Evolutionary and Structural Overview of Human Picornavirus Capsid Antibody Evasion

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Picornaviruses constitute one of the most relevant viral groups according to their impact on human and animal health. Etiologic agents of a broad spectrum of illnesses with a clinical presentation that ranges from asymptomatic to fatal disease, they have been the cause of uncountable epidemics throughout history. Picornaviruses are small naked RNA-positive single-stranded viruses that include some of the most important pillars in the development of virology, comprising poliovirus, rhinovirus, and hepatitis A virus. Picornavirus infectious particles use the fecal–oral or respiratory routes as primary modes of transmission. In this regard, successful viral spread relies on the capability of viral capsids to (i) shelter the viral genome, (ii) display molecular determinants for cell receptor recognition, (iii) facilitate efficient genome delivery, and (iv) escape from the immune system. Importantly, picornaviruses display a substantial amount of genetic variability driven by both mutation and recombination. Therefore, the outcome of their replication results in the emergence of a genetically diverse cloud of individuals presenting phenotypic variance. The host humoral response against the capsid protein represents the most active immune pressure and primary weapon to control the infection. Since the preservation of the capsid function is deeply rooted in the virus evolutionary dynamics, here we review the current structural evidence focused on capsid antibody evasion mechanisms from that perspective.

Keywords: picornavirus, capsid, antibody, genetic variability, structure, vaccine

PICORNAVIRUS HISTORICAL RELEVANCE

Picornaviruses have been pivotal in the foundations of virology. Original research on “ultra-filterable infectious agents” such as foot-and-mouth disease virus (FMDV) and poliovirus (PV) began the era of animal virology (Loeffler and Frosch, 1898; Eggers, 1999). The development of cell cultures for PV replication led to Salk’s inactivated and Sabin’s attenuated vaccines (Enders et al., 1949). The first animal virus engineered into an infectious clone (Racaniello and Baltimore, 1981) and the first virus synthesized outside the cell was PV (Molla et al., 1991). Although vast knowledge has been gained, picornaviruses still challenge our understanding. The still open fundamental questions and public health challenges picornaviruses pose reflect that we are far from a conclusive comprehension (Holm-Hansen et al., 2016; Li et al., 2017; Zarocostas, 2018). In the following review, we examine how these agents evade host antibodies (Abs) based on their biological and evolutionary properties, with the spotlight on human picornaviruses.
CLASSIFICATION AND CLINICAL IMPACT ON HUMAN HEALTH

Picornaviridae is a large family of vertebrate viruses that produce both clinically asymptomatic infections but often mild and fatal disease. Their current classification includes more than 30 genera and 75 species (Zell et al., 2017), including several human picornaviruses. The genus Enterovirus comprises seven species infecting humans (enterovirus A-to-D and rhinovirus A-to-C). This genus contains poliovirus (PV), coxsackieviruses A/B (CVA/B), enteroviruses (EV), echoviruses (E), and rhinoviruses (RV). Further serological classification results in hundreds of serotypes. Hepatitis A virus (HAV) is the sole human-virus species in the genus Hepatovirus. Other human picornaviruses include members of the genera Cardiovirus (Saffold virus—SAFV), Cosavirus (CoSV), Parechovirus (Ljungan virus—LV), Kubovirus (Aichi virus—AiV), and Salivirus (Salivirus A—SaVA) (Nielsen et al., 2013).

RVs are airborne pathogens, while other enteroviruses and HAV use the fecal–oral route (Yin-Murphy and Almond, 1996). RVs cause the common cold, the most prevalent infectious disease worldwide, resulting in uncountable lost days from school and work. Epidemics occur yearly with outbreaks in the winter and spring (Drysdale et al., 2017). PV infection targets the central nervous system, destroying nerves and motor neurons, resulting in paralytic poliomyelitis. Until the PV worldwide eradication program based on global vaccination, polio epidemics have been the cause of high morbimortality ( Minor, 2014). The so-called “non-polio enteroviruses” (CVs, EVs, and Es) cause several diseases with high morbimortality including meningitis, myocarditis, poliomyelitis-like syndrome, pancreatitis, and possibly the onset of diabetes. Outbreaks are common and have been considered to have pandemic potential (Zhang et al., 2015; Pons-Salort et al., 2018). Hepatitis produced by HAV is a mild disease producing liver damage usually leading to total recovery, but rare, severe cases are fatal in older age individuals. HAV produces large outbreaks probably due to its long 2 to 3 week incubation time (Jacobsen and Wiersma, 2010).

GENOME ORGANIZATION

Picornaviruses have single-stranded RNA positive-sense genomes (∼7–9 kb) that serve as mRNA for viral protein synthesis (Baltimore, 1971) (Figure 1A). Their RNA holds a single ORF encoding a polyprotein precursor for all viral proteins. Importantly, the 5′-end of the genome is covalently bound to the Viral-Protein-genome (VPg) (Crawford and Baltimore, 1983), the primer for viral RNA synthesis (Nomoto et al., 1977). Two untranslated regions (UTR), 5′UTR and 3′UTR, flank the ORF and contain virus-specific RNA secondary structural elements implicated in replication and providing host specificity (Kloc et al., 2018). The 5′UTR bears an internal ribosomal entry site (IRES) and polyadenylidine tract (PPT) that elicit host ribosomes and PPT-binding protein for translation (Martinez-Salas et al., 2018). The 3′UTR finishes in a poly(A) tail that mimics host mRNA tail conferring genome stability.

From 5′-to-3′, the ORF comprises three regions: (i) P1, encoding the structural capsid viral proteins (VP4–VP2–VP3–VP1), while, in some picornaviruses, also codifying a short leader L-protein; (ii) P2, encoding the viral non-structural proteins 2A—2B—2C; and (iii) P3 encoding the viral non-structural proteins 3A—3B—3C–3D (Palmenberg, 1990) (Figure 1B). The non-structural proteins’ central role is replication, translation, and hijacking host-cell machinery (Figure 2). In particular, 3D is the RNA-dependent RNA polymerase (RdRp or 3Dpol) that synthesizes the virus genome and 3B (which is VPg) acts as its primer, being the only non-structural protein in the virion (Palmenberg, 1990).

PICORNAVIRUS CAPSID ANATOMY

Picornaviruses were some of the first human viruses to be structurally defined at the atomic level (Rossmann et al., 1985). To date, several structures of human picornaviruses have been unveiled including HRV, PV, HAV, CVB, E, CVA, EV, SAFV, and AiV (Hogle et al., 1985; Rossmann et al., 1985; Filman et al., 1989, 1998; Muckelbauer et al., 1995; Zhao et al., 1996; Lentz et al., 1997; Hendry et al., 1999; Verdaguer et al., 2000, 2003; Stuart et al., 2002; Zhang et al., 2004; Venkataaraman et al., 2008; Plevka et al., 2010, 2012; Zocher et al., 2014; Liu et al., 2015, 2016; Ren et al., 2015; Zhu et al., 2015; Mullanipudi et al., 2016). Moreover, the structure of FMDV has been disclosed (Logan et al., 1993; Lea et al., 1994). All picornaviruses have a naked 30 nm icosahedral capsid composed of 60 identical tightly packed protomers (Figure 1E). Early in particle morphogenesis, immature protomers contain VP1 and VP3 together with VP0, the precursor of VP4 and VP2 (Jiang et al., 2014) (Figure 1B). Virus assembly likely goes through a dodecahedral pathway (Li et al., 2012), by the association of pentamers formed by five immature protomers leading to the icosahedral particle, defining 5-, 3-, and 2-fold symmetry axes. Upon genome encapsidation, VP0 is generally auto-catalytically cleaved into VP2 and VP4 generating the mature capsid (except for Parechovirus and Kubovirus) (Figure 2).

The larger proteins (VP1–3) form the external and internal capsid surface. These proteins have a common fold, the “jelly-roll,” formed by two 4-strand anti-parallel β-sheets and two helices (Figure 1D). Conversely, the small VP4 is located inside the capsid only and usually appears myristoylated at its N-terminus (Paul and Schultz, 1987; Belsham et al., 1991) (Figure 1C). The capsid external surface displays a rugged topography. Main surface features include (i) a principal protrusion built by the interaction of copies of VP1 forming a star-shaped 5-fold vertex, (ii) a 5-fold surrounding valley called the “canyon,” (iii) a VP2 loop proteolysis or the “puff,” (iv) a VP3 loop rise or the “knob,” and (v) a large 2-fold depression (Muckelbauer et al., 1995). Loop differences result in distinctive surface traits between picornaviruses. Finally, some picornaviruses exhibit a lipid molecule, the “pocket factor,” bound
in a cavity located inside the VP1 jelly-roll, which has been observed to play a role in particle stability (Figure 1C).

**RECEPTORS AND TROPISM**

Picornavirus cell infection starts with its attachment to cell receptors (Figure 2). Therefore, virus-receptor usage is critical for tropism and its evolution can change virus targets at the level of cells to host ranges. The capsid binding sites of picornavirus receptors can be used to classify them into canyon binders and non-canyon binders.

Canyon binders are members of the immunoglobulin superfamily including: (i) ICAM-1 used by HRV and CVA (Greve et al., 1989; Staunton et al., 1989; Tomassini et al., 1989; Kolatkar et al., 1999; Xiao et al., 2001; Baggen et al., 2018), (ii) the PV receptor (PVR) (Mendelsohn et al., 1989; He et al., 2000; Strauss et al., 2015), (iii) the coxsackie and
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FIGURE 2 | Picornavirus life cycle. (A, B) Picornavirus uses different receptors to enter the cell, some implicated in the signaling internalization (A), meanwhile others can act as carriers that transport the viral particle to meet the primary receptor (B). (C, D) This infection event can be impeded by the action of specific neutralizing antibodies that can destabilize the viral particle (C) or opsonize or stabilize the particle to impair receptor binding or conformational changes required for infection (D). (E) Once the virus enters the cell, the viral RNA delivery mechanism is triggered, and the viral genome (black wavy line) is released into the cytoplasm. (F) Upon removal of VPg (magenta oval), the genome starts the IRES-driven translation leading to the production of the viral polyprotein. (G) The proteolytic cascade produces all viral proteins, structural and non-structural. (H) Some proteins act by hijacking the host cellular systems such as the nuclear pore, the cell translation machinery, and the apoptotic systems and initiate the remodeling of the internal cell membranes. (I) The structural proteins assemble into the capsid intermediates, the protomer and the pentamer, and also procapsids (L). (J) The formed replication complex assembled from non-structural proteins and modified internal membranes firing the picornaviral genome replication by the 3D polymerase via RNA complementary (red wavy lines) and using VPg as a primer. (K) The new progeny genomes including eventual mutations (yellow stars). (M) Mature virions assemble from pentamers that surround and package the new viral genomes. Viral particles escape from the cell by cell lysis or budding within membranes that can protect the viral progeny (P). (N) Some progeny virus with mutations in their capsids (yellow star) may escape from to the action of specific NAbs. (O) Empty capsids can act as molecular decoys for Abs to protect the infecting particles from neutralization.

adenovirus receptor (CAR) used by CVBs (Bergelson, 1997; He et al., 2001; Organtini et al., 2014; Lee et al., 2016), (iv) α5β3 and α5β6 integrin used by some CVAs (Roivainen et al., 1994; Williams et al., 2004; Shakeel et al., 2013), and (v) the α2 subunit of VLA-2 used by E1 and E8 (Bergelson et al., 1992; Xing et al., 2004). Canyon binders’ apical domain engages in the binding into the canyon, triggering conformational changes essential for infection, leading to the altered-particle (A-particle) conformational state (Greve et al., 1989; Xing et al., 2004; Xiao et al., 2005; Shakeel et al., 2013; Organtini et al., 2014; Strauss et al., 2015). These changes have been observed to depend on the number of binding events that stimulate the viral particle (Lee et al., 2016). Engagement of several receptors is known to bring the viral 5-fold vertex close to the cell membrane (Bubeck et al., 2005).

Non-canyon binders attach to the virus surface elsewhere outside the canyon, tethering the virus to the cell surface eventually signaling for virus internalization. Importantly, they are diverse in molecular characteristics. These receptors include (i) the LDL-receptor used by HRV-C (Verdaguer et al., 2004); (ii) the decay-accelerating factor (DAF), receptor of many echoviruses and CVBs (Bergelson et al., 1994, 1995; Pettigrew et al., 2006; Plevka et al., 2010; Pan et al., 2011; Yoder et al.,...
Picornaviruses display a great potential for adaptation and evolution, which is primarily dictated by their high mutation rate. Viral progenies are huge in population size and they have short generation times. Thus, the RNA virus population is a dynamic cloud of mutants where the average-consensus sequence of all variants represents the “genotype.” The mutation rate is the number of genetic changes, such as point mutations, insertions, or deletions introduced during viral replication. The first mutation rate measurement on RNA viruses was reported for CVA, disclosing a value of 1 mutation every 10,000 nucleotides copied (Eggers and Tamm, 1965).

Natural selection may have shaped picornaviral mutation rates in response to extremely dynamic ecosystems (Elena and Sanjuan, 2005). Therefore, this natural low replicative fidelity results in populations that quickly adapt to unexpected changes in the environment, such as immune pressure (Andino and Domingo, 2015). These observations have been conceptualized in the light of quasispecies evolution (Domingo et al., 2012). This adaptive capacity can be impaired by altering viral mutation rates (Vignuzzi et al., 2006). Indeed, the first support of the role of replicative fidelity in viral pathogenesis was observed in picornaviruses. Two groups isolated the first antimutator variant of an RNA virus, by serially passaging PV in the presence of ribavirin (Pfleiffer and Kirkegaard, 2003; Vignuzzi et al., 2006). The resistant variant contained a 3Dpol single point mutant (G64S) relatively resistant to lethal mutagenesis leading to (i) populations with lower mutation rates, (ii) reduced genetic diversity, and (iii) attenuated phenotype in mice (Vignuzzi et al., 2006). Recently, it has been suggested that this attenuation could be partly an outcome of a decrease in the replication speed (Fitzsimmons et al., 2018). Moreover, genetic engineering of viral polymerases has also identified several low-fidelity variants, called mutator variants (Thompson et al., 2007; Gong and Peersen, 2010; Gnädig et al., 2012; Rozen-Gagnon et al., 2014).

Interestingly, recombination-deficient variants of PV have been identified. These viruses carry amino acid changes in the 3Dpol that reduce recombination without conferring other detectable replication deficiencies. These non-recombining viruses accumulate a higher number of detrimental mutations, presumably by an inability to purge deleterious mutations, and fewer beneficial mutations (Xiao et al., 2016, 2017). Lately, the combined approach of mathematical modeling and experimental evolution experiments have predicted the frequency of recombination of picornaviruses such as PV and EV71 (Stern et al., 2017; Woodman et al., 2018).

ANTIBODY RESPONSE AGAINST PICORNAVIRUS INFECTION

Innate immune response detects foreign RNA using sensing proteins such as RIG-I, MDA-5, and Toll-like Receptor-3. These mediators act in the early control of picornaval infection (Slater et al., 2012); (iii) P-selectin glycoprotein ligand-1 (PSGL-1) used by EV71 (Nishimura et al., 2009); and (iv) scavenger receptor B2 (ScarbB2), a receptor of EV71 (Yamayoshi et al., 2009; Zhou et al., 2018). This group possibly includes heparan sulfate used by several enteroviruses (Escribano-Romero, 2004; Zautner et al., 2006; McLeish et al., 2012; Tan et al., 2013; Nishimura et al., 2015) and cadherin-related family member 3 (CDHR3) used by HRV-C (Watters and Palmenberg, 2018). Non-canyon binders rarely induce substantial conformational changes due to their interaction, and some may not be essential for infection since few mutations allow or limit their usage (Pan et al., 2011; McLeish et al., 2012; Lee et al., 2013). This alternative receptor usage can modify the infection mechanism, as seen in the case of CVB3 binding to DAF which signals the trafficking of the attached virus to tight junctions where the virus can meet CAR (Goyne and Bergelson, 2006).
et al., 2010). Nevertheless, the adaptive response mediated by Abs plays the definitive role in the resolution of the infection. Several pieces of evidence support this view as (i) patients with agammaglobulinemia develop chronic infections (Wilfert et al., 1977; McKinney et al., 1987; Kainulainen et al., 2010; Bucciol et al., 2018), (ii) humoral response mediates the protective effect of picornavirus vaccines (Grant et al., 2017; Sun et al., 2017), (iii) mice lacking B-cells cannot clear enteroviral infections (Mena et al., 1999), and (iv) passive immunization with Abs is an efficient treatment of HAV infection (Stapleton, 1992), although their effectiveness for enteroviral neonatal infections is disputed (Yen et al., 2015). Therefore, it appears to be clear that an effective humoral immune response represents the final host weapon to shortcut the viral infection.

**ANTIBODY ESCAPE MUTANTS**

Picornaviruses’ high mutation rates permit the rapid escape from the immune system. The change of residues in the exterior capsid surface overcomes the intense pressure exerted by host Abs. These properties were used to locate capsid antigenic sites by testing neutralizing Abs (nAbs) escape mutants in vitro (Minor et al., 1986; Sherry et al., 1986; Stapleton and Lemon, 1987). Successful progeny virus rely on capsid functionality. Therefore, the preservation of the architecture and receptor binding restrict viable mutations. The exposed jelly-roll loops can accommodate mutations easier than secondary structure elements and protein–protein interacting surfaces (Murray et al., 1988; Usherwood and Nash, 1995). Several structural studies by cryoEM revealed the way nAbs bind to solvent-exposed loops of VP1–3 by interacting with critical residues. Complexes of viruses with Fab-Abs fragments can display 1-Fab:1-protomer ratio following the icosahedral symmetry. Nevertheless, when epitopes are close to the symmetry axis, lower binding ratios are observed due to Fab–Fab steric hindrance (Lee et al., 2013). Three mechanisms of neutralization are interpreted from these structures: (i) destabilization of the virion by triggering conformational changes upon Fab binding, which is accompanied by the “pocket factor” release when present (Smith et al., 1996; Plevka et al., 2014; Dong et al., 2017; Zheng et al., 2019), (ii) stabilization of virions by cross-linking of protomers to prevent conformational changes for infection (Ye et al., 2016), (iii) virus aggregation by antibody cross-linking particles (Mosser et al., 1989), and (iv) opsonization that can interfere with virus-cell attachment and receptor binding (Lee et al., 2013; Wang et al., 2017).

**THE CANYON HYPOTHESIS**

Hiding the receptor-interacting surface from Abs surveillance is the hypothetical function of the canyon (Rossmann, 1989). Therefore, the canyon is the result of an evolutionary process to preserve and protect the critical residues required for host-cell receptor recognition. Conversely, accessible areas can mutate to disguise the virus from the humoral immune response. Nevertheless, some observations have challenged this view, proposing the receptor-binding site topology as an uncoating mechanism that dictates receptor binding to trigger the uncoating event (Smith et al., 1996). These views are not strictly incompatible; hence, both pictures contribute to the paradigm of picornavirus capsid evolution. Here, capsid topology arose out of and continues to be shaped by the interplay of host environmental pressure, random genome mutations, and fixation of mutations when beneficial.

**PICORNAVIRUS ANTIBODY DECOY PARTICLES**

Picornaviruses are known to produce a significant amount of procapsids during the infectious cell cycle, which appears as an inefficient way to replicate (Shingler et al., 2015). This wastefulness looks aggravated considering each polyproteic translation event would lead to a single protomer. Finally, the so-called procapsid may be an off-pathway particle (Cifuente et al., 2013). Although counterintuitive in appearance, the function of the procapsid could be to act as an immune decoy to enhance the infectivity of mature virions providing an evolutive advantage (Shingler et al., 2015; Liu et al., 2016). In this regard, empty Dane particles also have been proposed to be decoy particles for the hepatitis B virus (Rydell et al., 2017).

Several picornavirus procapsids are larger particles compared to the mature virion and similar in shape to the A-particle. Procapsides have some viral epitopes more accessible and consequently can bind nAbs more efficiently. This phenomenon has been observed for the procapsid and mature virion of EV71, revealing that they are antigenically distinct (Shingler et al., 2015). Moreover, a non-nAb has been structurally solved in complex with procapsids but not disclosed for the mature virion (Hewat and Blaas, 2006), which suggests that procapsid may also lead to effects in the modulation of non-nAbs immune response.

**VACCINES**

Inactivated vaccines for PV, HAV, and FMDV are shown to prevent associated illnesses by inducing specific antibody defenses (Salk, 1957). Nevertheless, it was the live-attenuated oral PV vaccine (OPV) responsible for most of the success in controlling polio epidemics. PV attenuation was obtained by serial passage in cell lines from non-human hosts (Sabin, 1965). Rare cases of vaccination-derived paralytic disease can occur as well as vaccines shedding of virulent poliovirus revertant (Platt et al., 2014).

New ideas for picornavirus vaccines, currently under development, exploit evolutionary concepts including (i) codon usage, (ii) mutational robustness, (iii) modification of translational efficiency, and (iv) RdRp fidelity manipulation (Burns et al., 2006; Mueller et al., 2006; Coleman et al., 2008; Le Nouèn et al., 2014; Tulloch et al., 2014; McDonald et al., 2016). In this regard, synonymous codon deoptimization was first applied for PV (Burns et al., 2006; Mueller et al., 2006) and other picornviruses such as FMDV (Diaz-San Segundo et al., 2016). Interestingly, increasing the CpG and UpA dinucleotide frequencies using synonymous codon substitutions leads to
increased activation of the immune system (Tulloch et al., 2014). The maintenance of phenotype despite changing the genotype (mutations), or mutational robustness, can be altered by codon substitutions, correlating less robust viruses with attenuation in mice (Lauring et al., 2012). This approach was further extended using synonymous codons that upon mutation became stop codons (Moratorio et al., 2017). The increase in stop codon mutations in codon-engineered CVB3 during replication led to a loss of infectivity in vitro and attenuation in vivo. These new methods can improve the safety of already existing live-attenuated vaccines and can be broadly applied by re-coding any viral genome (Jorge et al., 2015).

Further elucidation of the mechanisms underlying these phenotypes could be used for rational codon rewiring in combination with increasing CpG and UpA frequencies to activate innate host immunity (Kumagai et al., 2008; Tulloch et al., 2014).

FINAL REMARKS

The control and eradication of pathogenic picornaviruses is an ongoing problem. Picornavirus evolution, ruled by high mutation rates and recombination, has made this viral group genetically and antigenically highly variable. Moreover, changes in virulence represent an unpredictable threat. Important lessons drawn from the polio eradication battle indicate the necessity of a new generation of vaccines (Agol et al., 2016). A holistic approach based on big data and mathematical modeling combining views from structural and evolutionary biology, cellular, and molecular immunology will allow a better understanding of picornavirus–host interactions. This knowledge could have the potential to foresee possible outbreaks and changes on viral virulence. Furthermore, these models may redefine the way new vaccines and antiviral therapies will be designed.

AUTHOR CONTRIBUTIONS

JC and GM conceived the review project, designed figures, wrote the manuscript, and approved it for publication. They both contributed equally to the work.

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REFERENCES

Agol, V., Cello, J., Chumakov, K., Ehrenfeld, E., and Wimmer, E. (2016). Eradicating polio: a balancing act. Science 351:348. doi: 10.1126/science.351.6271.348-b

Andino, R., and Domingo, E. (2015). Viral quasispecies. Virology 479–480, 46–51. doi: 10.1016/j.virology.2015.03.022

Arita, M., Zhu, S.-L., Yoshida, H., Yoneyama, T., Miyamura, T., and Shimizu, H. (2005). A sabin 3-derived poliovirus recombinant contained a sequence homologous with indigenous human enterovirus species C in the viral polymerase coding region. J. Virol. 79, 12650–12657. doi: 10.1128/JVI.79.20.12650-12657.2005

Baggen, J., Hurdiss, D. L., Zocher, G., Mistry, N., Roberts, R. W., Slager, J. J., et al. (2018). Role of enhanced receptor engagement in the evolution of a pandemic acute hemorrhagic conjunctivitis virus. Proc. Natl. Acad. Sci. U.S.A. 115, 397–402. doi: 10.1073/pnas.1713284115

Baltimore, D. (1971). Expression of animal virus genomes. Bacteriol. Rev. 35, 235–241.

Belsham, G. J., Abrams, C. C., King, A. M., Roosien, J., and Vlak, J. M. (1991). Myristoylation of foot-and-mouth disease virus capsid protein precursors is independent of other viral proteins and occurs in both mammalian and insect cells. J. Gen. Virol. 72, 747–751. doi: 10.1099/0022-1317-72-3-747

Bergelson, J. M. (1997). Isolation of a common receptor for coxsackie B viruses and adenoviruses 2 and 5. Science 275, 1320–1323. doi: 10.1126/science.275.5304.1320

Bergelson, J. M., Chan, M., Solomon, K. R., St John, N. F., Lin, H., and Finberg, R. W. (1994). Decay-accelerating factor (CD55), a glycosylphosphatidylinositol-anchored complement regulatory protein, is a receptor for several echoviruses. Proc. Natl. Acad. Sci. U.S.A. 91, 6245–6248.

Bergelson, J. M., Mohanty, J. G., Crowell, R. L., St John, N. F., Lublin, D. M., and Finberg, R. W. (1995). Coxackievirus B3 adapted to growth in RD cells binds to decay-accelerating factor (CD55). J. Virol. 69, 1903–1906.

Bergelson, J. M., Shepley, M. P., Chan, B. M., Hemler, M. E., and Finberg, R. W. (1992). Identification of the integrin VLA-2 as a receptor for echovirus 1. Science 255, 1718–1720.

Bessaud, M., Joffret, M.-L., Blondel, B., and Delpeyrroux, F. (2016). Exchanges of genomic domains between poliovirus and other cocirculating species C enteroviruses reveal a high degree of plasticity. Sci. Rep. 6:38831. doi: 10.1038/srep38831

Borderia, A. V., Isakov, O., Moratorio, G., Henningsson, R., Agüera-González, S., Organti, L., et al. (2015). Group selection and contribution of minority variants during virus adaptation determines virus fitness and phenotype. PLoS Pathog. 11:e1004838. doi: 10.1371/journal.ppat.1004838

Bubeck, D., Fillman, D. J., Cheng, N., Steven, A. C., Hogle, J. M., and Belnap, D. (2005). The structure of the poliovirus 13S cell entry intermediate at 10-angstrom resolution reveals the location of an externalized polypeptide that binds to membranes. J. Virol. 79, 7745–7755. doi: 10.1128/JVI.79.12.7745-7755.2005

Bucciol, G., Moens, L., Payne, K., Wollants, E., Mekahli, D., Levchenko, E., et al. (2018). Chronic Aichi virus infection in a patient with x-linked agammaglobulinemia. J. Clin. Immunol. 38, 748–752. doi: 10.1007/s10875-018-0558-z

Burns, C. C., Shaw, J., Campagnoli, R., Jorba, J., Vincent, A., Quay, J., et al. (2006). Modulation of poliovirus replicative fitness in HeLa cells by deoptimization of synonymous codon usage in the capsid region. J. Virol. 80, 3259–3272. doi: 10.1128/JVI.80.7.3259-3272.2006

Chen, Y. H., Du, W., Hagemeijer, M. C., Takvorian, P. M., Pau, C., Cali, A., et al. (2015). Phosphatidylinerse vesicles enable efficient en bloc transmission of enteroviruses. Cell 160, 619–630. doi: 10.1016/j.cell.2015.01.032

Cifuentes, J. O., Lee, H., Yoder, J. D., Shingler, K. L., Carnegie, M. S., Yoder, J. L., et al. (2013). Structures of the procapsid and mature virion of enterovirus 71 strain 1095. J. Virol. 87, 7637–7645. doi: 10.1128/JVI.03519-12

Coleman, J. R., Papamichail, D., Skiena, S., Fuchter, B., Wimmer, E., and Mueller, S. (2008). Virus attenuation by genome-scale changes in codon pair bias. Science 320, 1784–1787. doi: 10.1126/science.1155761

Combelas, N., Holmblat, B., Joffret, M.-L., Colbère-Garapin, F., and Delpeyrroux, F. (2011). Recombination between poliovirus and coxsackie A viruses of species C: a model of viral genetic plasticity and emergence. Viruses 3, 1460–1484. doi: 10.3390/v3081460

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Coyne, C. B., and Bergelson, J. M. (2006). Virus-induced Abd and Fyn kinase signals permit coxsackievirus entry through epithelial tight junctions. Cell 124, 119–131. doi: 10.1016/j.cell.2005.10.035

Crawford, N. M., and Baltimore, D. (1983). Genome-linked protein VPg of poliovirus is present as free VPg and VPg-PuP in poliovirus-infected cells. Proc. Natl. Acad. Sci. U.S.A. 80, 7452–7455.

Crotty, S., Cameron, C. E., and Andino, R. (2001). RNA virus error catastrophe: direct molecular test by using ribavirin. Proc. Natl. Acad. Sci. U.S.A. 98, 6895–6900. doi: 10.1073/pnas.101185598

Diaz-San Segundo, F. D.-S., Medina, G. N., Ramirez-Medina, E., Velazquez-Salinas, L., Koster, M., Grubman, M. J., et al. (2016). Synonymous deoptimization of foot-and-mouth disease virus causes attenuation in vivo while inducing a strong neutralizing antibody response. J. Virol. 90, 1298–1310. doi: 10.1128/JVI.02167-15

Domingo, E., Sheldon, J., and Perales, C. (2012). Viral quasispecies evolution. Microbiol. Mol. Biol. Rev. 76, 159–216. doi: 10.1128/MMBR.05203-11

Enders, J. F., Weller, T. H., and Robbins, F. C. (1949). Cultivation of the Lansing strain of poliomyelitis virus in cultures of various human embryonic tissues. Science 109, 85–87. doi: 10.1126/science.109.2822.85

Eggers, H. J., and Tamm, I. (1965). Coxsackie A9 virus: mutation specificity in type 3 poliovirus. J. Virol. 73, 4533–4535.

Eggers, H. J., and Tamm, I. (1965). Coxsackie A9 virus: mutation from drug dependence to drug independence. Science 148, 97–98. doi: 10.1126/science.148.3666.97

Elena, S. F., and Sanjuán, R. (2005). Adaptive value of high mutation rates of RNA viruses: separating causes from consequences. J. Virol. 79, 11555–11558. doi: 10.1128/JVI.79.24.11555-11558.2005

Enders, J. F., Weller, T. H., and Robbins, F. C. (1949). Cultivation of the Lansing strain of poliomyelitis virus in cultures of various human embryonic tissues. Science 109, 85–87. doi: 10.1126/science.109.2822.85

Escribano-Romero, E. (2004). Heparan sulphate mediates swine vesicular virus infection of human cells by promoting receptor clustering. J. Gen. Virol. 85, 802–811. doi: 10.1099/vir.0.19603-0

Fitzsimmons, W. J., Woods, R. J., McCrone, J. T., Woodman, A., Arnold, J. J., and Yennawar, M. (2018). A speed–fidelity trade-off determines the evolutionary landscape of foot-and-mouth disease virus. EMBO J. 37, 6249–6259. doi: 10.15252/embj.2018.1826249

Gong, P., et al. (2012). Interaction of coxsackievirus B3 with the full-length coxsackievirus–adenovirus receptor. Nat. Struct. Biol. 8, 874–878. doi: 10.1038/nstb.10-874

Hendry, E., Hatanaka, H., Fry, E., Smyth, M., Tate, J., Stanway, G., et al. (1999). The crystal structure of coxsackievirus A9: new insights into the uncoating mechanisms of enteroviruses. Structure 7, 1527–1538. doi: 10.1016/S0969-2126(99)83843-4

Holetz, E. A., and Blaa, D. (2006). Nonneutralizing human rhinovirus serotype 2–specific monoclonal antibody 2G2 attaches to the region that undergoes the most dramatic changes upon release of the viral RNA. J. Virol. 80, 12398–12401. doi: 10.1128/JVI.01399-06

Hogle, J. M., Chow, M., and Filman, D. J. (1985). Three-dimensional structure of poliovirus at 2.9 Å resolution. Science 229, 1358–1365. doi: 10.1126/science.2994218

Holm-Hansen, C. C., Midgley, S. E., and Fischer, T. K. (2016). Global emergence of enterovirus D68: a systematic review. Lancet Infect. Dis. 16, 64–75. doi: 10.1016/S1473-3099(15)30543-3

Jacobsen, K. H., and Wiersma, S. T. (2010). Hepatitis A Virus seroprevalence by age and world region, 1990 and 2005. Vaccine 28, 6653–6657. doi: 10.1016/j.vaccine.2010.08.037

Jiang, P., Liu, Y., Ma, H.-C., Paul, A. V., and Wimmer, E. (2014). Picornavirus morphogenesis. Microbiol. Mol. Biol. Rev. 78, 418–437. doi: 10.1128/MMBR.00012-14

Jorge, D. M., Mills, R. E., and Lauring, A. S. (2015). CodonShuffle: a tool for generating and analyzing synonymously mutated sequences. Virus Evol. 1:vev012. doi: 10.1093/virus/vev012

Kainulainen, L., Vuorinen, T., Rantakokko-Jalava, K., Osterback, R., and Rauskanen, O. (2010). Recurrent and persistent respiratory tract viral infections in patients with primary hypogammaglobulinemia. J. Allergy Clin. Immunol. 126, 120–126. doi: 10.1016/j.jaci.2010.04.016

Kirkegaard, K. (2017). Unconventional secretion of hepatitis A virus. Proc. Natl. Acad. Sci. U.S.A. 114, 6653–6655. doi: 10.1073/pnas.1701742114

Kloc, A., Rai, D. K., and Rieder, E. (2018). The roles of picornavirus untranslated regions in infection and innate immunity. Front. Microbiol. 9:485. doi: 10.3389/fmicb.2018.00485

Kolatkar, P. R., Bella, J., Olson, N. H., Bator, C. M., Baker, T. S., and Rossmann, M. G. (1999). Structural studies of two rhinovirus serotypes complexed with fragments of their cellular receptor. EMBO J. 18, 6249–6259. doi: 10.1093/emboj/18.2.6249

Kumagai, Y., Takeuchi, O., and Akira, S. (2008). TLR9 as a key receptor for the recognition of DNA. Adv. Drug Deliv. Rev. 60, 795–804. doi: 10.1016/j.addr.2007.12.004

Lauring, A. S., Acevedo, A., Cooper, S. B., and Andino, R. (2012). Codon usage determines the mutational robustness, evolutionary capacity, and virulence of an RNA virus. Cell Host Microbe 12, 623–632. doi: 10.1016/j.chom.2012.10.008

Le Nourou, C., Brock, L. G., Luongo, C., McCarty, T., Yang, L., and Mehedi, M. (2014). Attenuation of human respiratory syncytial virus by genome-scale codon-pair deoptimization. Proc. Natl. Acad. Sci. U.S.A. 111, 13169–13174. doi: 10.1073/pnas.1411290111

Lea, S., Hernández, J., Blakemore, W., Brocchi, E., Curry, S., Domingo, E., et al. (1994). The structure and antigenicity of a type C foot-and-mouth disease virus. Structure 2, 123–139.

Ledinko, N. (1963). Genetic recombination with poliovirus type 1. Virology 20, 107–119. doi: 10.1016/0042-6822(63)90145-4

Lee, H., Cifuentes, J. O., Ashley, R. E., Conway, J. F., Makhov, A. M., Tano, Y., et al. (2013). A strain-specific epitope of enterovirus 71 identified by cryo-electron microscopy of the complex with Fab from neutralizing antibody. J. Virol. 87, 11363–11370. doi: 10.1128/JVI.01926-13

Lee, H., Shingler, K. L., Organiti, L. J., Ashley, R. E., Makhov, A. M., Conway, J. F., et al. (2016). The novel asymmetric entry intermediate of a picornavirus captured with nanodiscs. EMBO J. 35, 26062–26069. doi: 10.15252/embj.2016-872649

Li, C., Wang, J. C.-Y., Taylor, M. W., and Zlotnick, A. (2012). In vitro assembly of an empty picornavirus capsid follows a dodecahedral path. J. Virol. 86, 13062–13069. doi: 10.1128/JVI.01033-12
Mueller, S., Papamichail, D., Coleman, J. R., Skiena, S., and Wimmer, E. (2006). Reduction of the rate of poliovirus protein synthesis through large-scale codon deoptimization causes attenuation of viral virulence by lowering specific infectivity. J. Virol. 80, 9687–9696. doi: 10.1128/JVI.00738-06
Mullapudi, E., Nováček, J., Pákllová, L., Kulich, P., Lindberg, A. M., van Kuppeveld, F. J. M., et al. (2016). Structure and genome release mechanism of the human cardiovascular virus 3 J. Virol. 90, 7628–7639. doi: 10.1128/JVI.01320-15
Murray, M. G., Bradley, J., Yang, X. F., Wimmer, E., Moss, E. G., and Raciuzzi, V. C. (1988). Poliovirus host range is determined by a short amino acid sequence in neutralization antigenic site I. Science 241, 213–215.
Nielsen, A. C., Gyhrs, L., Nielsen, L. P., Pedersen, C., and Böttiger, B. (2013). Gastroenteritis and the novel picornaviruses aichi virus, coxsavirus, sialovirus, and salivirus in young children. J. Clin. Virol. 57, 239–242. doi: 10.1016/j.jcv.2013.03.015
Nishimura, Y., McLaughlin, N. P., Pan, J., Goldstein, S., Hafenstein, S., Shimizu, H., et al. (2015). The suramin derivative NF449 interacts with the 5-fold vertex of the enterovirus A71 capsid to prevent virus attachment to PSGL-1 and heparan sulfate. PLOS Pathog. 11:e1005184. doi: 10.1371/journal.ppat.1005184
Nishimura, Y., Shimojima, M., Tano, Y., Miyamura, T., Wakita, T., and Shimizu, H. (2009). Human P-selectin glycoprotein ligand-1 is a functional receptor for enterovirus 71. Nat. Med. 15, 794–797. doi: 10.1038/nm.1961
Nomoto, A., Detjen, B., Pozzatti, R., and Wimmer, E. (1977). The location of the polio genome in viral RNAs and its implication for RNA synthesis. Nature 268, 208–213.
Organtini, L. I., Makhov, A. M., Conway, J. F., Hafenstein, S., and Carson, S. D. (2014). Kinetic and structural analysis of coxsackievirus B3 receptor interactions and formation of the A-particle. J. Virol. 88, 5755–5765. doi: 10.1128/JVI.00299-14
Palmenberg, A. C. (1990). Proteolytic processing of picornaviral polyprotein. Annu. Rev. Microbiol. 44, 603–623. doi: 10.1146/annurev.mi.44.100190.03131
Pan, J., Narayanan, B., Shah, S., Yoder, J. D., Cifers, J. O., Hafenstein, S., et al. (2011). Single amino acid changes in the virus capsid permit coxsackievirus B3 to bind decay-accelerating factor. J. Virol. 85, 7436–7443. doi: 10.1128/JVI.00503-11
Paul, A. V., and Schultz, A. (1987). Capsid protein VP4 of poliovirus is N-myristoylated. Proc. Natl. Acad. Sci. U.S.A. 84, 7827–7831.
Pettigrew, D. M., Williams, D. T., Kerrigan, D., Evans, D. J., Lea, S. M., and Platt, L. R., Estivariz, C. F., and Sutter, R. W. (2014). Vaccine-associated paralytic poliomyelitis: a review of the epidemiology and estimation of the global burden. J. Infect. Dis. 210, S380–S389. doi: 10.1093/infdis/jiu184
Plevka, P., Hafenstein, S., Harris, K. G., Cifuente, J. O., Zhan g, Y., Bowman, V. D., et al. (2010). Evolutionary dynamics and temporal/geographical correlates of recombination in the human enterovirus type 91, and 30. J. Virol. 84, 9292–9300. doi: 10.1128/JVI.00783-10
Meier, A., van der Sanden, S., Snijders, B. E. P., Jaramillo-Gutierrez, G., Bont, L., van der Ent, C. K., et al. (2012). Emergence and epidemic occurrence of enterovirus 68 respiratory infections in The Netherlands in 2010. Virology 423, 486–507. doi: 10.1016/j.virol.2011.11.021
Mena, L., Perry, C. M., Harkins, S., Rodrigo, F., Gebhard, J., and Whitton, J. L. (1999). The role of B lymphocytes in coxsackievirus B3 infection. Am. J. Pathol. 155, 1205–1215. doi: 10.1002/ajpath.1056223-6
Mendelsohn, C. L., Wimmer, E., and Racaniello, V. R. (1989). Cellular receptor for poliovirus: molecular cloning, nucleotide sequence, and expression of a new member of the immunoglobulin superfamily. Cell 56, 855–865.
Minor, P. (2014). The polio endgame. Hum. Vaccin. Immunother. 10, 2106–2108. doi: 10.4161/hvim.2015515.2014.981115
Minor, P. D., Ferguson, M., Evans, D. M., Almond, J. W., and Icenogle, J. P. (1986). Antigenic structure of polioviruses of serotypes 1, 2, and 3. J. Gen. Virol. 67(1), 7283–1291. doi: 10.1099/0022-1317-67-7-1283
Molla, A., Paul, A. V., and Wimmer, E. (1991). Cell-free, de novo synthesis of poliovirus. Science 254, 1647–1651.
Moratorio, G., Henningsson, R., Barbezante, C., Carrau, L., Borderia, A. V., and Blanc, H. (2017). Attenuation of RNA viruses by redirecting their evolution in sequence space. Nat. Microbiol. 2:17088. doi: 10.1038/nmicrobiol.2017.88
Mosser, A. G., Leippe, D. M., and Rueckert, R. R. (1989). “Neutralization of picornaviruses: support for the pentamer bridging hypothesis,” in Molecular Aspects of Picornavirus Infection and Detection, eds B. L. Semler and E. Ehrenfeld (Washington, DC: American Society for Microbiology (ASM)), 155–167.
Muckelbauer, J. K., Kremer, M., Minor, L., Diana, G., Dutko, F. J., Groarke, J., et al. (1995). The structure of coxsackievirus B3 at 3.5 Å resolution. Structure 3, 653–667. doi: 10.1016/S0969-2121(01)00205-5
Ren, J., Zhu, L., Hu, Z., Gao, Q., Yang, P., et al. (2015). Structures of coxsackievirus A16 capsids with native antigenicity: implications for particle expansion, receptor binding, and immunogenicity. J. Virol. 89, 10500–10511. doi: 10.1128/JVI.01012-15

Rooijen, M., Piirainen, L., Hovi, T., Virtanen, I., Rikkonen, T., Heino, J., et al. (1994). Entry of coxsackievirus A9 into host cells: specific interactions with αvβ3 integrin, the vitronectin receptor. Virology 203, 357–365. doi: 10.1006/viro.1994.1494

Rossmann, M. G. (1989). The canyon hypothesis. Hiding the host cell receptor attachment site on a viral surface from immune surveillance. J. Biol. Chem. 264, 14387–14390.

Rossmann, M. G., Arnold, E., Erickson, J. W., Frankenberger, E. A., Griffith, J. P., Hecht, H.-J., et al. (1985). Structure of a human common cold virus and functional relationship to other picornaviruses. Nature 317, 145–153. doi: 10.1038/317145a0

Roivainen, M., Piirainen, L., Hovi, T., Virtanen, I., Riikonen, T., Heino, J., et al. (2017). Hepatitis B Sabin, A. B. (1965). Oral poliovirus vaccine. History of its development and prospects for eradication of poliomyelitis. JAMA 194, 872–876.

Sherry, B., Mosser, A. G., Colonno, R. J., and Rueckert, R. R. (1986). Use of antibodies and rescues virus infection in vitro. J. Virol. 89, 1900–1908. doi: 10.1128/JVI.03098-14

Simon-Loriere, E., and Holmes, E. C. (2011). Why do RNA viruses recombine? Nat. Rev. Microbiol. 9, 617–626. doi: 10.1038/nrmicro2614

Slater, L., Bartlett, N. W., Haas, J. Z., Zhu, J., Message, S. D., Walton, R. P., et al. (2010). Co-ordinated role of TLR3, RIG-I and MDA5 in the innate response to rhinovirus in bronchial epithelium. PLoS Pathog 6, e1001178. doi: 10.1371/journal.ppat.1001178

Smith, T. J., Chase, E. S., Schmidt, T. J., Olson, N. H., and Baker, T. S. (1996). Neutralizing antibody to human rhinovirus 14 penetrates the receptor-binding canyon. Nature 383, 350–354. doi: 10.1038/383350a0

Stapleton, J. T. (1992). Passive immunization against hepatitis A. Vaccine 10, 545–547. doi: 10.1016/0264-410X(92)90541-Q

Stapleton, J. T., and Lemon, S. M. (1987). Neutralization escape mutants define a dominant immunogenic neutralization site on hepatitis A virus. J. Virol. 61, 491–498.

Staunton, D. E., Merluzzi, V. J., Rothlein, R., Barton, M., Marlin, S. D., and Springer, T. A. (1989). A cell adhesion molecule, ICAM-1, is the major surface receptor for rhinoviruses. Cell 56, 849–853. doi: 10.1016/0092-8674(89)90899-2

Strain, D. M., Ferraro, M. A., Williams, P. A., Harley, C., Shen, S., Stuart, D. I., et al. (2002). Determination of the structure of a decay accelerating factor-binding clinical isolate of echovirus 11 allows mapping of mutants with altered receptor requirements for infection. J. Virol. 76, 7694–7704. doi: 10.1128/JVI.76.15.7694-7704.2002

Sun, M., Li, C., Xu, W., Liao, G., Li, R., Zhou, J., et al. (2017). Immune serum from sabin inactivated poliovirus vaccine immunization neutralizes multiple individual wild and vaccine-derived polioviruses. Clin. Infect. Dis. 64, 1317–1325. doi: 10.1093/cid/cix110

Tan, C. W., Poh, C. L., Sam, L.-C., and Chan, Y. F. (2013). Enterovirus 71 uses cell surface heparan sulfate glycans as an attachment receptor. J. Virol. 87, 611–620. doi: 10.1128/JVI.02226-12

Thompson, A. A., Albertini, R. A., and Peersen, O. B. (2007). Stabilization of poliovirus polymerase by NTP binding and fingers–thumb interactions. J. Mol. Biol. 366, 1459–1474. doi: 10.1016/j.jmb.2006.11.070

Tomassini, J. E., Graham, D., DeWitt, C. M., Lineberger, D. W., Rodkey, J. A., and Colombo, R. J. (1989). CDNA cloning reveals that the major group rhinovirus receptor on HeLa cells is intercellular adhesion molecule 1. Proc. Natl. Acad. Sci. U.S.A. 86, 4907–4911.

Usherwood, E. J., and Nash, A. A. (1995). Lymphocyte recognition of picornaviruses. J. Gen. Virol. 76, 499–508. doi: 10.1099/0022-1317-76-3-499

Verdaguer, N., Blas, D., and Fita, I. (2000). Structure of human rhinovirus serotype 2 (HRV2). J. Mol. Biol. 300, 1179–1194. doi: 10.1006/jmbi.2000.4393

Verdaguer, N., Fita, I., Reithmayer, M., Moser, R., and Blas, D. (2004). X-ray structure of a minor group human rhinovirus bound to a fragment of its cellular receptor protein. Nat. Struct. Mol. Biol. 11, 429–434. doi: 10.1038/nsmb753

Watters, K., and Palmenberg, A. C. (2018). CDHR3 extracellular domains ECI-3 mediate rhinovirus C interaction with cells and as recombinative derivatives, are inhibitory to virus infection. PLoS Pathog. 14:e1007477. doi: 10.1371/journal.ppat.1007477

Williams, C. H., Kajander, J., Hyytiä, T., Jackson, T., Sheppard, D., and Stanway, G. (2004). Integrin alpha v beta 6 is an RGD-dependent receptor for coxsackievirus A9. J. Virol. 78, 6967–6973. doi: 10.1128/JVI.78.13.6967-6973.2004

Woodman, A., Lee, K.-M., Janissen, R., Gong, Y.-N., Dekker, N. H., Shi, S.-R., et al. (2018). Predicting intraserotypic recombination in enterovirus 71. J. Virol. 92:e02057-18. doi: 10.1128/JVI.02057-18

Xiao, Y., Rouzine, I. M., Bianco, S., Acevedo, A., Goldstein, E. F., Farkov, M., Brodsky, L., et al. (2017). RNA virus attenuation by codon pair deoptimization is an artefact of increases in OgP/Ua dinucleotide frequencies. Elife 3:e04531. doi: 10.7554/eLife.04531

Xiao, Y., Dolan, P. T., Goldstein, E. F., Li, M., Farkov, M., Brodsky, L., et al. (2017). RNA virus attenuation by codon pair deoptimization is an artefact of increases in OgP/Ua dinucleotide frequencies. Elife 3:e04531. doi: 10.7554/eLife.04531

Xiao, Y., Dolan, P. T., Goldstein, E. F., Li, M., Farkov, M., Brodsky, L., et al. (2017). RNA virus attenuation by codon pair deoptimization is an artefact of increases in OgP/Ua dinucleotide frequencies. Elife 3:e04531. doi: 10.7554/eLife.04531
required for virus spread and virulence. *Cell Host Microbe* 19, 493–503. doi: 10.1016/j.chom.2016.03.009

Xie, X., Wang, H., Zeng, J., Li, C., Zhou, G., Yang, D., et al. (2014). Foot-and-mouth disease virus low-fidelity polymerase mutants are attenuated. *Arch. Virol.* 159, 2641–2650. doi: 10.1007/s00705-014-1216-z

Xing, L., Huhtala, M., Pietiäinen, V., Käpylä, J., Vuorinen, K., Marjomäki, V., et al. (2004). Structural and functional analysis of integrin α2 I domain interaction with echovirus 1. *J. Biol. Chem.* 279, 11632–11638. doi: 10.1074/jbc.M312441200

Yamayoshi, S., Yamashita, Y., Li, J., Hanagata, N., Minowa, T., Takemura, T., et al. (2009). Scavenger receptor B2 is a cellular receptor for enterovirus 71. *Nat. Med.* 15, 798–801. doi: 10.1038/nm.1992

Ye, X., Fan, C., Ku, Z., Zuo, T., Kong, L., Zhang, C., et al. (2016). Structural basis for recognition of human enterovirus 71 by a bivalent broadly neutralizing monoclonal antibody. *PLoS Pathog.* 12:e1005454. doi: 10.1371/journal.ppat.1005454

Yen-Murphy, M., and Almond, J. W. (1996). “Picornaviruses,” in *Medical Microbiology*, ed S. Baron (Galveston, TX: University of Texas Medical Branch).

Zarocostas, J. (2018). WHO keeps polio on the international health emergency list. *Lancet* 392:2425. doi: 10.1016/S0140-6736(18)33115-5

Zocher, G., Mistry, N., Frank, M., Hähnlein-Schick, I., Ekström, J.-O., Arnborg, N., et al. (2014). A sialic acid binding site in a human picornavirus. *PLoS Pathog.* 10:e1004401. doi: 10.1371/journal.ppat.1004401

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