



What the pox? Review of poxviruses affecting humans

D.Jane Hata^{a,*}, Eleanor A. Powell^b, Meghan W. Starolis^c, Susan E. Realegeno^d

^a Department of Pathology and Laboratory Medicine, Mayo Clinic Florida, 4500 San Pablo Rd., Jacksonville, FL 32224 USA

^b Department of Pathology and Laboratory Medicine, University of Cincinnati College of Medicine, 234 Goodman St., Cincinnati, OH 45219, USA

^c Quest Diagnostics, 14225 Newbrook Dr., Chantilly, VA 20155, USA

^d Quest Diagnostics, 33608 Ortega Highway, San Juan Capistrano, CA 92675 USA

ARTICLE INFO

Keywords:

Poxvirus

Mpox

Orthopoxvirus

Zoonotic poxviruses

ABSTRACT

The re-emergence of human mpox with the multi-country outbreak and a recent report of borealpoX (previously Alaskapox) resulting in one death has heightened awareness of the significance of the *Poxviridae* family and their zoonotic potential. This review examines various poxviruses affecting humans, with discussion of less commonly encountered *Poxviridae* members, including pathogenesis, epidemiology, and diagnostic methods. Poxvirus treatment is beyond the intended scope of this review and will not be discussed.

1. Introduction

The family *Poxviridae* consists of large, double-stranded, enveloped DNA viruses with 10 significant genera in human disease [1,2]. Although members of the *Orthopoxvirus* (OPXV) genus are commonly cited, less frequently encountered genera such as *Parapoxvirus*, *Yatapoxvirus*, and *Molluscipoxvirus* have clinical significance. All poxviruses exhibit two distinct infecting particles – the mature virion and enveloped virion [3]. Disease manifestations vary from self-limiting localized dermal nodules in *Molluscipoxvirus* infections to high morbidity and mortality associated with human smallpox, caused by variola virus (VARV). Some members of *Poxviridae* are zoonotic, presenting risk of human infection via exposure to wild or domesticated mammals. This review discusses the four genera of *Poxviridae* in terms of structure, pathogenesis, epidemiology, and diagnostic methods.

2. Orthopoxvirus genus

2.1. Structure

Orthopoxviruses are among the largest mammalian viruses known (240 × 300 nm) [2,4,5]. The complex internal structure consists of two lateral bodies and a dumbbell-shaped nucleosome with a genome approximately 130 – 260 kb length. The large brick-shaped viral particles are encased with palisading surface tubules surrounded by a fragile outer envelope upon release from infected host cells [4]. Replication occurs solely in the host cell cytoplasm, notable among *Poxviridae* and

Asfarviridae viral families [3]. Among OPXVs, cowpox virus (CPXV) has the most conserved genome, as other OPXV species contain point mutations, deletions, and rearrangements which delineate them from the original progenitor CPXV sequence [5].

2.2. Pathogenesis

OPXVs exhibit significant variability of host cell tropism and host range. The cytopathic effect observed in OPXV-infected cells is a manifestation of rearrangement of host cell cytoskeletal components due to viral replication [5]. Although a single specific host cell receptor for poxviruses has not been identified, several glycosaminoglycans and laminins have been demonstrated to mediate adsorption of OPXV into susceptible cells [6].

Transmission of OPXV occurs primarily via exposure to skin lesions, respiratory droplets, aerosols, animal scratches, or fomites containing virus [7]. OPXV infections generally present as a macular rash or lesions progressing through vesicular and pustular stages with occasional ulcerations and epidermal hyperplasia. Intracytoplasmic inclusion bodies can be noted within tissue keratinocytes [7].

Lesions may occur in oropharyngeal, ocular, or mucocutaneous sites. Genital and anorectal infections have been a hallmark of the recent mpox outbreak. Lesions typically develop simultaneously on a given part of the body and may be singular or multiple [8,9], eventually developing crusts with desquamation over several weeks. Systemic symptoms may include fever, chills, lymphadenopathy, cough, and conjunctivitis. Leukocytosis, thrombocytopenia, and liver enzyme

* Corresponding author at: Department of Pathology and Laboratory Medicine, Mayo Clinic Florida, 4500 San Pablo Rd., Jacksonville, FL 32224 USA.

E-mail address: Hata.Donna@mayo.edu (D.Jane Hata).

<https://doi.org/10.1016/j.jcv.2024.105719>

Received 21 May 2024; Received in revised form 31 July 2024;

Available online 5 August 2024

1386-6532/© 2024 Elsevier B.V. All rights are reserved, including those for text and data mining, AI training, and similar technologies.



Fig. 1. Electron micrograph of *Vaccinia* (mulberry form) demonstrating surface tubular elements surrounding the brick shaped virion structure. Photo source: CDC Public Health Image Library (PHIL). Home - Public Health Image Library (PHIL) (cdc.gov).



Fig. 2. HE stain of skin containing *Variola* lesion (Courtesy Dr. Bobbi Pritt).

elevations are common [7].

2.3. Epidemiology

The OPXV genera includes vaccinia (VACV), cowpox (CPXV), camelpox, horsepox, borealpox (BRPV) and mpox (MPXV) viruses, which affect humans and multiple wild or domestic mammals, and VARV, affecting only humans. Edward Jenner first used CPXV in 1796 to induce VARV immunity [10]. VACV has been used to vaccinate individuals against smallpox since the 19th century [2,5]. OPXVs are commonly named after their zoological source of isolation, which may or may not be their natural reservoir; misalignment with the actual reservoir host or host range has contributed to confusion with the true origin of a given virus and/or its geographical distribution [2]. For instance, mpox, previously known as “monkeypox” was recently renamed to remove zoological and geographic references in efforts to reduce associated stigmas. The name “monkeypox” was a misnomer since it was originally discovered in primates housed in a research facility and not the primary

reservoir for infection, which are arboreal African rodents [11].

As a wide variety of OPXVs have been identified in vertebrate hosts, reservoir populations have the potential to provide a platform for zoonotic transmission to humans, with VACV, CPXV, and MPXV commonly cited [2]. Most zoonotic outbreaks are relatively limited in terms of size and scope with low mortality [7]. However, the mpox outbreak of 2022 resulted in over 88,000 cases and 140 deaths by the end of the global health emergency in May 2023 [12]. Prior to 2022, an increase in the number and magnitude of mpox outbreaks occurring in endemic regions had been observed [13,14]. Environmental/ecological influences, viral adaptation, and cessation of routine cross-protective smallpox vaccination are suggested as contributors to the increased number of cases [13,14]. The first fatal case of borealpox in an immunocompromised host was recently reported, and speaks to the need for ongoing OPXV awareness [15,16].

VARV carries great significance due to its historical impact, potential for rapid spread, and hypothetical use as a bioweapon. VARV was responsible for an estimated 300–500 million human deaths during the 20th century, impacting national economies, trade, and societal evolution [17]. *Variola major* can result in 30 % mortality, with *Variola minor* demonstrating a mortality rate of < 1 % [7]. Through quarantine and vaccine deployment, natural transmission of VARV has been eliminated worldwide since 1980 [2]. Excellent reviews of OPXVs are provided in references [2,5,7,18] and in-depth reviews of MPXV are also available [9,11].

3. Parapoxvirus genus

3.1. Structure

The *Parapoxvirus* (PPV) genus contains 6 viruses: Orf (ORFV), grey sealpox (GSEPV), bovine papular stomatitis (BPSV), pseudocowpox virus, and red deerpox, all of which have been documented in human infection. PPVs are enveloped viruses that are generally ovoid in shape and are 240–300 nm by 140–190 nm [19,20]. They contain a 140 kb dsDNA genome containing more than 100 genes [20].

3.2. Pathogenesis

All PPVs have a similar disease presentation in humans. Initially, one or a small number of dermal nodules develops after exposure to an infected animal. These nodules typically move through a maculopapular, targetoid, acute, regenerative, papilloma, and regression stage [21–23]. Each stage lasts approximately one week, with healing in four to six weeks. Localized lymphadenopathy may occur, but systemic illness is rare and is generally limited to immunosuppressed hosts. In immunosuppressed patients, nodules may be larger and persist longer [23]. Immunosuppressed patients (and rarely immunocompetent patients) may also develop erythema multiforme [23–25]. While parapoxvirus disease is mild in immunocompetent individuals, accurate diagnosis is essential to rule out other pathogens exhibiting overlapping symptoms [26].

3.3. Epidemiology

PPVs typically occur worldwide without geographic restriction. Each PPV has a distinct host reservoir and risk group. For instance, pseudocowpox is the causative agent for milker’s nodules, causing lesions on the udders and mucous membranes of cows, and infections in dairy workers [21,27]. BPSV also occurs in cows and has been associated with their handlers, but it does not cause visible lesions in cows [20]. ORFV is the causative agent for ecthyma contagiosum in humans. It causes papules, vesicles, and pustules around the mouth and nose of goats and sheep [23]. Transmission to humans has been associated with veterinarians, butchers, sheep shearers, and petting zoo attendees [23]. GSEPV is found in grey and harbor seals, and transmission to humans is

Table 1

Summary of selected poxviruses associated with human infection.

Genus	Virus	Size of Virus Particle	Size of complete Viral Genome	Host Range	Reservoir Host	Transmission	Disease
<i>Orthopoxvirus</i>	<i>Variola</i>	240 × 300 nm	130 - 260 KB	Humans	Humans	Contact, respiratory droplets, fomites	Smallpox
	<i>Vaccinia</i>		Approximately 200 KB	Multiple, including humans	Rodents	Contact, respiratory droplets, fomites	Generalized vaccinia, Vaccinia eczematum
	<i>Cowpox</i>		220 - 222 KB [68]	Humans, cattle, rodents, cats	Rodents	Contact	Cowpox
	<i>Camelpox</i> MPOX		205 KB [69] Approximately 200 KB [70]	Humans, camels Humans, various primates, rodents, squirrels	Dromedary camels Unknown; likely arboreal rodents [71]	Contact, fomites Contact, fomites, droplets	Camelpox MPOX
<i>Parapoxvirus</i>	Orf (ORFV)	240–300 × 140–190 nm [19]	Approximately 140 kb [20]	Humans, goats, and sheep Humans, cows	Goats and sheep Cows	Contact	Orf Bovine papular stomatitis
	Bovine papular stomatitis virus (BPSV)			Humans, grey and harbor seals	Seals		Sealpox
	Grey Sealpox virus (GSEPV)			Humans, Cows	Cows		Milker's nodule
	Pseudocowpox Red Deerpox			Humans, deer	Deer		Deerpox
<i>Yatapoxvirus</i>	Tanapoxvirus	280 nm x 220 nm	145 KB [34]	Humans, various primates	Unknown	Unknown, but suspected vector borne [36,37]	Tanapox
<i>Molluscipoxvirus</i>	Molluscum contagiosum virus (MCV)	320 nm × 250 nm × 200 nm	190 KB [44]	Humans	Humans	Contact, fomites	Molluscum contagiosum
Genus	Virus	Clinical Symptoms	Disease Duration	Mortality (human)	Biosafety Level (Clinical Specimens)	Diagnostic Methods*	Vaccine Available
<i>Orthopoxvirus</i>	<i>Variola</i>	Rash, macules, vesicles, pustules	24 h (death) -to 2–3 weeks	<i>Variola major</i> –30 % <i>Variola minor</i> -< 1 %	BSL 4 (CDC and public health labs only) BSL-2	Species- specific NAAT, OPXV antibody detection Species- specific NAAT, OPXV antibody detection	ACAM 2000, JYNNEOS (both Vaccinia strains) Vaccine strain
	<i>Vaccinia</i>	Rash, macules, vesicles, pustules, lymphadenopathy	4 –6 weeks (human)	0.0001 %: routine vaccination 1 %: eczema vaccinatum 33 %: vaccinia necrosum [72]			
	<i>Cowpox</i>	Hand lesions, lymphadenopathy, fever	2 - 4 weeks	Not reported	BSL-2	Species- specific NAAT, OPXV antibody detection	Cross protection possible with vaccinia virus vaccines
	<i>Camelpox</i>	Fever, hand lesions, nasal discharge, GI, respiratory distress	4 - 6 weeks	Up to 25 %	BSL-2	Genus-level NAAT	Cross protection possible with vaccinia virus vaccines
	MPOX	Lesions, fever, lymphadenopathy, hepatic necrosis	4 weeks	Clade I ~ 10 % Clade II ~ 1 %	BSL-2 with enhanced precautions	Species and clade specific NAAT, OPXV antibody detection	JYNNEOS, cross-protection with ACAM2000
<i>Parapoxvirus</i>	Orf (ORFV)	Lesions, localized lymphadenopathy	4–6 weeks	Not Reported	BSL-2	Species- specific NAAT Species- specific NAAT	No
	Bovine papular stomatitis virus (BPSV)					Species- specific NAAT	
	Grey Sealpox virus (GSEPV)					Species- specific NAAT	
	Pseudocowpox					Genus- level NAAT	
<i>Yatapoxvirus</i>	Red Deerpox Tanapoxvirus	Fever, lesions	6 weeks	Not reported	BSL-2	Species- specific NAAT	No
<i>Molluscipoxvirus</i>	Molluscum contagiosum virus (MCV)	Pearly pink or white rounded papules	6–9 months typically but as long as 3–4 years	Not reported	BSL-2	Species- specific NAAT	No

EM: Electron microscopy, NAAT: Nucleic Acid Amplification Test, IHC: Immunohistochemistry.

* EM, IHC, sequencing may be used for all genera.

associated with seal handlers and trainers [28]. In some cases infections in humans have resulted from seal bites [29]. Red deerpox virus causes disease in red deer and infection has been noted in deer hunters [30]. As the clinical features of infection are typically mild and self-limiting, cases are likely underreported.

4. Yatapoxvirus genus

4.1. Structure

The yatapoxvirus group contains Yaba monkey tumor virus (which does not infect humans) and Tanapoxvirus (TPV) [31]. Yaba-like disease virus is described in the literature, but is likely the same virus as TPV [32]. TPV is an enveloped brick-shaped virus that is approximately 280 nm by 220 nm with a 145 kb linear genome [33,34]. Studies of TPV show experimental infection in primary human dermal fibroblasts and monocytes but no replication in lymphocytes [35]. Inability to replicate in lymphocytes as well as improved replication at 35 °C as compared to 37 °C (i.e., below human body temperature) may contribute to lack of systemic symptoms during infection [35].

4.2. Pathogenesis

TPV infection is characterized by a short febrile illness of 3–4 days with headache, backache and singular lesions. Vesicles are smallpox-like and follow a similar disease course, but lesions are larger, firmer, and evolve slowly [36]. In the largest documented outbreak, 77 % of lesions were on the front or lower body, 79 % were on areas not covered by clothing, and 78 % were single lesions. Lesions are preceded by itching before erupting as a large (approximately 10 mm) macular lesion that may become nodular during the first week. Lesions are typically accompanied by regional lymphadenopathy, may ulcerate, and enlarge for 2 to 3 weeks before spontaneously resolving within 6 weeks [37].

4.3. Epidemiology

TPV was first identified in a small outbreak among children attending a single school in the Tana River valley (Kenya) in 1957, followed shortly by a 50-case outbreak in 1962 [36]. Both outbreaks followed large floods, which created swamps and isolated communities. By 1971, 16.3 % of Tana River valley residents had serologic evidence of infection indicative of ongoing transmission [38]. Subsequent surveillance in the Congo River valley found 357 TPV cases from 1979 to 1983, most within a 9-month period in 1980–1981 [37]. In this outbreak, 57 % of cases were in patients who lived within 300 m of a recent case, 20 % had a neighbor who was also infected, and 25 % lived in the same household as another infected person. Due to the association with floods and mosquito season, it is presumed that TPV is a mosquito-borne disease, though this has never been experimentally proven [36,37]. Beyond the Tana and Congo river valleys, TANV has also been detected in South Africa [33], a European traveler returning from Tanzania [39], a New Hampshire college student after caring for chimps in the Republic of Congo [40], and among imported macaques and their handlers in Texas, California, and Oregon [32,41].

5. Molluscipoxvirus genus

5.1. Structure

The genus *Molluscipoxvirus* has only one species, *Molluscum contagiosum virus* (MCV), which is responsible for a skin infection commonly seen in children [42]. The virion shape has been described as oval or brick-shaped, measuring approximately 200 - 300 nm [43]. This enveloped virus encodes for approximately 160 proteins and is approximately 190 kb in size [44]. There are four recognized subtypes (I/Ia, II, III, and IV) with type I being the most prevalent [42,44,45].

5.2. Pathogenesis

MCV infects epidermal keratinocytes which play a crucial role in skin repair and protective immunity [42,46]. Children ages 2–5 are most affected and present with pearly pink or white rounded papules measuring approximately 2–5 mm on areas of exposed skin except the palms and soles. Lesions on the face, trunk, extremities, skin folds, and genital area are usually from self-inoculation [45]. Typically, the patient has between 1 and 20 painless papules which have a characteristic central dimple [47]. Transmission in adults is mainly through sexual contact with lesions present in genital and perianal regions [45]. Skin lesions are self-limiting but can last for long periods of time, usually between 6 and 9 months but may take 3–4 years for resolution [47].

5.3. Epidemiology

MCV is species-specific to humans and ubiquitous worldwide but may be more prevalent in warm geographic areas [45]. The virus is spread person-to-person or via contaminated fomites [48]. A meta-analysis to understand global prevalence data showed an 8.28 % overall prevalence but was skewed by three high prevalence MCV reports (New Guinea at 21.8 % [49], Israel at 34.6 % [50], and Japan at 19.7 % [51,52]). With these outliers removed, prevalence was calculated at 2.8 % [52]. The overall prevalence in the United States is estimated to be 5.0 % [45,53]. Transmission of MCV can be observed year-round with notable hot spots of transmission within daycares and schools [53].

6. Diagnostic methods for poxvirus infections

6.1. Biosafety

Clinical specimens submitted for high-risk agents, such as VARV or MPXV Clade I viruses that are unfixed or not inactivated should be handled in a BSL-3 or higher facility by vaccinated personnel and preferably referred to a public health laboratory. Vaccinated individuals can perform work with replication-competent VACV and other human poxviruses in BSL-2 facilities. Additional safety practices and equipment can be considered based on a site-specific risk assessment [3,54]. Routine virus isolation or propagation of virus from clinical samples should not be attempted in clinical laboratories. VARV and MPXV Clade I are listed as select agents and subject to subsequent reporting and inactivation/destruction as outlined by the Federal Select Agent Program. Positive samples identified by MPXV Clade II specific NAATs are not subject to select agent regulations [55].

6.2. Diagnostic methods

Initial clinical suspicion for infection is determined by symptom presentation. Since poxvirus infections are uncommon, laboratory testing should be based on epidemiologic risk factors and in consultation with the public health department. Epidemiologic risk factors may include travel history to endemic areas, contact with a potentially infected person or animal, or recent vaccination history with ACAM2000, the smallpox vaccine comprised of replication competent vaccinia virus. Diagnostic testing of poxviruses in the United States is limited to public health laboratories, except for MPXV which is also available in some hospital and commercial laboratories. In addition to evaluating risk factors, characterizing dermatologic lesion appearance and progression is one of the key determinants for performing poxvirus specific testing. ORFV and MCV infections can be diagnosed without the need for laboratory testing, while other poxviruses infections generally require confirmation. Laboratory testing may also be indicated when lesions have an atypical appearance or cannot be differentiated from other infectious and non-infectious etiologies causing a rash illness.

A specialized algorithm for evaluating patients suspected of smallpox presenting with an acute, generalized vesicular or pustular rash illness

has been developed [56]. The clinical algorithm classifies patients into high, moderate, and low risk categories to strategically guide diagnostic testing to rule out other common infectious causes (e.g. varicella-zoster) before initiating VARV-specific testing. This reduces the risk of unnecessary testing that may lead to false-positive results in the context of disease eradication, but still allows for consideration of a potential bioterrorism event and the intentional release of VARV [56]. An OPXV laboratory testing algorithm is available for patients suspected of a smallpox vaccine adverse event related to ACAM2000 or inadvertent infection from close contact with a person's vaccination site [57].

Laboratory diagnostic methods for identifying and differentiating poxviruses have traditionally included virus isolation via cell culture, antigen testing, and electron microscopy (EM). However, direct detection using nucleic acid amplification tests (NAATs) are preferred as they are rapid with high sensitivity and specificity. Genome sequencing has also become an important tool for molecular epidemiology, as it can differentiate variants and identify resistance mutations in addition to species identification. Specimens submitted for initial testing include lesion material or fluid, which often contain a high viral burden depending on the stage of the lesion. Vigorously swabbed lesions, exudate or vesicular/pustule fluid, scab or crusts are also acceptable. Deroofing of crusts or fluid aspiration is not required and may pose an infection risk for the specimen collector [3].

6.2. Serology Testing

Antibody detection may be used to support diagnosis, evaluate exposure or immune status, or for serosurveillance. In the absence of lesion specimens, acute and convalescent sera can be collected several weeks apart to document seroconversion. Species-specific antibody assays are typically not offered due to significant cross-reactivity within the genera. An IgG/IgM ELISA using purified VACV is available for OPXVs through the CDC [58]. Serology testing for other poxviruses is not readily available.

An OPXV IgG result alone may be indicative of vaccination, or previous exposure/infection, whereas IgM can be detected soon after infection in both vaccinated and unvaccinated individuals [58]. Responses can be detected within a week after presentation of rash for both IgM and IgG [58] and IgG responses have been detected indefinitely in vaccinated individuals [59,60]. Alternatively, serum neutralization tests using plaque reduction have been developed to assess immune responses to OPXVs. Although a 4-fold increase in antibody titer between acute and convalescent sera can be diagnostic for infection, established thresholds correlated with protection against future infection have not been established [3].

6.3. Viral Isolation & microscopic identification: cell culture, immunohistochemistry (IHC), electron microscopy (EM)

Most poxviruses infecting humans will grow in a variety of cell lines and can exhibit cytopathic effects (CPE), except for MCV which cannot be propagated [61]. EM can be used to identify the presence of a poxvirus in cultured specimens or directly from specimens due to its unique brick-shaped virion structure surrounded by irregular short tubular elements (Fig. 1). Histologic examination of lesion biopsy samples stained with H&E or Giemsa stains can reveal perinuclear basophilic or cytoplasmic round or oval inclusion bodies (Fig. 2). Acidophilic-type inclusion bodies can also be found with CPXV infection. IHC can identify poxviruses only to the genus level using antibodies against genus specific antigens. Overall, these methods can be useful in identifying a novel poxvirus infection associated with a rash illness of unknown etiology [62].

6.4. Direct Detection (NAAT)

Real-time qPCR assays are available through the CDC for genus

specific detection of OPXV, PPV, and Yatapoxviruses [63]. A pan-poxvirus assay has also been developed and used for screening/identification of novel poxviruses in the *Chordopoxvirinae* subfamily [2,64]. Several species-specific assays are available within the OPXV and PPV genera, while TPV is the primary species available within the *Yatapoxvirus* genus [63]. An MCV-specific PCR assay is also available. [65]. Available OPXV specific assays include VARV, non-VARV, VACV, MPXV (generic and clade specific), and CPXV. *Parapoxvirus* specific assays available include ORFV, pseudocowpox virus, BPSV, and GSEPV [63].

During the 2022 global mpox outbreak, some clinical laboratories implemented PCR-based assays as laboratory developed tests targeting OPX, non-variola OPX, or MPXV (generic or clade specific) genes. Commercially available NAATs for OPXV and MPXV are also available under FDA emergency use authorization status. An OPXV real-time PCR primer and probe set was previously FDA approved for use in designated Laboratory Response Network (LRN) laboratories and was recently authorized for use in select commercial reference laboratories [66,67].

8. Conclusion

As presented in this review, continual evolution in the emergence and pathogenesis of *Poxviridae* family require ongoing surveillance efforts, in addition to enhancement of diagnostic methods. Clinical laboratories are encouraged to remain vigilant to best assess *Poxviridae* epidemiology and participate as active partners with medical providers in patient care and assessment.

Table 1

CRediT authorship contribution statement

D.Jane Hata: Writing – review & editing, Writing – original draft, Investigation, Formal analysis, Data curation, Conceptualization. **Eleanor A. Powell:** Writing – review & editing, Writing – original draft, Investigation, Formal analysis, Data curation, Conceptualization. **Meghan W. Starolis:** Writing – review & editing, Writing – original draft, Investigation, Formal analysis, Data curation, Conceptualization. **Susan E. Realegeno:** Writing – review & editing, Writing – original draft, Investigation, Formal analysis, Data curation, Conceptualization.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

D. Jane Hata and Eleanor Powell report no financial conflicts of interest. Meghan Starolis and Susan Realegeno disclose they are full-time employees and shareholders of Quest Diagnostics.

Funding

D. Jane Hata and Eleanor Powell report no financial conflicts of interest. Meghan Starolis and Susan Realegeno disclose they are full-time employees and shareholders of Quest Diagnostics.

Disclosures/Disclaimers

This Report is a product of the Pan-American Society for Clinical Virology (PASCV) Clinical Practice Committee (CPC) which was developed to provide guidance and recommendations to healthcare professionals. It is not clinical advice and is not intended to characterize all possible disease presentations, diagnostic methods or treatment.

Ethical Approval

Not required.

Acknowledgments

The authors thank the members of the PASCV Clinical Practice and Public Policy Committee for thoughtful critique and review of this document. We would also like to thank Dr. Bobbi Pritt for contribution of poxvirus images.

References

- [1] G.P. Oliveira, R.A.L. Rodrigues, M.T. Lima, B.P. Drumond, J.S. Abrahao, Poxvirus Host Range Genes and Virus-Host Spectrum: a Critical Review, *Viruses*. 9 (2017).
- [2] N.I.O. Silva, J.S. de Oliveira, E.G. Kroon, G.S. Trindade, B.P. Drumond, Here, There, and Everywhere: the Wide Host Range and Geographic Distribution of Zoonotic Orthopoxviruses, *Viruses*. 13 (2020).
- [3] Carroll K.C., Pfaller M.A., Landry M.L., McAdam A.J., Patel R., Richter S.S., et al. *Manual of Clinical Microbiology*. 12th edition. ed. Washington, DC: ASM Press; 2019. p. 2 volumes.
- [4] Baxby D. Poxviruses. In: Baron S, editor. *Medical Microbiology*. 4th ed. Galveston (TX) 1996.
- [5] S.N. Shchelkunov, Orthopoxvirus genes that mediate disease virulence and host tropism, *Adv. Virol.* 2012 (2012) 524743.
- [6] Q. Gong, C. Wang, X. Chuai, S. Chiu, Monkeypox virus: a re-emergent threat to humans, *Virol. Sin.* 37 (2022) 477–482.
- [7] A.L. MacNeill, Comparative Pathology of Zoonotic Orthopoxviruses, *Pathogens*. 11 (2022).
- [8] C.M. Kava, D.M. Rohraff, B. Wallace, J.L. Mendoza-Alonzo, D.W. Currie, A. E. Munsey, et al., Epidemiologic Features of the Monkeypox Outbreak and the Public Health Response - United States, May 17–October 6, 2022, *MMWR. Morb. Mortal. Wkly. Rep.* 71 (2022) 1449–1456.
- [9] J.P. Thornhill, S. Barkati, S. Walmsley, J. Rockstroh, A. Antinori, L.B. Harrison, et al., Monkeypox Virus Infection in Humans across 16 Countries — April–June 2022, *New England Journal of Medicine* 387 (2022) 679–691.
- [10] G.A. Shchelkunova, Shchelkunov S.N. Smallpox, Monkeypox and Other Human Orthopoxvirus Infections, *Viruses*. (2022) 15.
- [11] S. Elsayed, L. Bondy, W.P. Hanage, Monkeypox Virus Infections in Humans, *Clin. Microbiol. Rev.* 35 (2022) e0009222.
- [12] <https://www.nbcnews.com/health/health-news/mpox-who-ends-global-health-emergency-rcna83940> Accessed 11.28.2023.
- [13] A.M. McCollum, V. Shelus, A. Hill, T. Traore, B. Onoja, Y. Nakazawa, et al., Epidemiology of Human Mpox - Worldwide, 2018–2021, *MMWR. Morb. Mortal. Wkly. Rep.* 72 (2023) 68–72.
- [14] M.G. Reynolds, I.K. Damon, Outbreaks of human monkeypox after cessation of smallpox vaccination, *Trends. Microbiol.* 20 (2012) 80–87.
- [15] https://epi.alaska.gov/bulletins/docs/b2024_02.pdf. 2 ed.
- [16] <https://health.alaska.gov/dph/Epi/id/SiteAssets/Pages/Borealpox/Borealpox-FA-Q.pdf>. Accessed 7/25/2024.
- [17] I.V. Babkin, I.N. Babkina, N.V. Tikunova, An Update of Orthopoxvirus Molecular Evolution, *Viruses*. (2022) 14.
- [18] A.A. Mercer, A. Schmidt, Weber OF. Poxviruses. Basel, Birkhäuser, Boston, 2007.
- [19] T. Kassa, A Review on Human Orf: a Neglected Viral Zoonosis, *Res Rep Trop Med.* 12 (2021) 153–172.
- [20] K.F. Bowman, R.T. Barbery, L.J. Swango, P.R. Schnurrenberger, Cutaneous form of bovine papular stomatitis in man, *JAMA* 246 (1981) 2813–2818.
- [21] C.M. Davis, G. Musil, Milker's nodule. A clinical and electron microscopic report, *Arch. Dermatol.* 101 (1970) 305–311.
- [22] A.C.C. Esposito, M.F.S. Jorge, M.E.A. Marques, L.P.F. Abbade, Milker's nodules: classic histological findings, *An. Bras. Dermatol.* 92 (2017) 838–840.
- [23] H.J. Thompson, C.L. Harview, B. Swick, J.G. Powers, Orf Virus in Humans: case Series and Clinical Review, *Cutis* 110 (2022) 48–52.
- [24] N.S. Handler, M.Z. Handler, A. Rubins, S. Rubins, M. Septe, C.K. Janniger, et al., Milker's nodule: an occupational infection and threat to the immunocompromised, *J. Eur. Acad. Dermatol. Venereol.* 32 (2018) 537–541.
- [25] K. Wu, S. de Menezes, A. Robinson, Erythema Multiforme Induced by a "Milker's Nodule" Pseudocowpox Infection: a Case Report and Review of Literature, *Case Rep. Dermatol. Med.* 2021 (2021) 5584773.
- [26] Centers for Disease C, Prevention, Orf virus infection in humans—New York, Illinois, California, and Tennessee, 2004–2005, *MMWR. Morb. Mortal. Wkly. Rep.* 55 (2006) 65–68.
- [27] A.E. Friedman-Kien, W.P. Rowe, W.G. Banfield, Milker's nodules: isolation of a poxvirus from a human case, *Science* (1979) 140 (1963) 1335–1336.
- [28] B.D. Hicks, G.A. Worthing, Sealpox in captive grey seals (*Halichoerus grypus*) and their handlers, *J. Wildl. Dis.* 23 (1987) 1–6.
- [29] C. Clark, P.G. McIntyre, A. Evans, C.J. McInnes, S. Lewis-Jones, Human sealpox resulting from a seal bite: confirmation that sealpox virus is zoonotic, *Br. J. Dermatol.* 152 (2005) 791–793.
- [30] A.A. Roess, A. Galan, E. Kitces, Y. Li, H. Zhao, C.D. Paddock, et al., Novel deer-associated parapoxvirus infection in deer hunters, *N. Engl. J. Med.* 363 (2010) 2621–2627.
- [31] A.W. Downie, C. Espana, A comparative study of Tanapox and Yaba viruses, *J. Gen. Virol.* 19 (1973) 37–49.
- [32] A.W. Downie, C. Espana, Comparison of Tanapox virus and Yaba-like viruses causing epidemic disease in monkeys, *J. Hyg. (Lond)* 70 (1972) 23–32.
- [33] M. Birkhead, W. Grayson, A. Grobelaar, V. Msimang, N. Moolla, A. Mathee, et al., Tanapox, 29, *Emerg Infect Dis*, South Africa, 2023, pp. 1206–1209, 2022.
- [34] H.J. Lee, K. Essani, G.L. Smith, The genome sequence of Yaba-like disease virus, a yatapoxvirus, *Virology*. 281 (2001) 170–192.
- [35] S.H. Nazarian, J.W. Barrett, M.M. Stanford, J.B. Johnston, K. Essani, G. McFadden, Tropism of Tanapox virus infection in primary human cells, *Virology*. 368 (2007) 32–40.
- [36] A.W. Downie, C.H. Taylor-Robinson, A.E. Caunt, G.S. Nelson, P.E. Manson-Bahr, T. C. Matthews, Tanapox: a new disease caused by a pox virus, *Br. Med. J.* 1 (1971) 363–368.
- [37] Z. Jezek, I. Arita, M. Szczeniowski, K.M. Paluku, K. Ruti, J.H. Nakano, Human tanapox in Zaire: clinical and epidemiological observations on cases confirmed by laboratory studies, *Bull. World Health Organ.* 63 (1985) 1027–1035.
- [38] P.E. Manson-Bahr, A.W. Downie, Persistence of tanapox in Tana River valley, *Br. Med. J.* 2 (1973) 151–153.
- [39] A. Stich, H. Meyer, B. Kohler, K. Fleischer, Tanapox: first report in a European traveller and identification by PCR, *Trans. R. Soc. Trop. Med. Hyg.* 96 (2002) 178–179.
- [40] A.D. Dhar, A.E. Werchniak, Y. Li, J.B. Brennick, C.S. Goldsmith, R. Kline, et al., Tanapox infection in a college student, *N. Engl. J. Med.* 350 (2004) 361–366.
- [41] R.A. Crandell, H.W. Casey, W.B. Brumlow, Studies of a newly recognized poxvirus of monkeys, *J. Infect. Dis.* 119 (1969) 80–88.
- [42] N.B. Silverberg, Pediatric molluscum: an update, *Cutis* 104 (2019) 301–305. E1;E2.
- [43] A. Hussain, J. Kaler, G. Lau, T. Maxwell, Clinical Conundrums: differentiating Monkeypox From Similarly Presenting Infections, *Cureus*. 14 (2022) e29929.
- [44] A.L. Hammarin, Y. Eklund, M. Karlberg, M. Bogh, P. Sikora, [New subtype of molluscipoxvirus detected], *Lakartidningen*. (2016) 113.
- [45] R. Meza-Romero, C. Navarrete-Dechent, C. Downey, Molluscum contagiosum: an update and review of new perspectives in etiology, diagnosis, and treatment, *Clin. Cosmet. Investig. Dermatol.* 12 (2019) 373–381.
- [46] M. Piipponen, D. Li, N.X. Landen, The Immune Functions of Keratinocytes in Skin Wound Healing, *Int. J. Mol. Sci.* 21 (2020).
- [47] J.C. van der Wouden, R. van der Sande, E.J. Kruithof, A. Sollie, L.W. van Suijlekom-Smit, S. Koning, Interventions for cutaneous molluscum contagiosum, *Cochrane Database Syst. Rev.* 5 (2017) CD004767.
- [48] C.M. Hughes, I.K. Damon, M.G. Reynolds, Understanding U.S. healthcare providers' practices and experiences with molluscum contagiosum, *PLoS. One* 8 (2013) e76948.
- [49] R.J. Sturt, H.K. Muller, G.D. Francis, Molluscum contagiosum in villages of the West Sepik District of New Guinea, *Med. J. Aust.* 2 (1971) 751–754.
- [50] B. Oren, S.O. Wende, An outbreak of molluscum contagiosum in a kibbutz, *Infection* 19 (1991) 159–161.
- [51] S. Hayashida, N. Furusho, H. Uchi, S. Miyazaki, K. Eiraku, C. Gondo, et al., Are lifetime prevalence of impetigo, molluscum and herpes infection really increased in children having atopic dermatitis? *J. Dermatol. Sci.* 60 (2010) 173–178.
- [52] J.R. Olsen, J. Gallacher, V. Piguot, N.A. Francis, Epidemiology of molluscum contagiosum in children: a systematic review, *Fam. Pract.* 31 (2014) 130–136.
- [53] A.A. Hebert, N. Bhatia, J.Q. Del Rosso, Molluscum Contagiosum: epidemiology, Considerations, Treatment Options, and Therapeutic Gaps, *J. Clin. Aesthet. Dermatol.* 16 (2023) S4–S11.
- [54] Biosafety in Microbiological and Biomedical Laboratories, U.S. Department of Health and Human Services Public Health Service. Centers For Disease Control and Prevention National Institutes of Health HHS Publication, 6th Edition, 2020. No. (CDC) 300859.
- [55] Exclusion Guidance Document, Toxins CfDCaPoDoSaa, Toxins AaPHISADoASaa, 2019 editors.
- [56] J.F. Seward, K. Galil, I. Damon, S.A. Norton, L. Rotz, S. Schmid, et al., Development and experience with an algorithm to evaluate suspected smallpox cases in the United States, 2002–2004, *Clin. Infect. Dis.* 39 (2004) 1477–1483.
- [57] J. Cono, C.G. Casey, D.M. Bell, Centers for Disease C, Prevention. Smallpox vaccination and adverse reactions. Guidance for clinicians, *MMWR. Recomm. Rep.* 52 (2003) 1–28.
- [58] K.L. Karem, M. Reynolds, Z. Braden, G. Lou, N. Bernard, J. Patton, 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.* 12 (2005) 867–872.
- [59] D.D. Taub, W.B. Ershler, M. Janowski, A. Artz, M.L. Key, J. McKelvey, et al., Immunity from smallpox vaccine persists for decades: a longitudinal study, *Am. J. Med.* 121 (2008) 1058–1064.
- [60] E. Hammarlund, M.W. Lewis, S.G. Hansen, L.I. Strelow, J.A. Nelson, G.J. Sexton, et al., Duration of antiviral immunity after smallpox vaccination, *Nat. Med.* 9 (2003) 1131–1137.
- [61] G. McFadden, Poxvirus tropism, *Nat. Rev. Microbiol.* 3 (2005) 201–213.
- [62] Pritt B. *Atlas of Fundamental Infectious Diseases Histopathology*: college of American Pathologists; 2018.
- [63] C.M. Hughes, E.R. Lederman, M.G. Reynolds, I.K. Damon, R.R. Lash, S. E. Beekmann, et al., Clinical experience, infection control practices and diagnostic algorithms for poxvirus infections - an Emerging Infections Network survey, *BMC. Res. Notes*. 3 (2010) 46.
- [64] Y. Li, H. Meyer, H. Zhao, I.K. Damon, GC content-based pan-pox universal PCR assays for poxvirus detection, *J. Clin. Microbiol.* 48 (2010) 268–276.
- [65] J.P. Trama, M.E. Adelson, E. Mordechai, Identification and genotyping of molluscum contagiosum virus from genital swab samples by real-time PCR and Pyrosequencing, *J. Clin. Virol.* 40 (2007) 325–329.

- [66] T.A. Aden, P. Blevins, S.W. York, S. Rager, D. Balachandran, C.L. Hutson, 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.* 71 (2022) 904–907.
- [67] Y. Li, V.A. Olson, T. Laue, M.T. Laker, I.K. Damon, Detection of monkeypox virus with real-time PCR assays, *J. Clin. Virol.* 36 (2006) 194–203.
- [68] D. Diaz-Canova, C. Mavian, A. Brinkmann, A. Nitsche, U. Moens, M.I. Okeke, Genomic Sequencing and Phylogenomics of Cowpox Virus, *Viruses.* (2022) 14.
- [69] C.L. Afonso, E.R. Tulman, Z. Lu, L. Zsak, N.T. Sandybaev, U.Z. Kerembekova, et al., The genome of camelpox virus, *Virology.* 295 (2002) 1–9.
- [70] J.R. Kugelman, S.C. Johnston, P.M. Mulembakani, N. Kisalu, M.S. Lee, G. Koroleva, et al., Genomic variability of monkeypox virus among humans, Democratic Republic of the Congo, *Emerg. Infect. Dis.* 20 (2014) 232–239.
- [71] M. Curaudeau, C. Besombes, E. Nakoune, A. Fontanet, A. Gessain, A. Hassanin, Identifying the Most Probable Mammal Reservoir Hosts for Monkeypox Virus Based on Ecological Niche Comparisons, *Viruses.* (2023) 15.
- [72] <https://emedicine.medscape.com/article/231773-overview#a6>. Accessed 12/29/23.