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Mpox (formerly monkeypox): pathogenesis, prevention, and treatment

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In 2022, a global outbreak of Mpox (formerly monkeypox) occurred in various countries across Europe and America and rapidly spread to more than 100 countries and regions. The World Health Organization declared the outbreak to be a public health emergency of international concern due to the rapid spread of the Mpox virus. Consequently, nations intensified their efforts to explore treatment strategies aimed at combating the infection and its dissemination. Nevertheless, the available therapeutic options for Mpox virus infection remain limited. So far, only a few numbers of antiviral compounds have been approved by regulatory authorities. Given the high mutability of the Mpox virus, certain mutant strains have shown resistance to existing pharmaceutical interventions. This highlights the urgent need to develop novel antiviral drugs that can combat both drug resistance and the potential threat of bioterrorism. Currently, there is a lack of comprehensive literature on the pathophysiology and treatment of Mpox. To address this issue, we conducted a review covering the physiological and pathological processes of Mpox infection, summarizing the latest progress of anti-Mpox drugs. Our analysis encompasses approved drugs currently employed in clinical settings, as well as newly identified small-molecule compounds and antibody drugs displaying potential antiviral efficacy against Mpox. Furthermore, we have gained valuable insights from the process of Mpox drug development, including strategies for repurposing drugs, the discovery of drug targets driven by artificial intelligence, and preclinical drug development. The purpose of this review is to provide readers with a comprehensive overview of the current knowledge on Mpox.

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INTRODUCTION

Mpox (formerly monkeypox) is an emerging zoonotic disease caused by Mpox virus infection, which affects both humans and animals.^{1,2} The virus was first discovered in monkeys in 1958 and has since been detected in a variety of animal species.³ The first human case of Mpox infection was diagnosed in 1970 in the Republic of the Congo, located in Central Africa.^{4–6} Subsequently, Mpox has predominantly circulated in Central and West Africa, with transmission occurring between animals (primarily primates and rodents), as well as between animals and humans, and through human-to-human contact.^{7–9} In recent years, the rapid globalization, population movement, and deepening trade networks have contributed to the international dissemination of Mpox, resulting in outbreaks in various countries worldwide.^{10–12} Notably, in 2022, a global outbreak of Mpox affected 110 countries and regions.¹³ Although the World Health Organization declared that Mpox outbreaks no longer constituted an “a Public Health Emergency of International Concern in May 2023,”¹⁴ it is important to highlight that certain regions in Asia have experienced a rise in Mpox cases due to the virus’s rapid evolution and increased international travel (Fig. 1).^{15–17} Cases of Mpox virus infection have been reported in cities such as Beijing, Guangzhou, and Shenyang.¹⁸ With the continuous increase in the number of

infected patients, Mpox, a disease that was previously neglected, has re-entered the public attention.¹⁹ To effectively combat the disease, a renewed comprehension of Mpox is necessary. This review presents a comprehensive overview of Mpox, including its transmission patterns, pathogenesis, genome organization, and antiviral drugs that have been studied for their activity against Mpox over the past few decades, both in vivo and in vitro. Additionally, it provides helpful insights for the prevention and control of worldwide Mpox outbreaks by summarizing the valuable experiences gained from the development of anti-Mpox strategies, such as drug repurposing, drug target discovery, and the identification of potential drug targets.

TRANSMISSION

The natural hosts of Mpox virus include some rodents and primates in central Africa. Early human infections are typically linked to contact with infected animals, including exposure to mucous membranes, body fluids, tissues, or consumption of undercooked meat. Transmission can also occur through scratches or bites from infected animals.²⁰ Human-to-human transmission is believed to occur through direct contact with respiratory droplets from infected individuals.^{21–23} Furthermore, vertical transmission

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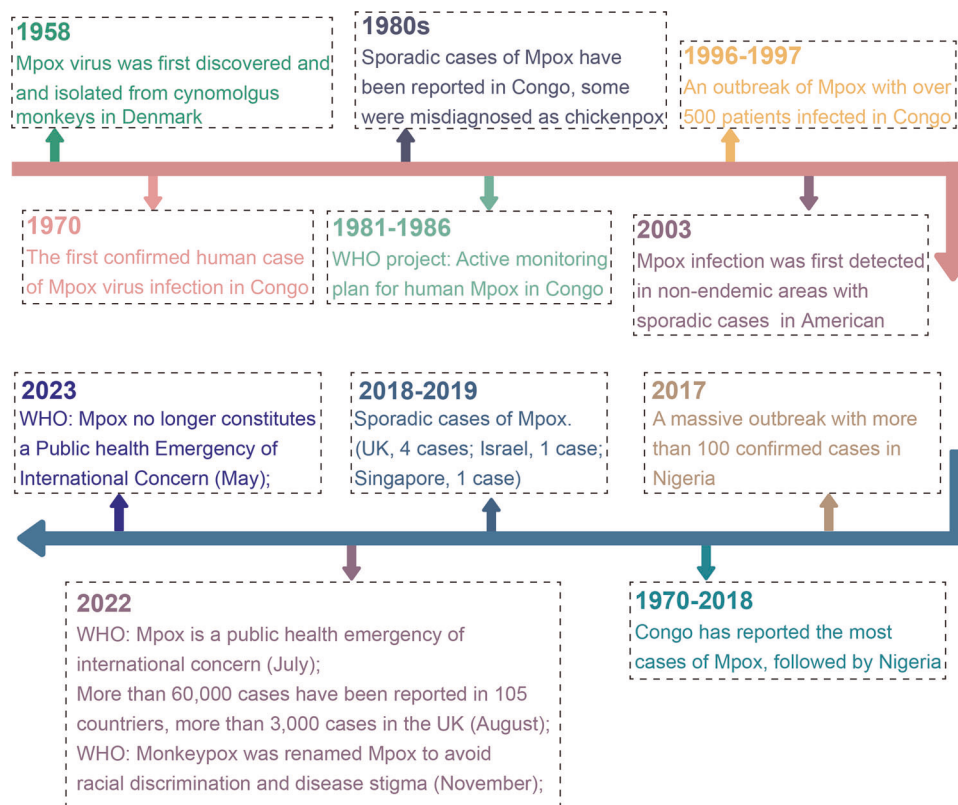


Fig. 1 The timeline of the historical review and major milestones in Mpox

of Mpox virus can occur from infected mothers to their newborns. (Fig. 2a)^{24,25} This recent outbreak of Mpox infection was the largest reported epidemic outside of Africa, unlike previous outbreaks. In the past, Mpox infection was only diagnosed after contact with infected animals or traveling to regions affected by Mpox.^{26–28} However, in this current epidemic, most Mpox cases are not associated with contact with infected animals or travel, but with sexual contact between individuals.²⁹ Over the past two years, the majority of reported cases of Mpox outbreaks have involved homosexual or bisexual males. A research study reported that 98% of cases were among homosexual or bisexual males, with 41% of them co-infected with human immunodeficiency virus (HIV). Additionally, 73% of the observed lesions occurred in the anal and genital regions.^{30,31} The incubation period of Mpox is ~7–14 days, with symptoms lasting for 14–21 days.^{32,33} The prolonged incubation period poses significant challenges for accurate diagnosis, potentially leading to delayed medical attention, disease progression, and further transmission of the virus.^{34,35}

PATHOGENESIS

Mpox is a self-limiting disease, and the severity of infection can be influenced by various factors, such as the specific viral strain, individual immune status, and potential complications that may arise.³⁶ Common early symptoms of Mpox virus infection include pain, fever, fatigue, and lymphadenectasis, with significant inguinal lymphadenectasis often observed.^{37–40} The presence of lymphadenectasis can help to distinguish Mpox virus infection from other orthopoxviruses infection.⁴¹ Furthermore, understanding the transmission mode is essential in establishing effective measures to combat Mpox. Following exposure to the respiratory secretions or body fluids of Mpox patients, the Mpox virus enters nearby tissues through mucous membranes (such as ocular, respiratory, oral, urethral, and rectal) or broken skin.^{42,43} It then

spreads throughout the body via tissue-resident immune cells and draining lymph nodes.^{42,44} This constitutes the latent period for Mpox virus infection, which typically lasts up to two weeks. Throughout this period, individuals infected with Mpox are generally asymptomatic and devoid of lesions. Following the latent period, individuals infected with Mpox virus begin to exhibit atypical symptoms, including fever and chills, headache, muscle pain, and lymphadenectasis. These initial prodromal symptoms of Mpox typically last for three days. After the fever and lymphadenectasis, rashes begin to appear on the head and face, and gradually spread throughout the body. The rash evolves from papules to vesicles and pustules, and ultimately forming crusts that heal, leaving behind scars. This progressive rash phase lasts about 2–4 weeks.^{43,45,46} In the current outbreak of Mpox among men who have sex with men (MSM), some unusual clinical signs have been observed with rashes appearing primarily around the genital or anal area and subsequently spreading throughout the body.^{27,47} Severe cases of Mpox virus infection can lead to complications such as hemorrhagic disease, necrotic disease, obstructive disease, inflammation of vital organs, and septicemia. The case fatality rate of Mpox in non-epidemic regions during 2022 was ~0.04%. (Fig. 2b)^{3,25,38,48–50} Immunocompromised individuals, including children, older adults, and those with immunodeficiencies (such as HIV patients and individuals using immunosuppressive medications), are more susceptible to experiencing these severe manifestations. In addition, immunocompromised individuals are more likely to contribute to the evolution of Mpox, making it increasingly adaptable to human hosts and resulting in widespread transmission (Fig. 2c).^{46,51–54}

VIRUS MORPHOLOGY AND GENOME

Mpox is caused by Mpox virus, a member of the genus *orthopoxvirus* in the family Poxviridae, is characterized by its brick-shaped or oval morphology with a diameter of

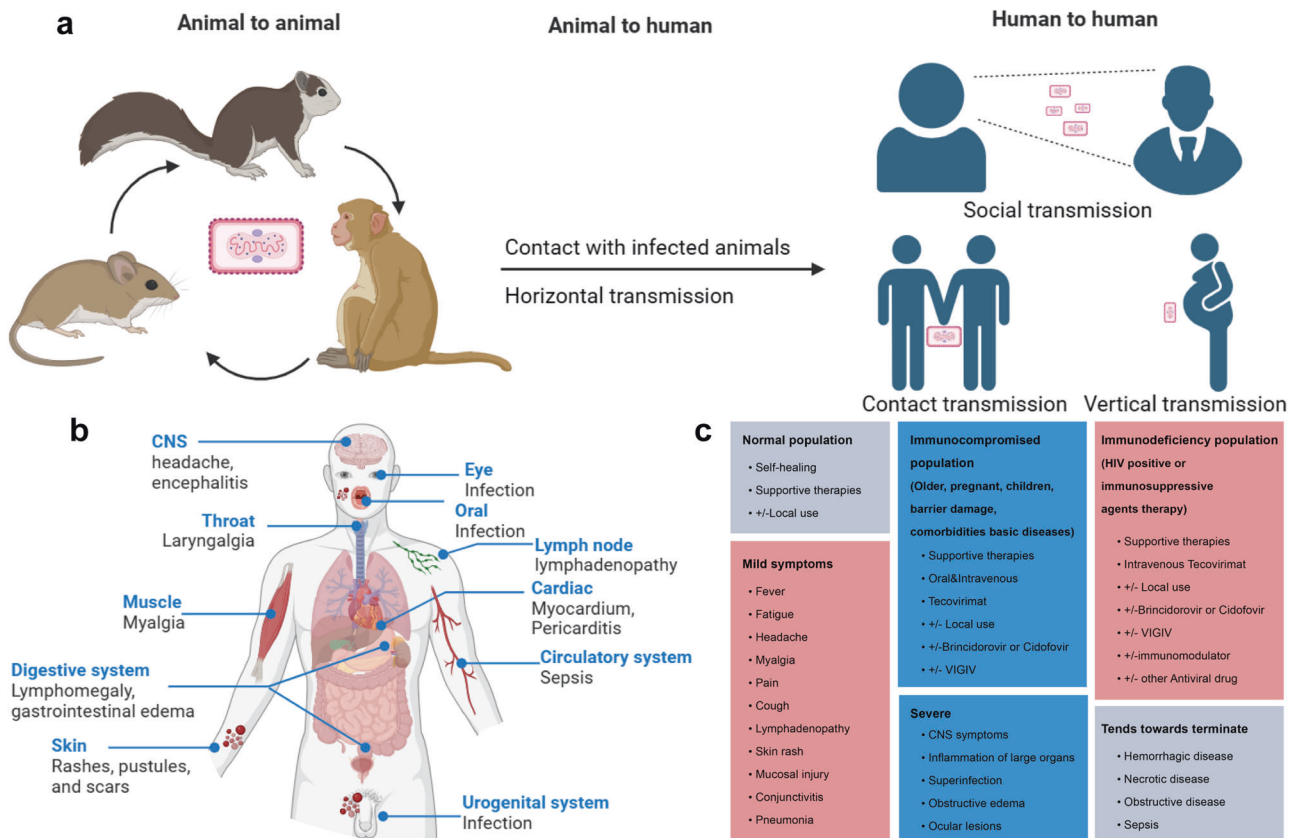


Fig. 2 The epidemiological characteristics, pathogenesis, clinical diagnosis, and treatment of Mpox. **a** The transmission of Mpox occurs through animal-to-animal, animal-to-human, and human-to-human routes. **b** Clinical symptoms typically manifest after Mpox infection. **c** Symptoms of Mpox infection may vary based on the immune status and clinical treatment options and clinical treatment options are listed

~200–250 nm.^{55,56} Its genome consists of a linear, double-stranded DNA with a length of ~197 kb and encoding about 180 proteins.⁵⁷ Additionally, Mpox virus possesses dumbbell-shaped nucleocapsid enveloped by ovoid lipid-containing particles. The genomic structure of Mpox virus closely resembles that of other orthopoxviruses, characterized by a highly conserved central core region, variable regions at the left and right ends, and a tandemly repeated inverted terminal repeat. (Fig. 3)^{58,59} The central core region of Mpox virus shares more than 90% sequence homology with other orthopoxviruses, particularly within the open reading frame (ORF) located between C10L and A25R.^{57,60} Species and strain-specific characteristics of orthopoxviruses are often found in the variable regions at the ends of the genome. A better understanding about these ORFs may provide insights into its host tropism, pathogenesis, and differences in immune regulation.²⁸ Based on a genomic and phylogenetic analysis conducted in 2022, the prevalent strain of Mpox virus was identified as belonging to the B.1 lineage of the West African clade. The B.1 lineage exhibits multiple mutations in genes associated with virulence, host recognition, and immune evasion.⁶¹ In comparison to previously obtained complete genome sequences of Mpox virus isolated in Nigeria from 2017 to 2018, the Mpox virus strains that emerged in 2022 exhibited a higher number of single nucleotide polymorphisms (SNPs). The Mpox virus strain isolated in 2022 exhibit ~50 SNPs, indicating an approximately 6–12-fold increase in the predicted substitution rate of Mpox virus compared to the strains isolated from 2018–2019 (1–2 nucleotide substitutions per genome every year).^{62,63} The functional significance of these mutations is yet to be fully understood, but this high mutation rate may help explain the sudden appearance and heightened transmissibility of Mpox virus in non-endemic regions.

THE LIFE CYCLE OF MPOX VIRUS AND THE DISCOVERY OF ANTI-MPOX VIRUS DRUGS

The process of Mpox virus infection and replication can be summarized into three distinct stages: 1) virus invasion; 2) virus replication and synthesis; 3) virus assembly, maturation and release.^{64–66} Targeting any stage of the Mpox virus lifecycle holds promise for the development of effective antiviral interventions against Mpox virus.

Anti-Mpox virus drugs targeting the invasive phase

The development of antiviral drugs begins with a thorough understanding of the complete life cycle of the virus (Fig. 4). In the early stages of Mpox virus infection, two distinct infectious viral particles are present: extracellular enveloped virions (EEV) and intracellular mature virions (IMV).⁶⁷ These viral particles vary in surface glycoprotein and envelope membrane composition, with IMV exhibiting a single-membrane structure and EEV possessing a double-membrane structure. IMV are released only upon cell lysis and enters host cells through direct fusion and endocytosis,^{68,69} while EEV enters via membrane fusion.^{70–72} IMV are the most abundant viral particles in terms of quantity, due to the absence of a lipid membrane, which gives them a simpler and more robust structure.⁷³ This enhances their resistance to external damage, prolonging their survival time outside the host. However, the exposed surface proteins of IMV trigger higher production of neutralizing antibodies and activate complement responses.^{74–76} Additionally, these exposed surface proteins enhance the recognition and inactivation of Mpox virus by immune cells. In contrast, EEV possesses an additional lipid membrane layer on their surface, enabling better intracellular dissemination.⁶⁹ The pox virus can utilize lipid rafts on the lipid membrane to enter host cells, and cholesterol is one of the important components responsible for maintain the structure and function of lipid rafts.^{77,78} Amphotericin B,

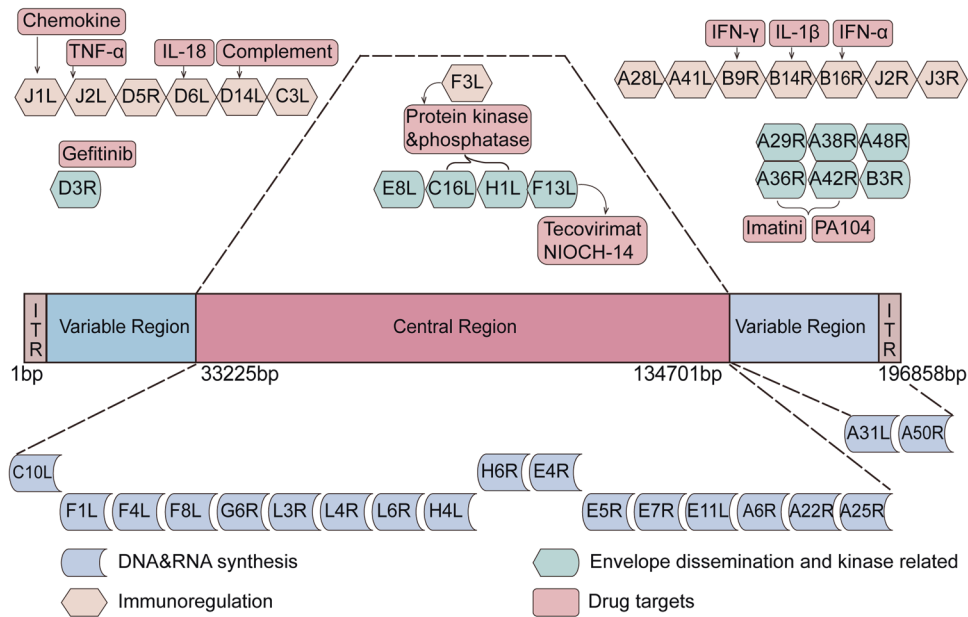


Fig. 3 The genome structure and potential antiviral targets of Mpox virus. The Mpox virus genome consists of a double-stranded linear DNA comprising approximately 196,858 base pairs. It consists of a central recognition region, two variable region, and two terminal inverted terminal repeats (ITRs) (Monkeypox virus strain Zaire, GenBank accession number: AF380138.1, web link: <https://www.ncbi.nlm.nih.gov/nuccore/17529780>). In the genome map, target genes implicated in the interaction between Mpox virus and antiviral drugs are listed. Most essential genes are located in the central region of the genome

a long-standing antibiotic used for the treatment of fungal infections, can sequester cholesterol within host cell membranes, disrupting the integrity of lipid raft and potentially inhibiting Mpox virus infection.⁷⁹ Additionally, cholesterol-lowering drugs such as statins and PCSK9 inhibitors may exhibit antiviral activity by modulating cellular cholesterol levels.⁸⁰ Mpox virus attaches to mucous membranes and damaged skin, where a high concentration of glycosaminoglycans (GAGs) are present. GAGs serve as primary attachment receptors for host cells. EEV particles of Mpox virus interact with GAGs and enter host cells. Marine sulfated polysaccharides are natural analogs of GAGs that competitively bind to the host cell membrane surface, thereby preventing the attachment and entry of Mpox virus.

Since no specific receptor for Mpox virus on the host cell membrane has been found so far, several envelope proteins that play a key role in the invasion of host cells by Mpox virus, may be attractive targets for the development of anti-Mpox virus drugs. He et al. evaluated the binding capacity of eight marine sulfated polysaccharides to the surface envelope protein A35R of Mpox virus using surface plasmon resonance technology. The research findings indicated that some sulfated polysaccharides exhibited competitive binding effects and anti-Mpox virus activity.⁸¹ In another study, Li et al. inoculated BALB/c mice with recombinant A35R protein and purified the A35R immune serum, which showed high neutralizing activity against two types of vaccinia virus (VACV)-EEV.⁸² Moreover, several IMV surface membrane proteins, including I5L, E8L, and A43R have been identified through whole-genome sequencing, and may facilitate the entry of Mpox virus into host cells through receptor and membrane fusion.^{57,60} Although the exact mechanisms of interaction between these proteins and the host are not fully understood, they could potentially serve as targets for future anti-Mpox virus discovery. Further research is needed to unravel the specific roles of these proteins in Mpox virus infection.

Antiviral drugs that influence viral replication and synthesis After IMV or EEV enter the host cell, the exposed viral core is transported to the periphery of the cell nucleus through microtubule structures at an average speed of 52 $\mu\text{m}/\text{min}$.⁸³ The

viral core consists of the central viral genome and an enveloped nucleocapsid. The mechanism of nucleocapsid uncoating involves ubiquitination of the nuclear capsid proteins and degradation by proteasomes.^{84–86} Once uncoating is completed, the Mpox virus genome begins efficient replication, rapidly amplifying like a factory.^{87–91}

Currently, researchers are devoted to developing anti-Mpox drugs by interfering with the DNA or RNA synthesis of the viral genome.⁹² Nucleoside analogs are chemical compounds that have a similar structure to naturally occurring nucleosides. These drugs competitively bind to the viral DNA or RNA polymerase, disrupting the replication process by causing termination of the DNA or RNA chain synthesis. Due to their ability to inhibit viral replication, these drugs often exhibit broad-spectrum antiviral activity.^{93,94} Cidofovir, a non-cyclic monophosphate nucleoside analog, can be used for the treatment of orthopoxviruses and demonstrate potent antiviral activity in vitro (Mpox virus, effective concentration half maximal (EC_{50}) = 2.52 $\mu\text{g}/\text{mL}$, Selectivity index (SI) = 15, in human embryonic lung fibroblasts) and in vivo (Mpox virus, 5 mg/kg, cynomolgus macaques, intraperitoneal injection; Mpox virus, 5 mg/kg, human, intravenous).^{95–98} Following the Mpox outbreak in 2022, Cidofovir was rapidly employed in clinical trials for the treatment of Mpox.^{99–101} However, Cidofovir is a divalent anion with low bioavailability. In patients with impaired renal function or undergoing renal replacement therapy, its metabolites can accumulate in proximal renal tubular cells, leading to kidney damage.^{102–104} In order to overcome the limitations of Cidofovir, its derivative Brincidofovir has been developed. Brincidofovir has been modified using lipid conjugation technology, resulting in improved cellular uptake and conversion capabilities, it was approved by the FDA in 2021 for the treatment of smallpox.¹⁰⁵ Unlike Cidofovir, Brincidofovir does not require metabolism through the renal anion transport system, thus exhibiting higher bioavailability and no significant nephrotoxicity in vitro (VACV, EC_{50} = 0.19 μM , in vero cells) and in vivo (Mpox virus, 10 mg/kg, mice, gastric gavage; Mpox virus, 200 mg, human, oral).^{105–109} However, Brincidofovir still presents some adverse reactions such as gastrointestinal reactions and liver function injury.^{101,110} Apart

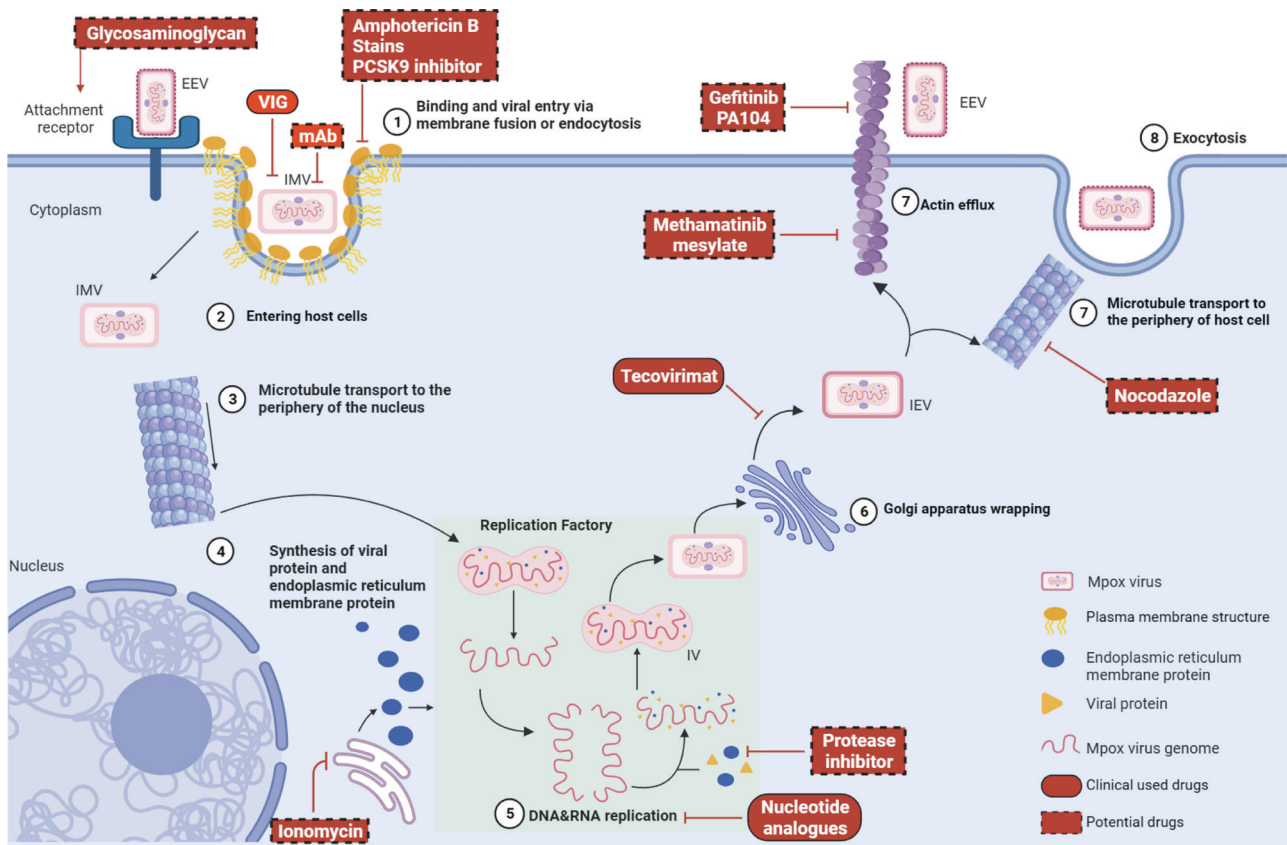


Fig. 4 The life cycle of Mpox virus replication in hosts and potential targets for anti-Mpox virus drugs. The complete life cycle of Mpox virus infection: from entry into host cells to excretion. Briefly, both EEV and IMV viral particles penetrate the host membrane through membrane fusion and endocytosis. Mpox virus viral particles utilize glycosaminoglycans as host receptor. IMV particles enter the cytoplasm and are transported to the perinuclear replication factory via microtubules. The released Mpox virus genome serves as a template for DNA replication. Furthermore, IMV are enveloped by the Golgi apparatus to form IEV, and are transported to the cell surface via actin or microtubules. Part of the important drugs targeting each stage of the replication process are listed. EEV extracellular enveloped virions, IMV intracellular mature virions, IEV intracellular enveloped virions, IV immature virion

from Brincidofovir, other compounds based on structural modifications of Cidofovir have been developed.^{111,112} For instance, NPP-669 is synthesized by linking a long-chain sulfonate to Cidofovir. This modification improves its solubility in water and affinity for lipid through alkyl chain modification. As a result, this structural modification enhances the metabolic stability and bioavailability while reducing nephrotoxicity. It has shown enhanced antiviral effectiveness in vitro (vaccinia, $EC_{50} = 8.95 \mu\text{M}$, in HFF cells) and in vivo (cytomegalovirus, 3 mg/kg, mice, intraperitoneal injection).¹¹³ Ribavirin, a well-known nucleoside analog, blocks viral nucleotide synthesis and thus inhibits viral replication and transmission. It has broad-spectrum antiviral efficacy against various DNA and RNA viruses, including Mpox virus. Studies have shown that ribavirin can impede the replication of orthopoxviruses in vitro (Mpox virus, $EC_{50} = 5.9 \mu\text{g/mL}$, in Vero cells) and in vivo (cowpox virus, 50 mg/kg, mice, subcutaneous injection).^{114,115} However, further clinical studies are needed to assess its effectiveness in Mpox patients are needed. Although nucleotide analogs possess potent antiviral effects, they also have the potential to induce viral resistance. Recently, resistant Mpox virus strains to Cidofovir have been identified.^{116–118} Consequently, researchers are currently focused on the development of new nucleotide analogs such as KAY-2-41, a novel guanosine analog developed by Sophie et al. This analog exhibits potent antiviral activity against VACV in vitro (VACV-WR, $EC_{50} = 0.8 \mu\text{M}$, SI = 18, in human embryonic lung cells) and in vivo (VACV-WR, 50 mg/kg, mice, intraperitoneal injection), remaining effective against Cidofovir-resistant strains.^{119,120} This discovery provides a

new therapeutic approach for nucleotide-resistant Mpox virus strains that are currently in use. However, further research and evaluation are needed for the clinical application of this novel nucleotide analog. The DNA-dependent RNA polymerase (DdRp) plays a crucial role in catalyzing the replication process of DNA viruses in the cytoplasm. Due to its biological significance, DdRp is considered a potential therapeutic target for Mpox virus. Through computer modeling of DdRp, along with techniques such as molecular dynamics simulations, docking, and computational screening, potential inhibitors of DdRp can be efficiently identified. Several small molecule compounds with inhibitory activity against DdRp have been discovered through computer-assisted drug design.^{121–123} However, further researches are needed to validate these identified candidate compounds, evaluate their safety and effectiveness, and ultimately progress them to the clinical application stage. The endoplasmic reticulum (ER) plays a crucial role in enveloping and stabilizing the viral genome. Electron microscopy observations have shown that the replication factory is surrounded by a significant amount of ER membrane. The ER plays a key role in the synthesis of viral membrane proteins.^{124,125} Subsequently, these membrane proteins, together with other viral structure proteins, enter into the viral factory, encapsulate the core genes, forming crescent-shaped structures.^{125,126} Moreover, the presence of the ER is important for maintaining the stability of the viral genome. Studies have indicated that ionomycin disrupts the integrity of ER in vitro, resulting in the inability of ER membrane proteins to enclose the exposed genome. This exposure triggers an immune response,

leading to the degradation of viral DNA and significantly impacting VACV DNA replication.¹²⁷ This discovery highlights the essential role of the ER in maintaining Mpx virus genome stability and facilitating viral replication. Based on these findings, compounds that effectively inhibit the formation of ER membrane proteins could also serve as potential antiviral drugs against Mpx virus.

Antiviral drugs that affect virus assembly, maturation, and release Within the replication factory, these crescent-shaped structures develop into ellipsoidal or spherical shapes, representing immature virion (IV) particles.^{56,127} IV particles undergo the proteolytic cleavage of several capsid proteins and the condensation of the core, resulting in the formation of mature virus particles known as IMV. These IMV are abundant within the host cells. As IMV proliferate, they cause the lysis of the host cells, subsequently releasing IMV viral particles. Additionally, a portion of the IMV exits the virus factory through the microtubule organizing center and becomes enveloped by the trans-Golgi network (TGN) or the nuclear membranes, forming intracellular enveloped virus (IEV).^{128–131} Compared to the single-layered membrane structure of IMV, IEV possesses a three-layered membrane structure. During the early stage of infection, a majority of IMV are enveloped to form IEV. However, in the later stages of infection, IMV become the predominant form, possibly due to the depletion of TGN and nuclear membranes.^{132,133} Once IEV reach the peripheral region of the cell, the viral envelope fuses with the host cell membrane, forming cell-associated enveloped viruses (CEV) through the process of exocytosis.¹³⁴ Virus particles remaining on the surface of the host cell are referred to as CEV, whereas those released into the extracellular environment are referred to as EEV.¹³⁵ The ratio of EEV to CEV depends on the specific virus strain and host cell type. While the mechanism by which EEV released from infected cells further infect neighboring cells is not fully understood, researchers have discovered that the actin tails can form and extend a long distance outside the cell. EEV can utilize actin tails to enter adjacent cells, establishing bridges between the actin tails and neighboring cells, thereby facilitating efficient viral spread.^{136–138}

In order to reach the cellular plasma membrane, the release of viruses requires the involvement of the actin cytoskeleton.¹³⁹ Currently, two mechanisms have been proposed to explain how IEV traverse the actin cytoskeleton. The first mechanism is actin polymerization-induced assembly. Upon viral infection, host cells trigger the polymerization of actin, resulting in the formation of filamentous structures known as actin tails. Failure to form actin tails hinders virus migration, adhesion, and intercellular spread. Several studies suggest that tyrosine phosphorylation plays a pivotal role in the formation of actin tails.^{140,141} The second mechanism is microtubule transport. IEV reach the cell surface through microtubule-mediated transport. Studies conducted by Hollinshead et al. observed the movement trajectory of viral particles labeled with green fluorescent protein.¹⁴² They found that viral particles co-localized with microtubules and exhibited an average velocity of 60 $\mu\text{m}/\text{min}$, consistent with the speed of microtubule transport. This speed far exceeds the transport rate of actin tails (2.8 $\mu\text{m}/\text{min}$).^{143,144} The movement of viral particles to the cell surface can be hampered by the microtubule-depolymerizing drug nocodazole in vitro.^{145–147} Based on this evidence, it is evident that microtubule transport plays a crucial role in the externalization of IEV to the cell surface. Disruption of microtubule structures may contribute to reducing the export of virus particles and inhibiting the spread of infection. As we mentioned earlier, the actin tails play an important role in the process of Mpx virus infecting of neighboring cells. Several drugs have been reported to inhibit the formation of actin tails, including the anti-cancer drug imatinib mesylate, which has shown anti-*orthopoxvirus* activity in vitro.¹⁴⁸ Furthermore, a compound named PA104, identified by Lalita, has been shown

to significantly inhibit the formation of actin tails, thereby reducing viral release and spread in vitro (VACV, $EC_{50} = 0.8 \mu\text{M}$, $SI > 800$, in BSC40 cells).¹⁴⁹ Epidermal growth factors (EGFs) encoded by orthopoxviruses, play a crucial role in intercellular virus transmission.^{150,151} EGF can activate the EGFR/MEK/FAK signaling pathway, promoting intercellular virus transmission and facilitating rapid movement of infected cells.¹⁵² This increases the likelihood of contact between infected and uninfected cells, ultimately enhancing the transmission efficiency of orthopoxviruses.^{153,154} Experimental findings demonstrate that the use of EGFR inhibitor gefitinib and MEK inhibitor effectively reduces the area of virus-infected plaques in vitro (VACV, $EC_{50} = 4.93 \mu\text{M}$, in Hep2 cells).¹⁵⁵ Notably, gefitinib reduces actin tail formation by 1.6-fold and decreases infected cell migration efficiency by fourfold.¹⁵³

The viral-encoded membrane proteins of orthopoxviruses also play a significant role in viral transmission. Research has demonstrated that the Mpx virus A36R protein is crucial role in intercellular virus transmission and the release of EEV into the surrounding environment.¹⁵⁶ Mohammad et al. identified three peptides that effectively targeting A36R have been identified and exhibit effective antiviral activity against Mpx virus. These peptides show a high affinity for A36R while being non-allergenic and non-toxic.¹⁵⁷ Furthermore, a study on vaccinia virus revealed that the absence of A33R and A34R proteins increases EEV production,¹⁵⁸ while the absence of A36R and B5R proteins decreases EEV production.¹⁵⁹ This suggests that Mpx virus-encoded proteins with homology with A36R or B5R could potentially be valuable antiviral targets for future therapeutic strategies against Mpx virus. The VP37 protein, encoded by the F13L gene, is a catalytic protein involved in the intracellular envelopment of mature viral particles.^{160–162} It is widely distributed and highly conserved among orthopoxviruses, playing a crucial role in the in the formation of IMV within the TGN.¹⁶⁰ It is essential for the virus's pathogenicity and infectivity. Deletion of the F13L gene hinders the membrane envelopment process of orthopoxviruses, limiting their further spread.¹⁶³ Tecovirimat, initially named ST-246, is a compound discovered through high-throughput screening (HTS) that exhibits potent antiviral activity against Mpx virus in vitro (Mpx virus, $EC_{50} = 0.01 \mu\text{M}$, in Vero cells) and in vivo (Mpx virus, 10 mg/kg, cynomolgus macaques, gavage; Mpx virus, human, 600 mg bid, oral).^{164–167} Tecovirimat functions by inhibiting the synthesis of the VP37 protein, thereby impeding the maturation process of orthopoxviruses and disrupting their envelopment and release.^{168–170} Tecovirimat primarily restricts intercellular virus spread without affecting the viral replication process. It has been identified as one of the most effective drugs against orthopoxviruses. Following the Mpx outbreak in 2022, the U.S. Food and Drug Administration granted emergency approval for Tecovirimat as a therapeutic drug for Mpx, demonstrating its promising clinical efficacy.¹⁷¹ Between May 2022 and February 2023, Germany reported 12 severe cases of Mpx, in patients with either severe immunosuppression due to HIV infection ($CD4 + T$ cell count below 200/ μL) or significant systemic involvement (over 100 skin lesions). These patients underwent treatment with tecovirimat, and clinical outcomes revealed complete recovery in all patients, with Tecovirimat exhibiting excellent tolerability.¹⁷² In addition to Tecovirimat, another compound called NIOCH-14, serving as a precursor to Tecovirimat, has shown specific antiviral activity against orthopoxviruses and shares structural similarity with Tecovirimat in vitro (Mpx virus, $EC_{50} = 0.013 \mu\text{g}/\text{mL}$, in Vero cells) and in vitro (Mpx virus, 40 mg/kg, marmot, oral). Unlike tecovirimat, NIOCH-14 offers a simpler synthesis route, thereby reducing costs and technical requirements. This natural advantage makes NIOCH-14 promising candidate for future wide-ranging applications.^{100,173–175} The current round of Mpx outbreak is highly mutable and still evolving. Although Tecovirimat is effective in current clinical use, it

may pose a risk of resistance in the future. Discovering drugs with a new mechanism of action could provide a solution to the drug resistance problem. PA104 suppressed the formation of extracellular virus particle and viral propagation by inhibiting actin tail formation. Its mechanism of action differed from that of Tecovirimat. Of note, PA104 has demonstrated the ability to inhibit the replication of Tecovirimat-resistant VACV strains *in vitro*.¹⁴⁹

IMMUNOTHERAPY IN MPOX

Mpox virus-induced immunopathology leads to adverse outcomes in clinical, and immunotherapy for Mpox has the potential to reduce severe cases. Antibody-based therapeutics, immune cell, immune effector molecules, and Modulation of cellular signal transduction are potential immunotherapies. Combination antiviral drugs with immunotherapy may be more effective and provide greater clinical benefit than single antiviral therapy alone.^{176–178}

Immune globulin and antibodies

Antibody-based therapeutics have shown significant progress in treating certain infectious diseases and currently being actively explored.^{176,179} Immune globulin, convalescent plasma, and neutralizing antibodies offer promising options as adjunctive treatments for cases with insufficient antiviral drug efficacy in severe patients.^{180–182} Notably, individuals who have been previously vaccinated with the smallpox vaccine produce more neutralizing antibodies that may be cross-protective against Mpox virus infection.¹⁸³ Thus, some countries have approved the intravenous administration of vaccinia immune globulin (VIGIV) for managing complications associated with smallpox vaccination.¹⁸⁴ For individuals with severe T cell functional immunodeficiency due to contraindications to smallpox vaccination, VIGIV can be considered as a prophylactic measure *in vitro* (5 mg, incubated with VACV) and *in vivo* (VACV, 400 mg/kg, mice, intravenous) and in one clinical case (6000 U/kg, single-dose

intravenous).^{185,186} Although convalescent plasma (CP) therapy shows therapeutic potential for other infectious viruses,^{187,188} there is currently no available literature regarding its use for the treatment of Mpox infection.¹⁷⁶

Li et al. demonstrated that monoclonal antibodies (mAbs) targeting the specific proteins (A29L and A35R) of the Mpox virus effectively neutralized orthopoxviruses, including VACV. These mAbs also showed protective effect in mice, resulting in reduced viral titers and alleviation of lung injury.⁸² Gilchuk et al. identified a large number of *orthopoxvirus*-specific mAbs from the blood cells of human subjects with a history of prior orthopoxvirus vaccination or infection, and of which 16 mAbs had neutralizing activity against Mpox were identified. Moreover, mAbs targeting A33, L1, A27, or H3 antigens exhibited the broadest cross-neutralizing activity against VACV and Mpox virus. *In vivo* experiments confirmed that a combination of mAbs with high neutralizing activity provides efficient protection against lethal doses of VACV infection in mice (VACV, 1.2 mg mAbs, intraperitoneal injection), compared to VIGIV. This protective effect was observed even in severe combined immune-deficiency mouse models.¹⁸¹ Therefore, mAbs drugs are most likely to provide effective clinical treatment outcomes in the development of anti-Mpox treatments compared to VIGIV and CP, which have uncertain efficacy.

Immune cells

Mpox virus enters the human body through mucous membranes or compromised skin, resulting in infection of resident immune cells and antigen-presenting cells in the tissues.^{189–191} Subsequently, Mpox virus rapidly replicates in draining lymph nodes and disseminates through the lymphatic system,^{192,193} explaining the characteristic lymph node enlargement observed in Mpox virus infections. Innate immune cells act as the first line of defense against viral infections and are primary targets for viral assault. During the early stages of Mpox virus infection, monocytes are recruited to the infection site and become early targets for viral infection.¹⁹⁴ The level of Mpox virus antigens detected in

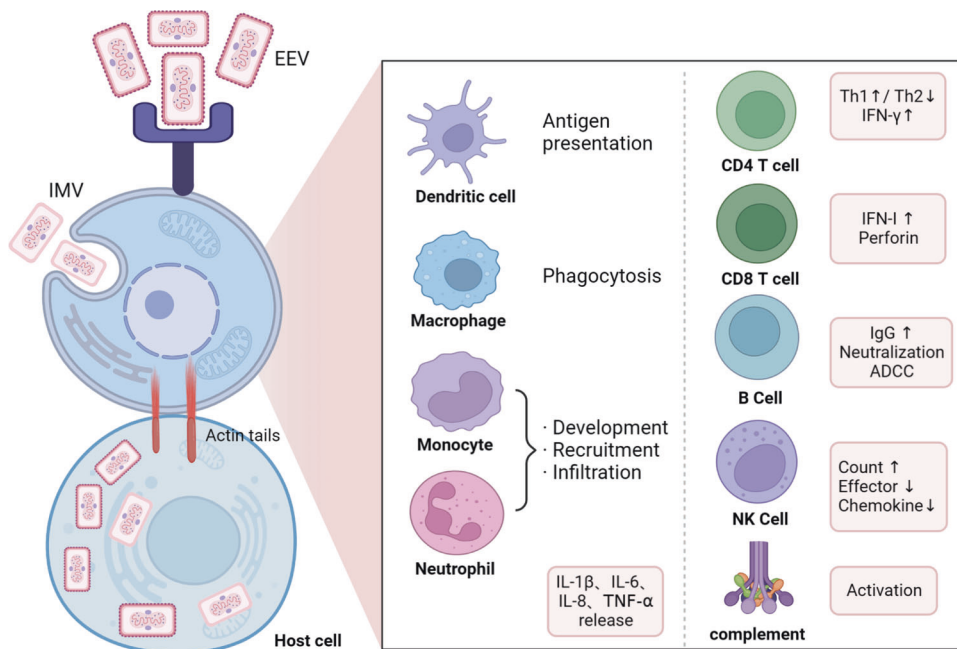


Fig. 5 The host cell immune response after Mpox virus infection. The Mpox induces specific and non-specific immune responses after infection. Briefly, upon entry of Mpox virus into host cells, mononuclear phagocytes and neutrophils initiate recruitment and increased proliferative infiltration, other antigen-presenting cells (such as dendritic cell) become activated, leading to the release of effector molecules and chemokines, while other cells (T cells, B cells, NK cells and the complement system) of the immune system also begin to exert their corresponding effector functions. IL interleukin, Th helper T cell, IFN Interferon, ADCC antibody-dependent cell-mediated cytotoxicity

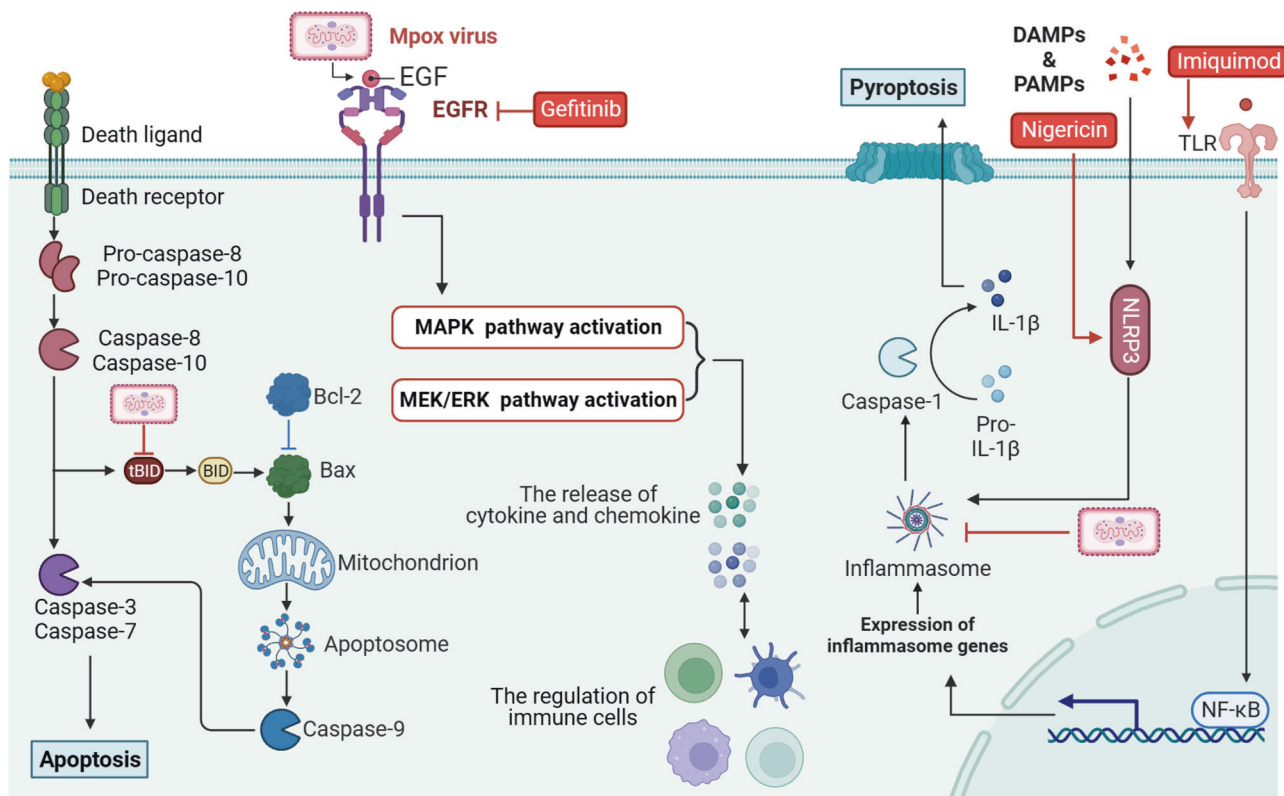


Fig. 6 Illustrates the signaling pathways associated with the targeted actions of certain drugs following Mpox virus infection. Upon infection, Mpox inhibits pyroptosis, impeding the formation of inflammasomes and activation of caspase-1. This blockade prevents pyroptosis and hampers the adequate activation of the immune response against Mpox infection. However, nigericin, an activator of NLRP3 can induce pyroptosis in host cells, making it a promising candidate for an anti-Mpox drug. Moreover, tBID, a protein involved in apoptosis, is suppressed upon Mpox virus infection, thereby inhibiting both intrinsic and extrinsic apoptotic pathways and ensuring the survival of Mpox virus within host cells. This mechanism can be exploited by employing apoptosis inducers as a strategy to combat Mpox virus. Furthermore, Mpox virus infection triggers the binding of EGF and EGFR, activation downstream MAPK and MEK signaling pathways, leading to the release of inflammatory and chemotactic factors, and modulation of immune cells. That is, EGFR inhibitors like gefitinib may exhibit significant anti-Mpox activity

monocytes can serve as an indicator of infection severity and prognosis. Additionally, natural killer (NK) cells play a crucial role in generating a robust immune response following Mpox virus infection. Despite an increase in the abundance of NK cells in Mpox virus-infected individuals, their migration, degranulation, and effector molecule release capabilities are significantly impaired. Patricia's study demonstrated that the injection of in vitro expanded NK cells into mice infected with vaccinia virus resulted in significantly prolonged mouse survival and enhanced secretion of IFN- γ by NK cells.^{195,196} T cells, another vital immune cell type, possess cytotoxic functions and the ability to regulate disease severity. Individuals with acquired immunodeficiency are at high risk of severe infections when co-infected with Mpox virus, often requiring active medical intervention rather than spontaneous resolution.^{197,198} Furthermore, individuals with compromised immune function are more susceptible to severe disease and mortality during Mpox virus infection. Other immune cell types, such as dendritic cells and innate lymphoid cells, also undergo alterations during Mpox virus infection.¹⁹⁹ Understanding the characteristics and transformations of diverse immune cells during Mpox virus infection is important for gaining insights into the immune response and developing immunotherapy.

Immune effector molecules and immunomodulators

The response of immune effector molecules during Mpox virus infection—plays a crucial role in disease progression and severity. At the beginning of infection, the Mpox virus can suppress the expression of chemokines, resulting in a decrease in effector

molecule expression like IFN- γ and TNF- α . This inhibition in T-cell activation hinders the initiation of humoral immune response, allowing the virus to evade the immune system much easier. However, severe Mpox infection often leads to a cytokine storm in later stages. This results in an increase in Th2-associated cytokines and a decrease in Th1-associated cytokines, characterized by increased expression of IL-2, IL-4, and IL-8 and a reduction in TNF- α , IL-2, and IL-12 (Fig. 5). By regulating these immune effectors, Mpox virus suppresses the antiviral immune response and disrupts the host immunity. Ribavirin, in addition to blocking viral nucleotide synthesis, also acts as an immunomodulator. It regulates T-cell polarization, and can enhance the release of interferon-gamma (IFN- γ) and T-bet, which are associated with Th1 response, in the serum of patients infected with hepatitis viruses. Simultaneously, ribavirin suppresses the release of GATA binding protein 3 and IL-4, which are related to Th2 response, promoting T-cell polarization towards Th1 and strengthening the antiviral action of the immune system.^{200–202} Ribavirin also stimulates the generation of central memory T-cells and Tregs.^{203,204} Pidotimod is an immunomodulator used as an adjunctive therapy for respiratory or urinary tract infections.²⁰⁵ It promotes non-specific and specific immune responses by activating NK cells, stimulating lymphocyte proliferation, and inducing the release of IL-1 β and IFN- γ .^{206–209} Thymosin is an exogenous polypeptide with immunoregulatory effects that promote T cell differentiation, development, and maturation.^{210,211} Additionally, Thymosin can indirectly enhance the immune responses of other immune cells.^{212,213} Nevertheless, the clinical use of pidotimod

Table 1. Anti-Mpox drugs and candidate compounds

	Name	Mechanism	Function	Clinical Use
Targeting virus intrusion	Amphotericin B	Isolate cholesterol and destroy lipid rafts	Restrict Mpox virus entry	☒
	Cholesterol lowering drugs			
	Glycosaminoglycan analog	Competitive binding to host cell membrane	Prevent the attachment and entry of Mpox virus	☒
Targeting virus replication	Cidofovir	Competitive binding of DNA or RNA polymerase	Interference with viral DNA or RNA synthesis	☒
	Brincidofovir			☒
	NPP-669			☒
	KAY-2-41			☒
	Trifluridine			☒
	Ribavirin			☒
	Ionomycin			Destroy the integrity of Endoplasmic reticulum
Targeting virus assembly, maturation and release	Nocodazole	Promote microtubule depolymerization	Inhibit the movement of viral particles to the cell surface	☒
	Imatinib mesylate	Tyrosinase inhibition	Inhibit the actin tail formation	☒
	PA104	Inhibit the actin tail formation	Inhibit Mpox virus efflux	☒
	Tecovirimat	Inhibit vp37 protein synthesis	Inhibit the maturation and budding release of orthopoxviruses	☒
	NIOCH-14			☒
	Gefitinib	EGFR inhibition	Inhibit the actin tail formation	☒
	MEK inhibitors	MEK inhibition	Inhibit the actin tail formation	☒
	A36R polypeptide	Anti Mpox virus-A36R	Inhibit Mpox virus transmission and release	☒
Immunoregulation	VIGIV	antigen binding	Prevent Mpox virus infection of target cells	☒
	Pidotimod	Enhancing specific and non-specific immunity	Enhance immune response	☒
	Thymopeptide			☒
	mAbs	Destroy virus particles	Prevent virus infection of cells	☒
	Nigericin	Activate IL-1 β and IL-18	Induced pyroptosis	☒
	Imiquimod	TLR agonists and local immune activity enhancer	Stimulate cytokine production and activate local immunity	☒

and thymosin preparations in Mpox virus-infected patients has not been reported and requires further investigation to validate their efficacy. Although immunomodulators cannot directly elicit an anti-Mpox virus effect, they have the potential to improve the immune system, which may help reduce the development of severe manifestations and decrease mortality rates.^{210,211,214} Exploring the combination of immunomodulators with other anti-Mpox virus medications may be a promising avenue for further investigation.

Modulation of the virus-induced cellular signal transduction

Mpox virus infection induces immune responses while also regulating cellular signal transduction.^{215,216} One example is the presence of a Mpox virus-encoded Bcl-2-like protein, which regulates the intrinsic apoptotic pathway. Additionally, the SPI-2 protein, encoded by the B12R gene,²¹⁷ inhibits both caspase-1 and caspase-8, thereby disrupting the pyroptosis or apoptosis pathway,^{218,219} respectively. However, active induction of pyroptosis can be achieved by using nigericin, an inflammasome activator and pyroptosis inducer, as a strategy against Mpox infection. In an in vitro study conducted by Chad et al., Hela cells were infected with vaccinia virus and treated with Nigericin in vitro (VACV, EC₅₀ = 7.9 nM, SI = 1038, in Hela cells).²²⁰ The findings demonstrated that Nigericin effectively reduced the viral titers and showed a stronger antiviral effect and lower EC₅₀ values compared to the control group treated with Cidofovir. Protein kinases play a key role in regulating signal transduction pathways.^{221,222} Raghav et al. conducted an analysis to explore the interactions between

Mpox virus and host proteins in order to further investigate the defense mechanisms triggered by Mpox infection. Their findings show the important role of the mitogen-activated protein kinase (MAPK) signaling pathway in the response to Mpox infection.²¹⁶ Inhibition of the thymidine kinase enzyme, which is activated by MAPK, led to a significant reduction in viral replication.²²³⁻²²⁵ This evidence supports the potential of targeted therapies against MAPK signaling pathway as a promising strategy to combat Mpox (Fig. 6).^{224,226}

PROSPECT AND CHALLENGES

With the global cessation of smallpox vaccination administration, the proportion of individuals with cross-immune protection against Mpox virus has rapidly declined, rendering Mpox a potential bioterrorism threat. While a few anti-Mpox drugs, such as Tecovirimat, have been clinically proven to be effective, relying solely on them would be unwise. Despite Mpox virus belonging to the DNA virus family, it exhibits significantly higher genomic variability due to increased nucleotide polymorphism. The rapid population mobility and increased international travel have facilitated the continuous spread of Mpox virus among populations, further increasing its potential for mutation.^{11,227,228} These factors contribute to increased variability, drug resistance, and the emergence of multidrug-resistant strains of Mpox virus.²²⁹ Moreover, currently available drugs face certain limitations that impede their clinical applications. For example, Cidofovir has low bioavailability and carries the risk of renal damage, while Cidofovir

and Brincidofovir pose potential threats to hematopoietic and liver functions. There is an urgent need to develop novel anti-Mpx virus drugs.²³⁰

The lengthy and costly nature of drug development, combined with numerous uncertainties, has led to the exploration of drug repurposing strategies as a more efficient and economical approach.^{110,231–233} HTS of marketed drugs or clinically established medications has the potential to expedite the identification of antiviral agents, thus saving valuable time.^{234–236} For instance, the potential antiviral drug ribavirin has demonstrated therapeutic effectiveness against Mpx infection. Similarly, the widely used EGFR inhibitor gefitinib has shown promising antiviral activity against Mpx virus in addition to its approved indication for late-stage non-small cell lung cancer. However, drug repurposing efforts still heavily rely on serendipitous discoveries. Historically, drug development has been predominantly confined to laboratory settings. However, advances in computer science and computational drug design have significantly accelerated the discoveries in drug repurposing.^{237–239} Computer-aided drug discovery (CADD) techniques encompass the following three main directions: 1) High-throughput library screening of small molecule libraries, such as the discovery and development of Tecovirimat based on the VP37 protein. 2) Structural optimization based on existing drugs, such as NPP669, which involves alkyl chain modifications based on Cidofovir, resulting in overall improved pharmacological properties compared to Cidofovir. 3) Directly targeting functional sites for novel drug design, such as DdRp, which usually serves as a target for antiviral CADD.

In recent years, there has been relatively little attention on the Mpx outbreak in endemic area. However, the rapid spread of Mpx in non-endemic regions and its global impact have brought it back into the public attention. This particular outbreak of Mpx appears to exhibit distinct epidemiological characteristics and transmission dynamics compared to previous outbreaks.⁶³ Previous knowledge suggested that the West African clade had weak transmission and pathogenicity compared to the Central African clade. However, the current situation reveals that the 2022-Mpx virus genome mutation and phylogenetic analysis indicate that this outbreak belongs to the B.1 lineage of the West African clade. The B1 lineage has exhibited mutations in virulence proteins, host recognition proteins, and immune evasion.²⁴⁰ APOBEC3 is an important enzyme that demonstrates antiviral activity against HIV, Hepatitis B Virus, Epstein-Barr virus, and other viruses through its functional cytidine deaminase activity.^{241,242} APOBEC3-mediated viral genome editing may be characterized by compatible substitutions GA>AA and TC>TT. Isidro et al. discovered a significant increase in G-to-A and C-to-T mutations in the recent Mpx isolates, with 46 SNPs showing mutation bias, among which 26 and 15 substitutions were GA>AA and TC>TT, respectively.^{63,243,244} These unusual mutation biases and the abundance of A: T bases in Mpx virus indicate that specific mutations driven by APOBEC3 may further reduce the pathogenicity and symptoms caused by Mpx virus infection, facilitating covert transmission within populations, and indirectly contributing to the global epidemic of Mpx. Although further experimental validation is necessary to confirm Mpx virus mutations mediated by APOBEC3, it is undeniable that APOBEC3 is a promising host antiviral protein for research. Understanding the mechanism of mutations driven by APOBEC3 may help reveal the mysteries behind the pathogenicity transition of Mpx.²⁴⁵ Additionally, the development of anti-Mpx drugs targeting APOBEC3 may provide a new direction for future drug development.

The development of multi-omics technologies and HTS techniques has enabled precise identification and characterization of various molecular targets of Mpx virus, which is crucial for the development of novel anti-Mpx virus drugs targeting new mechanisms. Furthermore, multi-omics technologies have revealed the gene expression patterns during Mpx infection

and identified specific receptors and pathways regulated during Mpx progression. By precisely modulating these receptors and pathways, it is possible to develop drugs for Mpx therapy. This study contributes to optimizing the chemical structure of drugs, enhancing their delivery and targeting, thereby improving treatment precision and reducing drug side effects (Table 1).

In recent years, AI, especially machine learning and deep learning methods, have increasingly been utilized in various stages of the drug development process, challenging the traditional paradigms of new drug discovery and design.²⁴⁶ By leveraging extensive compound data within libraries, AI enables efficient design and optimization of compounds targeting various Mpx virus homologous proteins. This approach facilitates the effective screening of optimal anti-Mpx virus drugs or the development of promising new candidate molecules, ultimately reducing the cost and time associated with drug development.^{247,248} However, it is important to note that no commercially available drugs have emerged from this approach yet, indicating the need for further technical advancements and breakthroughs in the field.²⁴⁹

In addition to the development of systemic anti-infective drugs, exploring local therapies for Mpx is crucial. Mpx infections can cause severe physiological and psychological trauma to the skin and eyes.²⁵⁰ This damage is often visibly evident and difficult to conceal. Skin lesions, for example, are a hallmark of Mpx infection and inflict immense pain on patients. The psychological trauma resulting from these skin injuries and subsequent scarring may surpass the physical harm. A distressing incident reported in 2017 by Dimie et al. highlighted the tragic suicide of a 34-year-old Mpx patient due to the psychological trauma endured post-infection.²⁵¹ Therefore, addressing skin lesions during the course of Mpx infection is essential. Notably, the topical cream imiquimod has demonstrated particular efficacy in treating Mpx-induced skin lesions.^{252,253} The exact mechanism of action of imiquimod in the treatment of Mpx infections is not fully understood, although several potential mechanisms have been proposed. Imiquimod acts as an agonist for Toll-like receptor 7 (TLR-7) and Toll-like receptor 8 (TLR-8), triggering the nuclear translocation and transcriptional activity of nuclear factor κ B (NF- κ B). This activation leads to the release of downstream pro-inflammatory cytokines, enhancing the immune response against Mpx. Additionally, imiquimod acts as a direct local immune stimulant by stimulating the production of various cytokines, including IFN- γ , TNF- α , IL-1 β , and IL-6. These cytokines play a crucial role in activating the innate immune system and promoting a localized immune response.^{254–256} Studies have shown that imiquimod can recruit plasmacytoid dendritic cells to the site of infection, thereby enhancing the antigen presentation process. Although ocular infections caused by Mpx are relatively rare, they may result in permanent visual impairment, including irreversible conditions such as corneal perforation. As a locally administered nucleoside analog, trifluridine eye drops are currently considered the most effective treatment for ocular Mpx infections. In addition to controlling Mpx proliferation in the eyes, trifluridine eye drops also help reduce the production of conjunctival secretions, thereby minimizing the risk of spreading the infection to others.

To address the widespread occurrence of Mpx, it is imperative to recognize it as a global public health concern. In addition to the development of therapeutic medications, emphasis must be placed on preventive measures. Prevention, especially among areas with active Mpx transmission and those in high-risk populations like HIV-infected MSM, is crucial.²⁵⁷ Vaccination is an effective strategy for preventing Mpx. Studies indicate that vaccinia vaccine can offer partial protection against Mpx infection.^{258,259} However, the use of smallpox vaccines for Mpx prevention in epidemic regions limited due to potential risks for immunocompromised individuals, particularly those co-infected

with HIV. First-generation vaccines like Dryvax and second-generation vaccines such as ACAM2000, which are live replicating vaccinia virus vaccines, can cause severe infections such as progressive vaccinia.²⁶⁰ The emergence of third-generation vaccines provides an alternative for this specific population. One example is Imvamune (also known as JYNNEOS) a non-replicating vaccinia vaccine that has been tested safe for HIV-infected patients. Animal models have shown that JYNNEOS also provides protection against Mpx.²⁶¹ In 2019, the FDA approved JYNNEOS for preventing Mpx infection in high-risk populations aged 18 and above.²⁶² Clinical evidence has demonstrated that JYNNEOS vaccination effectively prevents Mpx cases and reduce the incidence of severe illness.^{258,263,264} At the national and regional levels, enhancing public health investments, including environmental sanitation and disinfection, and establishing efficient case identification and contact tracing mechanisms, is essential. On an individual level, it is crucial to educate oneself about Mpx, maintain good personal hygiene, employ personal protective measures, and avoid contact with infection sources. Therefore, the most cost-effective method to reduce the incidence and transmission of Mpx is through implementing preventive measures rather than solely relying on the development of novel anti-Mpx drugs.

CONCLUSIONS

While some progress has been made in the development of drugs against Mpx, it is crucial to expedite the research progress. This will enable us to effectively combat potential long-term outbreaks and the emergence of drug-resistant Mpx virus strains. In the development of drugs against Mpx, the following aspects should be given priority: Firstly, improving the specificity and delivery efficiency of drugs is essential to ensure accurate targeting of the Mpx and efficient transmission to the infection site. Secondly, development anti-Mpx drugs that are less prone to resistance is necessary to prevent the gradual emergence of drug-resistant strains and ensuring sustained efficacy of treatment. Additionally, exploring the development of sequential and combination drug therapies should enhance effectiveness against different stages of Mpx infections and their variants. Lastly, attention should be paid to drug modifications to mitigate or eliminate toxicity, minimizing the adverse impact on patients during the treatment process. The early investment in drug development against Mpx is crucial in tackling the ongoing global Mpx outbreak. Accelerating progress in the development of effective anti-Mpx drugs will help prepare for future challenges and provide more reliable protection for public health.

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AUTHOR CONTRIBUTIONS

J.J.L., H.X., C.H.W., M.J.T., Y.Y., W.J.T. and L.S. contributed to the conception of the review, J.J.L., H.X., C.H.W. and M.J.T. collected the information and wrote the manuscript. C.C.W., F.Y. and L.J.Y. provided guidance on article writing and polishing. Y.Y., W.J.T. and L.S. contributed to the constructive discussions. All authors have read and approved the article.

ADDITIONAL INFORMATION

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REFERENCES

- McCollum, A. M. & Damon, I. K. Human monkeypox. *Clin. Infect. Dis.* **58**, 260–267 (2014).
- Otu, A. et al. Global human monkeypox outbreak: atypical presentation demanding urgent public health action. *Lancet Microbe* **3**, e554–e555 (2022).
- Cabanillas, B. et al. A compilation answering 50 questions on monkeypox virus and the current monkeypox outbreak. *Allergy* **78**, 639–662 (2023).
- Carrubba, S. et al. Novel severe oculocutaneous manifestations of human monkeypox virus infection and their historical analogues. *Lancet Infect. Dis.* **23**, e190–e197 (2023).
- Ladnyj, I. D., Ziegler, P. & Kima, E. A human infection caused by monkeypox virus in Basankusu Territory, Democratic Republic of the Congo. *Bull. World Health Organ.* **46**, 593–597 (1972).
- Foster, S. O. et al. Human monkeypox. *Bull. World Health Organ.* **46**, 569–576 (1972).
- Khodakevich, L., Jezek, Z. & Messinger, D. Monkeypox virus: ecology and public health significance. *Bull. World Health Organ.* **66**, 747–752 (1988).
- Khodakevich, L. et al. The role of squirrels in sustaining monkeypox virus transmission. *Trop. Geogr. Med.* **39**, 115–122 (1987).
- Doty, J. B. et al. Assessing Monkeypox virus prevalence in small mammals at the human-animal interface in the Democratic Republic of the Congo. *Viruses* **9**, 283 (2017).
- Bunge, E. M. et al. The changing epidemiology of human monkeypox—a potential threat? A systematic review. *PLoS Negl. Trop. Dis.* **16**, e0010141 (2022).
- Meo, S. A., Al-Khlaiwi, T., Al Jassir, F. F. & Meo, A. S. Impact of traveling on transmission trends of human monkeypox disease: worldwide data based observational analysis. *Front Public Health* **11**, 1029215 (2023).
- Costello, V. et al. Imported Monkeypox from International Traveler, Maryland, USA, 2021. *Emerg. Infect. Dis.* **28**, 1002–1005 (2022).
- Alakunle, E. F. & Okeke, M. I. Monkeypox virus: a neglected zoonotic pathogen spreads globally. *Nat. Rev. Microbiol.* **20**, 507–508 (2022).
- WHO Director-General's opening remarks at the media briefing, <https://www.who.int/director-general/speeches/detail/who-director-general-s-opening-remarks-at-the-media-briefing--11-may-2023> (2023).
- Nuzzo, J. B., Borio, L. L. & Gostin, L. O. The WHO declaration of Monkeypox as a global public health emergency. *JAMA* **328**, 615–617 (2022).
- Sabeena, S. The changing epidemiology of monkeypox and preventive measures: an update. *Arch. Virol.* **168**, 31 (2023).
- Papukashvili, V. et al. Strategy of developing nucleic acid-based universal monkeypox vaccine candidates. *Front. Immunol.* **13**, 1050309 (2022).
- China CDC: Monitoring of Monkeypox Epidemic in August 2023, https://www.chinacdc.cn/jkzt/crb/qt/szkb_13037/gwjszl_13092/202309/t20230908_269405.html (2023).
- Adetifa, I., Muyembe, J. J., Bausch, D. G. & Heymann, D. L. Mpx neglect and the smallpox niche: a problem for Africa, a problem for the world. *Lancet* **401**, 1822–1824 (2023).
- Harris, E. What to know about Monkeypox. *JAMA* **327**, 2278–2279 (2022).
- Walter, K. & Malani, P. N. What is Monkeypox? *JAMA* **328**, 222 (2022).
- Beeson, A. et al. Mpx respiratory transmission: the state of the evidence. *Lancet Microbe* **4**, e277–e283 (2023).
- Upadhayay, S. et al. Monkeypox infection: the past, present, and future. *Int. Immunopharmacol.* **113**, 109382 (2022).
- Adler, H. & Taggart, R. Monkeypox exposure during pregnancy: what does UK public health guidance advise? *Lancet* **400**, 1509 (2022).
- Billioux, B. J., Mbaya, O. T., Sejvar, J. & Nath, A. Potential complications of monkeypox. *Lancet Neurol.* **21**, 872 (2022).
- Durski, K. N. et al. Emergence of Monkeypox—West and Central Africa, 1970–2017. *Morb. Mortal. Wkly Rep.* **67**, 306–310 (2018).
- Wang, Y., Leng, P. & Zhou, H. Global transmission of monkeypox virus—a potential threat under the COVID-19 pandemic. *Front. Immunol.* **14**, 1174223 (2023).
- Kumar, P. et al. Recent advances in research and management of human Monkeypox virus: an emerging global health threat. *Viruses* **15**, 937 (2023).
- WHO: Multi-country monkeypox outbreak in non-endemic countries, <https://www.who.int/emergencies/disease-outbreak-news/item/2022-DON385> (2022).
- Del Rio, C. & Malani, P. N. Update on the Monkeypox outbreak. *JAMA* **328**, 921–922 (2022).
- Aden, D., Zaheer, S., Kumar, R. & Ranga, S. Monkeypox (Mpx) outbreak during COVID-19 pandemic-past and the future. *J. Med. Virol.* **95**, e28701 (2023).
- Guarner, J., Del Rio, C. & Malani, P. N. Monkeypox in 2022—what clinicians need to know. *JAMA* **328**, 139–140 (2022).
- Nolen, L. D. et al. Extended human-to-human transmission during a Monkeypox outbreak in the Democratic Republic of the Congo. *Emerg. Infect. Dis.* **22**, 1014–1021 (2016).
- Accordini, S. et al. People with asymptomatic or unrecognised infection potentially contribute to monkeypox virus transmission. *Lancet Microbe* **4**, e209 (2023).

35. Reda, A., El-Qushayri, A. E. & Shah, J. Asymptomatic monkeypox infection: a call for greater control of infection and transmission. *Lancet Microbe* **4**, e15–e16 (2023).
36. Mahmoud, A. & Nchasi, G. Monkeypox virus: a zoonosis of concern. *J. Med. Virol.* **95**, e27968 (2023).
37. Altindis, M., Puca, E. & Shapo, L. Diagnosis of monkeypox virus—an overview. *Travel. Med. Infect. Dis.* **50**, 102459 (2022).
38. Candela, C. et al. Human Monkeypox experience in a tertiary level hospital in Milan, Italy, between May and October 2022: epidemiological features and clinical characteristics. *Viruses* **15**, 667 (2023).
39. Liu, Q. et al. Clinical characteristics of Human Mpx (Monkeypox) in 2022: a systematic review and meta-analysis. *Pathogens* **12**, 146 (2023).
40. Gaspari, V. et al. Monkeypox outbreak 2022: clinical and virological features of 30 patients at the sexually transmitted diseases centre of Sant'Orsola Hospital, Bologna, Northeastern Italy. *J. Clin. Microbiol.* **61**, e0136522 (2023).
41. Shafaati, M. & Zandi, M. State-of-the-art on monkeypox virus: an emerging zoonotic disease. *Infection* **50**, 1425–1430 (2022).
42. Kumar, N., Acharya, A., Gendelman, H. E. & Byrareddy, S. N. The 2022 outbreak and the pathobiology of the monkeypox virus. *J. Autoimmun.* **131**, 102855 (2022).
43. Alakunle, E., Moens, U., Nchinda, G. & Okeke, M. I. Monkeypox virus in Nigeria: infection biology, epidemiology, and evolution. *Viruses* **12**, 1257 (2020).
44. Martínez-Fernández, D. E. et al. Human Monkeypox: a comprehensive overview of epidemiology, pathogenesis, diagnosis, treatment, and prevention strategies. *Pathogens* **12**, 947 (2023).
45. Malik, S. et al. Monkeypox Virus: a comprehensive overview of viral pathology, immune response, and antiviral strategies. *Vaccines* **11**, 1345 (2023).
46. Zahmatyar, M. et al. Human monkeypox: history, presentations, transmission, epidemiology, diagnosis, treatment, and prevention. *Front. Med.* **10**, 1157670 (2023).
47. Srivastava, S. et al. The Global Monkeypox (Mpx) outbreak: a comprehensive review. *Vaccines* **11**, 1093 (2023).
48. Fonti, M. et al. Monkeypox associated acute arthritis. *Lancet Rheumatol.* **4**, e804 (2022).
49. Rao, A. K. et al. Interim clinical treatment considerations for severe manifestations of Mpx - United States, February 2023. *Morb. Mortal. Wkly Rep.* **72**, 232–243 (2023).
50. Maqbool, K. U. et al. Cardiovascular manifestations of human Monkeypox virus: an updated review. *Curr. Probl. Cardiol.* **48**, 101869 (2023).
51. Harris, E. Severe form of Mpx identified in patients with advanced HIV. *JAMA* **329**, 968 (2023).
52. Laurenson-Schafer, H. et al. Description of the first global outbreak of mpx: an analysis of global surveillance data. *Lancet Glob. Health* **11**, e1012–e1023 (2023).
53. Fink, D. L. et al. Clinical features and management of individuals admitted to hospital with monkeypox and associated complications across the UK: a retrospective cohort study. *Lancet Infect. Dis.* **23**, 589–597 (2023).
54. Huhn, G. D. et al. Clinical characteristics of human monkeypox, and risk factors for severe disease. *Clin. Infect. Dis.* **41**, 1742–1751 (2005).
55. Li, H. et al. The evolving epidemiology of monkeypox virus. *Cytokine Growth Factor Rev.* **68**, 1–12 (2022).
56. Condit, R. C., Moussatche, N. & Traktman, P. In a nutshell: structure and assembly of the vaccinia virion. *Adv. Virus Res.* **66**, 31–124 (2006).
57. Shchelkunov, S. N. et al. Analysis of the monkeypox virus genome. *Virology* **297**, 172–194 (2002).
58. Garon, C. F., Barbosa, E. & Moss, B. Visualization of an inverted terminal repetition in vaccinia virus DNA. *Proc. Natl. Acad. Sci. USA* **75**, 4863–4867 (1978).
59. Wittek, R. et al. Inverted terminal repeats in rabbit poxvirus and vaccinia virus DNA. *J. Virol.* **28**, 171–181 (1978).
60. Shchelkunov, S. N. et al. Human monkeypox and smallpox viruses: genomic comparison. *FEBS Lett.* **509**, 66–70 (2001).
61. Andrei, G. & Snoeck, R. Differences in pathogenicity among the mpx virus clades: impact on drug discovery and vaccine development. *Trends Pharmacol. Sci.* **44**, 719–739 (2023).
62. Wang, L. et al. Genomic annotation and molecular evolution of monkeypox virus outbreak in 2022. *J. Med. Virol.* **95**, e28036 (2023).
63. Isidro, J. et al. Phylogenomic characterization and signs of microevolution in the 2022 multi-country outbreak of monkeypox virus. *Nat. Med.* **28**, 1569–1572 (2022).
64. Armstrong, J. A., Metz, D. H. & Young, M. R. The mode of entry of vaccinia virus into L cells. *J. Gen. Virol.* **21**, 533–537 (1973).
65. Dales, S. The uptake and development of vaccinia virus in strain L cells followed with labeled viral deoxyribonucleic acid. *J. Cell Biol.* **18**, 51–72 (1963).
66. Haller, S. L., Peng, C., McFadden, G. & Rothenburg, S. Poxviruses and the evolution of host range and virulence. *Infect. Genet. Evol.* **21**, 15–40 (2014).
67. Locker, J. K. et al. Entry of the two infectious forms of vaccinia virus at the plasma membrane is signaling-dependent for the IMV but not the EEV. *Mol. Biol. Cell* **11**, 2497–2511 (2000).
68. Schmidt, F. I., Bleck, C. K., Helenius, A. & Mercer, J. Vaccinia extracellular virions enter cells by macropinocytosis and acid-activated membrane rupture. *EMBO J.* **30**, 3647–3661 (2011).
69. Doms, R. W., Blumenthal, R. & Moss, B. Fusion of intra- and extracellular forms of vaccinia virus with the cell membrane. *J. Virol.* **64**, 4884–4892 (1990).
70. Moss, B. Membrane fusion during poxvirus entry. *Semin. Cell Dev. Biol.* **60**, 89–96 (2016).
71. Chang, A. & Metz, D. H. Further investigations on the mode of entry of vaccinia virus into cells. *J. Gen. Virol.* **32**, 275–282 (1976).
72. Janeczko, R. A., Rodriguez, J. F. & Esteban, M. Studies on the mechanism of entry of vaccinia virus in animal cells. *Arch. Virol.* **92**, 135–150 (1987).
73. Vanderplasschen, A., Hollinshead, M. & Smith, G. L. Intracellular and extracellular vaccinia virions enter cells by different mechanisms. *J. Gen. Virol.* **79**, 877–887 (1998).
74. Ichihashi, Y. Extracellular enveloped vaccinia virus escapes neutralization. *Virology* **217**, 478–485 (1996).
75. Law, M. & Smith, G. L. Antibody neutralization of the extracellular enveloped form of vaccinia virus. *Virology* **280**, 132–142 (2001).
76. Vanderplasschen, A. & Smith, G. L. A novel virus binding assay using confocal microscopy: demonstration that the intracellular and extracellular vaccinia virions bind to different cellular receptors. *J. Virol.* **71**, 4032–4041 (1997).
77. Simons, K. & Toomre, D. Lipid rafts and signal transduction. *Nat. Rev. Mol. Cell Biol.* **1**, 31–39 (2000).
78. Gee, Y. J., Sea, Y. L. & Lal, S. K. Viral modulation of lipid rafts and their potential as putative antiviral targets. *Rev. Med. Virol.* **33**, e2413 (2023).
79. Peruzzi, D., Fecchi, K., Venturi, G. & Gagliardi, M. C. Repurposing amphotericin B and its liposomal formulation for the treatment of human Mpx. *Int. J. Mol. Sci.* **24**, 8896 (2023).
80. Sekaran, S. & Sekar, S. K. R. Repurposing cholesterol lowering drugs in the treatment and management of monkeypox. *Int. J. Surg.* **109**, 60–61 (2023).
81. He, P. et al. SPR sensor-based analysis of the inhibition of marine sulfated glycans on interactions between Monkeypox virus proteins and glycosaminoglycans. *Mar. Drugs* **21**, 264 (2023).
82. Li, M. et al. Three neutralizing mAbs induced by MPXV A29L protein recognizing different epitopes act synergistically against orthopoxvirus. *Emerg. Microbes. Infect.* **12**, 2223669 (2023).
83. Mallardo, M., Schleich, S. & Krijnse Locker, J. Microtubule-dependent organization of vaccinia virus core-derived early mRNAs into distinct cytoplasmic structures. *Mol. Biol. Cell* **12**, 3875–3891 (2001).
84. Mercer, J. et al. RNAi screening reveals proteasome- and Cullin3-dependent stages in vaccinia virus infection. *Cell Rep.* **2**, 1036–1047 (2012).
85. Satheshkumar, P. S., Anton, L. C., Sanz, P. & Moss, B. Inhibition of the ubiquitin-proteasome system prevents vaccinia virus DNA replication and expression of intermediate and late genes. *J. Virol.* **83**, 2469–2479 (2009).
86. Lant, S. & Maluquer de Motes, C. Poxvirus interactions with the host ubiquitin system. *Pathogens* **10**, 1034 (2021).
87. Kates, J. & Beeson, J. Ribonucleic acid synthesis in vaccinia virus. I. The mechanism of synthesis and release of RNA in vaccinia cores. *J. Mol. Biol.* **50**, 1–18 (1970).
88. Mallardo, M. et al. Relationship between vaccinia virus intracellular cores, early mRNAs, and DNA replication sites. *J. Virol.* **76**, 5167–5183 (2002).
89. Katsafanas, G. C. & Moss, B. Colocalization of transcription and translation within cytoplasmic poxvirus factories coordinates viral expression and subjugates host functions. *Cell Host Microbe* **2**, 221–228 (2007).
90. Kieser, Q. et al. Cytoplasmic factories, virus assembly, and DNA replication kinetics collectively constrain the formation of poxvirus recombinants. *PLoS One* **15**, e0228028 (2020).
91. Peng, Q. et al. Structure of monkeypox virus DNA polymerase holoenzyme. *Science* **379**, 100–105 (2023).
92. Dsouza, L. et al. Antiviral activities of two nucleos(t)ide analogs against vaccinia, Mpx, and cowpox viruses in primary human fibroblasts. *Antiviral Res.* **216**, 105651 (2023).
93. Abdullah Al Awadh, A. Nucleotide and nucleoside-based drugs: past, present, and future. *Saudi J. Biol. Sci.* **29**, 103481 (2022).
94. Johnson, K. A. & Dangerfield, T. Mechanisms of inhibition of viral RNA replication by nucleotide analogs. *Enzymes* **49**, 39–62 (2021).
95. Andrei, G. & Snoeck, R. Cidofovir activity against poxvirus infections. *Viruses* **2**, 2803–2830 (2010).
96. Lebeau, I. et al. Activities of alkoxyalkyl esters of cidofovir (CDV), cyclic CDV, and (S)-9-(3-hydroxy-2-phosphonylmethoxypropyl)adenine against orthopoxviruses in cell monolayers and in organotypic cultures. *Antimicrob. Agents Chemother.* **50**, 2525–2529 (2006).
97. Stittelaar, K. J. et al. Antiviral treatment is more effective than smallpox vaccination upon lethal monkeypox virus infection. *Nature* **439**, 745–748 (2006).
98. Mailhe, M. et al. Clinical characteristics of ambulatory and hospitalized patients with monkeypox virus infection: an observational cohort study. *Clin. Microbiol. Infect.* **29**, 233–239 (2023).

99. Chenchula, S. et al. A systematic review to identify novel clinical characteristics of monkeypox virus infection and therapeutic and preventive strategies to combat the virus. *Arch. Virol.* **168**, 195 (2023).
100. Byrareddy, S. N. et al. Potential therapeutic targets for Mpx: the evidence to date. *Expert Opin. Ther. Targets* **27**, 419–431 (2023).
101. Shamim, M. A. et al. The use of antivirals in the treatment of human monkeypox outbreaks: a systematic review. *Int. J. Infect. Dis.* **127**, 150–161 (2023).
102. Kim, G. H. & Jun, J. B. Altered serum uric acid levels in kidney disorders. *Life* **12**, 1891 (2022).
103. Caetano-Pinto, P. et al. Amplifying the impact of kidney microphysiological systems: predicting renal drug clearance using mechanistic modelling based on reconstructed drug secretion. *Altex* **40**, 408–424 (2022).
104. Velioglu, A. et al. Topical cidofovir-related acute kidney injury in a kidney transplant recipient. *Clin. Transplant.* **36**, e14824 (2022).
105. Imran, M. et al. Oral brincidofovir therapy for monkeypox outbreak: a focused review on the therapeutic potential, clinical studies, patent literature, and prospects. *Biomedicines* **11**, 278 (2023).
106. Shamim, M. A. et al. Pharmacological treatment and vaccines in monkeypox virus: a narrative review and bibliometric analysis. *Front. Pharmacol.* **14**, 1149909 (2023).
107. Wang, B. et al. Disulfide-incorporated lipid prodrugs of cidofovir: synthesis, antiviral activity, and release mechanism. *Eur. J. Med. Chem.* **258**, 115601 (2023).
108. Stabenow, J. et al. A mouse model of lethal infection for evaluating prophylactics and therapeutics against Monkeypox virus. *J. Virol.* **84**, 3909–3920 (2010).
109. Adler, H. et al. Clinical features and management of human monkeypox: a retrospective observational study in the UK. *Lancet Infect. Dis.* **22**, 1153–1162 (2022).
110. Bojkova, D. et al. Repurposing of the antibiotic nitroloxline for the treatment of mpx. *J. Med. Virol.* **95**, e28652 (2023).
111. Eriksson, U. et al. Serine peptide phosphoester prodrugs of cyclic cidofovir: synthesis, transport, and antiviral activity. *Mol. Pharm.* **5**, 598–609 (2008).
112. Peterson, L. W. et al. Synthesis, transport and antiviral activity of Ala-Ser and Val-Ser prodrugs of cidofovir. *Bioorg. Med. Chem. Lett.* **21**, 4045–4049 (2011).
113. Lipka, E. et al. NPP-669, a novel broad-spectrum antiviral therapeutic with excellent cellular uptake, antiviral potency, oral bioavailability, preclinical efficacy, and a promising safety margin. *Mol. Pharm.* **20**, 370–382 (2023).
114. Baker, R. O., Bray, M. & Huggins, J. W. Potential antiviral therapeutics for smallpox, monkeypox and other orthopoxvirus infections. *Antiviral Res.* **57**, 13–23 (2003).
115. Smee, D. F., Bailey, K. W. & Sidwell, R. W. Treatment of cowpox virus respiratory infections in mice with ribavirin as a single agent or followed sequentially by cidofovir. *Antivir. Chem. Chemother.* **11**, 303–309 (2000).
116. Kannan, S. R. et al. Mutations in the monkeypox virus replication complex: potential contributing factors to the 2022 outbreak. *J. Autoimmun.* **133**, 102928 (2022).
117. Andrei, G. et al. Cidofovir resistance in vaccinia virus is linked to diminished virulence in mice. *J. Virol.* **80**, 9391–9401 (2006).
118. Kornbluth, R. S. et al. Mutations in the E9L polymerase gene of cidofovir-resistant vaccinia virus strain WR are associated with the drug resistance phenotype. *Antimicrob. Agents Chemother.* **50**, 4038–4043 (2006).
119. Duraffour, S. et al. KAY-2-41, a novel nucleoside analogue inhibitor of orthopoxviruses in vitro and in vivo. *Antimicrob. Agents Chemother.* **58**, 27–37 (2014).
120. Coen, N. et al. Antiherpetic activities of two novel 4'-thiothymidine derivatives, KAY-2-41 and KAH-39-149, are dependent on viral and cellular thymidine kinases. *Antimicrob. Agents Chemother.* **58**, 4328–4340 (2014).
121. Altayb, H. N. Fludarabine, a potential DNA-dependent RNA polymerase inhibitor, as a prospective drug against Monkeypox virus: a computational approach. *Pharmaceuticals* **15**, 1129 (2022).
122. Dutt, M. et al. Drug repurposing for Mpx: discovery of small molecules as potential inhibitors against DNA-dependent RNA polymerase using molecular modeling approach. *J. Cell Biochem.* **124**, 701–715 (2023).
123. Abduljalil, J. M., Elfiky, A. A. & Elgohary, A. M. Exploration of natural compounds against the human mpx virus DNA-dependent RNA polymerase in silico. *J. Infect. Public Health* **16**, 996–1003 (2023).
124. Tolonen, N., Doglio, L., Schleich, S. & Krijnsse Locker, J. Vaccinia virus DNA replication occurs in endoplasmic reticulum-enclosed cytoplasmic mini-nuclei. *Mol. Biol. Cell* **12**, 2031–2046 (2001).
125. Maruri-Avidal, L., Weisberg, A. S. & Moss, B. Direct formation of vaccinia virus membranes from the endoplasmic reticulum in the absence of the newly characterized L2-interacting protein A30.5. *J. Virol.* **87**, 12313–12326 (2013).
126. Liu, L., Cooper, T., Howley, P. M. & Hayball, J. D. From crescent to mature virion: vaccinia virus assembly and maturation. *Viruses* **6**, 3787–3808 (2014).
127. Greseth, M. D. & Traktman, P. The life cycle of the vaccinia virus genome. *Annu. Rev. Virol.* **9**, 239–259 (2022).
128. Tooze, J. et al. Progeny vaccinia and human cytomegalovirus particles utilize early endosomal cisternae for their envelopes. *Eur. J. Cell Biol.* **60**, 163–178 (1993).
129. Alzhanova, D. & Hruby, D. E. A trans-Golgi network resident protein, golgin-97, accumulates in viral factories and incorporates into virions during poxvirus infection. *J. Virol.* **80**, 11520–11527 (2006).
130. Sodeik, B. et al. Assembly of vaccinia virus: role of the intermediate compartment between the endoplasmic reticulum and the Golgi stacks. *J. Cell Biol.* **121**, 521–541 (1993).
131. Schmelz, M. et al. Assembly of vaccinia virus: the second wrapping cisterna is derived from the trans Golgi network. *J. Virol.* **68**, 130–147 (1994).
132. Sivan, G., Weisberg, A. S., Americo, J. L. & Moss, B. Retrograde transport from early endosomes to the trans-Golgi network enables membrane wrapping and egress of vaccinia virus virions. *J. Virol.* **90**, 8891–8905 (2016).
133. Bonifacino, J. S. & Rojas, R. Retrograde transport from endosomes to the trans-Golgi network. *Nat. Rev. Mol. Cell Biol.* **7**, 568–579 (2006).
134. Blasco, R. & Moss, B. Role of cell-associated enveloped vaccinia virus in cell-to-cell spread. *J. Virol.* **66**, 4170–4179 (1992).
135. Smith, G. L. & Law, M. The exit of vaccinia virus from infected cells. *Virus Res.* **106**, 189–197 (2004).
136. Roper, R. L., Wolffe, E. J., Weisberg, A. & Moss, B. The envelope protein encoded by the A33R gene is required for formation of actin-containing microvilli and efficient cell-to-cell spread of vaccinia virus. *J. Virol.* **72**, 4192–4204 (1998).
137. Roberts, K. L. & Smith, G. L. Vaccinia virus morphogenesis and dissemination. *Trends Microbiol.* **16**, 472–479 (2008).
138. Arakawa, Y. et al. The release of vaccinia virus from infected cells requires RhoA-Dia modulation of cortical actin. *Cell Host Microbe* **1**, 227–240 (2007).
139. Martinez-Quiles, N. et al. WIP regulates N-WASP-mediated actin polymerization and filopodium formation. *Nat. Cell Biol.* **3**, 484–491 (2001).
140. Frischknecht, F. et al. Actin-based motility of vaccinia virus mimics receptor tyrosine kinase signalling. *Nature* **401**, 926–929 (1999).
141. Masters, J. et al. Poxvirus infection rapidly activates tyrosine kinase signal transduction. *J. Biol. Chem.* **276**, 48371–48375 (2001).
142. Ward, B. M. Pox, dyes, and videotape: making movies of GFP-labeled vaccinia virus. *Methods Mol. Biol.* **269**, 205–218 (2004).
143. Cudmore, S., Cossart, P., Griffiths, G. & Way, M. Actin-based motility of vaccinia virus. *Nature* **378**, 636–638 (1995).
144. Hollinshead, M. et al. Vaccinia virus utilizes microtubules for movement to the cell surface. *J. Cell Biol.* **154**, 389–402 (2001).
145. Geada, M. M. et al. Movements of vaccinia virus intracellular enveloped virions with GFP tagged to the F13L envelope protein. *J. Gen. Virol.* **82**, 2747–2760 (2001).
146. Ward, B. M. & Moss, B. Visualization of intracellular movement of vaccinia virus virions containing a green fluorescent protein-B5R membrane protein chimera. *J. Virol.* **75**, 4802–4813 (2001).
147. Ward, B. M. & Moss, B. Vaccinia virus intracellular movement is associated with microtubules and independent of actin tails. *J. Virol.* **75**, 11651–11663 (2001).
148. Reeves, P. M. et al. Disabling poxvirus pathogenesis by inhibition of Abl-family tyrosine kinases. *Nat. Med.* **11**, 731–739 (2005).
149. Priyamvada, L. et al. Discovery of Retro-1 analogs exhibiting enhanced anti-vaccinia virus activity. *Front. Microbiol.* **11**, 603 (2020).
150. Eppstein, D. A. et al. Epidermal growth factor receptor occupancy inhibits vaccinia virus infection. *Nature* **318**, 663–665 (1985).
151. Carlin, C. R. Role of EGF receptor regulatory networks in the host response to viral infections. *Front. Cell. Infect. Microbiol.* **11**, 820355 (2021).
152. Yang, H. et al. Antiviral chemotherapy facilitates control of poxvirus infections through inhibition of cellular signal transduction. *J. Clin. Investig.* **115**, 379–387 (2005).
153. Beerli, C. et al. Vaccinia virus hijacks EGFR signalling to enhance virus spread through rapid and directed infected cell motility. *Nat. Microbiol.* **4**, 216–225 (2019).
154. Lai, K. M. & Lee, W. L. The roles of epidermal growth factor receptor in viral infections. *Growth Factors* **40**, 46–72 (2022).
155. Langhammer, S., Koban, R., Yue, C. & Ellerbrok, H. Inhibition of poxvirus spreading by the anti-tumor drug Gefitinib (Iressa). *Antiviral Res.* **89**, 64–70 (2011).
156. Wolffe, E. J., Weisberg, A. S. & Moss, B. Role for the vaccinia virus A36R outer envelope protein in the formation of virus-tipped actin-containing microvilli and cell-to-cell virus spread. *Virology* **244**, 20–26 (1998).
157. Miah, M. M., Tabassum, N., Afroj Zinnia, M. & Islam, A. Drug and anti-viral peptide design to inhibit the monkeypox virus by restricting A36R protein. *Bioinform. Biol. Insights* **16**, 1177932221141164 (2022).
158. McIntosh, A. A. & Smith, G. L. Vaccinia virus glycoprotein A34R is required for infectivity of extracellular enveloped virus. *J. Virol.* **70**, 272–281 (1996).

159. Herrera, E., Lorenzo, M. M., Blasco, R. & Isaacs, S. N. Functional analysis of vaccinia virus B5R protein: essential role in virus envelopment is independent of a large portion of the extracellular domain. *J. Virol.* **72**, 294–302 (1998).
160. Blasco, R. & Moss, B. Extracellular vaccinia virus formation and cell-to-cell virus transmission are prevented by deletion of the gene encoding the 37,000-Dalton outer envelope protein. *J. Virol.* **65**, 5910–5920 (1991).
161. Bryk, P., Brewer, M. G. & Ward, B. M. Vaccinia virus phospholipase protein F13 promotes rapid entry of extracellular virions into cells. *J. Virol.* **92**, e02145–17 (2018).
162. Schmutz, C., Payne, L. G., Gubser, J. & Wittek, R. A mutation in the gene encoding the vaccinia virus 37,000-M(r) protein confers resistance to an inhibitor of virus envelopment and release. *J. Virol.* **65**, 3435–3442 (1991).
163. Borrego, B., Lorenzo, M. M. & Blasco, R. Complementation of P37 (F13L gene) knock-out in vaccinia virus by a cell line expressing the gene constitutively. *J. Gen. Virol.* **80**, 425–432 (1999).
164. Merchinsky, M. et al. The development and approval of tecovirimat (TPOXX[®]), the first antiviral against smallpox. *Antiviral Res.* **168**, 168–174 (2019).
165. Warner, B. M. et al. In vitro and in vivo efficacy of tecovirimat against a recently emerged 2022 monkeypox virus isolate. *Sci. Transl. Med.* **14**, eade7646 (2022).
166. Russo, A. T. et al. Effects of treatment delay on efficacy of tecovirimat following lethal aerosol monkeypox virus challenge in cynomolgus macaques. *J. Infect. Dis.* **218**, 1490–1499 (2018).
167. O’Laughlin, K. et al. Clinical use of tecovirimat (TPOXX) for treatment of Monkeypox under an investigational new drug protocol—United States, May–August 2022. *Morb. Mortal. Wkly Rep.* **71**, 1190–1195 (2022).
168. Das, T. et al. Efficacy of smallpox approved tecovirimat (TPOXX) drug against Monkeypox: current update and futuristic prospects. *Int. J. Surg.* **109**, 1528–1530 (2023).
169. Li, D., Liu, Y., Li, K. & Zhang, L. Targeting F13 from monkeypox virus and variola virus by tecovirimat: molecular simulation analysis. *J. Infect.* **85**, e99–e101 (2022).
170. Frenois-Veyrat, G. et al. Tecovirimat is effective against human monkeypox virus in vitro at nanomolar concentrations. *Nat. Microbiol.* **7**, 1951–1955 (2022).
171. McQuiston, J. H. et al. The CDC domestic Mpx response - United States, 2022–2023. *Morb. Mortal. Wkly Rep.* **72**, 547–552 (2023).
172. Hermanussen, L. et al. Tecovirimat for the treatment of severe Mpx in Germany. *Infection* **5**, 1–6 (2023).
173. Shishkina, L. N. et al. Safety and pharmacokinetics of the substance of the anti-smallpox drug NIOCH-14 after oral administration to laboratory animals. *Viruses* **15**, 205 (2023).
174. Mazurkov, O. Y. et al. New effective chemically synthesized anti-smallpox compound NIOCH-14. *J. Gen. Virol.* **97**, 1229–1239 (2016).
175. Kabanov, A. S. et al. A comparative study of the antiviral activity of chemical compounds concerning the orthopoxviruses experiments in vivo. *Vopr. Virusol.* **58**, 39–43 (2013).
176. Bloch, E. M. et al. The potential role of passive antibody-based therapies as treatments for Monkeypox. *mBio* **13**, e0286222 (2022).
177. Saghazadeh, A. & Rezaei, N. Insights on Mpx virus infection immunopathogenesis. *Rev. Med. Virol.* **33**, e2426 (2023).
178. Li, H. et al. The land-scape of immune response to monkeypox virus. *EBioMedicine* **87**, 104424 (2023).
179. Shchelkunova, G. A. & Shchelkunov, S. N. Immunomodulating drugs based on poxviral proteins. *BioDrugs* **30**, 9–16 (2016).
180. Mack, T. M., Noble, J. Jr. & Thomas, D. B. A prospective study of serum antibody and protection against smallpox. *Am. J. Trop. Med. Hyg.* **21**, 214–218 (1972).
181. Gilchuk, I. et al. Cross-neutralizing and protective human antibody specificities to poxvirus infections. *Cell* **167**, 684–694.e689 (2016).
182. Galmiche, M. C., Goenaga, J., Wittek, R. & Rindisbacher, L. Neutralizing and protective antibodies directed against vaccinia virus envelope antigens. *Virology* **254**, 71–80 (1999).
183. Edghill-Smith, Y. et al. Smallpox vaccine-induced antibodies are necessary and sufficient for protection against monkeypox virus. *Nat. Med.* **11**, 740–747 (2005).
184. Yi Mohammadi, J. J., Franks, K. & Hines, S. Effectiveness of professional oral health care intervention on the oral health of residents with dementia in residential aged care facilities: a systematic review protocol. *JBI Database Syst. Rev. Implement. Rep.* **13**, 110–122 (2015).
185. Shearer, J. D., Siemann, L., Gerkovich, M. & House, R. V. Biological activity of an intravenous preparation of human vaccinia immune globulin in mouse models of vaccinia virus infection. *Antimicrob. Agents Chemother.* **49**, 2634–2641 (2005).
186. Thet, A. K. et al. The use of vaccinia immune globulin in the treatment of severe Mpx. virus infection in human immunodeficiency virus/AIDS. *Clin. Infect. Dis.* **76**, 1671–1673 (2023).
187. Denkinger, C. M. et al. Anti-SARS-CoV-2 antibody-containing plasma improves outcome in patients with hematologic or solid cancer and severe COVID-19: a randomized clinical trial. *Nat. Cancer* **4**, 96–107 (2023).
188. Marconato, M. et al. Antibodies from convalescent plasma promote SARS-CoV-2 clearance in individuals with and without endogenous antibody response. *J. Clin. Investig.* **132**, e158190 (2022).
189. Engelmayer, J. et al. Vaccinia virus inhibits the maturation of human dendritic cells: a novel mechanism of immune evasion. *J. Immunol.* **163**, 6762–6768 (1999).
190. Li, P. et al. Disruption of MHC class II-restricted antigen presentation by vaccinia virus. *J. Immunol.* **175**, 6481–6488 (2005).
191. Humrich, J. Y. et al. Vaccinia virus impairs directional migration and chemokine receptor switch of human dendritic cells. *Eur. J. Immunol.* **37**, 954–965 (2007).
192. Chahroudi, A. et al. Vaccinia virus tropism for primary hemolymphoid cells is determined by restricted expression of a unique virus receptor. *J. Virol.* **79**, 10397–10407 (2005).
193. Reynoso, G. V. et al. Lymph node conduits transport virions for rapid T cell activation. *Nat. Immunol.* **20**, 602–612 (2019).
194. Fischer, M. A. et al. CD11b⁺, Ly6G⁺ cells produce type I interferon and exhibit tissue-protective properties following peripheral virus infection. *PLoS Pathog.* **7**, e1002374 (2011).
195. Earl, P. L., Americo, J. L. & Moss, B. Natural killer cells expanded in vivo or ex vivo with IL-15 overcomes the inherent susceptibility of CAST mice to lethal infection with orthopoxviruses. *PLoS Pathog.* **16**, e1008505 (2020).
196. Song, H. et al. Monkeypox virus infection of rhesus macaques induces massive expansion of natural killer cells but suppresses natural killer cell functions. *PLoS One* **8**, e77804 (2013).
197. Adamo, S. et al. Memory profiles distinguish cross-reactive and virus-specific T cell immunity to mpx. *Cell Host Microbe* **31**, 928–936.e924 (2023).
198. Grifoni, A. et al. Defining antigen targets to dissect vaccinia virus and monkeypox virus-specific T cell responses in humans. *Cell Host Microbe* **30**, 1662–1670.e1664 (2022).
199. Lum, F. M. et al. Monkeypox: disease epidemiology, host immunity and clinical interventions. *Nat. Rev. Immunol.* **22**, 597–613 (2022).
200. Méndez-Lagares, G. et al. Memory T cell proliferation before hepatitis C virus therapy predicts antiviral immune responses and treatment success. *J. Immunol.* **200**, 1124–1132 (2018).
201. Zhang, M. et al. Circulating T follicular helper cells are associated with rapid virological response in chronic hepatitis C patients undergoing peginterferon therapy. *Int. Immunopharmacol.* **34**, 235–243 (2016).
202. Nakatsuka, K. et al. Ribavirin contributes to eradicate hepatitis C virus through polarization of T helper 1/2 cell balance into T helper 1 dominance. *World J. Hepatol.* **7**, 2590–2596 (2015).
203. Grubczak, K. et al. Effects of pegylated interferon alpha and ribavirin (pegIFN- α /RBV) therapeutic approach on regulatory T cells in HCV-monoinfected and HCV/HIV-coinfected patients. *Viruses* **13**, 1448 (2021).
204. Essa, S., Al-Attayah, R., Siddique, I. & Al-Nakib, W. Modulation of immune cell subsets by hepatitis C virus and antiviral therapy in early virological response HCV genotype 4-infected patients with compensated liver disease. *Med. Princ. Pract.* **30**, 168–177 (2021).
205. Riboldi, P., Gerosa, M. & Meroni, P. L. Pidotimod: a reappraisal. *Int. J. Immunopathol. Pharmacol.* **22**, 255–262 (2009).
206. Ding, L., Luo, K., Feng, C. G. & Oehlers, S. H. Pidotimod increases inflammation in wounded zebrafish embryos. *Fish Shellfish Immunol.* **120**, 429–433 (2022).
207. Zhao, N. et al. Pidotimod: a review of its pharmacological features and clinical effectiveness in respiratory tract infections. *Expert Rev. Anti-Infect. Ther.* **17**, 803–818 (2019).
208. Esposito, S. et al. Immunomodulatory activity of pidotimod administered with standard antibiotic therapy in children hospitalized for community-acquired pneumonia. *J. Transl. Med.* **13**, 288 (2015).
209. Santus, P. et al. Anti-inflammatory effects of immunostimulation in patients with COVID-19 pneumonia. *J. Clin. Med.* **10**, 5765 (2021).
210. Tao, N. et al. Thymosin α 1 and its role in viral infectious diseases: the mechanism and clinical application. *Molecules* **28**, 3539 (2023).
211. Binder, U. & Skerra, A. PASylated thymosin α 1: a long-acting immunostimulatory peptide for applications in oncology and virology. *Int. J. Mol. Sci.* **22**, 124 (2020).
212. Minutolo, A. et al. Thymosin alpha 1 restores the immune homeostasis in lymphocytes during post-acute sequelae of SARS-CoV-2 infection. *Int. Immunopharmacol.* **118**, 110055 (2023).
213. Chen, M. et al. Combination of gemcitabine and thymosin alpha 1 exhibit a better anti-tumor effect on nasal natural killer/T-cell lymphoma. *Int. Immunopharmacol.* **98**, 107829 (2021).
214. Carta, S., Silvestri, M. & Rossi, G. A. Modulation of airway epithelial cell functions by Pidotimod: NF- κ B cytoplasmic expression and its nuclear translocation are associated with an increased TLR-2 expression. *Ital. J. Pediatr.* **39**, 29 (2013).
215. Kindrachuk, J. et al. Systems kinomics demonstrates Congo Basin monkeypox virus infection selectively modulates host cell signaling responses as compared to West African monkeypox virus. *Mol. Cell Proteom.* **11**, M111.015701 (2012).

216. Kataria, R., Kaur, S. & Kaundal, R. Deciphering the complete human-monkeypox virus interactome: Identifying immune responses and potential drug targets. *Front. Immunol.* **14**, 1116988 (2023).
217. Banham, A. H. & Smith, G. L. Characterization of vaccinia virus gene B12R. *J. Gen. Virol.* **74**, 2807–2812 (1993).
218. Suraweera, C. D., Hinds, M. G. & Kvsanakul, M. Poxviral strategies to overcome host cell apoptosis. *Pathogens* **10**, 6 (2020).
219. Klaas, L. et al. Diversity of cell death signaling pathways in macrophages upon infection with modified vaccinia virus Ankara (MVA). *Cell Death Dis.* **12**, 1011 (2021).
220. Myskiw, C. et al. Nigericin is a potent inhibitor of the early stage of vaccinia virus replication. *Antiviral Res.* **88**, 304–310 (2010).
221. Nichols, R. J., Wiebe, M. S. & Traktman, P. The vaccinia-related kinases phosphorylate the N' terminus of BAF, regulating its interaction with DNA and its retention in the nucleus. *Mol. Biol. Cell* **17**, 2451–2464 (2006).
222. Boyle, K. A. & Traktman, P. Members of a novel family of mammalian protein kinases complement the DNA-negative phenotype of a vaccinia virus ts mutant defective in the B1 kinase. *J. Virol.* **78**, 1992–2005 (2004).
223. Rodrigues Garcia, D. et al. Design of inhibitors of thymidylate kinase from Variola virus as new selective drugs against smallpox: part II. *J. Biomol. Struct. Dyn.* **37**, 4569–4579 (2019).
224. Ajmal, A. et al. Computer-assisted drug repurposing for thymidylate kinase drug target in monkeypox virus. *Front. Cell Infect. Microbiol.* **13**, 1159389 (2023).
225. Khan, A. et al. Structure-based design of promising natural products to inhibit thymidylate kinase from Monkeypox virus and validation using free energy calculations. *Comput. Biol. Med.* **158**, 106797 (2023).
226. Pourhajbagher, M. & Bahador, A. Virtual screening and computational simulation analysis of antimicrobial photodynamic therapy using propolis-benzofuran A to control of Monkeypox. *Photodiagnosis Photodyn. Ther.* **41**, 103208 (2023).
227. Dao, T. L. et al. Corrigendum to “Infectious disease symptoms and microbial carriage among French medical students travelling abroad: a prospective study.” *Travel Med. Infect. Dis.* **34**, 102609 (2023).
228. Yang, C. et al. Travel before, during and after the COVID-19 pandemic: exploring factors in essential travel using empirical data. *J. Transp. Geogr.* **110**, 103640 (2023).
229. Kmiec, D. & Kirchoff, F. Monkeypox: a new threat? *Int. J. Mol. Sci.* **23**, 7886 (2022).
230. Harris, E. Global Monkeypox outbreaks spur drug research for the neglected disease. *JAMA.* **328**, 231–233 (2022).
231. Karim, M., Lo, C. W. & Einav, S. Preparing for the next viral threat with broad-spectrum antivirals. *J. Clin. Investig.* **133**, e170236 (2023).
232. Ezat, A. A. et al. The discovery of novel antivirals for the treatment of Mpx: is drug repurposing the answer? *Expert Opin. Drug Discov.* **18**, 551–561 (2023).
233. Mercorelli, B., Palù, G. & Loregian, A. Drug repurposing for viral infectious diseases: how far are we? *Trends Microbiol.* **26**, 865–876 (2018).
234. Ayon, N. J. High-throughput screening of natural product and synthetic molecule libraries for antibacterial drug discovery. *Metabolites* **13**, 625 (2023).
235. Dueñas, M. E. et al. Advances in high-throughput mass spectrometry in drug discovery. *EMBO Mol. Med.* **15**, e14850 (2023).
236. Bon, M., Bilsland, A., Bower, J. & McAulay, K. Fragment-based drug discovery—the importance of high-quality molecule libraries. *Mol. Oncol.* **16**, 3761–3777 (2022).
237. Bassani, D. & Moro, S. Past, present, and future perspectives on computer-aided drug design methodologies. *Molecules* **28**, 3906 (2023).
238. Sadybekov, A. V. & Katritch, V. Computational approaches streamlining drug discovery. *Nature* **616**, 673–685 (2023).
239. Vemula, D. et al. CADD, AI and ML in drug discovery: a comprehensive review. *Eur. J. Pharm. Sci.* **181**, 106324 (2023).
240. Luna, N. et al. Monkeypox virus (MPXV) genomics: a mutational and phylogenomic analyses of B.1 lineages. *Travel Med. Infect. Dis.* **52**, 102551 (2023).
241. Salamango, D. J. & Harris, R. S. Demystifying cell cycle arrest by HIV-1 Vif. *Trends Microbiol.* **29**, 381–384 (2021).
242. Azimi, F. C. & Lee, J. E. Structural perspectives on HIV-1 Vif and APOBEC3 restriction factor interactions. *Protein Sci.* **29**, 391–406 (2020).
243. Colson, P. et al. Sequencing of monkeypox virus from infected patients reveals viral genomes with APOBEC3-like editing, gene inactivation, and bacterial agents of skin superinfection. *J. Med. Virol.* **95**, e28799 (2023).
244. Dobrovolná, M., Brázda, V., Warner, E. F. & Bidula, S. Inverted repeats in the monkeypox virus genome are hot spots for mutation. *J. Med. Virol.* **95**, e28322 (2023).
245. Dumonteil, E., Herrera, C. & Sabino-Santos, G. Monkeypox virus evolution before 2022 outbreak. *Emerg. Infect. Dis.* **29**, 451–453 (2023).
246. Blanco-González, A. et al. The role of AI in drug discovery: challenges, opportunities, and strategies. *Pharmaceuticals* **16**, 891 (2023).
247. Chadaga, K. et al. Application of artificial intelligence techniques for Monkeypox: a systematic review. *Diagnostics* **13**, 824 (2023).
248. Cheng, K. et al. Talk with ChatGPT about the outbreak of Mpx in 2022: reflections and suggestions from AI dimensions. *Ann. Biomed. Eng.* **51**, 870–874 (2023).
249. Gentile, F., Oprea, T. I., Tropsha, A. & Cherkasov, A. Surely you are joking, Mr Docking! *Chem. Soc. Rev.* **52**, 872–878 (2023).
250. Doan, S. et al. Severe corneal involvement associated with Mpx infection. *JAMA Ophthalmol.* **141**, 402–403 (2023).
251. Ogoina, D., Mohammed, A., Yinka-Ogunleye, A. & Ihekweazu, C. A case of suicide during the 2017 monkeypox outbreak in Nigeria. *IJD Reg.* **3**, 226–227 (2022).
252. Hanna, E., Abadi, R. & Abbas, O. Imiquimod in dermatology: an overview. *Int. J. Dermatol.* **55**, 831–844 (2016).
253. Gupta, A. K., Browne, M. & Bluhm, R. Imiquimod: a review. *J. Cutan Med. Surg.* **6**, 554–560 (2002).
254. Skinner, R. B. Jr. Imiquimod as an immune response modulator in infectious conditions. *Postgrad. Med.* **112**, 8–16 (2002).
255. Hengge, U. R. & Cusini, M. Topical immunomodulators for the treatment of external genital warts, cutaneous warts and molluscum contagiosum. *Br. J. Dermatol.* **149**, 15–19 (2003).
256. Dahl, M. V. Imiquimod: a cytokine inducer. *J. Am. Acad. Dermatol.* **47**, S205–S208 (2002).
257. Roper, R. L. et al. Monkeypox (Mpx) requires continued surveillance, vaccines, therapeutics and mitigating strategies. *Vaccine* **41**, 3171–3177 (2023).
258. Schildhauer, S. et al. Reduced odds of Mpx-associated hospitalization among persons who received JYNNEOS Vaccine - California, May 2022–May 2023. *Morb. Mortal. Wkly Rep.* **72**, 992–996 (2023).
259. Sammartino, J. C. et al. Characterization of immune response against monkeypox virus in cohorts of infected patients, historic and newly vaccinated subjects. *J. Med. Virol.* **95**, e28778 (2023).
260. Saadh, M. J. et al. Progress and prospects on vaccine development against monkeypox infection. *Microb. Pathog.* **180**, 106156 (2023).
261. Abdelaal, A. et al. Preventing the next pandemic: is live vaccine efficacious against Monkeypox, or is there a need for killed virus and mRNA vaccines? *Vaccines* **10**, 1419 (2022).
262. FDA. Monkeypox update: FDA authorizes emergency use of JYNNEOS vaccine to increase vaccine supply, <https://www.fda.gov/news-events/press-announcements/monkeypox-update-fda-authorizes-emergency-use-jynneos-vaccine-increase-vaccine-supply> (2022).
263. Eustaquio, P. C., Salmon-Trejo, L. A. T., McGuire, L. C. & Ellington, S. R. Epidemiologic and clinical features of mpx in adults aged >50 years—United States, May 2022–May 2023. *Morb. Mortal. Wkly Rep.* **72**, 893–896 (2023).
264. Cohn, H. et al. Mpx vaccine and infection-driven human immune signatures: an immunological analysis of an observational study. *Lancet Infect. Dis.* **23**, 00352–00353 (2023).



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