FEBS Letters

FEBS PRESS science publishing by scientists

IN A NUTSHELL

Murine adenoviruses: tools for studying adenovirus pathogenesis in a natural host

Silvio Hemmi[1] and Katherine R. Spindler[2]

1 Institute of Molecular Life Sciences, University of Zürich, Switzerland
2 Department of Microbiology and Immunology, University of Michigan, Ann Arbor, MI, USA

Correspondence
K. R. Spindler, Department of Microbiology and Immunology, 1150 W Med Ctr Dr., University of Michigan, Ann Arbor MI 48109-5620, USA
Tel: +1-734-615-2727
E-mail: krspin@umich.edu

(Received 4 October 2019, revised 11 November 2019, accepted 22 November 2019, available online 6 December 2019)

doi:10.1002/1873-3468.13699

Edited by Urs Greber

Small laboratory animals are powerful models for investigating *in vivo* viral pathogenesis of a number of viruses. For adenoviruses (AdVs), however, species-specificity poses limitations to studying human adenoviruses (HAdVs) in mice and other small laboratory animals. Thus, this review covers work on naturally occurring mouse AdVs, primarily mouse adenovirus type 1 (MAdV-1), a member of the species *Murine mastadenovirus A*. Molecular genetics, virus life cycle, cell and tissue tropism, interactions with the host immune response, persistence, and host genetics of susceptibility are described. A brief discussion of MAdV-2 (member of species *Murine mastadenovirus B*) and MAdV-3 (member of species *Murine mastadenovirus C*) is included. We report the use of MAdVs in the development of vectors and vaccines.

Keywords: adenovirus vaccines; adenovirus vectors; alpha-defensins; host susceptibility to viruses; immunoproteasome; Ly6; mouse adenovirus; PKR; viral pathogenesis

Study of animal adenoviruses is of importance both because of the fundamental knowledge gained from comparative biology with human adenoviruses (HAdVs) and because of the ability to study the pathogenesis of an adenovirus (AdV) in its natural host using experimental infections, which is not possible for HAdVs. Moreover, AdV species-specificity limits the use of small animal models for the study of human AdV pathogenesis. The ease and relative low cost of studying mice in the laboratory is augmented by the wealth of genetically distinct inbred strains of *Mus musculus* and the ability to make transgenic mice to test the functional importance of mouse genes for virus infection. These are coupled with a rich supply of immunological reagents for studying the mouse host response.

Mouse adenovirus MAdV-1 (Ad-FL) [1] was among the first nonprimate adenoviruses identified in the 1950s and 1960s, along with canine hepatitis virus, CAdV-1 [2], fowl AdV-1 (CELO)[3], and bovine AdV [4]. MAdV-1 has a tropism for endothelial cells and cells of the monocyte/macrophage lineage, and it also infects astrocytes [5–7]. MAdV-1 infects tissues throughout the mouse, and the highest levels of virus are found in the brain, spinal cord, and spleen after intraperitoneal infection [5,6,8]. MAdV-1 causes encephalitis in susceptible mice [6,8,9], myocarditis [10–12], and respiratory infection [13,14]. A second mouse AdV, MAdV-2 (K87), was isolated from feces of laboratory mice in Japan in 1966 [15]. Both in cultured cells and in mice, MAdV-2 has a tropism for cells of the intestinal tract, but it does not cause apparent disease in mice [15–17]. MAdV-1 and -2 were isolated from laboratory mice, whereas a third type of mouse AdV, MAdV-3, was isolated from a liver from

Abbreviations
AdV, adenovirus; CAR, coxsackie-adenovirus receptor; Chr, Chromosome; CNS, central nervous system; CR, conserved region; Cul, Cullin; dpi, days postinfection; E, early; EnAd, enadenotucirev; GlcNAc, N-acetylglucosamine; IFN, interferon; INR, initiator; L, late; MAdV, mouse adenovirus; MHC, major histocompatibility complex; MLP, major late promoter; MMP, matrix metalloproteinase; NK, natural killer; PGE2, prostaglandin E2; QTL, quantitative trait locus; RGD, Arg-Gly-Asp; VA, virus-associated.

a striped field mouse, *Apodemus agrarius* [18]. MAdV-3 has a primary tropism for cardiac tissue, and it is present at high levels in myocardium, not in the brain, and at lower levels in most other organs [18]. MAdV-3 is genetically more similar to MAdV-1 than to MAdV-2.

The MAdV types discovered to date seem to have low significance as natural pathogens, and only a limited number of prevalence studies in the wild [19–21] and in commercial or research colonies [22–26] have been performed. These studies indicate a moderate serologic prevalence of MAdV-2 with considerable site-to-site variability. More recently, virome profiling studies have added more insights and provided evidence for new rodent adenoviruses [27–29]. The 'murine' adenoviruses that have been characterized to date do not infect infant rats [30], and thus this review covers only mouse adenoviruses.

MAdVs have similarities and differences compared to HAdVs that will be discussed below. We include comparisons of molecular genetics and gene expression, cell and tissue tropism, and viral persistence. The MAdV-mouse host system enables studies of viral pathogenesis that have no direct comparison to HAdV studies. We also discuss the use of MAdVs for vaccines and gene delivery vectors. Readers are also directed to previous reviews of mouse adenoviruses [31,32] for additional historical perspectives and further details.

Mouse adenovirus type 1

Isolation, physical, and molecular genetic properties

MAdV-1 was isolated by Hartley and Rowe when they were establishing Friend mouse leukemia virus in culture, and they subsequently isolated it from Swiss mice in a mouse colony, designating it 'M.Ad. virus strain FL' [1]. The virus has physical and serologic properties like HAdVs [1,31], and infected mice transmit the virus to uninoculated cage contacts, but there is no apparent transmission through the air or *via* bedding from cages of infected mice [1,33]. MAdV-1 has subsequently been studied in cell culture and mice. One isolate of MAdV-1 is in the American Type Culture Collection (Cat. no. VR550), deposited by Steven Larsen; we and others from 1981 onward have used an isolate obtained directly from Dr. Larsen, referred to as 'standard'. These two isolates have minor molecular and pathogenetic differences [34], and it is not known which strain (if either) was used prior to 1981.

MAdV-1 has a 30 944 bp double-stranded genome with 93 nt inverted terminal repeats [34–36]. The complete sequence is available as NC_000942.1 [37]. Another entry with in silico-derived annotations of the same sequence is AC_000012.1 [38]; it has some predicted genes not in agreement with published experimental evidence (e.g., in silico E1A annotation does not match transcription mapping and cDNA sequencing data) [31].

In broad terms, the genome structure of MAdV-1 is similar to that of HAdV-5. It encodes genes with sequence and functional similarity to HAdV early (E) regions 1–4, a major late promoter (MLP) with a tripartite leader, and late (L) genes encoding the major virion proteins and proteins involved in morphogenesis [reviewed in Ref. [31]]. The gene arrangement is like that of HAdVs. A terminal protein is associated with the 5′ end of each end of the genome [36]. In fine details, there are some differences between HAdVs and MAdV-1 in gene expression and function. For example, MAdV-1 does not encode virus-associated RNA (VA RNA) [37], which in HAdV infections counteracts the host protein kinase R (PKR) antiviral response [39].

Instead of two major isoforms of the E1A protein found in HAdV-5, MAdV-1 only has one 200 aa protein, corresponding to the larger (289 aa, '13S') HAdV-5 protein [40]. Although its overall sequence similarity to HAdV E1A proteins is low, it has about 40% similarity to conserved regions 1 (CR1), CR2, and CR3. MAdV-1 E1A has functional similarity to HAdV E1A; it interacts with cellular proteins pRb and p107 *via* its CR2 domain and is involved in the regulation of cell proliferation of quiescent cells [41]. MAdV-1 E1A, like HAdV E1A, interacts with a component of the mediator transcriptional complex, Sur2, now known as Med23, through the E1A CR3 domain [42]. This finding enabled the demonstration that Med23 is required for efficient replication of adenoviruses, which was unable to be tested directly using HAdVs.

Transcription mapping has not been done for MAdV-1 E2, but the predicted proteins have good sequence similarity with the three HAdV E2 proteins, which are involved in viral DNA synthesis. The DNA polymerase, DNA binding protein, and pTP, which is involved in protein priming of DNA synthesis, have 33–57% sequence similarity to HAdV proteins [37,43].

MAdV-1 E3 is distinct from the E3 regions of HAdVs. There are three mRNAs encoded that share 5′ and 3′ termini but differ in splicing of the third exon, such that the three predicted proteins share amino terminal sequences but have different carboxy-terminal domains [44]. Only one of the MAdV-1 E3

proteins is detected in wild-type virus-infected cells, the E3 gp11K protein [45]. As is true of the HAdVs, the E3 region of MAdV-1 is involved in viral pathogenesis [45,46]. MAdV-1 E3 functions are not directly comparable to those of HAdVs, however. For example, one of the first viral immune evasion strategies identified for any virus is downregulation of class I major histocompatibility complex (MHC) antigens on the surface of infected cells by HAdV-2 [47]. This function of HAdV E3 gp19K is not mimicked by MAdV-1 infection [48]. The MAdV-1 E3 proteins do not have sequence similarity to other known proteins.

Unlike the E4 mRNAs of HAdVs, which are 5′ and 3′ coterminal, the MAdV-1 E4 mRNAs have different 5′ ends but share 3′ ends [49]. Predicted proteins have sequence similarity to HAdV-2 E4orf6 (34K protein) (MAdV-1 protein originally identified as orf a/b), E4orf3 protein (MAdV-1 orf a/c), E4orf2, and E4orf6/7 (MAdV-1 orf d) [34,49]. Little has been reported about function of the MAdV-1 E4 proteins. However, when E4orf6 is introduced into human cells by transfection, it coimmunoprecipitates with Cullin 2 (Cul2), Elongin C, and MAdV-1 E1B 55K protein [50]. Importantly, mouse p53 is degraded when it and MAdV-1 E4orf6 and E1B 55K protein are coexpressed, indicating that E4orf6 (together with E1B 55K) functions as a ubiquitin ligase.

The MLP of MAdV-1 has features like the HAdV-1 MLP, including a TATA box and an inverted CAAT box, but it lacks a sequence that binds the transcription factor USF and it lacks the initiator (INR) sequence [51]. At late times, the MLP is functional in MAdV-1-infected cells, as demonstrated by RNAse protection assays of infected cell RNAs. MAdV-1 late mRNAs have a typical tripartite leader structure (C. Coombes, J. Boeke, L. Gralinski, and K. Spindler, unpublished).

Receptor studies indicate that the mouse coxsackie-adenovirus receptor (CAR) homolog of the human CAR used by many HAdVs as the attachment receptor is not used by MAdV-1 [52]. Transfection of mouse CAR into Chinese hamster ovary cells does not increase attachment of virus relative to untransfected cells, and purified HAdV-5 fiber knob does not compete with MAdV-1 binding. MAdV-1 penton base protein does not have an Arg-Gly-Asp (RGD) sequence that in HAdVs is important for interaction with entry receptors. However, the MAdV-1 fiber knob has an RGD motif in a sequence of the knob that represents an insertion relative to HAdV-1 fiber sequences [53]. This RGD sequence plays a role in MAdV-1 infection of cultured fibroblasts. Competition studies indicate that the fiber knob is the viral attachment protein. Cell surface heparan sulfate is important for infection, and α_v integrin acts as a primary receptor for MAdV-1. MAdV-1 associates with factor X and factor IX, but this does not result in increased cellular attachment, unlike the case for HAdV [54].

MAdV-1 pathogenesis—Tropism, adaptive immune responses

In contrast to HAdVs, which have an epithelial tropism, MAdV-1 primarily infects endothelial cells and monocytes/macrophages, and astrocytes can also be infected [5–8,55,56]. The virus causes a pantropic infection; high levels of virus found in the central nervous system (CNS) lead to increased permeability of the blood–brain barrier, accompanied by altered tight junction-protein expression and encephalitis [5,6,8,56–58]. MAdV-1 CNS infection is characterized by viral brain loads that correlate with disease severity and induction of matrix metalloproteinase (MMP) activity [7,59]. Enzyme activity of MMP2 and MMP9, which are induced in brains during microbial infection or neurological disease, is increased in mice and cultured cells upon MAdV-1 infection. This is not accompanied by increases in MMP mRNA levels, indicating that the MMP activation is a post-transcriptional event. MAdV-1 does infect epithelial cells and infects the respiratory tract when inoculated intranasally, and neonatal mice are more susceptible than adults to respiratory infection [13,14]. MAdV-1 also causes myocarditis that is accompanied by myocyte and endothelial necrosis when inoculated intraperitoneally or intranasally [10–12]. Many mouse primary cell types, cell strains, and established cell lines can support MAdV-1 replication *in vitro*, including fibroblasts (3T6, 3T12 and L929) [35,60], endothelial cells (MBMEC) [57], preadipocyte cells (3T3-L1) [61], epithelial cells (LA-4, MLE-12 and MLE-15; J.B. Weinberg, unpublished), cardiac myocytes [12] macrophages/monocytes [55], and tumor cells such as lung adenoma (LA-4), renal adenocarcinoma (RAG) [48], and rectal carcinoma (CMT-93) cells [62].

Mice infected with MAdV-1 develop adaptive immune responses, both cell-mediated and humoral. These responses are important for host survival, because RAG-1$^{-/-}$ mice and SCID mice (deficient in T cells and B cells) are more susceptible to infection than wild-type mice [56,63]. Virus-specific cytotoxic T cells can be detected 4 days postinfection (dpi), peak at 10 dpi, and then decrease in numbers [64–67]. Studies with immunodeficient mice have shown that T cells are required for long-term survival of infection and contribute to immunopathology during the acute phase

of infection [68]. CD8 T cells, but not CD4 T cells, are required for efficient clearance of MAdV-1 from the lungs and for virus-induced inflammation and weight loss [69]. These effects of CD8 T cells are not mediated exclusively by IFN-γ, perforin, or Fas/FasL [69,70]. MAdV-1-induced humoral responses have been studied in outbred and inbred mice. Sublethal infection of outbred mice leads to high neutralizing antibody titers 2 weeks after infection that increase for a year before declining [71]. Infection of inbred mice leads to B cell proliferation in the spleen 10 dpi, stimulating predominantly IgG2a antibody [72–74]. Using B-cell-deficient mice, Moore et al. showed that B cells help prevent disseminated MAdV-1 infection, and they produce T-cell-independent antiviral IgM [63]. In studies using a model in which mice are made immunodeficient by cyclophosphamide, humoral immunity is important for protection from MAdV-1 disseminated infection, and survival correlates with presence of MAdV-1-specific antibodies [75].

MAdV-1 pathogenesis—Innate immune responses

The innate immune response to MAdV-1 infection is dependent on immune cells and cytokines. Macrophages are infected by the virus [5,55]. Peritoneal and splenic macrophages from infected mice express viral early and late genes, and peritoneal macrophages produce infectious virus [5,55]. However, the level of virus production is low, suggesting that replication is inefficient or only some cells can produce virus. Bone marrow-derived macrophages and CD11c+ cells (predominantly dendritic cells) infected ex vivo express MAdV-1 mRNAs and proteins [55]. Depletion of macrophages by clodronate liposome treatment increases MAdV-1 infection severity [55]. Macrophages are thus targets and effectors in MAdV-1 infection. In contrast, depletion of mice of their natural killer (NK) cells either genetically or biochemically does not alter MAdV-1 brain viral loads, indicating that in contrast to other viral infections, NK cells are not required for control of MAdV-1 infections in the brain [76].

Interferons (IFNs) are major antiviral cytokines. *In vitro*, wild-type MAdV-1 infections are resistant to the effects of type I and type II IFN [77]. In contrast, E1A mutant infections are sensitive to both types of IFN, and expression of E1A in the absence of other viral gene products rescues vesicular stomatitis virus from the effects of type I IFN. The results indicate that MAdV-1 uses E1A to inhibit expression of IFN-stimulated genes. IFNγ is not a major antiviral cytokine in MAdV-1 respiratory infection or myocarditis [12,78]. However, it is necessary for cardiac inflammation induced by intranasal infection of neonatal mice [12]. The virus replicates in both neonates and adult mice, but only neonates develop myocarditis, and there is higher IFN-γ expression in neonates compared to adults. Thus IFN-γ is a proinflammatory mediator in AdV-induced myocarditis. IFN-γ is important for the induction of the immunoproteasome, an inducible form of the proteasome involved in protein degradation and generation of peptides for MHC class I presentation [79]. Intranasal inoculation of MAdV-1 results in significantly increased immunoproteasome activity in the lung and heart compared to uninfected mice [80], and this is dependent on IFNγ. The immunoproteasome is important for survival of neonatal mice infected with MAdV-1 [60].

The role of interleukin 1β (IL-1β) in MAdV-1 encephalitis was examined in mice lacking the IL-1 receptor. IL-1β is a proinflammatory cytokine that contributes to inflammation in the CNS. Surprisingly, the lack of IL-1 signaling in the mutant mice results in increased mortality and inflammation during MAdV-1 infection compared to control mice; this is accompanied by an increase in transcription of type I IFN-stimulated genes [81]. Thus IL-1β, although proinflammatory, protects mice from some of the pathogenic effects of MAdV-1 CNS infections. A similar result is found in herpes simplex virus 1-induced encephalitis, in which IL-1β acts synergistically with tumor necrosis factor alpha [82].

IL-17 is another proinflammatory cytokine; it is produced by a subset of helper T cells known as Th17 cells. Pulmonary infection of mice by MAdV-1 results in increased IL-17 mRNA and protein compared to uninfected mice [83]. IL-17 knockout mice have less recruitment of neutrophils to the lung than wild-type mice, but peak viral loads, clearance of virus, and protective immunity do not differ. This indicates that the Th17 responses during respiratory infection are not required for viral control or for pulmonary inflammation.

Prostaglandin E_2 (PGE_2) is a lipid mediator that increases during many viral infections, and it can have effects on many immune cell types. Production of PGE_2 increases during MAdV-1 respiratory infection, promoting production of a variety of cytokines [84]. However, mice deficient in PGE_2 production do not differ from wt mice in virus replication, virus-induced lung inflammation, or protective immunity.

Alpha-defensins are short antimicrobial peptides with antibacterial activity [85]. These peptides can also neutralize viruses, and α-defensin binding to HAdV-5 blocks *in vitro* infection by stabilizing the capsid and

preventing uncoating of the virion [86–88]. *In vivo* evidence that defensins are a protective host response for AdV infection comes from study of MAdV-1 infection of mice lacking functional enteric α-defensin processing [62]. When mice lacking functional α-defensins in their small intestine are orally infected with MAdV-1, there is a dose-dependent increase in susceptibility compared to wild-type control mice. However, when the mice are infected intraperitoneally, there is no difference between mutant and wild-type mice, indicating that the defensin protective effect is specific to the small intestine. The effect is not dependent on the host intestinal microbiota. Viral loads in the defensin-deficient mice are higher in brain, spleen, and ileum only at late times after infection, suggesting that rather than directly delaying viral dissemination, α-defensins act indirectly to protect orally infected mice. Histological and humoral response assays indicate that the adaptive immune response to MAdV-1 infection, particularly the neutralizing antibody response, is delayed in the absence of functional α-defensins.

A major innate response to virus infection is PKR activation. PKR is an interferon-stimulated kinase that is activated by binding to double-stranded RNA produced in infections by DNA and RNA viruses [89–92]. Activation of PKR leads to phosphorylation of eukaryotic translation initiation factor eIF2a, which halts protein synthesis [93,94]. Viruses have evolved a variety of ways to circumvent this antiviral host response, including inhibiting PKR phosphorylation, sequestering PKR, dephosphorylating eIF2a, and degrading PKR [95]. HAdVs encode VA RNAs that sequester PKR by binding it as a monomer, preventing its autophosphorylation/activation [96]. However, MAdV-1 does not encode a VA RNA [37], and it circumvents the antiviral PKR response by a mechanism not previously shown for DNA viruses. MAdV-1 degrades PKR in multiple cell types infected *in vitro* [97]. PKR degradation has not been previously observed for DNA viruses. Inhibiting the proteasome blocked MAdV-1-induced PKR degradation, indicating that the degradation likely proceeds by a proteasomal mechanism. The viral gene(s) involved in PKR degradation have not been identified, but the data point toward an early viral gene.

MAdV-1 pathogenesis—Persistence and host genetics of susceptibility

Like HAdVs, MAdV-1 persists in the host after acute infection [32,33,98]. MAdV-1 is detected in brains, spleens, and kidneys of outbred mice up to 55 weeks after intraperitoneal infection [33], and in urine for up to 2 years [71]. After intranasal inoculation, MAdV-1 viral genomes can be detected in the hearts of inbred mice at 9 weeks postinfection [12]. This persistence in the heart can lead to cardiomyocyte hypertrophy. It is not known whether MAdV-1 also persists in lymphocytes, as has been found for HAdVs [99–101], including in lymphocytes of the gut lamina propria [102].

Different strains of outbred and inbred mice differ in their susceptibility to MAdV-1 [6,8,9]. Adult SJL/J mice are highly susceptible to the virus, whereas other inbred strains, such as BALB/cJ, are resistant [9]. The H-2^S haplotype of SJL/J mice is not associated with susceptibility, which was shown to be a quantitative trait [103]. Genetic mapping using an interstrain backcross identified a major quantitative trait locus (QTL) on mouse chromosome (Chr) 15, $Msq1^{SJL}$, that accounts for a significant portion of the total trait variance between SJL/J and BALB/cJ strains. Fine mapping localized the QTL to a region of Chr 15 that encodes genes of the hematopoietic cell surface-expressed LY6 family [104]. Further mapping was unable to identify specific *Ly6* genes involved in MAdV-1 susceptibility (Spindler, unpublished), but interestingly, *Ly6E* is among other IFN-stimulated genes shown to enhance the replication of several enveloped viruses, by targeting a late viral entry step [105,106]. Interval-specific congenic mice for the $Msq1^{SJL}$ locus were used to examine the contribution of the locus to disease phenotypes of intraperitoneally infected mice [9,58,104]. The locus is responsible for a subset of the physiological phenotypes that correlate with MAdV-1-induced encephalitis. Disease phenotype differences between the congenic mice and parental SJL mice indicate that there are additional host genes involved in CNS disease in mice.

Mouse adenoviruses type 2 and 3

The genomic sequence of MAdV-2 (HM049560.1) has been determined and compared to those of MAdV-1 and MAdV-3 (EU835513.1) [18,37,107,108]. Phylogenetic analyses showed that the three mouse AdVs have a close common ancestor, but MAdV-1 and MAdV-3 are closer to each other than to MAdV-2. Strikingly, although the number of predicted genes is almost the same, the MAdV-2 genome size of 35 203 bp is considerably larger than the genomes of MAdV -1 and MAdV-3 (30, 944 and 30 570 bp, respectively). This is mainly due to larger genes and ORFs in MAdV-2, although there are also some differences in the number of ORFs for the early regions E1, E3, and E4. A peculiar feature was noted when analyzing the leader

sequence of the late mRNA transcripts: in MAdV-1 and -3, as in most AdVs, the leader consists of three spliced sequences (tripartite sequence), whereas in MAdV-2, it is a bipartite leader sequence [109]. A bipartite leader is also found in fowl AdV type 10 [110]. It has been hypothesized that MAdV-2 is a genuine mouse AdV that has continuously coevolved with *M. musculus*, whereas MAdV-1 may have switched to this host from some other species and is still in an adaption process, including remodeling of the genetic content [107]. This could also explain the elevated pathogenicity of MAdV-1 in the house mouse.

The receptors for MAdV-2 and -3 are not known. Just as in MAdV-1 and -3, no RGD motif is found in penton base of MAdV-2 [107]. However, the RGD motif present in the fiber knob of MAdV-1 and -3 is lacking in the fiber knob of MAdV-2, which has little sequence identity (10–16%) to AdV fibers of known structures. A high-resolution crystal structure of the carboxy-terminus of the MAdV-2 fiber reveals a domain with the typical AdV fiber head topology and a domain containing two triple β-spiral repeats of the shaft domain [111]. The fiber head contains a monosaccharide N-acetylglucosamine (GlcNAc) binding site that allows binding to GlcNAc-containing mucin glycans, potentially representing a target in the mouse gut. As reported for MAdV-1, mouse CAR does not serve as receptor for MAdV-2 or -3. Studies with mouse CAR-transfected B16 melanoma cells did not reveal increased MAdV-2 or -3 infection (M. Bieri and S. Hemmi, unpublished). This is consistent with the fact that few of the residues important for CAR binding are conserved in the MAdV-2 fiber head [111]. Competition studies with recombinant MAdV-2 fiber knob demonstrated that fiber knob is the attachment protein [111,112], paralleling competition results for MAdV-1 fiber knob discussed above [53]. However, the receptors for MAdV-1 and MAdV-2 on cultured cells are not the same, because purified MAdV-2 fiber knob does not inhibit MAdV-1 infection [112].

As discussed above, enteric α-defensins have potent antiviral effects on *in vivo* and *in vitro* MAdV-1 infection. For enteric MAdV-2, α-defensins have the opposite effect and enhance infection of mouse colon CMT-93 cells and stem cell-derived small intestinal enteroids [112]. The enteroid experiments accurately predicted increased MAdV-2 shedding in the feces of wild-type mice compared to mice lacking functional α-defensins. These results are in line with *in vitro* findings for human enteric viruses and suggest that some viruses have evolved to use these host α-defensin proteins to enhance their replication.

MAdVs as vaccines and vectors

Only a few studies to date have used wild-type or recombinant MAdVs for therapeutic approaches. This may be due in part to the relatively fastidious growth of the viruses and the difficulty in generating recombinant vectors. Robinson et al. engineered an oncolytic MAdV-1 in which a small deletion in the E1A rendered the protein deficient for binding to pRb, thereby resembling the human AdVΔ24 vector that has been shown to replicate selectively in cancer cells defective for the pRb pathway [113]. This MAdV-1 was engineered to express the immune stimulatory GM-CSF and tested in immunocompetent syngeneic tumor models. Compared to the parental vectors, the MAdV-1 vector with GM-CSF was more effective in reducing tumor growth in the low immunogenic Pan02 tumor model and induced a greater systemic antitumor immune response. This system represents substantial progress for testing armed oncolytic viruses, since oncolytic HAdVs cannot productively infect mouse cells, and effects of immunostimulatory transgenes on overall virus potency, virus dissemination, and vector safety cannot be readily assessed in immunocompromised models, such as nude or SCID mice.

Genetically modified mouse cells (and possibly mice) capable of supporting HAdV replication represent an alternative immunocompetent mouse model. They would have the advantage that the vast number of oncolytic HAdV vectors could be tested directly. Initial studies showed that coinfection of HAdV-C5 and MAdV-1 or heterologous overexpression of HAdV-C5 L4-100K can partially complement late protein expression, which has been identified as a bottleneck for HAdV-C5 replication in mouse cells [114]. A second detailed complementation study used the HAdV-B-derived oncolytic virus, enadenotucirev (EnAd), in coinfections either with MAdV-1 or with EnAd viruses containing 24 different MAdV-1 ORFs [113,115]. These transcomplementations with MAdV-1 genes failed to rescue EnAd replication. Thus, the mechanisms preventing productive replication of HAdVs in mouse cells seem to vary for the different HAdV species, and more work needs to be done to understand and overcome these hurdles.

Oral replication-competent vaccines against HAdV-4 and -7 have long been used to immunize the US military against severe respiratory infection caused by these viruses [116]. MAdV-1 has recently been examined as a model to study oral replication-competent AdV vaccines *in vivo* in a natural host [117].

Intranasal, intraperitoneal, and natural MAdV-1 infection generate neutralizing antibodies [1,6,60,63]. Oral infection of C57BL/6 mice, which have intermediate susceptibility to MAdV-1 [104], leads to a systemic infection with moderate bowel pathogenesis and antiviral neutralizing antibody responses [62]. In the MAdV-1 vaccine study, when BALB/c mice, which are more resistant to MAdV-1 infection [104], were inoculated orally, there was only a subclinical infection that also generated a virus-specific neutralizing antibody response [62,117]. Although clinical signs of disease were not seen after oral infection of the BALB/c mice, sporadic shedding of virus in feces occurred, as measured by qPCR. Importantly, oral immunization with MAdV-1 protected against homologous virus challenge, similar to the HAdV-4 and -7 immunizations, with generation of an adaptive immune response. However, the oral administration of MAdV-1 alone led to a systemic infection, even at low doses. Whether this occurs in humans is not known; the authors note that the HAdV oral vaccines have been primarily used on a specific population of young, healthy adults. It would be interesting to test whether the enteric MAdV-2 gives rise to similar levels of neutralizing antibodies, perhaps in the absence of systemic spread. The findings of disseminated infection after MAdV-1 oral infection should be considered if HAdVs are developed for broader vaccination.

Conclusions and Perspectives

Mouse adenoviruses are important because they enable the study of adenoviruses in their natural hosts. Studies of mouse adenoviruses have revealed tropisms and pathologies distinct from those seen in HAdV infections. They have also increased our knowledge of adenoviral pathogenesis and revealed new mechanisms of virus response to the host response in the 'arms race' between viruses and cells. While humanized mouse models [118] and organoid systems [119,120] may enable study of HAdV pathogenesis, their complexity and costs may limit their use. Study of mouse AdV pathogenesis in its natural host, with all the benefits of mouse genetics and immunological reagents, will continue to advance our understanding of viral–host interactions.

Acknowledgements

We thank Jason Weinberg for helpful comments on the manuscript. This work was supported by SNF grant number 31003A_146286 to SH and by NIH R01AI133935 to KRS.

References

1 Hartley JW and Rowe WP (1960) A new mouse virus apparently related to the adenovirus group. *Virology* **11**, 645–647.
2 Evans CA, Dowell M and Green RG (1950) The effects of viruses on intraocular tissues; infections with the virus of fox encephalitis (canine hepatitis). *J Infect Dis* **86**, 1–11.
3 Yates VJ and Fry DE (1957) Observations on a chicken embryo lethal orphan (CELO) virus. *Am J Vet Res* **18**, 657–660.
4 Klein M, Earley E and Zellat J (1959) Isolation from cattle of a virus related to human adenovirus. *Proc Soc Exp Biol Med* **102**, 1–4.
5 Kajon AE, Brown CC and Spindler KR (1998) Distribution of mouse adenovirus type 1 in intraperitoneally and intranasally infected adult outbred mice. *J Virol* **72**, 1219–1223.
6 Guida JD, Fejer G, Pirofski LA, Brosnan CF and Horwitz MS (1995) Mouse adenovirus type 1 causes a fatal hemorrhagic encephalomyelitis in adult C57BL/6 but not BALB/c mice. *J Virol* **69**, 7674–7681.
7 Ashley SL, Pretto CD, Stier MT, Kadiyala P, Castro-Jorge L, Hsu T-H, Doherty R, Carnahan KE, Castro MG, Lowenstein PR *et al.* (2017) Matrix metalloproteinase activity in infections by an encephalitic virus, mouse adenovirus type 1. *J Virol* **91**, e01412–16.
8 Kring SC, King CS and Spindler KR (1995) Susceptibility and signs associated with mouse adenovirus type 1 infection of adult outbred Swiss mice. *J Virol* **69**, 8084–8088.
9 Spindler KR, Fang L, Moore ML, Hirsch GN, Brown CC and Kajon A (2001) SJL/J mice are highly susceptible to infection by mouse adenovirus type 1. *J Virol* **75**, 12039–12046.
10 Blailock ZR, Rabin ER and Melnick JL (1967) Adenovirus endocarditis in mice. *Science* **157**, 69–70.
11 Blailock ZR, Rabin ER and Melnick JL (1968) Adenovirus myocarditis in mice: an electron microscopic study. *Exp Mol Pathol* **9**, 84–96.
12 McCarthy MK, Procario MC, Twisselmann N, Wilkinson JE, Archambeau AJ, Michele DE, Day SM and Weinberg JB (2015) Proinflammatory effects of interferon gamma in mouse adenovirus 1 myocarditis. *J Virol* **89**, 468–479.
13 Weinberg JB, Stempfle GS, Wilkinson JE, Younger JG and Spindler KR (2005) Acute respiratory infection with mouse adenovirus type 1. *Virology* **340**, 245–254.
14 Procario MC, Levine RE, McCarthy MK, Kim E, Zhu L, Chang C-H, Hershenson MB and Weinberg JB (2012) Susceptibility to acute mouse adenovirus type 1 respiratory infection and establishment of protective immunity in neonatal mice. *J Virol* **86**, 4194–4203.

15 Hashimoto K, Sugiyama T and Saski S (1966) An adenovirus isolated from the feces of mice. I. Isolation and identification. *Jpn J Microbiol* **10**, 115–125.
16 Takeuchi A and Hashimoto K (1976) Electron microscope study of experimental enteric adenovirus infection in mice. *Infect Immun* **13**, 569–580.
17 Hashimoto K, Sugiyama T, Yoshikawa M and Sasaki S (1970) Intestinal resistance in the experimental enteric infection of mice with a mouse adenovirus. I. Growth of the virus and appearance of a neutralizing substance in the intestinal tract. *Jpn J Microbiol* **14**, 381–395.
18 Klempa B, Kruger DH, Auste B, Stanko M, Krawczyk A, Nickel K f, Uberla K and Stang A (2009) A novel cardiotropic murine adenovirus representing a distinct species of mastadenoviruses. *J Virol* **83**, 5749–5759.
19 Becker SD, Bennett M, Stewart JP and Hurst JL (2007) Serological survey of virus infection among wild house mice (*Mus domesticus*) in the UK. *Lab Anim* **41**, 229–238.
20 Singleton GR, Smith AL, Shellam GR, Fitzgerald N and Müller WJ (1993) Prevalence of viral antibodies and helminths in field populations of house mice (*Mus domesticus*) in southeastern Australia. *Epidemiol Infect* **110**, 399–417.
21 Smith AL, Singleton GR, Hansen GM and Shellam G (1993) A serologic survey for viruses and Mycoplasma pulmonis among wild house mice (*Mus domesticus*) in southeastern Australia. *J Wildl Dis* **29**, 219–229.
22 Dammann P, Hilken G, Hueber B, Köhl W, Bappert MT and Mähler M (2011) Infectious microorganisms in mice (*Mus musculus*) purchased from commercial pet shops in Germany. *Lab Anim* **45**, 271–275.
23 Grove KA, Smith PC, Booth CJ and Compton SR (2012) Age-associated variability in susceptibility of Swiss Webster mice to MPV and other excluded murine pathogens. *J Am Assoc Lab Anim Sci* **51**, 789–796.
24 Henderson KS, Perkins CL, Havens RB, Kelly MJ, Francis BC, Dole VS and Shek WR (2013) Efficacy of direct detection of pathogens in naturally infected mice by using a high-density PCR array. *J Am Assoc Lab Anim Sci* **52**, 763–772.
25 Marx JO, Gaertner DJ and Smith AL (2017) Results of survey regarding prevalence of adventitial infections in mice and rats at biomedical research facilities. *J Am Assoc Lab Anim Sci* **56**, 527–533.
26 Williams SH, Che X, Garcia JA, Klena JD, Lee B, Muller D, Ulrich W, Corrigan RM, Nichol S, Jain K *et al.* (2018) Viral diversity of house mice in New York City. *MBio* **9**, e01354–17.
27 Phan TG, Kapusinszky B, Wang C, Rose RK, Lipton HL and Delwart EL (2011) The fecal viral flora of wild rodents. *PLoS Pathog* **7**, e1002218.
28 Wu Z, Lu L, Du J, Yang L, Ren X, Liu B, Jiang J, Yang J, Dong J, Sun L *et al.* (2018) Comparative analysis of rodent and small mammal viromes to better understand the wildlife origin of emerging infectious diseases. *Microbiome* **6**, 178.
29 Zheng XY, Qiu M, Ke X-M, Guan W-J, Li J-M, Huo S-T, Chen S-W, Zhong X-S, Zhou W, Xiong Y-Q *et al.* (2016) Detection of novel adenoviruses in fecal specimens from rodents and shrews in southern China. *Virus Genes* **52**, 417–421.
30 Smith AL and Barthold SW (1987) Factors influencing susceptibility of laboratory rodents to infection with mouse adenovirus strains K87 and FL. *Arch Virol* **95**, 143–148.
31 Spindler KR, Moore ML and Cauthen AN (2007) Mouse adenoviruses. In The Mouse in Biomedical Research (Fox JG, Davisson MT, Quimby FW, Barthold SW, Newcomer CE and Smith AL, eds). pp. 49–65. Academic Press: New York, NY.
32 Smith K and Spindler KR (1999) Murine adenovirus. In Persistent Viral Infections (Ahmed R and Chen I, eds), pp. 477–484. John Wiley & Sons, New York, NY.
33 Smith K, Brown CC and Spindler KR (1998) The role of mouse adenovirus type 1 early region 1A in acute and persistent infections in mice. *J Virol* **72**, 5699–5706.
34 Ball AO, Beard CW, Villegas P and Spindler KR (1991) Early region 4 sequence and biological comparison of two isolates of mouse adenovirus type 1. *Virology* **180**, 257–265.
35 Ball AO, Williams ME and Spindler KR (1988) Identification of mouse adenovirus type 1 early region 1: DNA sequence and a conserved transactivating function. *J Virol* **62**, 3947–3957.
36 Temple M, Antoine G, Stahl S, Winnacker Ernst-L and Delius H (1981) Replication of mouse adenovirus strain FL DNA. *Virology* **109**, 1–12.
37 Meissner JD, Hirsch GN, LaRue EA, Fulcher RA and Spindler KR (1997) Completion of the DNA sequence of mouse adenovirus type 1: sequence of E2B, L1, and L2 (18–51 map units). *Virus Res* **51**, 53–64.
38 Davison AJ, Benko M and Harrach B (2003) Genetic content and evolution of adenoviruses. *J Gen Virol* **84**, 2895–2908.
39 Kitajewski J, Schneider RJ, Safer B, Munemitsu SM, Samuel CE, Thimmappaya B and Shenk T (1986) Adenovirus VAI RNA antagonizes the antiviral action of interferon by preventing activation of the interferon-induced eIF-2 alpha kinase. *Cell* **45**, 195–200.
40 Ball AO, Beard CW, Redick SD and Spindler KR (1989) Genome organization of mouse adenovirus type 1 early region 1: a novel transcription map. *Virology* **170**, 523–536.

41 Smith K, Ying B, Ball AO, Beard CW and Spindler KR (1996) Interaction of mouse adenovirus type 1 early region 1A protein with cellular proteins pRb and p107. *Virology* **224**, 184–197.
42 Fang L, Stevens JL, Berk AJ and Spindler KR (2004) Requirement of Sur2 for efficient replication of mouse adenovirus type 1. *J Virol* **78**, 12888–12900.
43 Cauthen AN and Spindler KR (1996) Sequence of the mouse adenovirus type-1 DNA encoding the 100-kDa, 33-kDa and DNA binding proteins. *Gene* **168**, 183–187.
44 Beard CW, Ball AO, Wooley EH and Spindler KR (1990) Transcription mapping of mouse adenovirus type 1 early region 3. *Virology* **175**, 81–90.
45 Beard CW and Spindler KR (1996) Analysis of early region 3 mutants of mouse adenovirus type 1. *J Virol* **70**, 5867–5874.
46 Cauthen AN, Brown CC and Spindler KR (1999) In vitro and in vivo characterization of a mouse adenovirus type 1 early region 3 mutant. *J Virol* **73**, 8640–8646.
47 Burgert H-G and Kvist S (1985) An adenovirus type 2 glycoprotein blocks cell surface expression of human histocompatibility class I antigens. *Cell* **41**, 987–997.
48 Kring SC and Spindler KR (1996) Lack of effect of mouse adenovirus type 1 infection on the cell surface expression of major histocompatibility complex class I antigens. *J Virol* **70**, 5495–5502.
49 Kring SC, Ball AO and Spindler KR (1992) Transcription mapping of mouse adenovirus type 1 early region 4. *Virology* **190**, 248–255.
50 Gilson T, Blanchette P, Ballmann MZ, Papp T, Pénzes JJ, Benkő M, Harrach Band Branton PE (2016) Using the E4orf6-based E3 ubiquitin ligase as a tool to analyze the evolution of adenoviruses. *J Virol* **90**, 7350–7367.
51 Song B, Hu S-l, Darai G, Spindler KR and Young CS (1996) Conservation of DNA sequence in the predicted major late promoter regions of selected mastadenoviruses. *Virology* **220**, 390–401.
52 Lenaerts L, Daelemans D, Geukens N, De Clercq E and Naesens L (2006) Mouse adenovirus type 1 attachment is not mediated by the coxsackie-adenovirus receptor. *FEBS Lett* **580**, 3937–3942.
53 Raman SR, Hsu TH, Ashley SL and Spindler KR (2009) Integrin and heparan sulfate usage as receptors for mouse adenovirus type 1. *J Virol* **83**, 2831–2838.
54 Lenaerts L, McVey JH, Baker AH, Denby L, Nicklin S, Verbeken E and Naesens L (2009) Mouse adenovirus type 1 and human adenovirus type 5 differ in endothelial cell tropism and liver targeting. *J Gene Med* **11**, 119–127.
55 Ashley SL, Welton AR, Harwood KM, Rooijen NV and Spindler KR (2009) Mouse adenovirus type 1 infection of macrophages. *Virology* **390**, 307–314.
56 Charles PC, Guida JD, Brosnan CF and Horwitz MS (1998) Mouse adenovirus type-1 replication is restricted to vascular endothelium in the CNS of susceptible strains of mice. *Virology* **245**, 216–228.
57 Gralinski LE, Ashley SL, Dixon SD and Spindler KR (2009) Mouse adenovirus type 1-induced breakdown of the blood-brain barrier. *J Virol* **83**, 9398–9410.
58 Hsu T-H, Althaus IW, Foreman O and Spindler KR (2012) Contribution of a single host genetic locus to mouse adenovirus type 1 infection and encephalitis. *MBio* **3**, e00131–12.
59 Tirumuru N, Pretto CD, Castro Jorge LA and Spindler KR (2016) Mouse adenovirus type 1 early region 1A effects on the blood-brain barrier. *mSphere* **1**, e00079-16.
60 Chandrasekaran A, Adkins LJ, Seltzer HM, Pant K, Tryban ST, Molloy CT and Weinberg JB (2019) Age-dependent effects of immunoproteasome deficiency on mouse adenovirus type 1 pathogenesis. *J Virol* **93**, e00569-19.
61 Molloy CT, Adkins LJ, Griffin C, Singer K and Weinberg JB (2018) Mouse adenovirus type 1 infection of adipose tissue. *Virus Res* **244**, 90–98.
62 Gounder AP, Myers ND, Treuting PM, Bromme BA, Wilson SS, Wiens ME, Lu W, Ouellette AJ, Spindler KR, Parks WC *et al*. (2016) Defensins potentiate a neutralizing antibody response to enteric viral infection. *PLoS Pathog* **12**, e1005474.
63 Moore ML, McKissic EL, Brown CC, Wilkinson JE and Spindler KR (2004) Fatal disseminated mouse adenovirus type 1 infection in mice lacking B cells or Bruton's tyrosine kinase. *J Virol* **78**, 5584–5590.
64 Inada T and Uetake H (1978) Cell-mediated immunity assayed by ^{51}Cr release test in mice infected with mouse adenovirus. *Infect Immun* **20**, 1–5.
65 Inada T and Uetake H (1978) Nature and specificity of effector cells in cell-mediated cytolysis of mouse adenovirus-infected cells. *Infect Immun* **22**, 119–124.
66 Inada T and Uetake H (1978) Cell-mediated immunity to mouse adenovirus infection: macrophage migration inhibition test. *Microbiol Immunol* **22**, 391–401.
67 Inada T and Uetake H (1980) Cell-mediated immunity to mouse adenovirus infection: Blocking of macrophage migration inhibition and T cell-mediated cytolysis of infected cells by anti-S antigen or anti-alloantigen serum. *Microbiol Immunol* **24**, 525–535.
68 Moore ML, Brown CC and Spindler KR (2003) T cells cause acute immunopathology and are required for long-term survival in mouse adenovirus type 1-induced encephalomyelitis. *J Virol* **77**, 10060–10070.
69 Molloy CT, Andonian JS, Seltzer HM, Procario MC, Watson ME and Weinberg JB (2017) Contributions of CD8 T cells to the pathogenesis of mouse adenovirus type 1 respiratory infection. *Virology* **507**, 64–74.

70 Adkins LJ, Molloy CT and Weinberg JB (2018) Fas activity mediates airway inflammation during mouse adenovirus type 1 respiratory infection. *Virology* **521**, 129–137.
71 van der Veen J and Mes A (1973) Experimental infection with mouse adenovirus in adult mice. *Arch Gesamte Virusforsch* **42**, 235–241.
72 Coutelier J-P, Coulie PG, Wauters P, Heremans H and van der Logt JT (1990) *In vivo* polyclonal B-lymphocyte activation elicited by murine viruses. *J Virol* **64**, 5383–5388.
73 Coutelier J-P, van der Logt JT, Heessen FW, Vink A and van Snick J (1988) Virally induced modulation of murine IgG antibody subclasses. *J Exp Med* **168**, 2373–2378.
74 Coutelier J-P, van der Logt JT, Heessen FW, Warnier G and Van Snick J (1987) IgG2a restriction of murine antibodies elicited by viral infections. *J Exp Med* **165**, 64–69.
75 Lenaerts L, Kelchtermans H, Geboes L, Matthys P, Verbeken E, De Clercq E and Naesens L (2008) Recovery of humoral immunity is critical for successful antiviral therapy in disseminated mouse adenovirus type 1 infection. *Antimicrob Agents Chemother* **52**, 1462–1471.
76 Welton AR, Gralinski LE and Spindler KR (2008) Mouse adenovirus infection of natural killer cell-deficient mice. *Virol* **373**, 163–170.
77 Kajon AE and Spindler KR (2000) Mouse adenovirus type 1 replication in vitro is resistant to interferon. *Virology* **274**, 213–219.
78 Anderson VE, Nguyen Y and Weinberg JB (2009) Effects of allergic airway disease on mouse adenovirus type 1 respiratory infection. *Virology* **391**, 25–32.
79 McCarthy MK and Weinberg JB (2015) The immunoproteasome and viral infection: a complex regulator of inflammation. *Front Microbiol* **6**, 21.
80 McCarthy MK, Malitz DH, Molloy CT, Procario MC, Greiner KE, Zhang L, Wang P, Day SM, Powell SR and Weinberg JB (2016) Interferon-dependent immunoproteasome activity during mouse adenovirus type 1 infection. *Virology* **498**, 57–68.
81 Castro-Jorge L, Pretto CD, Smith AB, Foreman O, Carnahan KE and Spindler KR (2017) A protective role for IL-1 signaling during mouse adenovirus type 1-induced encephalitis. *J Virol* **91**, e02106–16.
82 Sergerie Y, Rivest S and Boivin G (2007) Tumor necrosis factor-alpha and interleukin-1 beta play a critical role in the resistance against lethal herpes simplex virus encephalitis. *J Infect Dis* **196**, 853–860.
83 McCarthy MK, Zhu L, Procario MC and Weinberg JB (2014) IL-17 contributes to neutrophil recruitment but not to control of viral replication during acute mouse adenovirus type 1 respiratory infection. *Virology* **456–457**, 259–267.
84 McCarthy MK, Levine RE, Procario MC, McDonnell PJ, Zhu L, Mancuso P, Crofford LJ, Aronoff DM and Weinberg JB (2013) Prostaglandin E2 induction during mouse adenovirus type 1 respiratory infection regulates inflammatory mediator generation but does not affect viral pathogenesis. *PLoS ONE* **8**, e77628.
85 Lehrer RI and Lu W (2012) alpha-Defensins in human innate immunity. *Immunol Rev* **245**, 84–112.
86 Smith JG, Silvestry M, Lindert S, Lu W, Nemerow GR and Stewart PL (2010) Insight into the mechanisms of adenovirus capsid disassembly from studies of defensin neutralization. *PLoS Pathog* **6**, e1000959.
87 Smith JG and Nemerow GR (2008) Mechanism of adenovirus neutralization by human alpha-defensins. *Cell Host Microbe* **3**, 11–19.
88 Nguyen EK, Nemerow GR and Smith JG (2010) Direct evidence from single-cell analysis that human {alpha}-defensins block adenovirus uncoating to neutralize infection. *J Virol* **84**, 4041–4049.
89 Meurs E, Chong K, Galabru J, Thomas NB, Kerr IM, Williams BRG and Hovanessian AG (1990) Molecular cloning and characterization of the human double-stranded RNA-activated protein kinase induced by interferon. *Cell* **62**, 379–390.
90 Feng GS, Chong K, Kumar A and Williams BR (1992) Identification of double-stranded RNA-binding domains in the interferon-induced double-stranded RNA-activated p68 kinase. *Proc Natl Acad Sci USA* **89**, 5447–5451.
91 McCormack SJ, Thomis DC and Samuel CE (1992) Mechanism of interferon action: Identification of a RNA binding domain within the N-terminal region of the human RNA-dependent P1/eIF-2 alpha protein kinase. *Virol* **188**, 47–56.
92 Patel RC and Sen GC (1992) Identification of the double-stranded RNA-binding domain of the human interferon-inducible protein kinase. *J Biol Chem* **267**, 7671–7676.
93 Wek RC (2018) Role of eIF2alpha kinases in translational control and adaptation to cellular stress. *Cold Spring Harb Perspect Biol* **10**, a032870.
94 Farrell PJ, Balkow K, Hunt T, Jackson RJ and Trachsel H (1977) Phosphorylation of initiation factor elF-2 and the control of reticulocyte protein synthesis. *Cell* **11**, 187–200.
95 Dzananovic E, McKenna SA and Patel TR (2018) Viral proteins targeting host protein kinase R to evade an innate immune response: a mini review. *Biotechnol Genet Eng Rev* **34**, 33–59.
96 Mathews MB and Shenk T (1991) Adenovirus virus-associated RNA and translational control. *J Virol* **65**, 5657–5662.
97 Goodman DE, Pretto CD, Krepostman TA, Carnahan KE and Spindler KR (2019) Enhanced replication of

mouse adenovirus type 1 following virus-induced degradation of protein kinase R (PKR). *MBio* **10**, e00668–19.
98 Ginder DR (1964) Increased susceptibility of mice infected with mouse adenovirus to *Escherichia coli*-induced pyelonephritis. *J Exp Med* **120**, 1117–1128.
99 Garnett CT, Erdman D, Xu W and Gooding LR (2002) Prevalence and quantitation of species C adenovirus DNA in human mucosal lymphocytes. *J Virol* **76**, 10608–10616.
100 Garnett CT, Talekar G, Mahr JA, Huang W, Zhang Y, Ornelles DA and Gooding LR (2009) Latent species C adenoviruses in human tonsil tissues. *J Virol* **83**, 2417–2428.
101 Horvath J, Palkonyay L and Weber J (1986) Group C adenovirus DNA sequences in human lymphoid cells. *J Virol* **59**, 189–192.
102 Kosulin K, Geiger E, Vécsei A, Huber W-d, Rauch M, Brenner E, Wrba F, Hammer K, Innerhofer A, Pötschger U *et al.* (2016) Persistence and reactivation of human adenoviruses in the gastrointestinal tract. *Clin Microbiol Infect* **22**, 381 e1–381 e8.
103 Welton AR, Chesler EJ, Sturkie C, Jackson AU, Hirsch GN and Spindler KR (2005) Identification of quantitative trait loci for susceptibility to mouse adenovirus type 1. *J Virol* **79**, 11517–11522.
104 Spindler KR, Welton AR, Lim ES, Duvvuru S, Althaus IW, Imperiale JE, Daoud AI and Chesler EJ (2010) The major locus for mouse adenovirus susceptibility maps to genes of the hematopoietic cell surface-expressed LY6 family. *J Immunol* **184**, 3055–3062.
105 Schoggins JW, Wilson SJ, Panis M, Murphy MY, Jones CT, Bieniasz P and Rice CM (2011) A diverse range of gene products are effectors of the type I interferon antiviral response. *Nature* **472**, 481–485.
106 Mar KB, Rinkenberger NR, Boys IN, Eitson JL, McDougal MB, Richardson RB and Schoggins JW (2018) LY6E mediates an evolutionarily conserved enhancement of virus infection by targeting a late entry step. *Nat Commun* **9**, 3603.
107 Hemmi S, Vidovszky MZ, Ruminska J, Ramelli S, Decurtins W, Greber UF and Harrach B (2011) Genomic and phylogenetic analyses of murine adenovirus 2. *Virus Res* **160**, 128–135.
108 Smith AL, Winograd DF and Burrage TG (1986) Comparative biological characterization of mouse adenovirus strains FL and K87 and seroprevalence in laboratory rodents. *Arch Virol* **91**, 233–246.
109 Ramke M, Lee JY, Dyer DW, Seto D, Rajaiya J and Chodosh J (2017) The 5'UTR in human adenoviruses: Leader diversity in late gene expression. *Sci Rep* **7**, 618.
110 Sheppard M, Werner W, McCoy RJ and Johnson MA (1998) The major late promoter and bipartite leader sequence of fowl adenovirus. *Arch Virol* **143**, 537–548.
111 Singh AK, Nguyen TH, Vidovszky MZ, Harrach B, Benkő M, Kirwan A, Joshi L, Kilcoyne M, Berbis MÁ, Cañada FJ *et al.* (2018) Structure and N-acetylglucosamine binding of the distal domain of mouse adenovirus 2 fibre. *J Gen Virol* **99**, 1494–1508.
112 Wilson SS, Bromme BA, Holly MK, Wiens ME, Gounder AP, Sul Y and Smith JG (2017) Alpha-defensin-dependent enhancement of enteric viral infection. *PLoS Pathog* **13**, e1006446.
113 Robinson M, Li B, Ge Y, Ko D, Yendluri S, Harding T, VanRoey M, Spindler KR and Jooss K (2009) Novel immunocompetent murine tumor model for evaluation of conditionally replication-competent (oncolytic) murine adenoviral vectors. *J Virol* **83**, 3450–3562.
114 Young AM, Archibald KM, Tookman LA, Pool A, Dudek K, Jones C, Williams SL, Pirlo KJ, Willis AE, Lockley M *et al.* (2012) Failure of translation of human adenovirus mRNA in murine cancer cells can be partially overcome by L4–100K expression *in vitro* and *in vivo*. *Mol Ther* **20**, 1676–88.
115 Lei J, Jacobus EJ, Taverner WK, Fisher KD, Hemmi S, West K, Slater L, Lilley F, Brown A, Champion B *et al.* (2018) Expression of human CD46 and trans-complementation by murine adenovirus 1 fails to allow productive infection by a group B oncolytic adenovirus in murine cancer cells. *J Immunother Cancer* **6**, 55.
116 Top FH Jr (1975) Control of adenovirus acute respiratory disease in U.S. Army trainees. *Yale J Biol Med* **48**, 185–195.
117 Goffin E, Javaux J, Destexhe E, Pretto CD, Spindler KR, Machiels Band Gillet L (2019) Oral vaccination with replication-competent adenovirus in mice reveals dissemination of the viral vaccine beyond the gastrointestinal tract. *J Virol* **93**, e00237-19.
118 Wahl A, De C, Abad Fernandez M, Lenarcic EM, Xu Y, Cockrell AS, Cleary RA, Johnson CE, Schramm NJ, Rank LM *et al.* (2019) Precision mouse models with expanded tropism for human pathogens. *Nat Biotechnol* **37**:1163–1173.
119 Dutta D and Clevers H (2017) Organoid culture systems to study host-pathogen interactions. *Curr Opin Immunol* **48**, 15–22.
120 Holly MK and Smith JG (2018) Adenovirus infection of human enteroids reveals interferon sensitivity and preferential infection of goblet cells. *J Virol* **92**, e00250-18.