Review

ANNALS OF MEDICINE & SURGERY

OPEN

Comprehensive overview of human monkeypox: epidemiology, clinical features, pathogenesis, diagnosis and prevention

Eman Ali, MBBS[a,*], Ayesha Sheikh, MBBS[a], Rabia Owais, MBBS[a], Asim Shaikh, MBBS[b], Unaiza Naeem, MBBS[a]

Abstract

Monkeypox (MPX) is a zoonotic disease caused by the monkeypox virus (MPXV), belonging to the orthopoxvirus genus with a presentation resembling smallpox making it historically challenging to distinguish the disease from smallpox clinically. Since a British citizen brought MPX into the country on 6 May 2022, there have been concerns about the re-emergence of the human MPXV. Since then, the WHO has reported 92 confirmed cases and 28 suspected cases in 13 nations where MPXV was not endemic. WHO declared MPX a 'public health emergency of international concern' on 23 July 2022. MPXV can spread either through human-human contact or animal-human contact. Respiratory droplets, direct contact with bodily fluids, contaminated patient surroundings or objects, and skin sores from an infected person have all been linked to the disease's transmission from one person to another. Fever, headache, lethargy, asthenia, enlargement of the lymph nodes, weariness, back pain, and myalgia are some of the symptoms that last from 2 to 5 weeks. It can be diagnosed using a range of diagnostic methods, including electron microscopy, Immunoglobulin M, enzyme-linked immunosorbent assay, polymerase chain reactions, histological analysis, immunofluorescent antibody testing, virus isolation, etc. Smallpox immunization before infection may lessen clinical symptoms and is around 85% effective in protecting from the MPXV.

Keywords: epidemiology, monkeypox virus, orthopoxviruses, prevention, re-emergence

Introduction

The zoonotic illness known as monkeypox (MPX), which has been extensively reported on, is caused by the monkeypox virus (MPXV), a member of the genus Orthopoxvirus and family Poxviridae. Along with the vaccinia virus, cowpox virus, and the variola virus, which is now extinct, MPXV is one of the four orthopoxvirus species that can prove to be fatal in human beings[1,2]. It can be communicated through direct or indirect contact with contaminated fomites, respiratory droplets, body fluids, and skin lesions of sick animals that can spread the illness[3]. Clinical indicators of this illness include a pustular rash coupled with other clinical signs including adenopathy and maculopapular rash, particularly on the soles of the feet and the palms of the hands, but they are typically less severe than those of smallpox[4,5]. A corneal infection with subsequent vision loss, respiratory distress, bronchopneumonia, gastrointestinal involvement, dehydration, sepsis, and encephalitis are all potential complications[6].

HIGHLIGHTS

- WHO declared monkeypox a 'public health emergency of international concern' on 23 July 2022.
- Administration of a specific postexposure vaccine or pharmaceutical intervention should be decided by the bedside physician after a thorough cost-benefit analysis and strict exclusion of absolute contraindications.

The WHO declared the worldwide MPX outbreak a public health emergency of international concern on 23 July and provided advice to all nations to prevent the spread and manage the pandemic[7]. Since early May 2022, there has been an increase in cases worldwide, including those from countries where the disease is not endemic. On 17 August, the WHO president announced that there had been more than 35 000 cases in 92 countries, along with 12 fatalities[8]. The WHO European Region reported the vast majority of laboratory-confirmed cases (2933-/3413; 86%). Cases are also reported in the Eastern Mediterranean Region (15/3413), the Western Pacific Region (11/3413), the African Region (73/3413, 2%), and the Americas Region (381/3413, 11%). During the second quarter of 2022, Nigeria reported one fatality[7].

Geographical distribution and epidemiology

As an epidemic of a pox-like disease in monkeys, the MPXV was originally discovered in Denmark in 1959. A child in the

[a]Dow University of Health Sciences and [b]Department of Internal Medicine, Aga Khan University, Karachi, Pakistan

Sponsorships or competing interests that may be relevant to content are disclosed at the end of this article.

*Corresponding author. Address: Department of Internal Medicine, Dow University of Health Sciences, Baba-e-Urdu Road, Saddar, Karachi, Pakistan. Tel: +923362394255. E-mail: Eman.Ali21@dmc.duhs.edu.pk (E. Ali)

Annals of Medicine & Surgery (2023) 85:2767–2773

Received 15 September 2022; Accepted 17 April 2023

Published online 3 May 2023

http://dx.doi.org/10.1097/MS9.0000000000000763

Democratic Republic of the Congo (DRC) was diagnosed with the first documented case of smallpox in humans in 1970[9,10]. The West African and Congo Basins are the two clades of human MPXV that have been identified (Central Africa). The DRC and the Central African Republic, which are both located in Central Africa, have been the primary victims of the Congo basin clade, which is linked to greater rates of illness, mortality, and transmission in humans[11,12]. The 10 African countries with data on human MPX cases include the DRC, the Central African Republic, Nigeria, Cameroon, the Ivory Coast, Liberia, Sierra Leone, South Sudan, and Gabon[13]. On 6 May 2022, a MPX epidemic was confirmed in the United Kingdom (UK), with the index case being a British national who contracted the disease in Nigeria, where it is endemic[14]. In 13 countries where MPXV had not previously been widely distributed, including the United Kingdom, Italy, the Netherlands, Portugal, Spain, Canada, France, Germany, Sweden, Australia, Belgium, and the United States, WHO has reported 92 confirmed cases and 28 suspected cases with no reported deaths[15,16]. The United Arab Emirates (UAE) reported the first infection case in the middle east on May 24[17]. MPX, a disease generally found in central and western Africa, is recently being reported globally for the first time in fifty years.

Figure 1 highlights all countries that have seen MPX outbreaks recently[7]. The epidemiological characteristics of the human MPXV from 1970 to 2022 are shown in Table 1[18].

Virology

The zoonotic infectious disease known as MPX is caused by the MPXV, a double-stranded DNA virus of the Poxviridae family and Orthopoxvirus genus, which can cause a systemic illness that ranges in clinical severity from moderate to lethal[19]. A large family of viruses known as poxiridae completes its whole life cycle in the cytoplasm of infected cells. Eighteen species in this family infect vertebrates, with Orthopoxvirus being the most significant because of its potential for zoonotic spread. The other three are the cowpox virus, the variola minor virus, and the now eradicated variola major virus, which was the primary cause of smallpox. MPXV is one of four orthopoxvirus species that are harmful to humans[20]. The MPXV virus particles are defined by the appearance of egg-shaped or brick-shaped particles encased in a geometrically corrugated lipoprotein outer membrane and range in size from 200 to 250 nm, according to electron microscopy data[21]. MPXV is of the utmost medical significance since it causes a human infection with smallpox-like clinical signs and fatality rates[22].

There are two ways that MPXV might spread: through animal-human contact or human-human contact. While touching MPX-infected animals is assumed to be the primary source of animal-to-human transmission, the precise mechanism(s) by which this occurs is unknown. This can happen directly (by touch, bite, or scratch) or indirectly (via inhalation). The respiratory tract, injured skin, or mucous membranes are assumed to be the routes through which the virus enters the body (eyes, nose, or mouth). Transmission of the disease from one person to another has been associated to respiratory droplets, coming into direct contact with bodily fluids, contaminated patient environments or objects, and skin sores from an infected person[23]. Nearly 28% of the instances included secondary human-to-human transmission; tertiary and quaternary transmission chains are rare[22]. The findings of the study by Reynolds *et al.*[24] indicate that the mode of infection may affect how the illness caused by MPXV manifests. It has been noted that animals can transmit aerosols[25]. Additionally, there is evidence of nosocomial transmission[26]. Moreover, recent data suggests there has been some involvement of sexual transmission in the further spread of this precarious disease. In Italy, four cases of

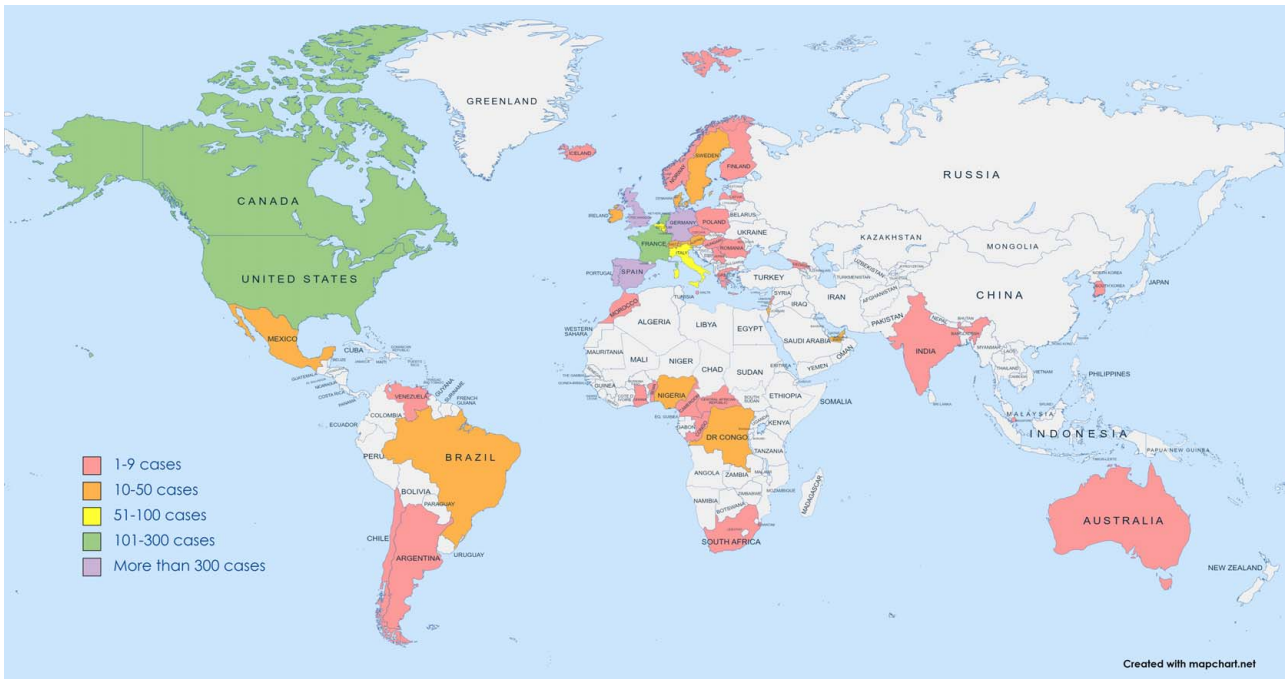

Figure 1. Current global prevalence of Monkeypox[7].

Table 1

Updated epidemiology of monkeypox from 1970 to 2022[18]

Features	1970–1979	1981–1986	1996–1997	2003	2017–2019	2022
Location	Central and western Africa	Democratic Republic of Congo	Democratic Republic of Congo	Central USA	Nigeria	United Kingdom
Epidemiological setting	Passive surveillance	Active surveillance	Outbreak	Outbreak	Outbreak	Outbreak
Number of reported cases	47	338	419	81	424	1735
Laboratory confirmed	87	100	Not known	40	37	Not known
Median age (y)	4	Not known	Not known	27	31	36
Suspected primary sources	Not known	Forest animals	Not known	Prairie dog, Gambian giant	Not known	Import from Nigeria in a UK resident
Primary cases (%)	91	72	22	100	Not known	Not known

young adult men were reported who had sex without using a condom. The patients did not require any specific antiviral medications because they were in good clinical condition. Seminal fluid biological samples tested positive for MPXV DNA. There is no proof of sexual transmission for several other viruses that have been discovered in semen. However, it is necessary to look at the possibilities of MPX viral sexual transmission[27]. However, a growing number of scientists now believe that the main driver of MPX transmission worldwide is likely sexual activity between men, including both anal and oral sex[28]. Since the current worldwide MPX outbreak began in Europe, the vast majority of cases have been discovered in men who have had sex with men (MSM). From 17 May to 22 June, the first 5 weeks of the MPX outbreak in the Madrid region saw 508 cases, with 427 patients reporting condomless sex or sex with many partners in the 21 days preceding the onset of symptoms, the majority of whom were MSM[29].

Mutations

Studies published in 2020 showed that MPXV lacked the viral ability to propagate widely among humans. Minor changes in viral proteins could still happen, aggravating future occurrences of human-human transmission, as the H1N1 virus outbreak revealed[30]. The MPXV strain that is generating the current outbreak in countries where it is not widespread likely diverged from the one that caused an epidemic in 2018–2019. The most comprehensive analysis of the virus' genetic makeup to date, is genetic research by Portuguese scientists to sequence the current strain's genome. The initial strain (first identified in 1958) and the present strain differ by 50 single nucleotide polymorphisms, and numerous mutations increased the virus's capacity for transmission. The strain is a member of West African virus clade 3, which has a lower mortality rate than the Congo Basin clade[31]. One of the virus-fighting enzyme groups that the researchers speculate may have contributed to the genesis of many of the mutations they found is the apolipoprotein B mRNA-editing enzyme, catalytic polypeptide 3 (APOBEC3) family. When viruses copy their genetic code, these enzymes prevent the absolute replication of said code, which often leads to the disintegration of the virus. The virus can occasionally survive the encounter and acquire some genetic changes. Additionally, compared to MPXV strains sequenced 4 years ago, the MPXV-WA lineage strains linked to the current outbreak differ by about 40 nucleotide mutations, raising the possibility that the evolutionary rate has increased from one mutation per genome per year to 12 mutations per genome per year[32]. Future research on the role of APOBEC3 in MPXV diversification is therefore necessary. Functional studies are also needed to determine whether this mutational mechanism causes MPXV adaptive evolution toward different phenotypic traits, such as increased transmissibility.

Clinical manifestations and symptomatology

The illness starts with a fever before rashes appear, and the incubation period for MPX is 10–21 days. Symptoms include fever, headache, lethargy, asthenia, lymph node enlargement, fatigue, back pain, and myalgia. The lasting duration of symptoms ranges from 2–5 weeks[13]. Clinical signs of MPXV include lesions and symptoms that are comparable to smallpox[18]. More than 90% of patients had considerable lymphadenopathy 1–2 days before the rash, which is an essential distinction between human MPX and smallpox. It can affect the lymph nodes in the submandibular, cervical, postauricular, axillary, or inguinal regions, and presentation can be unilateral or bilateral[33]. These lesions have a diameter of roughly 0.5 cm, yet some of them have the potential to double in size and enlarge to 1 cm[34]. The initial week following the appearance of the rash marks the inception of the infectious phase[35]. Beginning with macules, papules, vesicles, and pustules, the rash goes through several stages before concluding with crusts and scabs that break off during recovery. The rash may go through several stages at once. Around isolated lesions, erythema, and/or hyperpigmentation of the skin are usually observed. The detached scab may be considerably smaller than the original lesion. Inflammation of the vaginal, conjunctival, and pharyngeal mucosae is also possible[13–23].

Table 2 highlights all the common clinical signs and symptoms associated with MPXV[24].

Diagnosis, laboratory sample collection, and testing methods

Due to the clinical similarities between MPX, chickenpox, and smallpox, it is essential to consider minor diagnostic criteria to rule out these common diseases. Differential diagnosis is the key to curbing this disease, especially in light of the exponentially rising incidence of vaccine hesitancy globally[36]. The evaluation criteria for differential diagnosis for patients with MPX include an incubation period of 7–17 days, a prodrome period of 1–4 days, and major symptoms including fever, malaise, headache, and lymphadenopathy with moderate severity[37]. Since there are no clinically tested antiviral medications available for

Table 2

Clinical signs and symptoms due to monkeypox virus[24]

Systems involved	Signs and symptoms
Gastrointestinal	Diarrhea, Nausea, Vomiting.
Upper respiratory	Runny nose, Sore throat.
Lower respiratory	Wheeze, Cough, Respiratory Distress.
General respiratory	Combined symptoms from the upper and lower respiratory categories.
Systemic	Lymphadenopathy, Back Pain, Fever, Headache, Sweating, Chills, Abdominal Ache, Muscle Ache.

the treatment of MPX, rapid quarantine and ring vaccines are the only effective public protective measures once the identification of the disease agent is verified.

As the virus is easily transmitted from person to person through aerosol particles and direct contact, it is advised to collect specimens such as scabs or other cutaneous tissues with respiratory precautions and handle them with caution[38]. At least two scabs or samples from vesicles should be collected in separate sterile containers by using a sterile scalpel or 26-gage needle to unroof the lesions[39]. A sterile cotton or polyester swab should be used to vigorously scrub the vesicle's base. The material should then be put on a clean microscope slide and allowed to air dry. Because dilution can alter the outcomes of subsequent tests, the swabbed sample should not be kept in transport media. Only dry swabs or swabs in viral transport media from lesions or lesion crusts are currently accepted for testing at the CDC. Swabs in bacterial preservation media may cause polymerase chain reactions (PCR) inhibition and are not recommended[40]. Swabs should be placed in viral transport media like Virocult® MW951S or VCMTM MW910S. Scab scrapings should be taken only if no other lesions are present and sent in a dry sterile tube. Scraping testing may take longer than a lesion swab. Σ MM™, MWMM, or MW0250 can be used to inactivate specimens[41]. When transporting the sample to the CDC (or comparable national reference laboratory) for additional diagnostic testing, it should be kept on dry ice or at a temperature of – 20°C[22].

The MPXV can be detected using a variety of diagnostic methods, such as electron microscopy, Immunoglobulin M, enzyme-linked immunosorbent assay, PCR, histopathological analysis, immunofluorescent antibody tests, virus isolation, etc. Several nucleic acid amplification tests have been developed to detect MPX. A prospective study, which evaluated the performance characteristics of the Viasure MPVX PCR Assay demonstrated excellent performance characteristics with 91.2% of paired swabs generating concordant results while 8.8% generating nonconcordant results[42]. Additional TaqMan probe-based real-time PCR assays targeting both MPX clades, as well as a generic MPX assay, have been reported[43]. Diagnostic devices and commercial tests for coronavirus disease 2019 (COVID-19) such as the DASH SARS-CoV-2/S Test, Novel Coronavirus (2019-nCoV) Nucleic Acid Diagnostic Kit (PCR-Fluorescence Probing), and Amplitude Solution with the TaqPath COVID-19 High-Throughput Combo Kit with sensitivity and specificity can be utilized for MPX diagnostic purposes[44]. Furthermore, a self-contained cartridge (Cepheid GeneXpert) was recently developed as an alternative to traditional PCR detection methods. Laboratory Response Network (LRN) laboratories in the United States and other locations around the world offer diagnostic testing for orthopoxviruses, including MPX[45]. On 17 May 2022, the Massachusetts Department of Public Health contacted the CDC about a suspected case of MPX, a disease caused by the Orthopoxvirus MPXV. Specimens were collected and tested using the NVO assay by the Massachusetts Department of Public Health public health laboratory with LRN testing capability. In the month of June, 68 LRN laboratories across the country had the capacity to test ~8000 NVO tests per week. LRN laboratories tested 2009 specimens from suspected MPX cases. Out of 730 (36.3%) of the specimens from 395 patients tested positive for NVO, the CDC confirmed MPX in NVO-positive specimens from 159 persons[46].

Moreover, IgM enzyme-linked immunosorbent assay detects recent infection with orthopoxviruses and, in this case, MPXV. Of 37 confirmed cases, 29 either had or developed an IgG response and 34 developed an IgM response regardless of vaccination status[47]. Methods for detecting the presence of Orthopoxvirus in suspected samples include immunohistochemical staining for orthopoxvirus antigens, electron microscopy visualization, and serum studies for anti-orthopoxvirus IgM (indicating recent exposure). The use of these tests in clinical practice is, however, extremely rare[48].

There are point-of-care diagnostics that can be utilized in very basic settings with low training requirements, which can be extremely beneficial for lower-income countries which owing to limitations in resources are unable to provide effective diagnostic and therapeutic strategies to patients[4–49]. However, one of the greatest methods for differentiating between poxvirus and herpes virus infection is the use of immunohistochemical analysis, which makes use of monoclonal and polyclonal antibodies. In the past, viral diagnosis has benefited greatly from electron microscopy[50]. This approach, however, is unable to distinguish between distinct orthopoxvirus species. Although it is possible to isolate the smallpox virus from blood, especially during the prodromal viremic period, there is scarce information on the use of blood cultures for MPXV isolation. In some circumstances, blood collection for paired acute-phase and convalescent-phase serum samples can be helpful[18].

In conclusion, the most accurate techniques for identifying and classifying orthopoxviruses are DNA-based testing, such as PCR with subsequent sequencing[39]. Due to the close antigenic relationships among the surface antigens of the orthopoxviruses, serological testing for MPXV antigens is challenging. A virus-neutralizing test using hyper-immune reference sera and a haemagglutination-inhibition assay using chicken erythrocytes are some of the serological techniques that are presently available[51].

Role of Vaccination in Prevention

The findings suggest that smallpox immunization before infection may improve clinical symptoms and provide protection against the MPXV[52].

At present, there are three smallpox immunizations in the United States Strategic National Stockpile: JYNNEOSTM (otherwise called IMVAMUNE, IMVANEX, and MVA-BN) originated by the Danish biotechnology company, Bavarian Nordic, based in Kvistgaard, Denmark, and ACAM2000® are authorized for smallpox; the Aventis Pasteur Smallpox Vaccine, under an investigational new drug protocol, could be utilized for

Table 3
Current vaccines available for MPXV

Country	Vaccines	Manufacturer	Mechanism of action	Doses	Indication
USA	Jynneos[53,54]	Bavarian Nordic.	Modified vaccinia Ankara strain vaccine (MVA-BN)	Two doses, at least 28 days.	Prevention of smallpox and monkeypox disease in adults 18 years of age and older determined to be at high risk for smallpox or monkeypox infection.
USA	ACAM2000[53,54]	Emergent BioSolutions	Live-attenuated smallpox vaccine	1 dose, percutaneous route (scarification), booster every 3 years.	Active immunization against smallpox and monkeypox disease for persons determined to be at high risk for smallpox infection.
Canada	Imvamune[54,55]	Bavarian Nordic A/S	Live vaccine produced from Modified Vaccinia Ankara, attenuated (MVA), nonreplicating.	Two doses, s.c. - booster in previously vaccinated.	Active immunization against smallpox, monkeypox and related orthopoxvirus infection and disease in adults greater than or equal to 18 years determined to be at high risk for exposure.
Europe	IMVANEX[54–56]	Bavarian Nordic A/S	Live vaccine produced from Modified Vaccinia Ankara, attenuated (MVA), nonreplicating.	Two doses, s.c. -booster in previously vaccinated.	Active immunization against monkeypox and smallpox disease for persons 18 years of age and older.
USA	APSV: Aventis Pasteur smallpox vaccine[54–57]	SP	Live replicating vaccinia virus, derived from NY City Board of health strain of vaccinia.	One dose scarification.	Strategic National Stockpile-Used in smallpox and monkeypox emergency.
Canada	Smallpox vaccine (dried and frozen liquid formulation)[54–58].	SP	Live vaccinia virus, derived from NYCBH.	Single dose, scarification	released in emergency situation for active immunization against monkeypox and smallpox.

smallpox. The potential vaccinations against MPXV have been highlighted in Table 3.

Certain people at risk of occupational contact with the orthopoxvirus are advised to get vaccinated by the Advisory Committee of Immunization Practices[59]. Clinical laboratory staff, laboratory staff, and members of designated response teams at risk of occupational exposure to the orthopoxvirus are advised to get vaccinated. These individuals perform diagnostic tests for the orthopoxvirus. ACAM2000® users and caregivers of patients afflicted with replicative orthopoxvirus may also receive vaccinations based on shared clinical decisions. Long-term, close contact with the person exhibiting symptoms is necessary for the spread of MPX. Short-term interactions with the use of suitable personal protective equipment in compliance with standard safety precautions do not pose a high risk and typically do not warrant postexposure treatment (PEP)[60].

In order to evaluate exposure risk and make wise choices regarding PEP, the CDC has developed the necessary guidance. To help avoid disease, the CDC advises receiving the first immunization within 4 days after exposure.The vaccination can lessen disease symptoms when given 4–14 days after exposure, but it cannot prevent the disease onset[60]. High-degree exposures include unprotected skin-to-mucous membrane contact, skin lesions, bodily fluids (e.g. any sexual contact, unintentional splashes of patient saliva into the eyes or oral cavity) and being inside a patient's room or within 6 feet of a patient during any procedures that may create aerosols from oral secretions should be well-monitored as well as receive the PEP vaccination.

Patients typically heal without the need for medical attention. To reduce gastrointestinal fluid losses, those who experience gastrointestinal symptoms (such as vomiting or diarrhea) will need oral or intravenous rehydration[6]. Additionally, vaccinia immunoglobulin has received FDA approval for the treatment of certain vaccine-related side effects[61]. Administration of a specific PEP vaccine or pharmaceutical intervention should be decided by the bedside physician after a thorough cost-benefit analysis and strict exclusion of absolute contraindications.

Conclusion

MPXV emergence appears to be a recurring problem in COVID-19 pandemic regions. Due to the lack of disease-specific vaccines along with the availability of only symptomatic treatment, management of MPX is challenging. In addition, more trials should be conducted to assess the efficacy of the vaccines. Comprehensive research to address emerging challenges must be performed, together with the establishment of full surveillance systems, for the prevention and management of future MPX epidemics. The best strategy to control the increase would be to lay a great emphasis on primary prevention at the community level.

Informed consent.

This study did not involve any volunteers or patients, hence no consent was needed.

Funding

No funding was acquired for this paper.

Conflict of interests.

None.

Acknowledgments

None.

References

[1] Shchelkunov SN, Marennikova SS, Moyer RW. Orthopoxviruses Pathogenic for Humans. Springer, US.
[2] Sklenovská N, Van, Ranst M. Emergence of monkeypox as the most important orthopoxvirus infection in humans. Front Public Health 2018;6:241.
[3] Grant R, Nguyen LL, Breban R. Modelling human-to-human transmission of monkeypox. Bull World Health Organ 2020;98:638–40.
[4] McCollum AM, Damon IK. Human monkeypox. Clin Infect Dis 2014;58:260–7.
[5] Vaughan A, Aarons E, Astbury J, *et al*. Human-to-human transmission of monkeypox virus, United Kingdom, October 2018. Emerg Infect Dis 2020;26:782–5.
[6] Reynolds MG, McCollum AM, Nguete B, *et al*. Improving the care and treatment of monkeypox patients in low-resource settings: applying evidence from contemporary biomedical and smallpox biodefense research. Viruses 2017;9:380.
[7] Organization WH. Multi-country monkeypox outbreak: situation update World Health Organization; 2022. https://www.who.int/emergencies/disease-outbreak-news/item/2022-DON396
[8] Nations WU. Monkeypox cases top 35,000: WHO 2022. https://news.un.org/en/story/2022/08/1124892
[9] Khodakevich L, Jezek Z, Kinzanzka K. Isolation of monkeypox virus from wild squirrel infected in nature. Lancet 1986;1:98–9.
[10] Radonić A, Metzger S, Dabrowski PW, *et al*. Fatal monkeypox in wild-living sooty mangabey, Côte d'Ivoire, 2012. Emerg Infect Dis 2014;20:1009–11.
[11] Likos AM, Sammons SA, Olson VA, *et al*. A tale of two clades: monkeypox viruses. J Gen Virol 2005;86(Pt 10):2661–72.
[12] Berthet N, Descorps-Declère S, Besombes C, *et al*. Genomic history of human monkey pox infections in the Central African Republic between 2001 and 2018. Sci Rep 2021;11:13085.
[13] Petersen E, Kantele A, Koopmans M, *et al*. Human Monkeypox: epidemiologic and clinical characteristics, diagnosis, and prevention. Infect Dis Clin North Am 2019;33:1027–43.
[14] Organization WH. Monkeypox - United Kingdom of Great Britain and Northern Ireland 2022. https://www.who.int/emergencies/disease-outbreak-news/item/2022-DON383
[15] Organization WH. Multi-country monkeypox outbreak in non-endemic countries 2022. https://www.who.int/emergencies/disease-outbreak-news/item/2022-DON385
[16] Velavan TP, Meyer CG. Monkeypox 2022 outbreak: an update. Trop Med Int Health 2022;27:604–5.
[17] J. L. Monkeypox: Israel, Switzerland and Austria confirm cases 2022. https://www.bbc.com/news/health-61540474
[18] Di Giulio DB, Eckburg PB. Human monkeypox: an emerging zoonosis. Lancet Infect Dis 2004;4:15–25.
[19] Beer EM, Rao VB. A systematic review of the epidemiology of human monkeypox outbreaks and implications for outbreak strategy. PLoS Negl Trop Dis 2019;13:e0007791.
[20] Quarleri J, Delpino MV, Galvan V. Monkeypox: considerations for the understanding and containment of the current outbreak in non-endemic countries. Geroscience 2022;44:2095–103.
[21] McFadden G. Poxvirus tropism. Nat Rev Microbiol 2005;3:201–13.
[22] Preiser W. FF. ZJ. Human Monkeypox1 ed. 1988.
[23] Hutin YJ, Williams RJ, Malfait P, *et al*. Outbreak of human monkeypox, Democratic Republic of Congo, 1996 to 1997. Emerg Infect Dis 2001;7:434–8.
[24] Reynolds MG, Yorita KL, Kuehnert MJ, *et al*. Clinical manifestations of human monkeypox influenced by route of infection. J Infect Dis 2006;194:773–80.
[25] Wenner HA, Macasaet FD, Kamitsuka PS, *et al*. Monkey pox. I. Clinical, virologic and immunologic studies. Am J Epidemiol 1968;87:551–66.
[26] Ellis CK, Carroll DS, Lash RR, *et al*. Ecology and geography of human monkeypox case occurrences across Africa. J Wildl Dis 2012;48:335–47.
[27] Antinori A, Mazzotta V, Vita S, *et al*. Epidemiological, clinical and virological characteristics of four cases of monkeypox support transmission through sexual contact, Italy, May 2022. Euro Surveill 2022;27:2200421.
[28] Sex between men, not skin contact, is fueling monkeypox, new research suggests nbcnews.com 2022. https://www.nbcnews.com/nbc-out/out-health-and-wellness/sex-men-not-skin-contact-fueling-monkeypox-new-research-suggests-rcna43484
[29] Iñigo Martínez J, Gil Montalbán E, Jiménez Bueno S, *et al*. Monkeypox outbreak predominantly affecting men who have sex with men, Madrid, Spain, 26 April to 16 June 2022. Euro Surveill 2022;27:2200471.
[30] Le QM, Sakai-Tagawa Y, Ozawa M, *et al*. Selection of H5N1 influenza virus PB2 during replication in humans. J Virol 2009;83:5278–81.
[31] Soucheray S. Virus causing monkeypox outbreak has mutated to spread easier 2022. https://www.cidrap.umn.edu/news-perspective/2022/06/virus-causing-monkeypox-outbreak-has-mutated-spread-easier
[32] Genomic epidemiology of monkeypox virus, Next Strain 2022. https://nextstrain.org/monkeypox/hmpxv1
[33] Brown K, Leggat PA. Human monkeypox: current state of knowledge and implications for the future. Trop Med Infect Dis 2016;1:8.
[34] Breman JG, Kalisa R, Steniowski MV, *et al*. Human monkeypox, 1970-79. Bull World Health Organ 1980;58:165–82.
[35] Jezek Z, Marennikova SS, Mutumbo M, *et al*. Human monkeypox: a study of 2,510 contacts of 214 patients. J Infect Dis 1986;154:551–5.
[36] Organization WH. Ten threats to global health in 2019 2019. https://www.who.int/news-room/spotlight/ten-threats-to-global-health-in-2019
[37] Nalca A, Rimoin AW, Bavari S, *et al*. Reemergence of monkeypox: prevalence, diagnostics, and countermeasures. Clin Infect Dis 2005;41:1765–71.
[38] Mbala PK, Huggins JW, Riu-Rovira T, *et al*. Maternal and fetal outcomes among pregnant women with human monkeypox infection in the Democratic Republic of Congo. J Infect Dis 2017;216:824–8.
[39] Murray EJB PR, Jorgenson JH, Pfaller MA, *et al*. Manual of clinical microbiology 8th edition. Diagn Microbiol Infect Dis 2003;47:625–6.
[40] Guidelines for Collecting and Handling Specimens for Mpox Testing 2022. https://www.cdc.gov/poxvirus/mpox/clinicians/prep-collection-specimens.html
[41] Using the Right Specimen Collection Product for Monkeypox 2022. https://www.rapidmicrobiology.com/news/using-the-right-specimen-collection-product-for-monkeypox#:~:text=Monkeypox%20is%20diagnosed%20by%20performing,or%20%CE%A3%20VCM%E2%84%A2%20MW910S
[42] Kulesh DA, Loveless BM, Norwood D, *et al*. Monkeypox virus detection in rodents using real-time 3'-minor groove binder TaqMan assays on the Roche LightCycler. Lab Invest 2004;84:1200–8.
[43] Li Y, Zhao H, Wilkins K, *et al*. Real-time PCR assays for the specific detection of monkeypox virus West African and Congo Basin strain DNA. J Virol Methods 2010;169:223–7.
[44] Current antigen and molecular tests with FDA EUA Status. John Hopkins Center for Health Security. https://www.centerforhealthsecurity.org/covid-19TestingToolkit/molecular-based-tests/current-molecular-and-antigen-tests.html
[45] Minhaj FS, Ogale YP, Whitehill F, *et al*. Monkeypox Outbreak - Nine States, May 2022. MMWR Morb Mortal Wkly Rep 2022;71:764–9.
[46] Aden TA, Blevins P, York SW, *et al*. Rapid diagnostic testing for response to the monkeypox outbreak - laboratory response network, United States, May 17-June 30, 2022. MMWR Morb Mortal Wkly Rep 2022;71:904–7.
[47] Karem KL, Reynolds M, Braden Z, *et al*. Characterization of acute-phase humoral immunity to monkeypox: use of immunoglobulin M enzyme-linked immunosorbent assay for detection of monkeypox infection during the 2003 North American outbreak. Clin Diagn Lab Immunol 2005;12:867–72.
[48] Riabinin VA, Shundrin LA, Kostina EV, *et al*. [An oligonucleotide microarray for detection and discrimination of orthopoxviruses based on oligonucleotide sequences of two viral genes]. Mol Gen Mikrobiol Virusol 2006;4:23–30.
[49] Townsend MB, MacNeil A, Reynolds MG, *et al*. Evaluation of the Tetracore Orthopox BioThreat® antigen detection assay using laboratory grown orthopoxviruses and rash illness clinical specimens. J Virol Methods 2013;187:37–42.

[50] Gentile M, Gelderblom HR. Rapid viral diagnosis: role of electron microscopy. New Microbiol 2005;28:1–12.
[51] The current status of human monkeypox: memorandum from a WHO meeting. Bull World Health Organ 1984;62:703–13.
[52] Heymann DL, Szczeniowski M, Esteves K. Re-emergence of monkeypox in Africa: a review of the past six years. Br Med Bull 1998;54:693–702.
[53] Rizk JG, Lippi G, Henry BM, *et al*. Prevention and treatment of monkeypox. Drugs 2022;82:957–63.
[54] Marion F, Gruber P, VP Public Health & Regulatory Science, International AIDS Vaccine Initiative (IAVI). Currently Available Evidence on Monkeypox Vaccines 2022. https://cdn.who.int/media/docs/default-source/blue-print/marion-gruber_current-evidence_who-monkeypox-vaccine-research_2aug2022.pdf?sfvrsn=568221ee_3
[55] Keckler MS, Salzer JS, Patel N, *et al*. IMVAMUNE(®) and ACAM2000 (®) Provide different protection against disease when administered postexposure in an intranasal monkeypox challenge prairie dog model. Vaccines (Basel) 2020;8:396.
[56] EMA's Emergency Task Force advises on intradermal use of Imvanex / Jynneos against monkeypox 2022. https://www.ema.europa.eu/en/news/emas-emergency-task-force-advises-intradermal-use-imvanex-jynneos-against-monkeypox
[57] Talbot TR, Stapleton JT, Brady RC, *et al*. Vaccination success rate and reaction profile with diluted and undiluted smallpox vaccine: a randomized controlled trial. Jama 2004;292:1205–12.
[58] NACI Rapid Response: Interim guidance on the use of Imvamune in the context of monkeypox outbreaks in Canada 2022. https://www.canada.ca/en/public-health/services/immunization/national-advisory-committee-on-immunization-naci/guidance-imvamune-monkeypox.html
[59] 2022 Outbreak Cases and Data 2023. https://www.cdc.gov/poxvirus/mpox/response/2022/index.html
[60] Monitoring and Risk Assessment for Persons Exposed in the Community 2022. https://www.cdc.gov/poxvirus/mpox/clinicians/monitoring.html
[61] Wittek R. Vaccinia immune globulin: current policies, preparedness, and product safety and efficacy. Int J Infect Dis 2006;10:193–201.