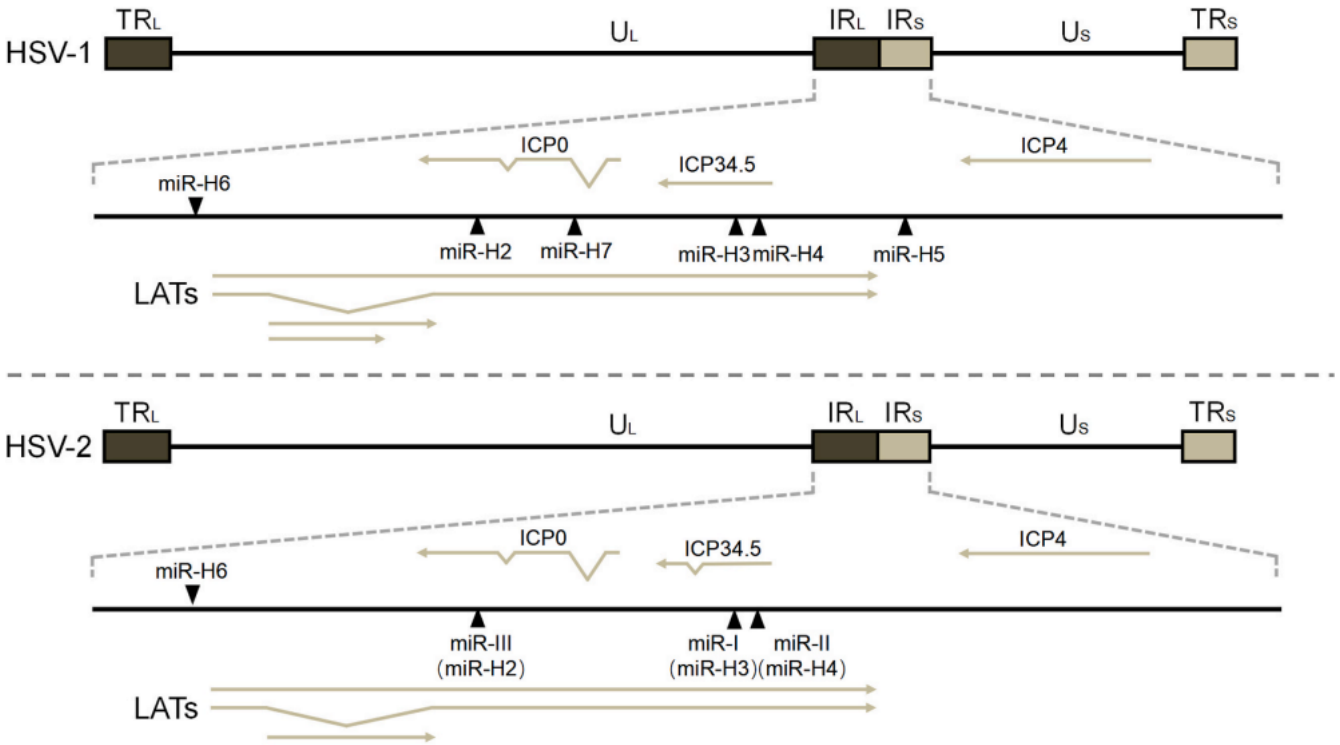
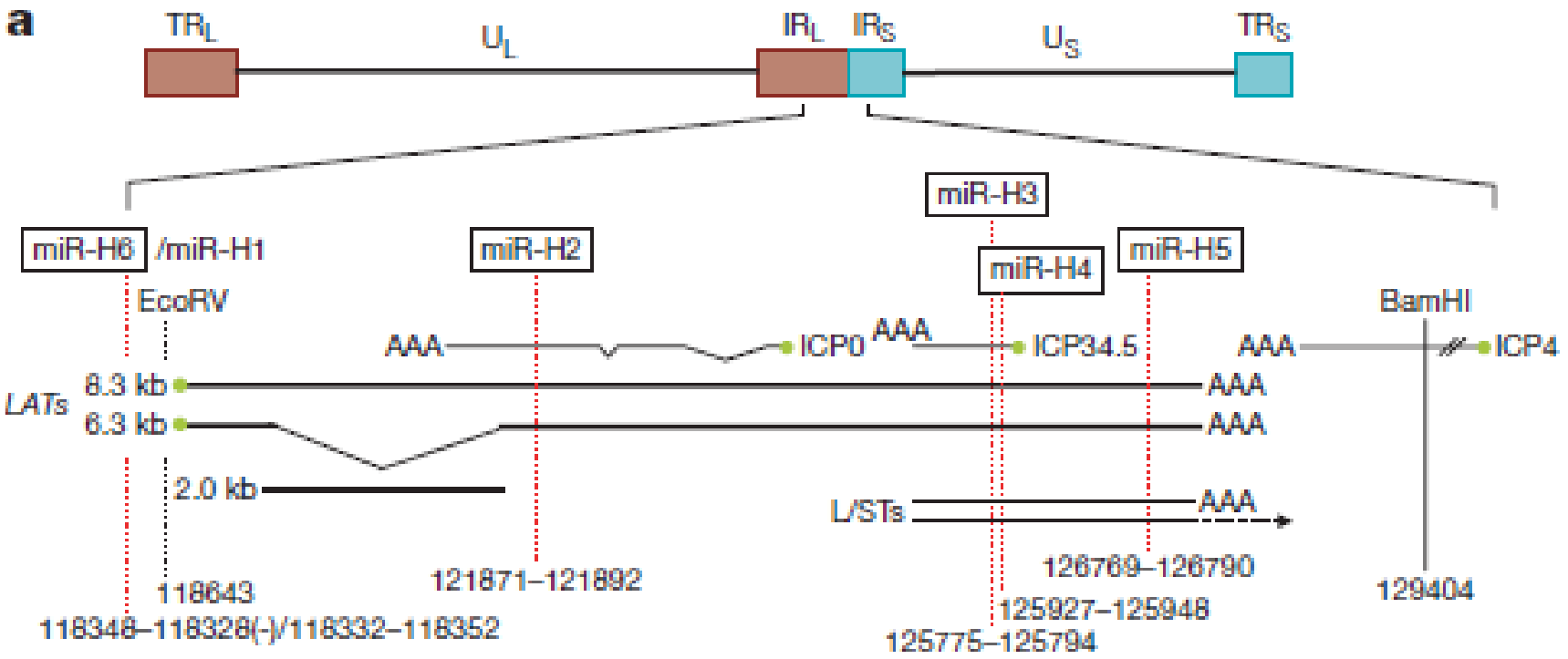


Fig. 1. Map of the LAT gene locus showing the positions of LATs and miRNAs readily detectable during latency. HSV-1 and HSV-2 genomes are shown in the prototype orientation. TR_L, IR_L, TR_S, IR_S, U_L and U_S denote long terminal repeat, long internal repeat, short terminal repeat, short internal repeat, long unique and short unique regions, respectively. The internal repeat regions are expanded to show the genomic positions of the different transcripts and miRNAs.

Mechanisms of HSV gene regulation during latency and reactivation
Hui Fu, Dongli Pan Virology 2025

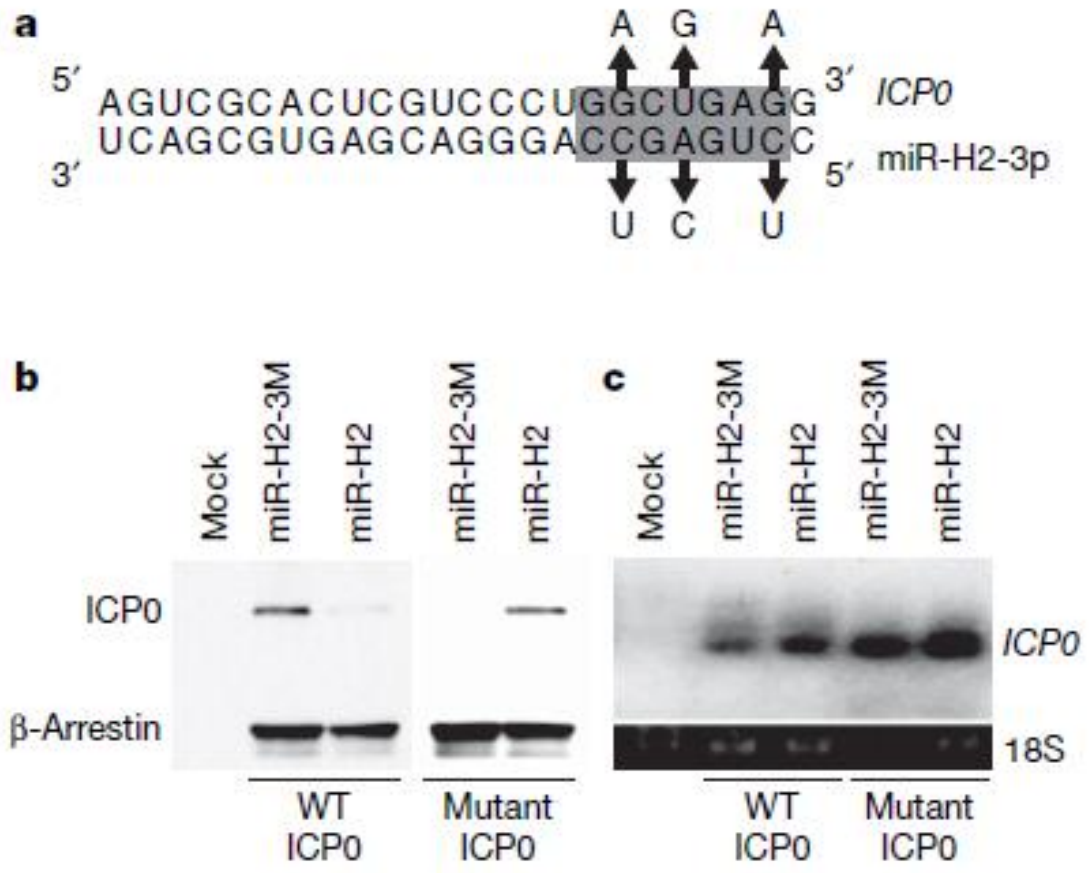
The exonic regions of LAT function as a primary miRNA precursor



miRNAs readily detected during latency include miR-H2, miR-H3, miR-H4, miR-H5, miR-H6 and miR-H7

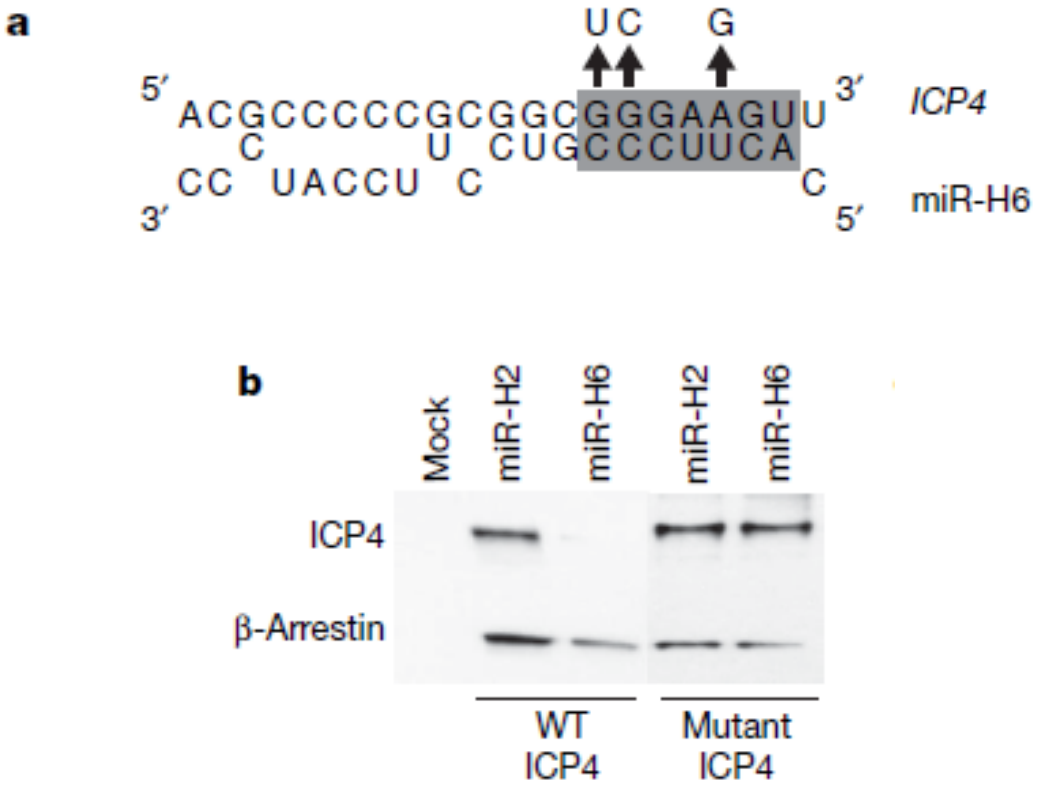
J.L. Umbach et al., Nature 2008, 454: 780-783
Hui Fu, Dongli Pan Virology 2025

Downregulation of ICP0 protein expression by HSV-1 miR-H2



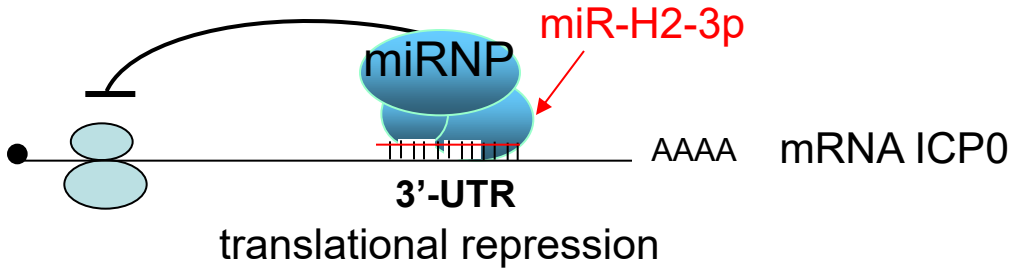
J.L. Umbach et al., Nature 2008, 454: 780-783

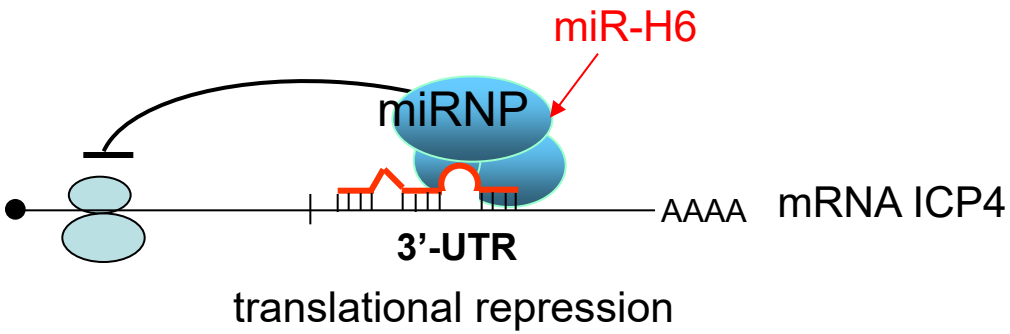
Downregulation of ICP4 protein expression by HSV-1 miR-H6



J.L. Umbach et al., Nature 2008, 454: 780-783

miR-H2-3p: derived from the antisense transcript of the viral mRNA encoding the immediate early protein ICP0



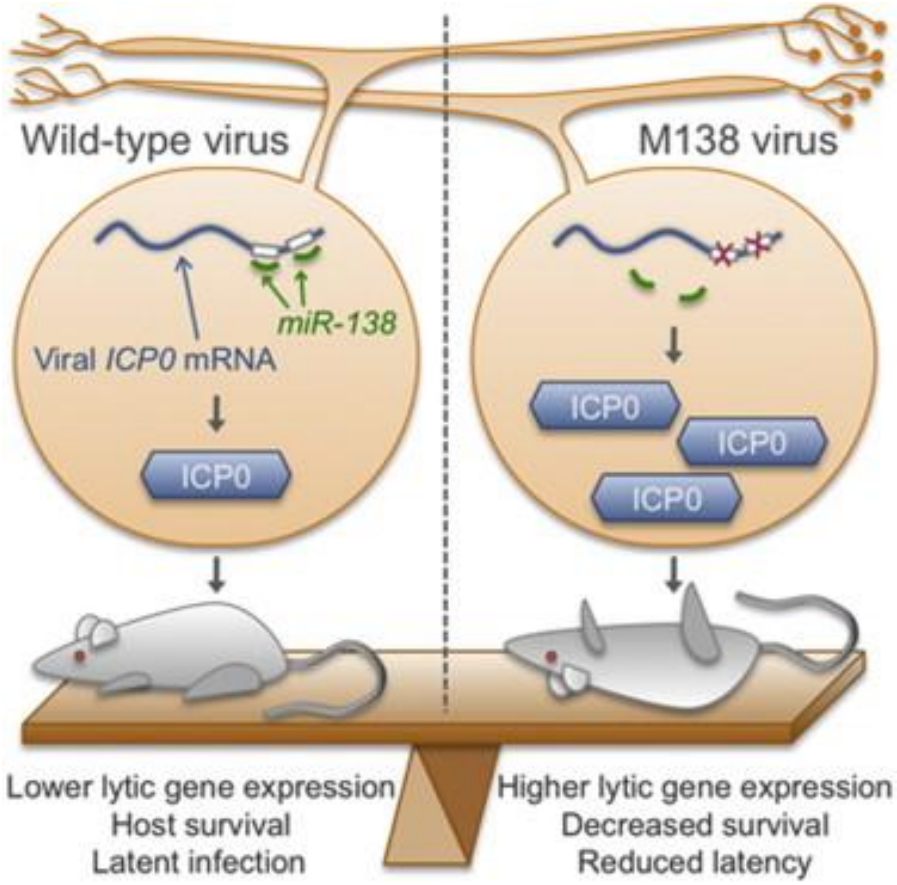


miR-H2-3p and miR-H6 negatively regulate 2 immediate early proteins

Important

- for entry into latency
- to inhibit the transition from latency to lytic cycle.

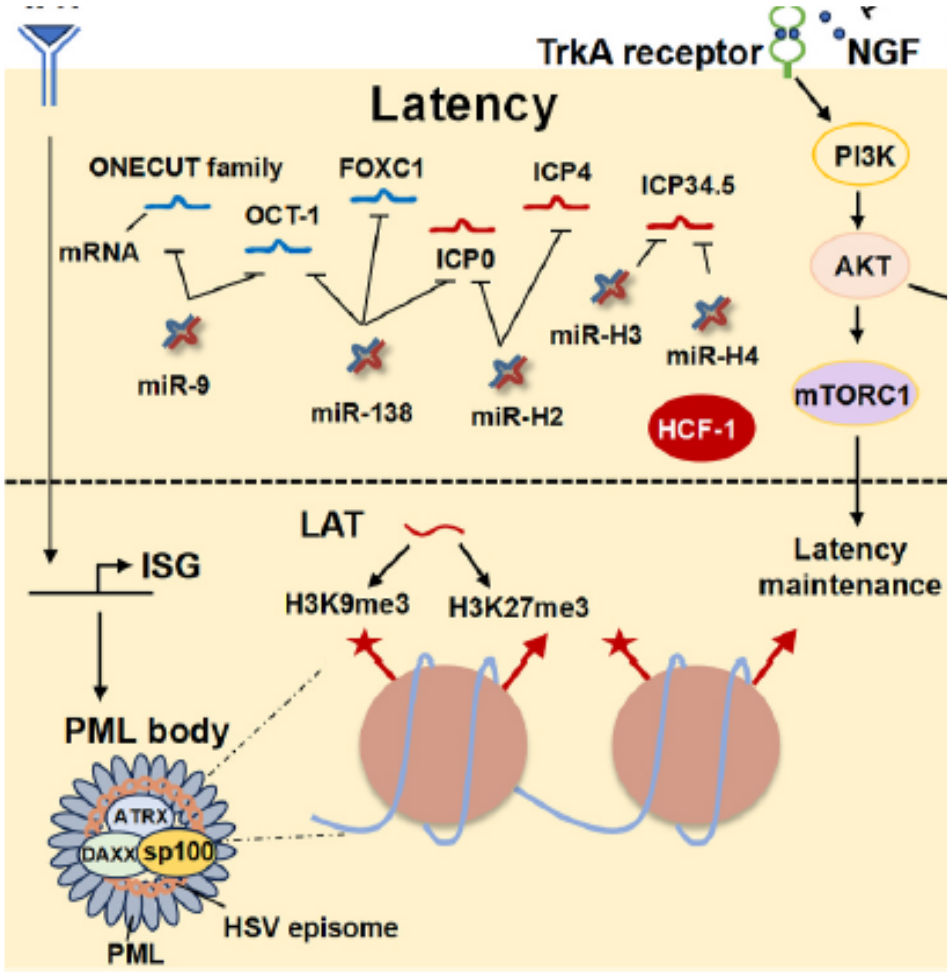
A Neuron-Specific Host MicroRNA Targets Herpes Simplex Virus-1 ICP0 Expression and Promotes Latency



After infecting peripheral sites, herpes simplex virus (HSV) invades the nervous system and initiates latent infection in sensory neurons. Establishment and maintenance of HSV latency require host survival, and entail repression of productive cycle ("lytic") viral gene expression. We find that a neuron-specific microRNA, miR-138, represses expression of ICP0, a viral transactivator of lytic gene expression. A mutant HSV-1 (M138) with disrupted miR-138 target sites in *ICP0* mRNA exhibits enhanced expression of ICP0 and other lytic proteins in infected neuronal cells in culture. Following corneal inoculation, M138-infected mice have higher levels of*ICP0* and lytic transcripts in trigeminal ganglia during establishment of latency, and exhibit increased mortality and encephalitis symptoms. After full establishment of latency, the fraction of trigeminal ganglia harboring detectable lytic transcripts is greater in M138-infected mice. Thus, miR-138 is a neuronal factor that represses HSV-1 lytic gene expression, promoting host survival and viral latency.

Cell Host and Microbe, Volume 15, Issue 4, 2014, 446 - 456

Formation of latent viral chromatin

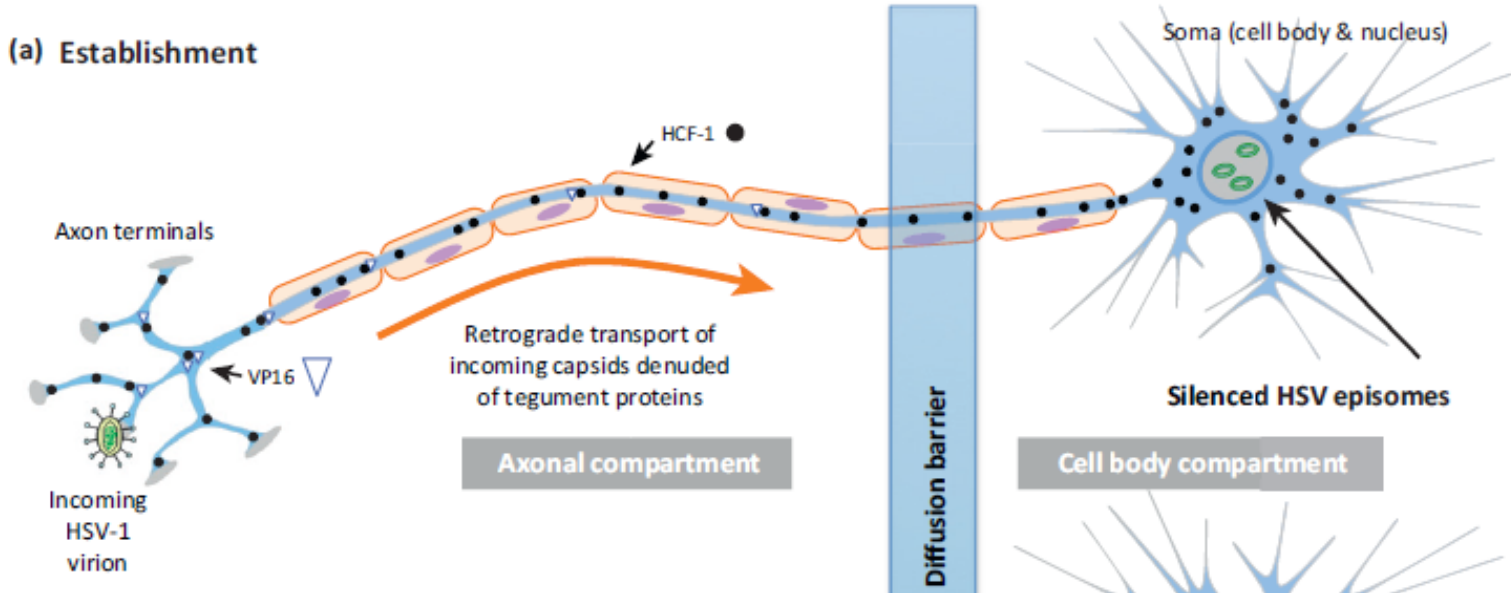


Concomitant with the establishment of latency is progressive **enrichment of H3K9me3 and H3K27me3** modifications **on viral lytic promoters**

H3K9me3 is a marker of **constitutive heterochromatin** while **H3K27me3** is a hallmark of **facultative heterochromatin** and might contribute to a “poised” viral chromatin ready for reactivation.

In contrast to lytic promoters, the LAT promoter is enriched with histone H3 acetylation and **H3K4me3** modifications indicative of **active chromatin** during latency

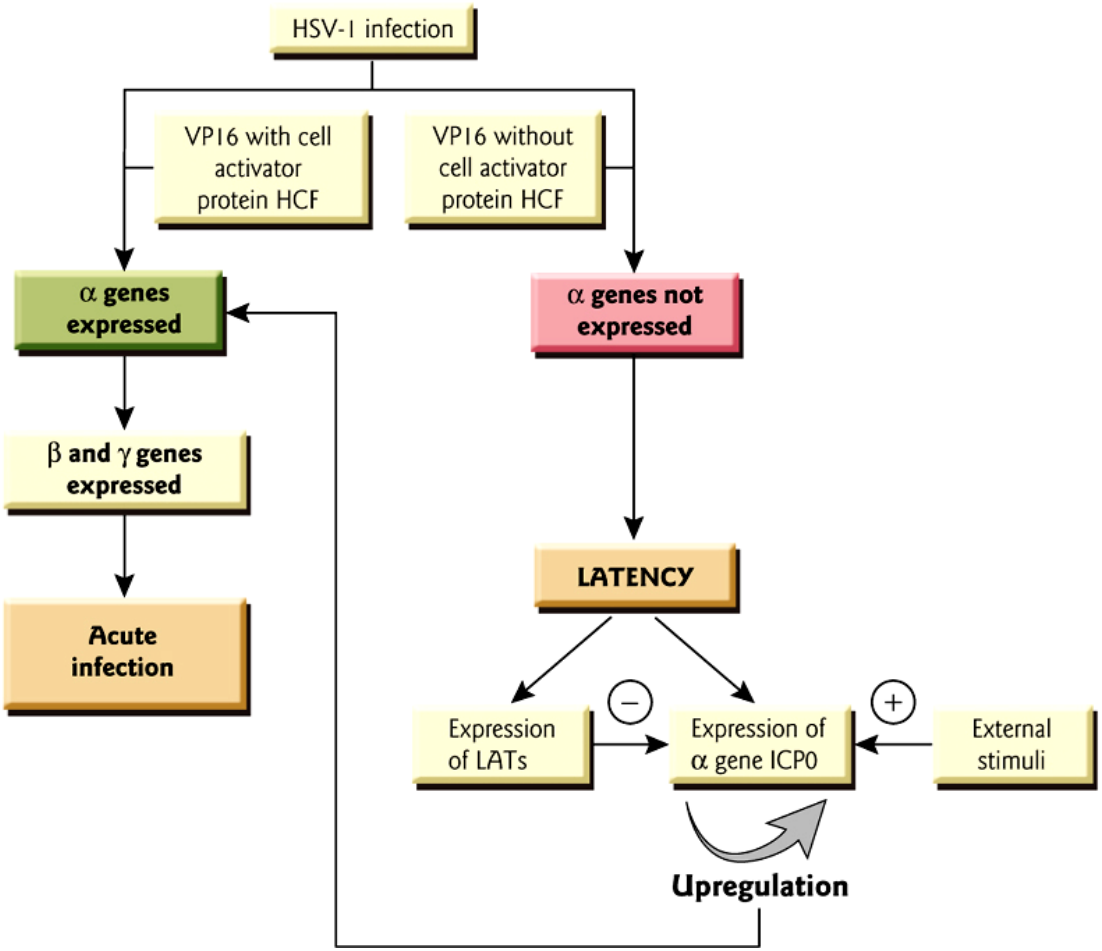
Reduced availability of the VP16-induced complex in latently infected neurons



The tegument protein VP16 (blue triangles) dissociates from the capsid almost immediately after release into the cytoplasm, and translocates to the nucleus with very low efficiency. Additionally, HCF-1 is mainly localized to the Golgi apparatus in normal or latently-infected neurons and re-localized to the nucleus only upon stimulation that results in reactivation (Whitlow and Kristie, 2009).
Furthermore, OCT-1 is poorly expressed in sensory neurons at least in part due to high expression of neuronal miRNAs miR-138 and miR-9 that target Oct-1 mRNA

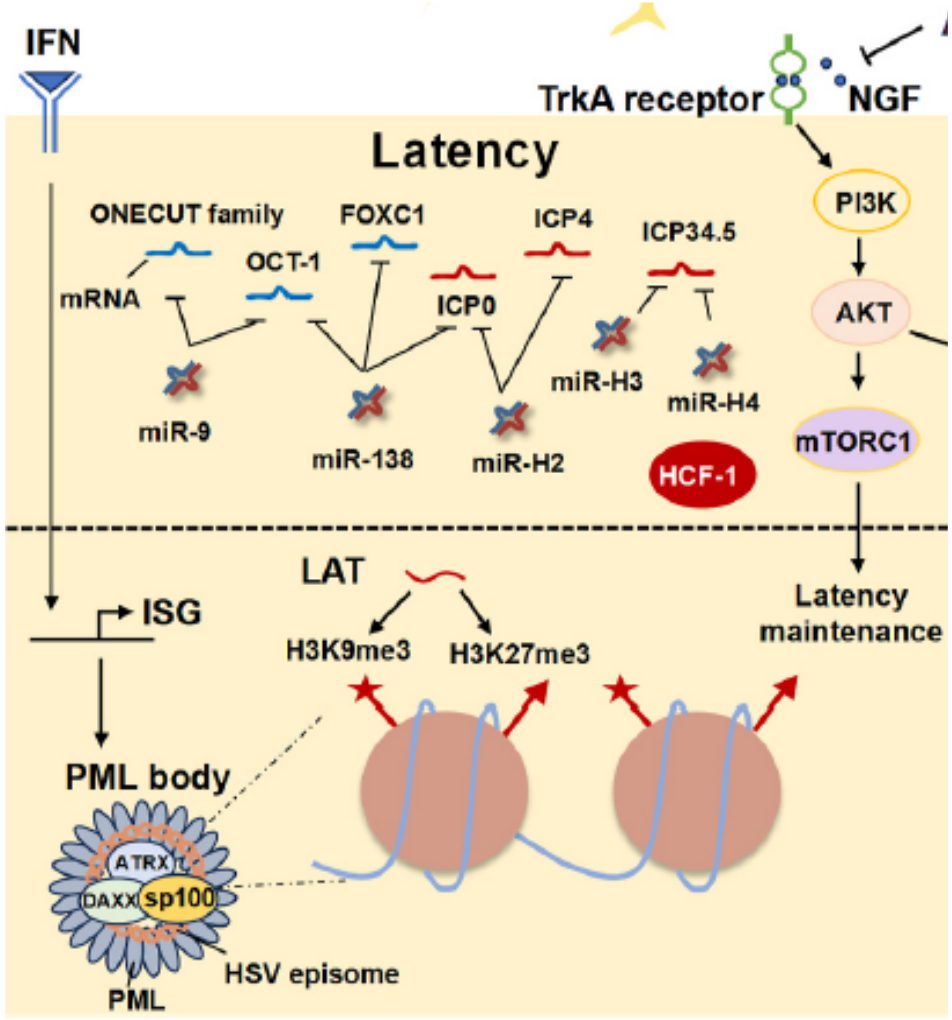
A.C. Wilson et al. Trends in Microbiology 2012, 20: 604-611

Latent Infections by HSV 1



From Dimmock, Easton and Leppard, "Introduction to Modern *Virology"* (2007). Blackwell Publishing

Role of intrinsic and innate immunity in viral gene repression during latency



IFN-stimulated levels of the nuclear DNA sensor IFI16 can stabilize virus-associated heterochromatin and repress viral gene expression in wild type HSV-1 infected fibroblasts (Sodroski and Knipe, 2023). Interestingly, although many host restriction factors are constitutively expressed, expression of some PML-NB proteins such as PML and Sp100 is further stimulated by IFN (Gr¨otzinger et al., 1996; Stadler et al., 1995). Accordingly, type I IFN treatment promotes the formation of PML-NBs, which persist for days after IFN removal and colocalize with HSV-1 genomes to restrict reactivation in a neuronal culture model

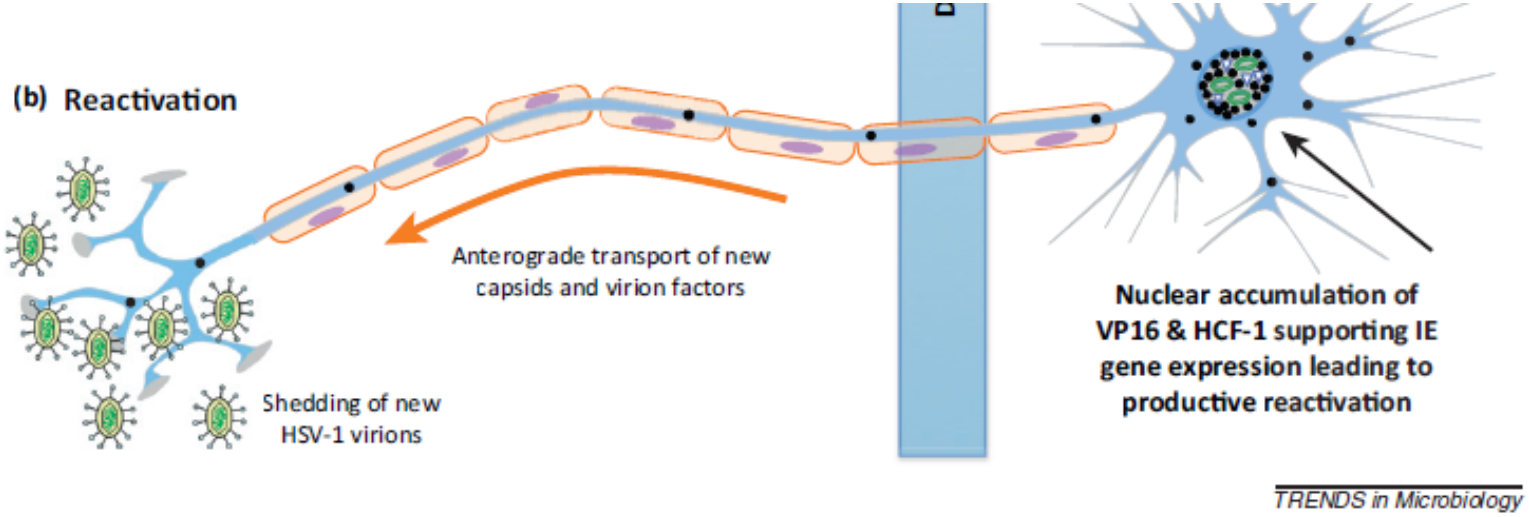
Reactivation

A study in a primary neuronal culture model identified two phases of reactivation from latency (Kim et al., 2012).

The first phase (also called the animation phase) is characterized by genome-wide de-repression of lytic genes without viral DNA synthesis or particle production.

The second phase exhibits the ordered gene expression program similar to de novo lytic infection

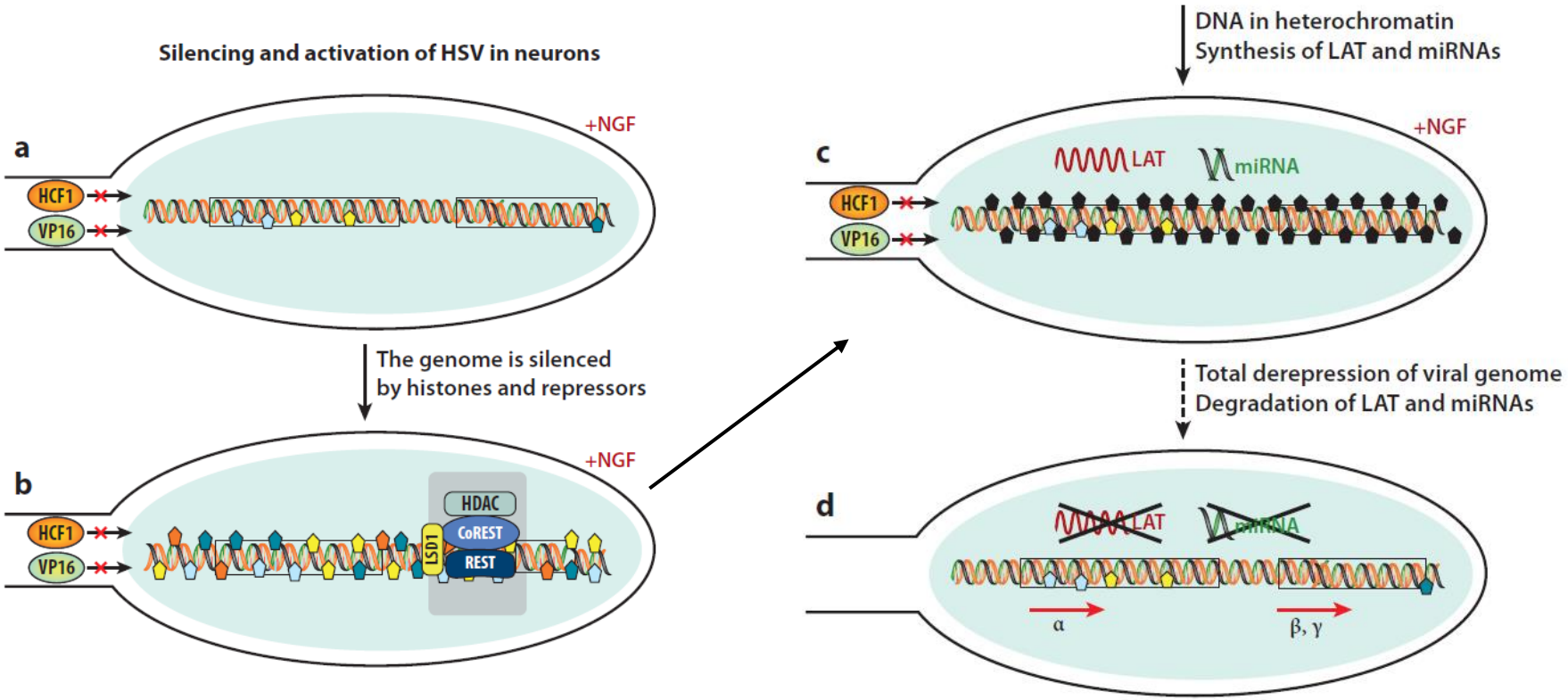
Latent Infections by HSV 1



Reactivation stimuli can elicit many changes in the neuron, including nuclear accumulation of HCF-1 and VP16, which is synthesized de novo along with other viral regulatory proteins. Stimulation of viral lytic transcription by VP16 leads to viral DNA amplification and synthesis of virion proteins. Capsids are transported in an anterograde fashion to the axonal termini where they mature and are then released, bringing the HSV-1 life cycle full circle.

A.C. Wilson et al. Trends in Microbiology 2012, 20: 604-611

A model of silencing and activation of HSV in neurons



Possible involvment of JNK in reactivation

Annu. Rev. Microbiol. 2013. 67:355–74

A model of silencing and activation of HSV in neurons

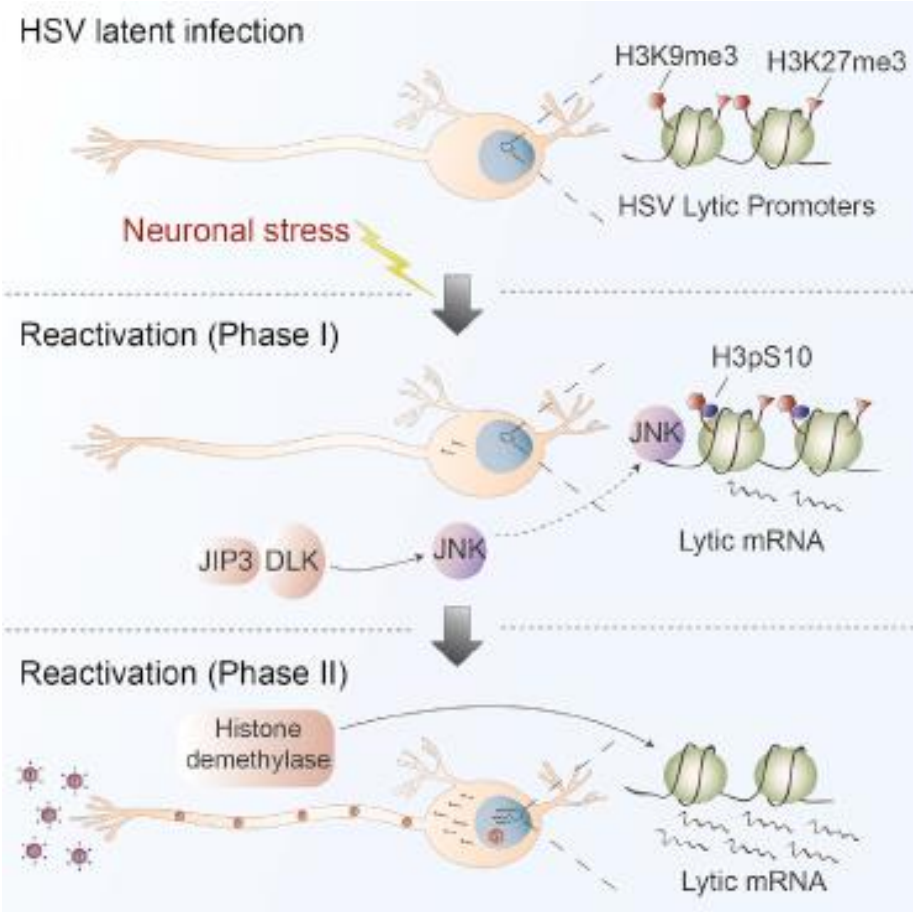
(a) The model is based on the proposed evidence that HCF1, VP16, or both are retained in the axons or cytoplasm and are not translocated to the neuronal nucleus.

(b) In the absence of HCF1 and VP16 viral DNA is progressively silenced by histones and histone-modifying enzymes and becomes encased in heterochromatin. The HDAC1/CoREST/LSD1/REST complex initiates the repression of the viral genome in peripheral neurons.

(c) Over time, LATs and viral miRNAs abound. Viral DNA equilibrates between total silencing and sporadic gene expression. The function of HDACs, LATs, and miRNAs is to maintain the DNA silence and to degrade small amounts of transcripts, whose expression may shift the dynamic state to activation of the viral genome.

(d) The stress of a specific type or of a high magnitude causes the entire genome to become derepressed and all viral genes are expressed at once. LATs and miRNAs are degraded.

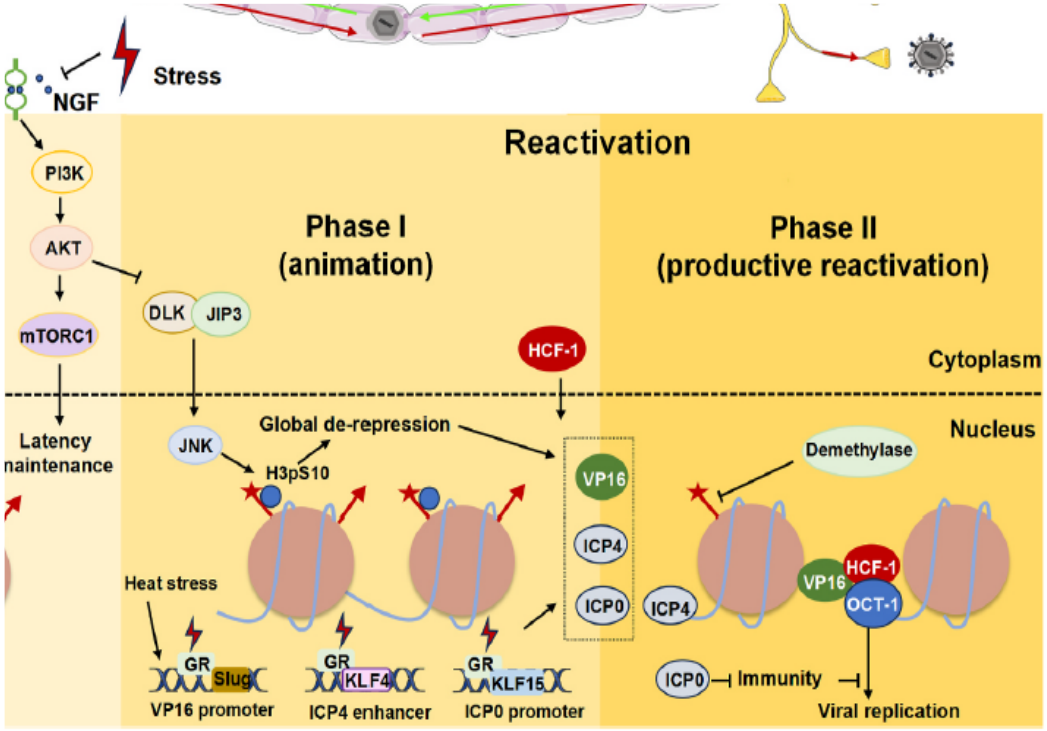
Stress Flips a Chromatin Switch to Wake Up Latent Virus



Initially, the episomal viral genome is coated with repressive chromatin marks such as H3K9me3 at promoters. As stress increases, the JNK signaling pathway is activated and phosphorylation on H3S10 allows transcription of viral genes without the removal of H3K9me3, passing the threshold for reactivation. With an increase in transcription of viral genes such as **VP16**, the reactivation enters Phase II with the re-writing of active marks, such as H3K9ac, on the viral genomes, viral replication, and progeny production.

Cliffe et al., 2015, Cell Host & Microbe 18, 649–658 December 9, 2015

(NGF) deprivation can trigger reactivation, indeed the continuous signaling through the PI3-K-AKT pathway triggered by NGF binding to the TrkA receptor tyrosine kinase is essential for latency maintenance



Phase 1 represents widespread de-repression of viral genes, Meanwhile, HCF-1 relocalizes from the cytoplasm of sensory neurons to the nucleus. The VP16 synthesized during phase 1 and the nuclear HCF-1 then stimulates phase 2, ultimately leading to the production of new viral DNA and infectious particles

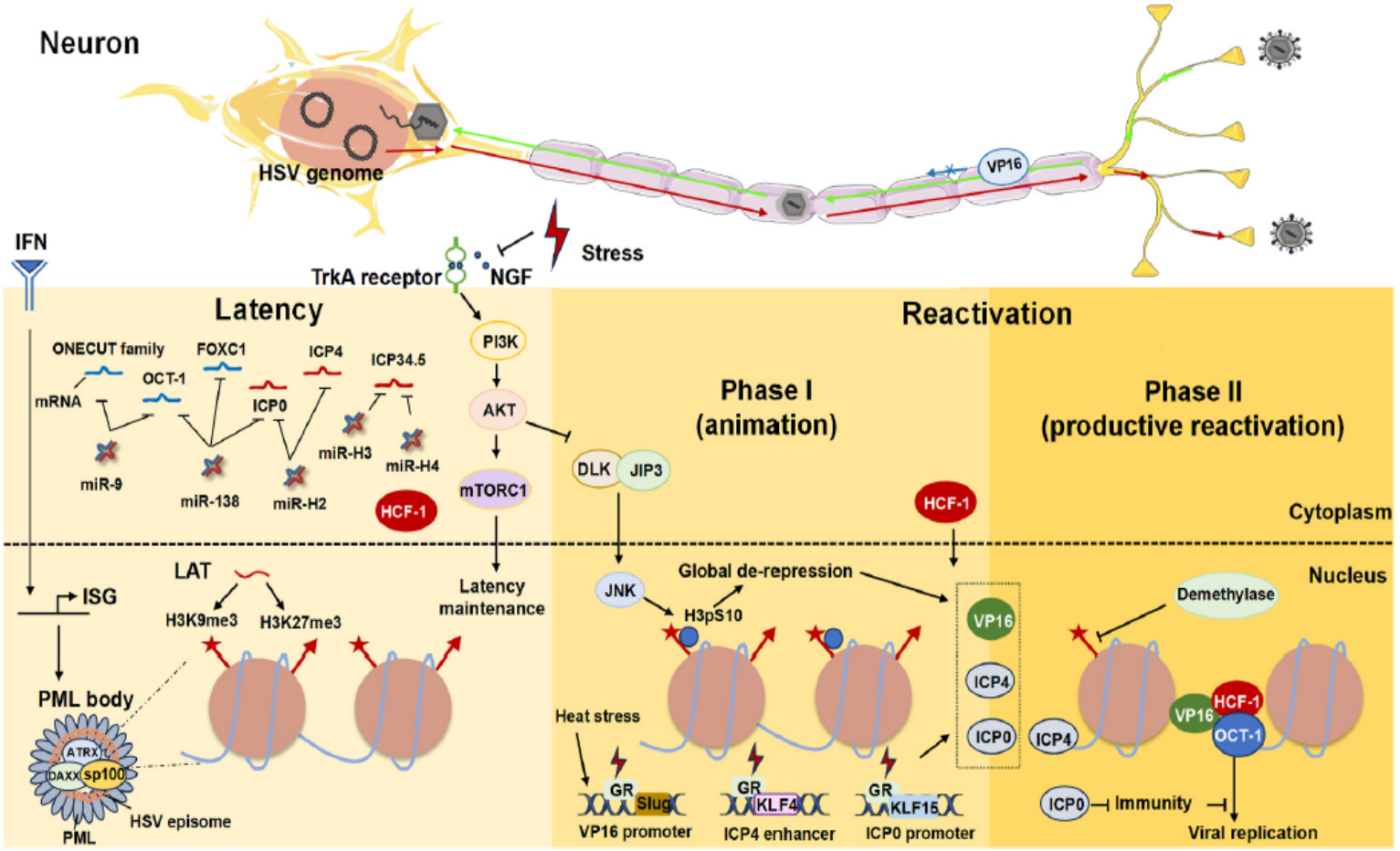


Fig. 3. Mechanisms of regulation of viral genes during latency and reactivation in neurons. At the top a sensory neuron is depicted with the movements of capsids indicated by arrows. A blocked arrow indicates dissociation of the VP16 during entry along the exon. The lower parts depict events during latency and the two phases of reactivation. During latency, highly expressed viral and host miRNAs targets viral and host genes important for the lytic cycle while LAT, interferon and PML-NB components promote formation of viral heterochromatin. The AKT-mTORC1 pathway is important for latency maintenance. Some stress signals deprive NGF resulting in the inhibition of the AKT pathway leading to phase 1 of reactivation through the DLK/JNK pathway. JNK causes a methyl/phosphor switch favorable for global de-repression. Meanwhile heat stress and hormones may specifically activate certain lytic gene promoters. The lytic proteins expressed during phase 1 then act together with host demethylases to stimulate phase 2. Some graphic elements were obtained from Server Medical Art (SMART) (https://smart.servier.com).

Da consultare

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The nuclear-cytoplasmic shuttling of virion host shutoff RNase is enabled by pUL47 and an embedded nuclear export signal and defines the sites of degradation of AU-rich and stable cellular mRNAs

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D.M. Knipe, P. Howley, D.E. Griffin, R.A. Lamb, M.A. Martin, V.R. Racaniello, B. Roizman (Eds.), New York, N.Y. (6th edition), Wolters Kluwer/Lippincott-Williams and Wilkins (2013), pp. 1823–1897

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HSV-1 gene expression from reactivated ganglia is disordered and concurrent with suppression of latency associated transcript and miRNAs

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Modulation of reactivation of latent herpes simplex virus 1 in ganglionic organ cultures by p300/CBP and STAT3

Proc. Natl. Acad. Sci. USA (2013), p. E2621 [Epub ahead of print Jun 20, 2013]

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HSV carrying WT REST establishes latency but reactivates only if the synthesis of REST is suppressed

Proc. Natl. Acad. Sci. USA, 110 (6) (2013), pp. 498–E506