Trends in Microbiology

CellPress

Review

Herpesviruses and Their Host Cells: A Successful Liaison

Barbara Adler,[1] Christine Sattler,[2] and Heiko Adler[3,*]

During a long history of coevolution, herpesviruses have reached a fine-tuned balance with their hosts, allowing them to successfully persist and spread to new hosts without causing too much damage. Only under certain circumstances, as in neonates or immunocompromised individuals, they may cause serious diseases. The delicate balance between herpesviruses and their hosts results from interactions of a great variety of viral and cellular factors which together shape the tropism for a particular host, tissue, or cell. Understanding these interactions will provide insight into the viral life cycle and cell biology in general. Moreover, it will also facilitate comprehension of herpesvirus pathogenesis, enabling the development of new strategies to combat herpesviruses in cases where they cause disease.

Herpesviruses: A Strategy of 'Travel and Hide'

Primary herpesvirus infection generally results in a productive infection which is subsequently limited by the host immune response, leaving behind latently infected cells which persist in the host [1]. Latency can be defined as carriage of the virus genome in the absence of virus production but the ability of the virus to reactivate and to re-enter the lytic cycle. During latency, only restricted sets of viral genes are expressed and the viral genomes mostly persist as episomes in the nuclei of infected cells. In some cases, viral genomes can also integrate into the host genome.

During their life cycle, herpesviruses usually infect different cell types in various tissues. Subclassification of herpesviruses is partially based on their cell and tissue **tropism** (see Glossary). α-Herpesviruses, such as herpes simplex virus (HSV) or varicella zoster virus (VZV), become latent in cells of the nervous system. β-Herpesviruses, including human cytomegalovirus (HCMV), are characterized by a very broad cell tropism when productively infecting cells and become latent in progenitors of the hematopoietic cell system. γ-Herpesviruses, such as Epstein–Barr virus (EBV) or Kaposi sarcoma-associated herpesvirus (KSHV), show a more restricted cell tropism and are characterized by their ability to transform latently infected cells and induce tumors in their infected hosts.

Usually, the portal of entry for a specific herpesvirus is not the site of latency. Thus, the incoming virus has to travel to the site of latency using either migrating cells as vehicles for dissemination or, in the case of α-herpesviruses, cell protrusions of nerve cells. Often, reactivating virus also uses the same routes back to ensure horizontal spread from productively infected cells. Understanding the interplay of viral and host cell factors during the different phases of the viral life cycle will not only provide insights into disease pathogenesis but also be the basis for the development of new antiviral drugs and herpesvirus-based vaccine or gene-therapy vectors. For these reasons, herpesvirus infection of different cell and tissue types is an area of intensive research using established and new techniques or screening methods (Table 1).

Trends

Herpesvirus host cells are defined by their susceptibility to productive or latent infection.

Herpesvirus host cells contribute to navigation of viruses through the infected host, either directly as vehicles or indirectly by shaping the glycoprotein content of viral envelopes.

Herpesviruses can manipulate their host cells by changing their differentiation status.

Herpesviruses stand out by a highly redundant equipment with regulatory proteins or noncoding RNAs. This redundancy stands in the way of clearing a herpesvirus infection.

The development of new antiherpesviral drugs or vaccines, and the application of herpesviruses as oncolytic agents, vaccine- or gene-therapy vectors depends on understanding interactions between viral and host cell factors.

[1]Max von Pettenkofer-Institute, Department of Virology, Ludwig-Maximilians-University Munich, Pettenkoferstraße 9A, D-80336 Munich, Germany
[2]Helmholtz Zentrum München – German Research Center for Environmental Health (GmbH), Institute of Lung Biology and Disease, Comprehensive Pneumology Center, Ingolstädter Landstr. 1, D-85764 Neuherberg, Germany
[3]Comprehensive Pneumology Center, Research Unit Lung Repair and Regeneration, Helmholtz Zentrum München – German Research Center for Environmental Health (GmbH), Marchioninistrasse 25, D-81377 Munich, Germany, and University Hospital Grosshadern, Ludwig-Maximilians-University, Munich, Germany[4]

Table 1. Methods and Techniques Used to Study the Interaction of Viral and Host Factors

Methods	Selected Applications	Refs
Computational biology approaches	Generation of integrated networks for virus (KSHV)–host interactions analysing sequence-based functional annotation and expression, RNAi- and experimental data	[58]
Classical loss of function/gain of function analyses	Determination of the role of THY-1 in HCMV infection via downregulation, antibody block, knockout and overexpression	[59]
Classical protein–protein interaction analyses like yeast two hybrid screens	Identification of cellular interaction partners of HSV-1 proteins by a genome-wide virus–host protein interaction screen	[60]
Integrative genome-wide approaches like high-throughput **RNAi** screens	Investigation of the functional role of cellular proteins in HSV-1 replication via siRNA-mediated depletion of host factors	[60,61]
Comprehensive **proteomic** analyses like **SILAC**-based quantitative proteomics	Global phosphorylation patterns in signaling pathways modulated by the EBV protein kinase BGLF4	[62]
Mass-Spec-based proteomics	Identification of an interaction between HSV-1 ICP0 and the cellular protein RanBP10 by **tandem affinity purification (TAP)** and **mass spectrometry**	[63]
Quantitative temporal viromics	Systematic quantitative analysis of temporal changes in host and viral proteins during HCMV infection by multiplexed tandem-mass-tag-based mass spectrometry	[64]
Subcellular fractionation combined with quantitative proteomics	Identification of Hsp70 isoforms as constituents of the KSHV replication and transcription compartments (RTCs)	[65]
Single-cell mass cytometry (CyTOF)	Analysis of concurrent changes in multiple host cell factors at the single cell level to follow phenotypic remodeling of T cells infected with VZV	[9]
High-resolution chromatin immunoprecipitation and deep sequencing (ChIP-Seq)	Analysis of protein–DNA interactions by combining chromatin immunoprecipitation with next-generation DNA sequencing to analyze the dynamic changes in CTCF and cohesin binding during KSHV reactivation	[28]
cre/*loxP*-system	Tracking MCMV and MHV-68 host cells *in vivo* by infecting mice cell type-specifically expressing Cre-recombinase with floxed reporter viruses or by infecting mice carrying floxed cellular genes with Cre-expressing viruses	[33,66]

Herpesvirus Tropism: Factors Influencing Herpesvirus Infection of Different Cell and Tissue Types

An important concept for understanding the mechanisms of infection of different cell and tissue types is the concept of tropism. This concept has been comprehensively reviewed by Heise and Virgin [2]. Briefly, tropism is the capacity of a virus to infect specific cells, tissues or species, and is determined by both susceptibility and permissiveness. A host cell is susceptible if it has the proper receptor(s), allowing the virus to enter the cell, and it is permissive if it allows viral replication, that is, it supports productive infection. Thus, tropism is determined by many factors of both the virus and the host. Although essential for infection, passage through the cellular membrane barrier is just the first step to successful infection. Several events that occur after binding and entry exert profound effects on the further progress of the infection. For example, the host cell is armed with molecules which can directly inhibit viral replication, induce antiviral innate

[4]Member of the German Center of Lung Research (DZL)

*Correspondence: h.adler@helmholtz-muenchen.de (H. Adler).

immune responses or cell death. Viruses can establish productive infection only when they find ways to counteract these lines of defense. The large genomes of herpesviruses, which code for hundreds of different genes, provide a highly redundant pool of proteins and noncoding RNAs to neutralize and overcome these barriers.

Virus–host cell interactions are extremely fine-tuned: differentiation or transformation of a cell may completely abrogate infection or generate new permissive host cells. The same is true for latency establishment. Differentiation may favor a latent stage or lead to reactivation from latency. One rare side-effect of this virus–host cell interaction is the capability of γ-herpesviruses to establish a latent state of infection favoring outgrowth of host cells and tumor development.

In summary, viral and cellular factors shape the course and outcome of an infection, including the decision whether the virus undergoes lytic replication or enters latency (Figure 1, Key Figure). These factors can either act in all cells or only in particular cell types or in cells in a particular condition (Table 2).

In the following sections, we discuss in more detail some selected examples of recent research of interactions between herpesviruses and host cellular factors.

Vehicles for Spread

Usually, host cells either become productively infected – and virus is spread to neighbouring cells – or a host-cell-dependent restriction of viral lytic genes and an induction of viral latent genes results in a nonproductive latent infection. There is also an in-between for herpesviruses. Herpesviruses use migrating host cells as vehicles to reach distant locations in their hosts. Usually, these cells are not overly permissive for the viral lytic cycle. Additionally, they are protected from virus-induced cell death and home to specific tissues. Virus replication in motile cells can, for example, be restricted by epigenetic regulation. Equine herpesvirus type 1 (EHV-1) replicates initially in epithelial cells but then infects monocytic cells for dissemination through the body. In these monocytic cells, EHV-1 replication is restricted and delayed, an effect which is mediated by histone deacetylases [3]. HCMV systemic spread is mediated by infection of circulating blood monocytes. To promote the survival of the infected monocytes, HCMV induces antiapoptotic proteins [4–6]. Additionally, it has been shown that infection activates a proinflammatory state which favors migration to organs where the monocytes differentiate to long-lived tissue macrophages which support the viral lytic cycle and thus establish infection in different organs [7]. Often, instead of infecting cells intrinsically programmed to migrate to sites of virus latency or reactivation, herpesvirus infection reprograms homing of host cells (Figure 2A): EBV, for example, infects naive B cells in the lymphoepithelium of the tonsils and transforms them by a specialized latency program to home to germinal centers, where they differentiate into memory B cells harboring a quiescent viral genome [8]. For VZV, it has been demonstrated that infection of T cells in tonsil lymphoid tissues remodels the surface of the infected T cells to enhance homing to skin sites of replication [9,10]. For murine cytomegalovirus (MCMV), it has been described that, in the peripheral blood of infected mice, only patrolling monocytes – but not inflammatory monocytes – are infected [11]. Under the aspect of a herpesvirus reprogramming, it will be of interest to find out whether MCMV programs monocytes to acquire a patrolling phenotype or whether it preferentially infects patrolling monocytes.

Herpesvirus Navigation

Herpesvirus spread can be the result of virus progeny randomly spreading infection from an infected cell to neighbouring host cells or transport through the bloodstream via infected cells, but it may also be the result of a host-cell-dependent virus modification addressing the virus progeny to specific host cells (Figure 2B). For EBV and HCMV, models of navigated virus spread have been studied in detail [12,13]. Both viruses code for alternative gH/gL glycoprotein

Glossary

Cre/*loxP*-system: a site-specific recombination system, consisting of the enzyme Cre recombinase and a pair of short target sequences, called *loxP* sites. Cre catalyzes DNA recombination between the *loxP* sites, and recombination between a pair of directly repeated *loxP* sites results in the deletion of the intervening DNA (the so-called floxed sequence).
Mass spectrometry: an analytical technique that ionizes chemical species and sorts the ions based on their mass-to-charge ratio.
Proteomics: large-scale study of proteins.
RNA interference (RNAi): a biological process in which RNA molecules inhibit gene expression.
SILAC: stable isotope labeling with amino acids in cell culture. The technique is based on mass spectrometry that detects differences in protein abundance among samples using nonradioactive isotopic labeling. Cells are differentially labeled by growing them in medium either containing normal amino acids or amino acids labeled with stable, nonradioactive heavy isotopes. Metabolic incorporation of the amino acids into proteins results in a mass shift of the corresponding peptides. When two samples are combined, the relative protein abundance is reflected by the ratio of peak intensities in the mass spectrum.
Single-cell mass cytometry: a combination of flow cytometry and mass spectrometry, also called cytometry by time-of-flight (CyTOF).
Tandem affinity purification (TAP): a purification technique for studying protein–protein interactions, applying a protein with a designed tag, the TAP tag.
Tropism: the capacity of a virus to infect or damage specific cells, tissues, or species.

Key Figure

The Interplay of Viral and Host Factors Shapes the Course and Outcome of Infection

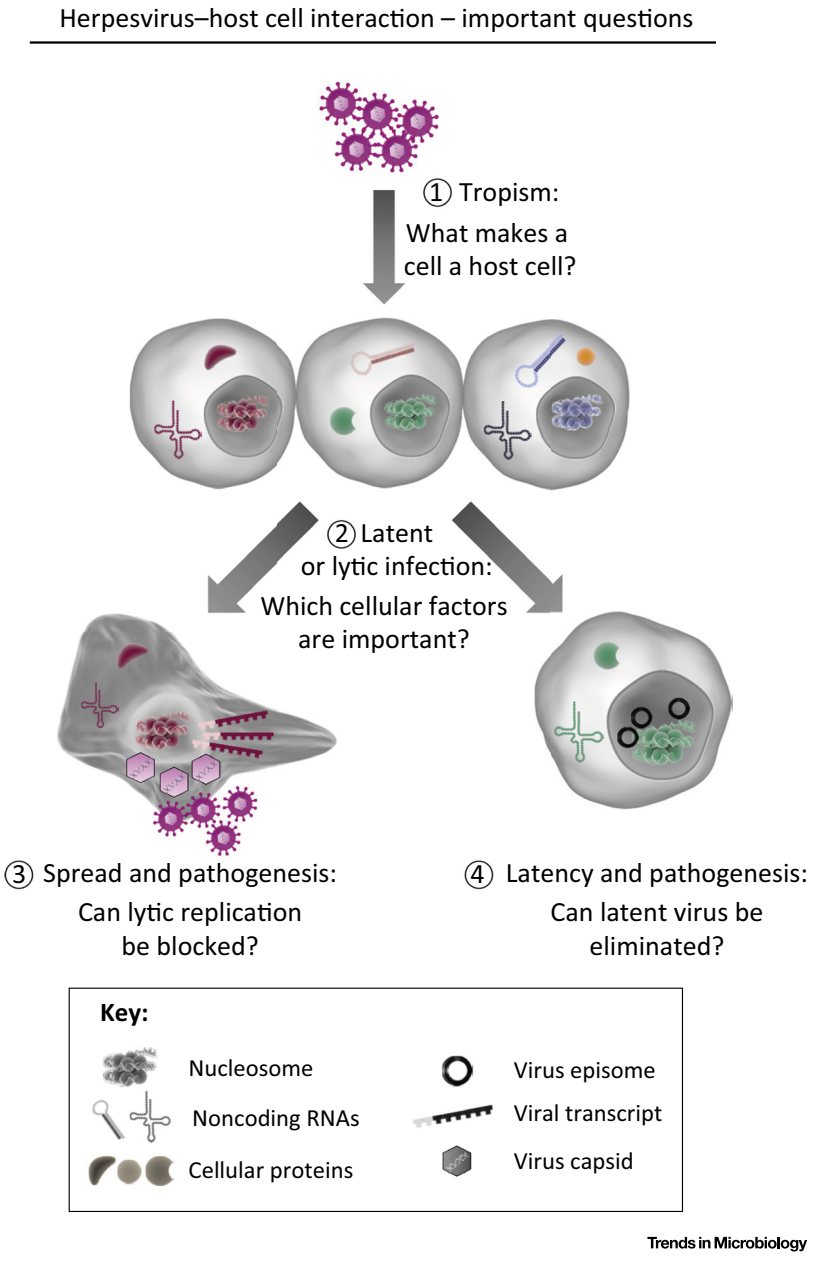

Figure 1. A great variety of viral and cellular (host) factors influence the infection of cells or tissues during the different phases of the viral life cycle. They may facilitate or prevent infection and influence the decision whether the virus replicates lytically or enters latency.

Table 2. Factors Influencing Cell and Tissue Tropism of Herpesviruses

Viral factors	Refs
Envelope glycoproteins	[67–69]
Noncoding RNAs (including miRNAs)	[44,45,47]
Origins of lytic replication (oriLyts)	[51]
Antiapoptotic genes	[70]
Genes counteracting host cell defense mechanisms (immune evasion genes)	[71–78]
Genes influencing cell cycle or proliferation	[79,80]
Genes interfering with epigenetic silencing	[63,81]
Cellular (host) factors	**Refs**
Glycocalyx, receptors, and signaling coreceptors	[67,82]
Noncoding RNAs (including miRNAs)	[41,42,48]
Proteins interacting with oriLyts	[51]
Interferons and other cytokines	[30,32–34]
Sensors of viral infection	[23,83,84]
Cell cycle proteins	[62,85,86]
Proteins regulating apoptosis	[87–90]
Transcription factors	[91]
DNA damage response proteins	[5]
Proteins involved in epigenetic gene regulation including chromatin assembly, histone modifications, and DNA methylation	[21–28,63,92–98]
Autophagy and xenophagy	[99–101]
Ubiquitination and NEDDylation	[102–105]
Chaperones	[65]
Post-translational modification proteins	[106,107]
Proteins regulating translation	[108,109]
Proteins regulating cell differentiation	[36–38]
Proteins regulating energy homeostasis	[110]
Oncogenes, including p53, SV40 large T antigen, and H-Ras	[111–114]
Structural proteins of virological synapses	[115]

complexes promoting recognition of specific host cell receptors. EBV particles released from epithelial cells predominantly contain trimeric gH/gL/gp42 complexes which recognize the B cell surface protein HLA class II and are directed toward infection of B cells. In B cells, the viral protein gp42 is retained and degraded, resulting in virus particles predominantly containing gp42-negative dimeric gH/gL complexes which promote entry into epithelial cells via integrin receptors [14]. Based on this, it has been proposed that latently infected memory B cells migrate to the lymphoepithelium of tonsils where they can differentiate to plasma cells and activate the EBV lytic cycle. Virus is then directed to neighbouring epithelial cells which produce B-cell-tropic virus for horizontal transmission to B cells in a new host [8,12]. Indeed, virus found in saliva of EBV-infected humans is gH/gL/gp42-rich [15].

HCMV codes for a gH/gL/gO complex which drives infection of cells expressing platelet-derived growth factor receptor alpha (PDGFR-α) [16], and for a gH/gL/pUL(128,130,131A) complex which drives infection of most PDGFR-α-negative cells through a still unknown receptor. In cell culture, fibroblasts infected with HCMV release virions with high or low amounts of gH/gL/pUL

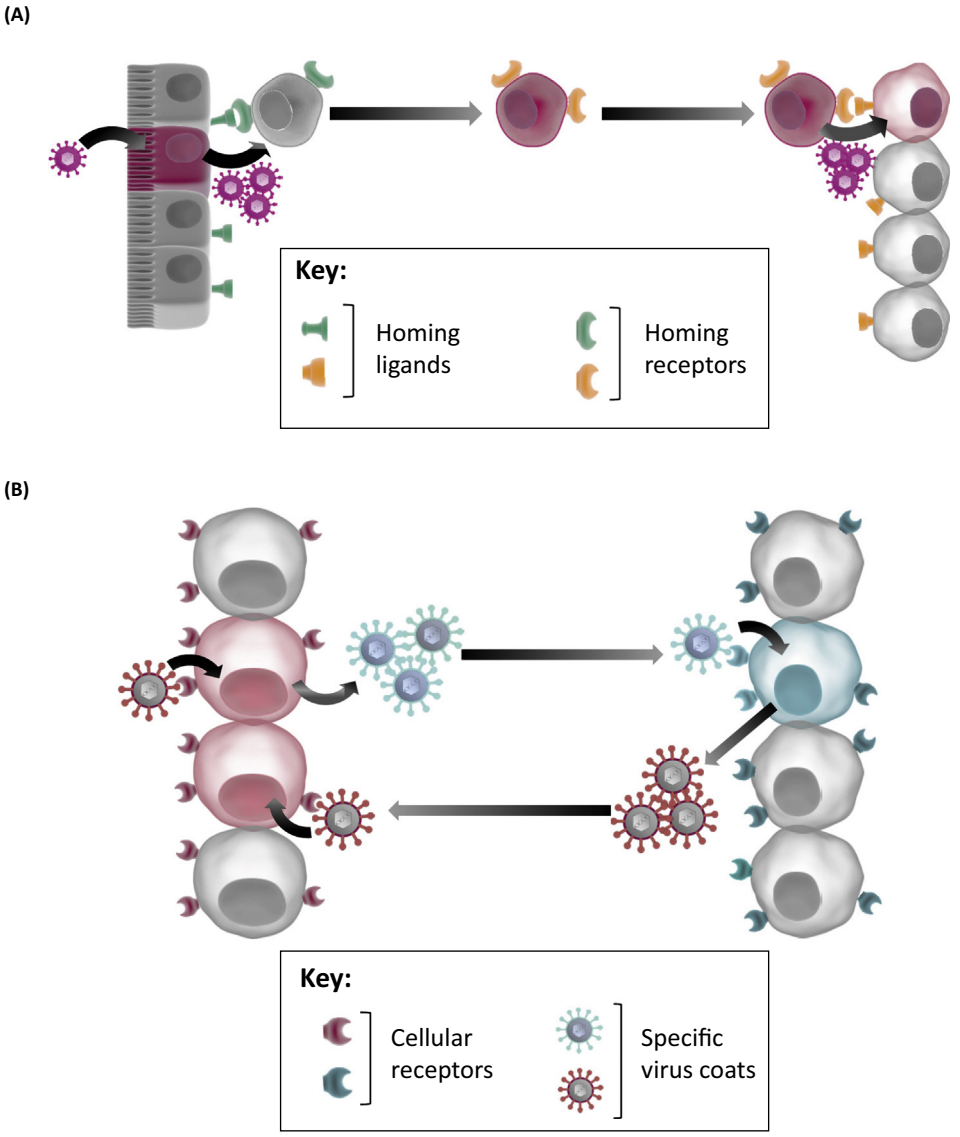

Figure 2. Infection Is Navigated by Virus Differentiating Its Host Cell and by Cells Shaping the Virus Surface. (A) Migrating cells which home to infected tissues (left side) become infected in the vicinity of their target tissues. Infection changes the cells' differentiation state, which drives homing of these newly infected cells to different target tissues (right side). (B) Different host cells differently shape the virion coat and thereby switch the viral cell tropism. Virus particles released from one cell type are thus targeted to a second cell type and vice versa.

(128,130,131A), a particle mixture which can readily infect all host cells of HCMV. Endothelial cells, in contrast, only release virions with low amounts of gH/gL/pUL(128,130,131A), which do not infect endothelial cells [13]. gH/gL/pUL(128,130,131A)-dependent cell-associated spread to neighbouring cells is possible from fibroblasts and endothelial cells. Thus, HCMV host cells shape the tropism of the virus which has to bridge long distances such as during horizontal transmission. Application of new techniques allows the ability to follow herpesvirus navigation *in vivo*. Direct cell contacts of infected and uninfected cells can be visualized by multiphoton intravital microscopy. To track the origin of virus traveling long distance, cell type-dependent labelling has been performed by infecting transgenic mice expressing cell type-specific cre with

viruses carrying lox-P-flanked reporter genes [17,18]. Virus navigation by employing the host cell machinery to shape the receptor-binding virion surface appears an efficient strategy of some herpesviruses to achieve directed, and if necessary, long-distance traveling to cells which are hideouts for latent virus, virus production sites, or first target cells in new hosts.

Latent or Lytic Cycle: The Host Cell Decides

After primary infection, the lifelong herpesvirus infection is characterized by virus persistence in a latent stage interrupted by sporadic periods of lytic replication. During lytic replication, infectious progeny is produced whereas, during latency, the viral genome remains 'silent' inside the infected host cell. The decision between lytic replication and latency is predominantly influenced by host cell factors.

Epigenetic Mechanisms

Epigenetic gene regulation is usually defined as heritable changes in the activity and expression of genes, caused by chromosomal changes, including DNA methylation, histone modifications, and nucleosome positioning, which do not alter the DNA sequence [19]. It has emerged only recently that both viral and host cell factors that are involved in epigenetic gene regulation play an important role in the regulation of herpesvirus latency. Consequently, the epigenetic regulation of infections by all classes of herpesviruses is an area of intensive research [20–23]. Epigenetic regulation often is cell-type-specific, and thus, may also influence the decision between lytic or latent infection in a cell-type-specific manner. Here, we highlight some recent findings in the field: HSV undergoes lytic infection in epithelial cells while it establishes a latent infection in sensory neurons [23]. It has been shown that, in dividing cells like epithelial cells or fibroblasts, HSV DNA associates with histones within a few hours whereas in nondividing neurons, the association with histones and Polycomb proteins, a family of proteins involved in modification and remodeling of chromatin, takes much longer [24]. It has been speculated that this might be due to smaller pools of histones in nondividing neurons [23]. Differences in Polycomb proteins between subtypes of neurons are also discussed as factors influencing the anatomical preferences of HSV-1 for the orofacial region and of HSV-2 for the genital region [25]. In hematopoietic stem cells, HCMV latency is achieved by recruitment of KRAB-associated protein 1 (KAP1) together with heterochromatin protein 1 (HP1) and the histone methyltransferase SET domain, bifurcated 1 (SETDB1), to the viral genome which finally results in transcriptional silencing [26]. After reactivation, KAP1 remains associated with the viral genome but its heterochromatin-inducing activity is counteracted by phosphorylation mediated by the mammalian target of rapamycin (mTOR). CCCTC binding factor (CTCF) is a cellular DNA-binding protein important for the regulation of genomic chromatin boundaries, control of transcription, and long-range DNA interactions [27]. Together with the cohesin component Rad21, CTCF acts as a potent restriction factor for KSHV replication [28]. Knockdown of these proteins by siRNAs increased the production of infectious virus. As CTCF and cohesin contribute to cell-type-specific chromatin organization and function [29], it would be highly interesting to see whether it affects virus infection in an equally cell-type-specific manner.

Cytokines

Already earlier studies have demonstrated a cell-type-specific regulation of gammaherpesvirus latency by interferon-gamma (IFN-γ) [30]. While macrophages were responsive to IFN-γ-mediated suppression of MHV-68 reactivation, B cells were not. Interleukin-21 (IL-21) is produced by T follicular helper cells and is important for the germinal center reaction and for the generation of long-lived plasma cells. MHV-68-infected B cells pass through the germinal center reaction to become latently infected [31]. Consequently, it has recently been shown that IL-21 is a critical factor for the efficient establishment of latency in B cells [32]. In IL-21R-deficient mice, that is, in the absence of IL-21 signaling, fewer infected spleen cells gained access to the germinal center B cell population, and the infected cells showed reduced expansion and reduced reactivation. It is likely

that this effect is specific for B cells and does not affect other latently infected cells like macrophages, although this has not yet been tested. New data demonstrate cell-type-dependent effects also for type I interferons (IFN-I). Using Mx1-cre mice to tag floxed MHV-68 genomes in IFN-I responding cells, Tan *et al.* [33] demonstrated that the impact of IFN-I on viral replication was strongly cell-type-dependent. In epithelial cells, MHV-68 infection induced only a weak IFN-I response, allowing virus replication. In contrast, in macrophages and B cells, it induced a strong IFN-I response which suppressed replication in macrophages, but due to counteracting viral evasion mechanisms, not in B cells. Consequently, a shift from lytically infected macrophages to latently infected B cells was promoted. For HSV-1, new data have shown that neuronal IFN-I signaling is required to suppress viral replication and protect from pathogenesis [34]. IFN-driven innate responses in neural tissues were much more important than IFN-I signaling in the immune system and in peripheral tissues. However, whether this neuronal IFN-I signaling is also involved in the regulation of HSV latency is currently not clear and needs further investigation.

Cell Differentiation

The stage of differentiation or maturation of a cell may facilitate or prevent efficient infection. For HCMV, the most established link between cell differentiation and the decision to go latent or lytic is myeloid differentiation [35]. Progenitors of the myeloid lineage are hosts for latent HCMV, and differentiation to macrophages or dendritic cells results in reactivation of the virus. Recently, Berger *et al.* investigated the transition from restriction of HCMV infection in human embryonic stem cells (hESC) toward susceptibility in hESC-derived neural precursors [36]. Using protocols for controlled induction of differentiation of hESC into neural precursors, they discovered PDGFR-α as a determinant of HCMV susceptibility. For MHV-68, the transcription factors Blimp-1 and interferon regulatory factor 4 (IRF4) are both required for maintenance of MHV-68 latency and for virus reactivation [37,38]. Both factors promote plasma cell differentiation which is a prerequisite for reactivation of MHV-68 from B cells. Efficient reactivation, in turn, serves to renew the viral latency reservoirs.

Latent or Lytic Cycle: The Role of Noncoding RNAs

Noncoding RNAs (ncRNAs), including microRNAs (miRNAs), are produced by both host cells and herpesviruses. miRNAs are approximately 22-nucleotide long ncRNAs, which silence gene expression post-transcriptionally by binding to the 3′ untranslated regions of target mRNAs [39]. Both cellular and viral miRNAs are predestined to cell-type-specifically influence infection of cells and tissues: (i) they can be expressed in a cell or tissue-type-specific manner or only at a particular time of the viral life cycle (spatiotemporal expression), and (ii) the abundance of target mRNAs might alter their effect [40]. For example, a neuron-specific host miRNA, miR-138, has been shown to repress expression of ICP0, the HSV-1 transactivator of lytic gene expression [41]. Thereby, miR-138 acts as a neuron-specific factor to promote host cell survival and HSV latency by repressing viral lytic replication. miR-155, a host miRNA mainly expressed in hematopoietic cells, is critical for reactivation of murine gammaherpesvirus 68 (MHV-68) from latency [42]. A KSHV long ncRNA called polyadenylated nuclear (PAN) RNA is essential for virion production [43]. It binds relocalized poly(A)-binding protein C1 (PABPC1) in KSHV-infected B cells and is required for the expression of late viral genes [44]. A highly abundant ncRNA of EBV, EBV-encoded RNA 2 (EBER2), localizes to specific regions of the viral genome and facilitates, through RNA–RNA interactions, the binding of the host transcription factor PAX5 [45]. This interaction seems to be necessary for efficient lytic replication since knockdown of EBER2 decreased lytic replication. A long ncRNA of HCMV (RNA4.9) represses transcription of lytic genes by interacting with components of the Polycomb repression complex and the major immediate early promoter, thereby most likely regulating latency in CD14-positive monocytes and CD34-positive hematopoietic cells [46]. Herpesvirus saimiri (HVS) U-rich RNA 1 (HSUR 1), an ncRNA expressed in HVS-infected T cells, was found to degrade host miR-27 in a sequence-specific and binding-dependent manner [47]. Murine cytomegalovirus

(MCMV) also targets miR-27 for degradation by a similar antisense RNA-based mechanism [43], and this activity has been shown to be important for efficient virus replication *in vivo* [48].

Redundancy Can Overcome Host Restrictions

Herpesvirus genomes code for a large number of viral proteins which, in the majority of cases, are neither structural proteins nor needed for viral replication in cultured cells. Through interaction with cellular proteins, they can influence the biology of their host cells, for example, as immunoregulatory proteins which interfere with the innate or adaptive antiviral immune response, as antiapoptotic proteins or regulators of cellular antiapoptotic proteins, or as regulators of cellular transcription factors. Often, herpesviruses express a redundant pool of proteins with identical or overlapping functions. An example of redundant genomic structures are herpesviruses with more than one lytic origin of replication (oriLyt). OriLyts are defined sites on the viral genome where herpesvirus lytic DNA replication is initiated. While some herpesviruses, for example HCMV, have a single oriLyt, others such as MHV-68 have multiple oriLyts [49,50]. In a recent study, the role of the oriLyts of MHV-68 was examined [51]. The working hypothesis was that the presence of two oriLyts can overcome cell-type-specific restrictions of lytic replication by cellular proteins. Loss of either of the two oriLyts was well tolerated in some cell types while it resulted in reduced fitness in others. DNA-affinity purification in combination with **mass spectrometry** revealed distinct sets of cellular proteins interacting with the right oriLyt in different cell types. There were 193 proteins exclusively detected in extracts of TCMK-1 epithelial cells, and 37 proteins exclusively detected in extracts of NIH3T3 fibroblasts. For example, Hexim1 was found only in TCMK-1 cells while Rbbp4 was found only in NIH3T3 cells [51]. Functional assays showed that, in the absence of the left oriLyt, Hexim1 exerted an inhibitory effect on lytic replication in TCMK-1 but not in NIH3T3 cells. Consequently, downregulation of Hexim1 in TCMK-1 cells enhanced replication while upregulation of Hexim1 in NIH3T3 cells inhibited virus replication of this mutant. Thus, depending on the cellular host, overexpression and downregulation of Hexim1 either reduced or enhanced the replication of the mutant lacking the left oriLyt, indicating that Hexim1 is a rate-limiting cellular protein in a situation where only one oriLyt is present. However, the virus can overcome this restriction by the presence of two oriLyts, indicating that two oriLyts are an advantage for optimal virus fitness. Similarly, Rbbp4 supported

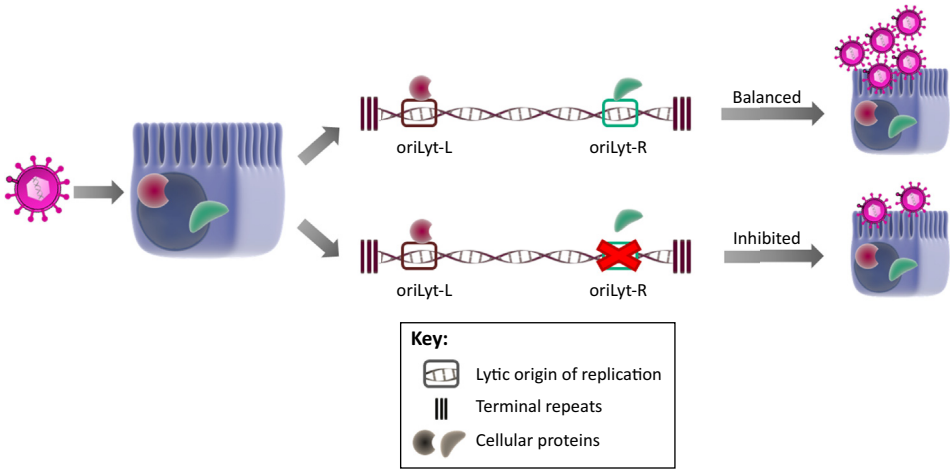

Figure 3. Redundancy Can Overcome Host Restrictions: Two Lytic Origins of Replication Are Better Than One. Cellular proteins may support (green) or inhibit (red) lytic replication originating at a given oriLyt. Thus, in situations where only one oriLyt is present (as indicated by the red x), cellular proteins may be rate limiting. The virus can overcome potential negative effects by the presence of two oriLyts.

lytic replication originating at the right oriLyt while it strongly inhibited lytic replication originating at the left oriLyt. Thus, Rbbp4, like Hexim1, was shown to be a rate-limiting cellular protein in situations where only one oriLyt is present. Again, the virus can overcome potential negative effects by the presence of two oriLyts. Taken together, these data suggested that the presence of multiple oriLyts enables herpesviruses which carry more than one oriLyt to efficiently replicate in the presence of varying sets of activating or inhibitory cellular proteins, thus assuring optimal fitness in different cell or tissue types (Figure 3).

Concluding Remarks and Future Perspectives

Investigation of factors influencing herpesvirus infection of different cells and tissues will continue to be an area of intense research. Understanding virus–host cell interactions will result in new antiviral drugs directed against both viral and cellular targets, and, as virus research has always done, reveal new insights into cell biology. To fight herpesvirus infections, eliminating latent virus would be an invaluable milestone (see Outstanding Questions). Thus, strategies to either inhibit reactivation from latency ('locking in latency') [20,23] or, the opposite approach, to induce reactivation and lytic replication ('forcing out of latency') are currently discussed [20,26]. Locking in latency might be valuable when reactivation from latency has the potential to harm the host, for example, reactivation of HSV in the peripheral or central nervous system [23]. Forcing out of latency by transient activation of HCMV lytic gene expression has been shown to enable killing of latently infected, that is, normally immunologically invisible cells, by cytotoxic T lymphocytes [52], and has been suggested as a way to purge latently infected cells from HCMV-positive transplants by *ex vivo* treatment prior to engraftment [26]. The better we understand herpesvirus–host cell interactions, the more accurate our risk–benefit evaluations will become when applying herpesviruses as oncolytic agents [53,54], and vectors for vaccination or tumor therapy [55,56]. Finally, any knowledge on herpesvirus–host cell interactions will contribute to the elaboration of the concept that herpesviruses, as constituents of the so-called virome, may shape the host immune response and even be beneficial for the host [57].

Outstanding Questions

Is redundancy or latency, or a combination of both, the secret of success of a herpesvirus infection?

It is possible to treat a herpesvirus infection, but will it be possible to cure it and also remove latent virus?

Are latent herpesviruses key players of human or animal viromes?

Acknowledgments

The authors acknowledge all of their colleagues for their contributions. Our review is neither comprehensive nor complete, and we apologize to those whose work could not be cited. Work in the lab of B.A. was funded by grants from the Deutsche Forschungsgemeinschaft (AD131/3-3 and AD131/4-1), and work in the lab of H.A. was funded by grants from the Bundesministerium fuer Bildung und Forschung (NGFNplus, PIM-01GS0802-3), from the Wilhelm Sander-Stiftung (2009.046.1+2), and by intramural funding for Environmental Health Projects of the Helmholtz Zentrum München – German Research Center for Environmental Health (GmbH).

References

1. Roizman, B. and Pellett, P.E. (2001) The family Herpesviridae: a brief introduction. In *Fields – Virology* (4th edn) (Knipe, D.M. *et al.*, eds), pp. 2381–2397, Lippincott Williams & Wilkins
2. Heise, M.T. and Virgin, H.W. (2013) Pathogenesis of viral infection. In *Fields – Virology* (6th edn) (Knipe, D.M. *et al.*, eds), pp. 254–285, Lippincott Williams & Wilkins
3. Laval, K. *et al.* (2015) Equine herpesvirus type 1 replication is delayed in CD172a+ monocytic cells and controlled by histone deacetylases. *J. Gen. Virol.* 96, 118–130
4. Reeves, M.B. *et al.* (2012) Human cytomegalovirus activation of ERK and myeloid cell leukemia-1 protein correlates with survival of latently infected cells. *Proc. Natl. Acad. Sci. U.S.A.* 109, 588–593
5. Collins-McMillen, D. *et al.* (2015) Human cytomegalovirus promotes survival of infected monocytes via a distinct temporal regulation of cellular Bcl-2 family proteins. *J. Virol.* 90, 2356–2371
6. Peppenelli, M.A. *et al.* (2016) Human cytomegalovirus stimulates the synthesis of select Akt-dependent antiapoptotic proteins during viral entry to promote survival of infected monocytes. *J. Virol.* 90, 3138–3147
7. Stevenson, E.V. *et al.* (2014) HCMV reprogramming of infected monocyte survival and differentiation: a Goldilocks phenomenon. *Viruses* 6, 782–807
8. Thorley-Lawson, D.A. (2015) EBV Persistence – introducing the virus. *Curr. Top. Microbiol. Immunol.* 390, 151–209
9. Sen, N. *et al.* (2015) Single cell mass cytometry reveals remodeling of human T cell phenotypes by varicella zoster virus. *Methods* 90, 85–94
10. Sen, N. and Arvin, A.M. (2016) Dissecting the molecular mechanisms of the tropism of varicella-zoster virus for human T cells. *J. Virol.* 90, 3284–3287
11. Daley-Bauer, L.P. *et al.* (2014) Cytomegalovirus hijacks CX3CR1(hi) patrolling monocytes as immune-privileged vehicles for dissemination in mice. *Cell Host Microbe* 15, 351–362
12. Hutt-Fletcher, L.M. (2007) Epstein–Barr virus entry. *J. Virol.* 81, 7825–7832
13. Scrivano, L. *et al.* (2011) HCMV spread and cell tropism are determined by distinct virus populations. *PLoS. Pathog.* 7, e1001256

14. Chesnokova, L.S. *et al.* (2009) Fusion of epithelial cells by Epstein–Barr virus proteins is triggered by binding of viral glycoproteins gHgL to integrins alphavbeta6 or alphavbeta8. *Proc. Natl. Acad. Sci. U.S.A.* 106, 20464–20469
15. Jiang, R. *et al.* (2006) Epstein–Barr virus shed in saliva is high in B-cell-tropic glycoprotein gp42. *J. Virol.* 80, 7281–7283
16. Kabanova, A. *et al.* (2016) Platelet-derived growth factor-alpha receptor is the cellular receptor for human cytomegalovirus gHgLgO trimer. *Nat. Microbiol.* 1, 16082
17. Sacher, T. *et al.* (2008) The major virus-producing cell type during murine cytomegalovirus infection, the hepatocyte, is not the source of virus dissemination in the host. *Cell Host Microbe* 3, 263–272
18. Sacher, T. *et al.* (2011) Shedding light on the elusive role of endothelial cells in cytomegalovirus dissemination. *PLoS. Pathog.* 7, e1002366
19. Paluch, B.E. *et al.* (2016) Epigenetics: A primer for clinicians. *Blood Rev.* 30, 285–295
20. Lieberman, P.M. (2016) Epigenetics and genetics of viral latency. *Cell Host Microbe* 19, 619–628
21. Tempera, I. and Lieberman, P.M. (2014) Epigenetic regulation of EBV persistence and oncogenesis. *Semin. Cancer Biol.* 26, 22–29
22. Kumar, A. and Herbein, G. (2014) Epigenetic regulation of human cytomegalovirus latency: an update. *Epigenomics* 6, 533–546
23. Knipe, D.M. (2015) Nuclear sensing of viral DNA, epigenetic regulation of herpes simplex virus infection, and innate immunity. *Virology* 479-480, 153–159
24. Cliffe, A.R. *et al.* (2013) Kinetics of facultative heterochromatin and polycomb group protein association with the herpes simplex viral genome during establishment of latent infection. *MBio* 4, e00590–e612
25. Watson, Z. *et al.* (2013) Role of polycomb proteins in regulating HSV-1 latency. *Viruses* 5, 1740–1757
26. Rauwel, B. *et al.* (2015) Release of human cytomegalovirus from latency by a KAP1/TRIM28 phosphorylation switch. *eLife* 4, e06068
27. Pentland, I. and Parish, J.L. (2015) Targeting CTCF to control virus gene expression: a common theme amongst diverse DNA viruses. *Viruses* 7, 3574–3585
28. Li, D.J. *et al.* (2014) CTCF and Rad21 act as host cell restriction factors for Kaposi's sarcoma-associated herpesvirus (KSHV) lytic replication by modulating viral gene transcription. *PLoS Pathog.* 10, e1003880
29. Hou, C. *et al.* (2010) Cell type specificity of chromatin organization mediated by CTCF and cohesin. *Proc. Natl. Acad. Sci. U.S.A.* 107, 3651–3656
30. Steed, A. *et al.* (2007) Gamma interferon blocks gammaherpesvirus reactivation from latency in a cell type-specific manner. *J. Virol.* 81, 6134–6140
31. Frederico, B. *et al.* (2014) A murid gamma-herpesvirus exploits normal splenic immune communication routes for systemic spread. *Cell Host Microbe* 15, 457–470
32. Collins, C.M. and Speck, S.H. (2015) Interleukin 21 signaling in B cells is required for efficient establishment of murine gammaherpesvirus latency. *PLoS Pathog.* 11, e1004831
33. Tan, C.S. *et al.* (2016) Type I interferons direct gammaherpesvirus host colonization. *PLoS Pathog.* 12, e1005654
34. Rosato, P.C. and Leib, D.A. (2015) Neuronal interferon signaling is required for protection against herpes simplex virus replication and pathogenesis. *PLoS Pathog.* 11, e1005028
35. Sinclair, J.H. and Reeves, M.B. (2013) Human cytomegalovirus manipulation of latently infected cells. *Viruses* 5, 2803–2824
36. Berger, A.A. *et al.* (2015) Transition toward human cytomegalovirus susceptibility in early human embryonic stem cell-derived neural precursors. *J. Virol.* 89, 11159–11164
37. Matar, C.G. *et al.* (2014) Murine gammaherpesvirus 68 reactivation from B cells requires IRF4 but not XBP-1. *J. Virol.* 88, 11600–11610
38. Siegel, A.M. *et al.* (2010) Blimp-1-dependent plasma cell differentiation is required for efficient maintenance of murine gammaherpesvirus latency and antiviral antibody responses. *J. Virol.* 84, 674–685
39. Eulalio, A. *et al.* (2008) Getting to the root of miRNA-mediated gene silencing. *Cell* 132, 9–14
40. Arvey, A. *et al.* (2010) Target mRNA abundance dilutes microRNA and siRNA activity. *Mol. Syst. Biol.* 6, 363
41. Pan, D. *et al.* (2014) A neuron-specific host microRNA targets herpes simplex virus-1 ICP0 expression and promotes latency. *Cell Host Microbe* 15, 446–456
42. Crepeau, R.L. *et al.* (2016) miR-155 is necessary for efficient gammaherpesvirus reactivation from latency, but not for latency establishment. *J. Virol.* 90, 7811–7821
43. Tycowski, K.T. *et al.* (2015) Viral noncoding RNAs: more surprises. *Genes Dev.* 29, 567–584
44. Borah, S. *et al.* (2011) A viral nuclear noncoding RNA binds re-localized poly(A) binding protein and is required for late KSHV gene expression. *PLoS Pathog.* 7, e1002300
45. Lee, N. *et al.* (2015) EBV noncoding RNA binds nascent RNA to drive host PAX5 to viral DNA. *Cell* 160, 607–618
46. Rossetto, C.C. *et al.* (2013) Cis and trans acting factors involved in human cytomegalovirus experimental and natural latent infection of CD14 (+) monocytes and CD34 (+) cells. *PLoS Pathog.* 9, e1003366
47. Cazalla, D. *et al.* (2010) Down-regulation of a host microRNA by a Herpesvirus saimiri noncoding RNA. *Science* 328, 1563–1566
48. Marcinowski, L. *et al.* (2012) Degradation of cellular mir-27 by a novel, highly abundant viral transcript is important for efficient virus replication in vivo. *PLoS Pathog.* 8, e1002510
49. Roizman, B. *et al.* (2007) Herpes simplex viruses. In *Fields – Virology* (5th edn) (Knipe, D.M. *et al.*, eds), pp. 2501–2601, Lippincott Williams & Wilkins
50. Mocarski, J. (2007) Comparative analysis of herpesvirus-common proteins. In *Human Herpesviruses: Biology, Therapy, and Immunoprophylaxis* (Arvin, A.M. *et al.*, eds), p. 44, Cambridge University Press
51. Sattler, C. *et al.* (2016) Multiple lytic origins of replication are required for optimal gammaherpesvirus fitness *in vitro* and *in vivo*. *PLoS Pathog.* 12, e1005510
52. Krishna, B.A. *et al.* (2016) Transient activation of human cytomegalovirus lytic gene expression during latency allows cytotoxic T cell killing of latently infected cells. *Sci. Rep.* 6, 24674
53. Menotti, L. *et al.* (2012) The molecular basis of herpesviruses as oncolytic agents. *Curr. Pharm. Biotechnol.* 13, 1795–1803
54. Campadelli-Fiume, G. *et al.* (2011) Rethinking herpes simplex virus: the way to oncolytic agents. *Rev. Med. Virol.* 21, 213–226
55. Barouch, D.H. and Picker, L.J. (2014) Novel vaccine vectors for HIV-1. *Nat. Rev. Microbiol.* 12, 765–771
56. Glorioso, J.C. (2014) Herpes simplex viral vectors: late bloomers with big potential. *Hum. Gene Ther.* 25, 83–91
57. Virgin, H.W. (2014) The virome in mammalian physiology and disease. *Cell* 157, 142–150
58. Zaman, A. *et al.* (2013) Kaposi's sarcoma: a computational approach through protein-protein interaction and gene regulatory networks analysis. *Virus Genes* 46, 242–254
59. Li, Q. *et al.* (2015) THY-1 cell surface antigen (CD90) has an important role in the initial stage of human cytomegalovirus Infection. *PLoS Pathog.* 11, e1004999
60. Griffiths, S.J. *et al.* (2013) A systematic analysis of host factors reveals a Med23-interferon-lambda regulatory axis against herpes simplex virus type 1 replication. *PLoS Pathog.* 9, e1003514
61. Griffiths, S.J. (2013) Screening for host proteins with pro- and antiviral activity using high-throughput RNAi. *Methods Mol. Biol.* 1064, 71–90
62. Li, R. *et al.* (2015) Phosphoproteomic profiling reveals Epstein-Barr virus protein kinase integration of DNA damage response and mitotic signaling. *PLoS Pathog.* 11, e1005346
63. Sato, Y. *et al.* (2016) Cellular transcriptional coactivator RanBP10 and herpes simplex virus 1 ICP0 interact and synergistically promote viral gene expression and replication. *J. Virol.* 90, 3173–3186
64. Weekes, M.P. *et al.* (2014) Quantitative temporal viromics: an approach to investigate host–pathogen interaction. *Cell* 157, 1460–1472

65. Baquero-Perez, B. and Whitehouse, A. (2015) Hsp70 isoforms are essential for the formation of Kaposi's sarcoma-associated herpesvirus replication and transcription compartments. *PLoS Pathog.* 11, e1005274
66. Sacher, T. *et al.* (2008) Conditional gene expression systems to study herpesvirus biology in vivo. *Med. Microbiol. Immunol.* 197, 269–276
67. Campadelli-Fiume, G. *et al.* (2012) Viral and cellular contributions to herpes simplex virus entry into the cell. *Curr. Opin. Virol.* 2, 28–36
68. Gardner, T.J. and Tortorella, D. (2016) Virion glycoprotein-mediated immune evasion by human cytomegalovirus: a sticky virus makes a slick getaway. *Microbiol. Mol. Biol. Rev.* 80, 663–677
69. Heldwein, E.E. (2016) gH/gL supercomplexes at early stages of herpesvirus entry. *Curr. Opin. Virol.* 18, 1–8
70. Banerjee, S. *et al.* (2016) The modulation of apoptotic pathways by gammaherpesviruses. *Front. Microbiol.* 7, 585
71. Laura, M.V. *et al.* (2015) KSHV latent protein LANA2 inhibits sumo2 modification of p53. *Cell Cycle* 14, 277–282
72. Sifford, J.M. *et al.* (2015) Murine gammaherpesvirus 68 LANA and SOX homologs counteract ATM-driven p53 activity during lytic viral replication. *J. Virol.* 90, 2571–2585
73. Taylor, K.E. *et al.* (2014) Novel roles of cytoplasmic ICP0: proteasome-independent functions of the RING finger are required to block interferon-stimulated gene production but not to promote viral replication. *J. Virol.* 88, 8091–8101
74. Zenner, H.L. *et al.* (2013) Herpes simplex virus 1 counteracts tetherin restriction via its virion host shutoff activity. *J. Virol.* 87, 13115–13123
75. Zhang, G. *et al.* (2016) Cytoplasmic isoforms of Kaposi sarcoma herpesvirus LANA recruit and antagonize the innate immune DNA sensor cGAS. *Proc. Natl. Acad. Sci. U.S.A.* 113, E1034–E1043
76. Kumari, P. *et al.* (2015) Herpesviruses: interfering innate immunity by targeting viral sensing and interferon pathways. *Rev. Med. Virol.* 25, 187–201
77. Sun, C. *et al.* (2015) Evasion of innate cytosolic DNA sensing by a gammaherpesvirus facilitates establishment of latent infection. *J. Immunol.* 194, 1819–1831
78. Feng, P. *et al.* (2013) Evasion of adaptive and innate immune response mechanisms by gamma-herpesviruses. *Curr. Opin. Virol.* 3, 285–295
79. Nascimento, R. *et al.* (2012) Virus manipulation of cell cycle. *Protoplasma* 249, 519–528
80. Wei, F. *et al.* (2016) Cell cycle regulatory functions of the KSHV oncoprotein LANA. *Front. Microbiol.* 7, 334
81. Johnson, K.E. *et al.* (2014) IFI16 restricts HSV-1 replication by accumulating on the hsv-1 genome, repressing HSV-1 gene expression, and directly or indirectly modulating histone modifications. *PLoS Pathog.* 10, e1004503
82. Chan, G. *et al.* (2012) Human cytomegalovirus induction of a unique signalsome during viral entry into monocytes mediates distinct functional changes: a strategy for viral dissemination. *J. Leukoc. Biol.* 92, 743–752
83. Chan, Y.K. and Gack, M.U. (2016) Viral evasion of intracellular DNA and RNA sensing. *Nat. Rev. Microbiol.* 14, 360–373
84. Luecke, S. and Paludan, S.R. (2015) Innate recognition of alphaherpesvirus DNA. *Adv. Virus Res.* 92, 63–100
85. Caffarelli, N. *et al.* (2013) Cyclin A degradation by primate cytomegalovirus protein pUL21a counters its innate restriction of virus replication. *PLoS Pathog.* 9, e1003825
86. Spector, D.H. (2015) Human cytomegalovirus riding the cell cycle. *Med. Microbiol. Immunol.* 204, 409–419
87. Austgen, K. *et al.* (2012) Multiple defects, including premature apoptosis, prevent Kaposi's sarcoma-associated herpesvirus replication in murine cells. *J. Virol.* 86, 1877–1882
88. Jurak, I. and Brune, W. (2006) Induction of apoptosis limits cytomegalovirus cross-species infection. *EMBO J.* 25, 2634–2642
89. Keyes, L.R. *et al.* (2012) Cyclophilin A is required for efficient human cytomegalovirus DNA replication and reactivation. *J. Gen. Virol.* 93, 722–732
90. Lee, S.H. *et al.* (2012) BclAF1 restriction factor is neutralized by proteasomal degradation and microRNA repression during human cytomegalovirus infection. *Proc. Natl. Acad. Sci. U.S.A.* 109, 9575–9580
91. Murata, T. *et al.* (2013) Contribution of myocyte enhancer factor 2 family transcription factors to BZLF1 expression in Epstein–Barr virus reactivation from latency. *J. Virol.* 87, 10148–10162
92. Zhou, G. *et al.* (2013) The role of the CoREST/REST repressor complex in herpes simplex virus 1 productive infection and in latency. *Viruses* 5, 1208–1218
93. Roizman, B. *et al.* (2011) Checkpoints in productive and latent infections with herpes simplex virus 1: conceptualization of the issues. *J. Neurovirol.* 17, 512–517
94. Gunther, T. *et al.* (2014) Influence of ND10 components on epigenetic determinants of early KSHV latency establishment. *PLoS Pathog.* 10, e1004274
95. Wagenknecht, N. *et al.* (2015) Contribution of the major ND10 proteins PML, hDaxx and Sp100 to the regulation of human cytomegalovirus latency and lytic replication in the monocytic cell line THP-1. *Viruses* 7, 2884–2907
96. Kim, Y.E. and Ahn, J.H. (2015) Positive role of promyelocytic leukemia protein in type I interferon response and its regulation by human cytomegalovirus. *PLoS Pathog.* 11, e1004785
97. Sun, R. *et al.* (2014) Kaposi's sarcoma-associated herpesvirus-encoded LANA interacts with host KAP1 to facilitate establishment of viral latency. *J. Virol.* 88, 7331–7344
98. Moser, J.M. *et al.* (2005) Role of B-cell proliferation in the establishment of gammaherpesvirus latency. *J. Virol.* 79, 9480–9491
99. Rosato, P.C. and Leib, D.A. (2015) Neurons versus herpes simplex virus: the innate immune interactions that contribute to a host-pathogen standoff. *Future Virol.* 10, 699–714
100. Park, S. *et al.* (2016) Autophagy genes enhance murine gammaherpesvirus 68 reactivation from latency by preventing virus-induced systemic inflammation. *Cell Host Microbe* 19, 91–101
101. McFadden, K. *et al.* (2016) Metabolic stress is a barrier to Epstein–Barr virus-mediated B-cell immortalization. *Proc. Natl. Acad. Sci. U.S.A.* 113, E782–E790
102. Huffmaster, N.J. *et al.* (2015) Dynamic ubiquitination drives herpesvirus neuroinvasion. *Proc. Natl. Acad. Sci. U.S.A.* 112, 12818–12823
103. Greene, W. *et al.* (2012) The ubiquitin/proteasome system mediates entry and endosomal trafficking of Kaposi's sarcoma-associated herpesvirus in endothelial cells. *PLoS Pathog.* 8, e1002703
104. Sato, Y. *et al.* (2016) Ubiquitin-specific protease 9X in host cells interacts with herpes simplex virus 1 ICP0. *J. Vet. Med. Sci.* 78, 405–410
105. Hughes, D.J. *et al.* (2015) NEDDylation is essential for Kaposi's sarcoma-associated herpesvirus latency and lytic reactivation and represents a novel anti-KSHV target. *PLoS Pathog.* 11, e1004771
106. Sloan, E. *et al.* (2015) Analysis of the SUMO2 Proteome during HSV-1 Infection. *PLoS Pathog.* 11, e1005059
107. Brown, J.R. *et al.* (2016) SUMO ligase protein inhibitor of activated STAT1 (PIAS1) is a constituent promyelocytic leukemia nuclear body protein that contributes to the intrinsic antiviral immune response to herpes simplex virus 1. *J. Virol.* 90, 5939–5952
108. McKinney, C. *et al.* (2013) A new role for the cellular PABP repressor Paip2 as an innate restriction factor capable of limiting productive cytomegalovirus replication. *Genes Dev.* 27, 1809–1820
109. Strang, B.L. *et al.* (2012) Host cell nucleolin is required to maintain the architecture of human cytomegalovirus replication compartments. *MBio* 3, e00301–e00311
110. Cheng, F. *et al.* (2016) Suppression of Kaposi's sarcoma-associated herpesvirus infection and replication by 5'-AMP-activated protein kinase. *J. Virol.* 90, 6515–6525
111. Xu, S. *et al.* (2016) Expression of oncogenic alleles induces multiple blocks to human cytomegalovirus infection. *J. Virol.* 90, 4346–4356

112. Leidal, A.M. *et al.* (2012) Evasion of oncogene-induced senescence by gammaherpesviruses. *Curr. Opin. Virol.* 2, 748–754

113. Maruzuru, Y. *et al.* (2013) Roles of p53 in herpes simplex virus 1 replication. *J. Virol.* 87, 9323–9332

114. Maruzuru, Y. *et al.* (2016) p53 is a host cell regulator during herpes simplex encephalitis. *J. Virol.* 90, 6738–6745

115. Liu, Y. *et al.* (2015) Tetherin restricts HSV-2 release and is counteracted by multiple viral glycoproteins. *Virology* 475, 96–109