

Concurrent Clade I and Clade II Monkeypox Virus Circulation, Cameroon, 1979–2022

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During 1979–2022, Cameroon recorded 32 laboratory-confirmed mpox cases among 137 suspected mpox cases identified by the national surveillance network. The highest positivity rate occurred in 2022, indicating potential mpox re-emergence in Cameroon. Both clade I (n = 12) and clade II (n = 18) monkeypox virus (MPXV) were reported, a unique feature of mpox in Cameroon. The overall case-fatality ratio of 2.2% was associated with clade II. We found mpox occurred only in the forested southern part of the country, and MPXV phylogeographic structure revealed a clear geographic separation among concurrent circulating clades. Clade I originated from eastern regions close to neighboring mpox-endemic countries in Central Africa; clade II was prevalent in western regions close to West Africa. Our findings suggest that MPXV re-emerged after a 30-year lapse and might arise from different viral reservoirs unique to ecosystems in eastern and western rainforests of Cameroon.

Monkeypox virus (MPXV) is an emerging zoonotic *Orthopoxvirus* causing mpox in humans, a disease similar to the eradicated smallpox (1). Since identification in a monkey in 1958 (2) and a human in 1970 (3), MPXV-associated outbreaks have occurred primarily in rural rainforests in countries of Central and West Africa (4–6).

Mpox is characterized by an influenza-like syndrome accompanied by adenopathy and maculopapular rashes typically developing on the palms of the hands and soles of the feet (4,7). For infected persons, supportive care and antiviral treatments, including cidofovir and tecovirimat, are provided (4). Cross-immunity with smallpox vaccination and a new generation of smallpox vaccines equally offer some protection (8–10). However, after smallpox vaccination was discontinued in the early 1980s, herd immunity gradually declined, enabling re-emergence of mpox, which is highlighted by the increased number of cases in Africa during the past 3 decades (4,8,11–13). Since early 2022, case counts have surged, and ≈1,215 confirmed mpox cases and 219 deaths were reported in Africa by December 28, 2022 (14). Before April 2022, mpox cases in the Western Hemisphere were typically reported from exposure to the exotic pet trade and international travel (15–20). Since then, MPXV-associated outbreaks have occurred worldwide, affecting >100 countries outside Africa (4,21) and becoming a global public health concern.

Primary MPXV transmission can occur through direct contact with body fluids or skin lesions of infected animals or indirectly via contaminated fomites. Similar contact with an infected person or with infected respiratory droplets might also lead to human-to-human secondary transmission, the main transmission mode of the 2022 global outbreak (4,22). Historically, primary zoonotic transmission was more common and mostly involved an at-risk population of hunters, butchers, and bushmeat handlers; secondary transmission was rare, but nosocomial and household transmission have been described (3,13,23–25).

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Phylogenetic studies report 2 distinct MPXV clades: clade I, prevalent in Central Africa, and clade II, endemic to West Africa (5,6,26–28). However, Cameroon is an exception, and both clades concurrently circulate in the country (6,29). Clade I is further subdivided into lineages 1–5 and clade II into subclades IIa and IIb; clade IIb is responsible for the multicountry outbreak that began in 2022 (27,28,30). Globally, MPXV lethality rates vary from 1% to 10%, and clade I is known to have higher mortality rates than clade II (4,24,25). The MPXV animal reservoir has not yet been identified, but the virus can infect a wide range of mammals, and *Funisciurus* squirrels and *Graphiurus lorraineus* mice are thought to be the most probable MPXV reservoirs (31–33).

In Cameroon, only 4 confirmed mpox cases were documented before the 2022 outbreak, 1 in each 1979, 1980, 1989, and 30 years later in 2018 (29,34–36). According to public health reports, more cases could have occurred and been undocumented in the country, particularly during 2018–2021, and especially in 2022, during which an mpox outbreak of unprecedented magnitude occurred and had recurrent clusters of cases (37). However, whether those infections were associated with importations from neighboring countries or from occurrence of indigenous primary or secondary transmission remains unclear (29). Overall, data on the epidemiologic features of MPXV occurrence and transmission dynamics in Cameroon are scarce. We investigated the clinical, epidemiologic, and molecular features of MPXV-associated outbreaks in Cameroon.

Methods

Sample Location

Cameroon is in central Africa and is divided into 10 administrative regions. Cameroon is known as Africa in miniature for its diverse agroecologic background: the steppe and savanna in the Far North, North, and Adamawa regions; the coastal zones in the Littoral and Southwest regions; mountain highlands in the Northwest and West regions; and the rainforest in the Centre, South, Southwest, and East regions (38). Cameroon has 3 major tropical forests: the Congo Basin Forest that extends across the East, South, and Centre regions; the Guinea moist forest in the western and Adamawa regions; and the Cameroonian Highlands forests in the Northwest and Southwest regions. Those forests are crossed by several waterways, including the Sanaga River, the largest river in Cameroon (33–40; J. Thia, master's thesis, University of Canterbury, 2014,

https://www.researchgate.net/publication/272494772_The_plight_of_trees_in_disturbed_forest_conservation_of_Montane_Trees_Nigeria).

Sample Collection

We defined a suspected case as ≥ 1 clinical signs or symptoms, including headache, asthenia, adenopathy, myalgia associated with fever, or gradually developing rashes spreading to other parts of the body, including the soles of the feet and palms of the hands. We defined a probable case as clinical manifestations without virologic confirmation but an epidemiologic link with another probable or confirmed case. A confirmed case was any case with laboratory-confirmed MPXV.

We recorded epidemiologic data, including demographic and clinical information, for all suspected cases during 1979–2022. We collected a 5-mL blood sample, vesicle swab, crust samples, or a combination of samples, from case-patients who consented to be tested. We shipped samples under a triple packaging system to the Centre Pasteur du Cameroun (CPC), which is the national reference laboratory for mpox diagnosis in Cameroon. We excluded patients from whom a sample could not be collected.

Laboratory Confirmation of MPXV Infection

At CPC, samples were received, processed, and inactivated in the Biosafety Level 3 laboratory. We purified total DNA by using the QIAamp DNA Mini Kit (QIAGEN, <https://www.qiagen.com>) according to the manufacturer's instructions. We tested purified DNA for MPXV by the generic real-time PCR Taqman assay, as previously described (41). For positive samples displaying a cycle threshold (Ct) value < 37 , we performed further genotyping by using real-time PCRs specifically targeting MPXV clade I and II (41).

We further amplified a subset of 8 positive samples from the 2022 outbreak that had Ct values ≤ 20 by using a PCR targeting a portion of the MPXV A-type inclusion (ATI) gene, according to a previously described protocol (42). We used a 1% green-stained agarose gel to reveal resulting amplicons, which we sent to Inqaba Biotechnical Industries (Pretoria, South Africa), a commercial service provider, for Sanger sequencing.

Phylogenetical Analyses

We assembled newly determined sequences and corrected by using CLC Main Workbench software (QIAGEN). We aligned resulting consensus sequences by using MAFFT version 7 (<https://mafft.cbrc.jp>)

and an extended dataset of 56 MPXV reference genomes from GenBank (Appendix Tables 1, 2, <https://wwwnc.cdc.gov/EID/article/30/3/23-0861-App1.pdf>). We submitted final alignments to the software-integrated Model Finder program (IQ-TREE, <http://www.iqtree.org>) to select the best evolutionary model based on Bayesian and Akaike information criterion. We used IQ-TREE version 1.6.12 (<http://www.iqtree.org>) to infer maximum-likelihood phylogenetic trees on MPXV *ATI* sequences based on the Hasegawa-Kishino-Yano plus amino acid substitution model, applying 1,000 bootstrap replicates. We submitted newly determined sequences to GenBank (accession nos. OR038717–24) (Appendix Table 2).

Statistical Analysis and Mapping

To provide a complete picture of the epidemiology of mpox in Cameroon, we added the 4 previously documented mpox cases from Cameroon to our dataset, along with available information collected from the literature and Ministry of Health archives (29,34–36). We summarized sociodemographic and clinical characteristics by using frequencies for categorical variables; we used median and interquartile range (IQR) for quantitative variables. We compared PCR-confirmed cases with nonconfirmed suspected cases by using Pearson χ^2 or Fisher exact tests for categorical variables and Wilcoxon test for quantitative variables. We used univariate logistic regression to identify factors associated with MPXV infection and estimate crude odds ratios (ORs) and 95% CIs. We were unable to infer multivariable analysis models, which failed to converge because too many data were missing (Tables 1, 2). We considered $p < 0.05$ statistically significant and $p < 0.07$ marginally significant. We performed all analyses in R version 4.1 (The R Foundation for Statistical Computing, <https://www.r-project.org>). We used Quantum GIS version 3.30.1 (QGIS, <https://qgis.org>) to analyze and map mpox cases by health zones and geographic data.

Ethics

Sample collection and laboratory analyses were conducted within the framework of the Cameroon national surveillance program. Under that program, we obtained written or oral informed consent from all persons with suspected mpox after we provided detailed information and explanations of the sampling purpose. We obtained informed consent from parents or recognized guardians for persons <15 years of age.

Results

Within the mpox surveillance system in Cameroon, during 1979–2022, we identified 137 suspected mpox cases, including 74 (54.41%) among male and 62 (45.59%) among female persons; 1 case had missing data for sex (Table 1). The median age of case-patients was 11 years (range 2 weeks–75 years; IQR 4–27 years); nearly half (48.18%) were ≤ 10 years of age (Table 1).

Molecular Diagnostic Results

Mpox virus generic PCR showed 32 (23.36%) laboratory-confirmed mpox cases of 137 patients tested during 1979–2022 in Cameroon (Table 1; Figure 1, panel A; Appendix Table 3). Before 2018, only 3 sporadic cases were confirmed as human MPXV infection. After a 30-year gap without reported mpox cases, the surveillance system continuously identified new mpox cases during 2018–2022. Among suspected cases, only 1 was found in 2018 and 1 in 2019. In 2020 and 2021, 5 laboratory-confirmed cases were recorded each year. During 2022, mpox cases dramatically increased to 17 confirmed cases among 84 suspected cases (Figure 1, panel A; Appendix Table 3).

Genotyping of real-time PCR results identified 12 (9%) patients infected with MPXV clade I and 18 (13%) infected with MPXV clade II among 137 suspected cases; 2 (1%) historic confirmed cases lacked clade determination results (Table 1; Figure 1, panel B; Appendix Tables 2, 3). Among all laboratory-confirmed cases, only 1 death was recorded, in a patient infected with MPXV clade II. Ministry of Health investigation records indicated 2 additional patient deaths among persons with typical mpox clinical manifestations who were epidemiologically linked to 2 confirmed case-patients infected with a clade II MPXV strain. However, no specimens were collected before death; thus, we considered those probable cases. Including the probable cases, the overall case-fatality ratio (CFR) in Cameroon was 2.2% (3/139) among confirmed and suspected cases, and all deaths were associated with viral clade II.

Epidemiology and Clinical Characteristics of Confirmed Mpox Cases

Univariable analysis revealed no statistically significant difference in increased likelihood of infection by sex: 21/74 (28.38%) male and 11/62 (17.74%) female persons had confirmed MPXV infection (Table 1). MPXV-confirmed case-patients had a median age of 21.5 years (range 2 weeks–52 years; IQR 8.5–32.25 years). MPXV infection was more prevalent

among adults ≥ 20 years of age; in all, 35.56% had confirmed MPXV infection, compared with 17.78% among younger MPXV-confirmed case-patients ($p = 0.025$). However, we saw no statistically significant

Table 1. Molecular diagnostic and epidemiologic characteristics of suspected and confirmed mpox cases in a study of concurrent clade I and clade II monkeypox virus circulation, Cameroon, 1979–2022*

Epidemiologic characteristics	MPXV real-time PCR, no. (%)		Crude OR (95% CI)	p value
	Positive	Negative		
Total no. (%), n = 137	32 (23.36)	105 (76.64)		
MPXV clades				
Clade I, n = 12	12 (100.00)	NA	NA	1
Clade II, n = 18	18 (100.00)	NA	NA	
Sex				
M, n = 74	21 (28.38)	53 (71.62)	Referent	0.142
F, n = 62	11 (17.74)	51 (82.26)	1.84 (0.81–4.19)	
Age				
Minimum	0	0	NA	0.075
1st quartile	8.5	4	NA	
Median	21.5	10	NA	
Mean	21.69	15.64	1.02 (1.00–1.05)	
3rd quartile	32.25	22.5	NA	
Maximum	52	75	NA	
Age group, y				
0–10, n = 66	10 (15.15)	56 (84.85)	Referent	0.101
11–20, n = 24	6 (25.00)	18 (75.00)	1.87 (0.60–5.85)	
21–30, n = 18	6 (33.33)	12 (66.67)	2.8 (0.85–9.19)	
>30, n = 27	10 (37.04)	17 (62.96)	3.29 (1.17–9.24)	
Born before 1980				
Y, n = 124	27 (45.45)	97 (54.55)	Referent	0.092
N, n = 11	5 (21.77)	6 (78.23)	3.07 (0.86–10.88)	
Born before 2002				
Y, n = 90	16 (17.78)	74 (82.22)	Referent	0.025
N, n = 45	16 (35.56)	29 (64.44)	2.55 (1.13–5.77)	
Occupation				
Underage/none, n = 29	6 (20.69)	23 (79.31)	Referent	0.067
Pupil/student, n = 54	8 (14.81)	46 (85.19)	0.67 (0.21–2.15)	
Health worker, n = 4	2 (50.00)	2 (50.00)	3.07 (0.84–11.17)	
Farmer, n = 18	8 (44.44)	10 (55.56)	3.83 (0.44–33.11)	
Others†, n = 14	5 (35.71)	9 (64.28)	2.13 (0.52–8.77)	
Contact with human case				
Y, n = 57	17 (29.82)	40 (70.18)	Referent	0.304
N, n = 56	10 (17.86)	46 (82.14)	0.51 (0.21–1.24)	
Unknown, n = 3	1 (33.33)	2 (66.67)	0.17 (0.10–13.86)	
Contact with animal				
Y, n = 37	12 (32.43)	25 (67.57)	Referent	0.143
N, n = 69	11 (15.94)	58 (84.06)	0.40 (0.15–1.0)	
Unknown, n = 7	2 (28.57)	5 (71.43)	0.88 (0.14–4.93)	
Contact with wild or domestic animal				
Domestic animal, n = 13	3 (23.08)	10 (76.92)	Referent	0.06
Wild animal, n = 13	6 (46.15)	7 (53.85)	2.86 (0.53–15.47)	
No contact, n = 69	11 (15.94)	58 (84.06)	0.63 (0.15–2.67)	
Travel history				
Y, n = 21	5 (23.81)	16 (76.19)	Referent	0.831
N, n = 96	25 (26.04)	71 (73.96)	0.89 (0.29–2.67)	
Geographic distribution				
Adamawa, n = 1	0	1 (100.00)	Referent	0.831
Centre, n = 32	11 (34.36)	21 (65.63)	0 to ∞	
East, n = 23	3 (13.04)	20 (86.96)	0 to ∞	
Far-North, n = 2	0	2 (100.00)	0 to ∞	
Littoral, n = 4	1 (25)	3 (75.00)	0 to ∞	
North, n = 1	0	1 (100.00)	0 to ∞	
North-West, n = 25	6 (24.00)	19 (76.00)	0 to ∞	
South, n = 9	1 (11.00)	8 (88.89)	0 to ∞	
South-west, n = 39	10 (25.64)	29 (74.36)	0 to ∞	
Other‡	0	1	0 to ∞	

*Bold text indicates statistical significance. Some categories might not add to 100% because of missing data. Missing data were not accounted for in the statistical analysis. MPXV, monkeypox virus; NA, not applicable; OR, odds ratio.

†Others include teachers, traders, driver or motorbiker, housewife, informal, retired.

‡Equatorial Guinea.

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difference for adults born before 1980 than for the rest of the population ($p = 0.092$). Larger datasets would be needed to confirm the observed trend.

MPXV infection was mostly associated with occupational activities involved in farming (OR 3.83, 95% CI 0.44–33.11) (Table 1). Similarly, potential nosocomial

Table 2. Clinical characteristics of suspected and confirmed mpox cases in a study of concurrent clade I and clade II monkeypox virus circulation, Cameroon, 1979–2022*

Characteristics	MPXV RT-PCR, no. (%)		Crude OR (95% CI)	p value
	Positive	Negative		
Total no. (%), n = 137	32 (23.36)	105 (76.64)		
Active skin lesions				
Lesions, n = 124	29 (23.39)	95 (76.61)	Referent	0.289
No lesions, n = 10	1 (10.00)	9 (90.00)	2.75 (0.33–22.6)	
Lesion progress				
Diffuse, n = 23	5 (21.74)	18 (78.26)	Referent	0.329
Head to limbs, n = 25	4 (16.00)	21 (84.00)	0.69 (0.16–2.95)	
Limbs to head, n = 15	6 (40.00)	9 (60.00)	2.4 (0.57–10.04)	
Others, n = 24	4 (16.67)	20 (83.33)	0.72 (0.17–3.1)	
Lesions at the same stage				
Y, n = 43	13 (30.23)	30 (69.77)	Referent	0.195
N, n = 53	10 (18.87)	43 (81.13)	1.86 (0.72–4.8)	
Lesions of the same size				
Y, n = 49	13 (26.53)	36 (73.47)	Referent	0.546
N, n = 47	10 (21.28)	37 (78.72)	1.34 (0.52–3.43)	
Lesions deep				
Y, n = 42	11 (26.19)	31 (73.81)	Referent	0.767
N, n = 51	12 (23.53)	39 (76.47)	1.15 (0.45–2.97)	
Fever before rash				
Y, n = 86	22 (25.58)	64 (74.42)	Referent	0.149
N, n = 30	4 (13.33)	26 (86.67)	2.23 (0.7–7.12)	
Missing	6	15	NA	
Headache				
Y, n = 51	15 (29.41)	36 (70.59)	Referent	0.1
N, n = 61	10 (16.39)	51 (83.61)	2.13 (0.86–5.26)	
Cough				
Y, n = 38	13 (34.21)	25 (65.79)	Referent	0.066
N, n = 76	14 (18.42)	62 (81.58)	2.3 (0.95–5.59)	
Vomiting, nausea				
Y, n = 15	4 (26.67)	11 (73.33)	Referent	0.722
N, n = 98	22 (22.45)	76 (77.55)	1.26 (0.36–4.34)	
Chills, sweat				
Y, n = 48	18 (37.50)	30 (62.50)	Referent	0.003
N, n = 66	9 (13.34)	57 (86.36)	3.8 (1.52–9.48)	
Lymphadenopathy				
Y, n = 29	12 (41.38)	17 (58.62)	Referent	0.009
N, n = 84	14 (16.37)	70 (83.33)	3.53 (1.38–9.00)	
Sore throat when swallowing				
Y, n = 28	16 (51.14)	12 (42.86)	Referent	<0.001
N, n = 85	10 (11.76)	75 (88.24)	10 (3.69–27.12)	
Oral ulcer				
Y, n = 18	11 (61.11)	7 (38.89)	Referent	<0.001
N, n = 95	15 (15.79)	80 (84.21)	8.38 (2.80–25.09)	
Itchy lesions				
Y, n = 75	19 (25.33)	56 (74.67)	Referent	0.397
N, n = 43	8 (18.60)	35 (81.40)	1.48 (0.59–3.75)	
Unknown	5	14	NA	
General fatigue				
Y, n = 62	20 (32.25)	42 (67.74)	Referent	0.016
N, n = 52	7 (13.46)	45 (86.54)	3.06 (1.17–7.98)	
Myalgia				
Y, n = 29	9 (31.03)	20 (68.97)	Referent	0.244
N, n = 84	17 (20.24)	67 (79.76)	1.77 (0.69–4.59)	
Unknown	6	18	NA	
Conjunctivitis				
Y, n = 14	2 (14.29)	12 (85.71)	Referent	0.385
N, n = 99	24 (24.24)	75 (75.76)	0.52 (0.11–2.49)	

*Bold text indicates statistical significance. Missing data is for patients who did not provide an answer. Unknown is for persons who replied that they did not know. Some categories might not add to 100% because of missing data. Missing data were not accounted for in the statistical analysis. MPXV, monkeypox virus; NA, not applicable; OR, odds ratio.

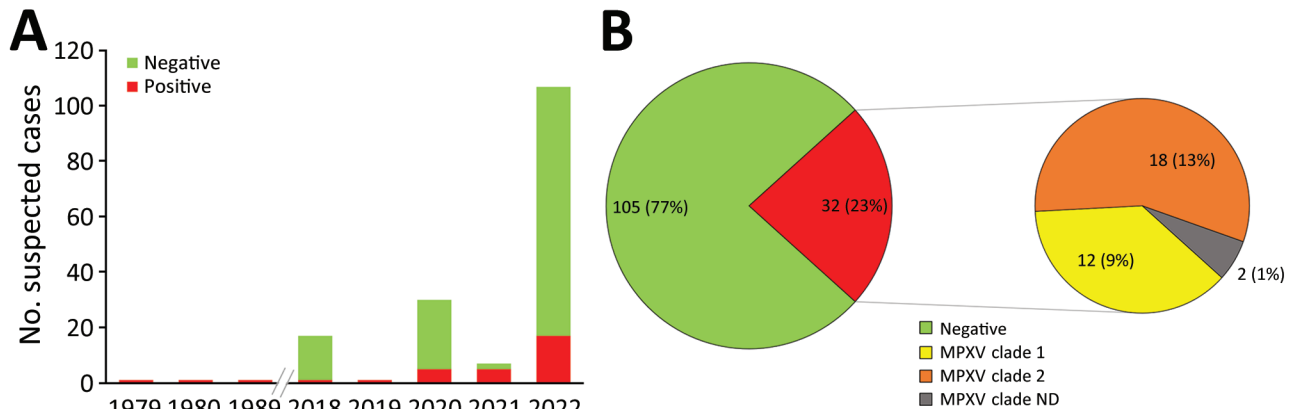


Figure 1. Mpox cases in a study of concurrent clade I and clade II MPXV circulation, Cameroon, 1979–2022. A) Epidemiologic curve of 137 suspected mpox cases. A 30-year gap occurred between the first 3 reported mpox cases and the consecutive cases since 2018, demonstrating increased surveillance in the country. The peak in 2022 corresponds to the worldwide alert raised on mpox, which led to enhanced mpox surveillance in Cameroon. B) Mpox genotyping results showing both clade I and clade II MPXV were identified. MPXV, monkeypox virus; ND, not determined.

transmission was identified in health workers (OR 3.07, 95% CI 0.84–11.17). Other activities, including teaching, trading, or driving, when considered together, also appeared to be potential risk activities for secondary MPXV transmission (OR 2.13, 95% CI 0.52–8.77). However, we found no association for secondary transmission in the 29.82% of MPXV-confirmed cases reporting past contact with persons who had mpox-like clinical signs (Table 1). Because mpox is typically zoonotic, we also assessed antecedent of animal exposures. We observed no association with unspecified animal contacts but observed a higher risk among confirmed cases (6/13 [46.15%]) who reported contact with wild animals (OR 2.86, 95% CI 0.53–15.47) compared with persons reporting contact with domestic animals or having no contact with animals (Table 1). Among wild animal contact, study participants frequently mentioned squirrels, bats, caterpillars, pangolins, rats, porcupines, and monkeys.

As expected from the case definition criterium requiring skin rashes, almost all (124/137 [90.5%]) MPXV-suspected cases had active skin lesions (Table 2; Figure 2; Appendix Table 3). However, we observed no specific difference for lesion progress, deepness, size, or stage among MPXV-confirmed cases compared with MPXV-negative persons (Table 2). Maculopapular lesions were more prevalent in confirmed cases who had lesions on their palms and soles (Figure 2). Clinical data identified cough (OR 2.3, 95% CI 0.95–5.59), chills or sweat (OR 3.8, 95% CI 1.52–9.48), lymphadenopathy (OR 3.53, 95% CI 1.38–9.00), sore throat when swallowing (OR 10, 95% CI 3.69–27.12), mouth ulcers (OR 8.38, 95% CI 2.8–25.09), and general fatigue (OR 3.06, 95% CI 1.17–7.98) as potential

symptoms associated with MPXV infection in Cameroon (Table 2; Figure 2). Among all suspected case-patients, ≈26% who reported experiencing fever before skin rashes developed were confirmed for MPXV infection, but we saw no difference between confirmed cases with or without fever. In addition, MPXV-confirmed or -negative cases did not experience differences in headache (Table 2). We noted little difference in clinical severity in cases infected with clade I compared with those infected with clade II (Appendix Table 4). The same was true for the exposure route; we found no association between zoonotic or human-to-human transmission and a specific infecting viral clade (Appendix Table 4). However, because considerable data were missing (Tables 1, 2) we were unable to perform a multivariable analysis. Therefore, concluding interpretations of the epidemiologic and clinical features of mpox infection in Cameroon are difficult to draw.

Geographic and Phylogenetic Analysis

Reported suspected mpox cases originated from 8 administrative regions of Cameroon (Table 1; Figure 3). Most (97.08%) suspected cases were reported from the southern part of the country where all confirmed cases also originated. In particular, 1 (3.13%) case was confirmed in Littoral, 1 (3.13%) in the South, 3 (9.38%) in the East, 6 (18.75%) in the Northwest, 10 (31.25%) in the Southwest, and 11 (34.88%) in the Centre regions (Table 1; Figure 3; Appendix Table 3). Of note, a unique case confirmed in the Littoral region was originally from the Southwest and sought healthcare in Littoral. Genotyping of real-time PCR revealed that all clade I MPXV infections were



Figure 2. Maculopapular lesions in mpxv patients from a study of concurrent clade I and clade II monkeypox virus circulation, Cameroon, 1979–2022. A–E) Deep maculopapular lesions of different sizes spread from the head (A, C) to hands (B) and diffuse to the soles of the feet (D) the palm of the hand (E). F) Lesions, including oral lesions and mouth ulcers, in a 3-month-old male baby.

confirmed in patients from the Centre, South, and East regions; all but 1 of clade II MPXV samples were recovered from patients from the Littoral, Northwest, and Southwest regions. Indeed, a clade II MPXV detected in the Centre region was an internally displaced person (IDP) originally from the Northwest region (Table 1; Appendix Table 3). The distribution of mpxv cases points toward geographic segregation of the 2 viral clades in Cameroon. Those findings indicate a strong geographic association of MPXV genotypes in southern Cameroon, and that MPXV clade II is associated with the western part and the clade I with the eastern part of the country.

We obtained partial MPXV *ATI* gene sequences from 8 mpxv-confirmed cases from 4 regions of Cameroon. We derived the newly determined sequences from samples collected in the Northwest (CPC code 22V-0972), Southwest (CPC codes 22V-07739, 22V-07911, 22V-07968), Centre (CPC codes 22V-05210, 22V-04865, 22V-4639), and South (CPC code 22V-6957) regions. Maximum-likelihood phylogenetic analysis of the 942 nt consensus sequences, including reference sequences (Appendix Tables 1–3), revealed that the 8 MPXV genomes from Cameroon segregated into clade I and clade II. As expected from the geographic association of MPXV isolates we report, MPXV clade I from the Centre

and South regions grouped reliably with reference counterparts previously reported from countries in Central Africa, and clade II sequences from the Northwest and Southwest regions grouped consistently with strains from West Africa (Figure 4). Clade II strains from Cameroon clustered reliably within subclade IIb with 83% bootstrap support (Figure 4). Altogether, genotypic and phylogenetic analysis confirmed the concurrent circulation of both MPXV clades I and II in Cameroon with a striking geographic segregation.

Discussion

We examined the clinical, epidemiologic, and molecular patterns of MPXV infection in Cameroon over a 44-year period (1979–2022) as part of mpxv surveillance in the country. During 1979–2022, a total of 137 persons were suspected of having mpxv, and 32 were confirmed to be MPXV infected. Three persons died (CFR 2.2%) and death was associated with MPXV clade II. That CRF is much lower than those reported in previous studies of MPXV clade I that showed CFRs of 7%–10% (13,43). Overall, CFRs are lower among patients infected with clade II, including in the 2022 global outbreak settings (4,25). We were not able to collect information on potential underlying conditions of case-patients to determine whether

immunocompromising conditions contributed to death, which would have worsened the clinical disease manifestations, as highlighted by others (44). In addition, fatal cases associated with clade I potentially escaped the national surveillance system in Cameroon, which is new and still being improved.

We found that both primary zoonotic and secondary human-to-human MPXV transmission occurs in Cameroon, including nosocomial transmission affecting health workers. Our results are consistent with reports describing secondary transmission chains, in-

cluding intrafamilial transmission and occupational transmission through trade, transportation, hunting, and healthcare in endemic countries (24,43,45,46). This study highlights a common MPXV acquisition pathway in endemic countries, interspecies transmission, and wild animals are presumed reservoirs of the virus (31,32,47). Distinguishing between primary and secondary transmission is difficult because both could occur. Additional data and further investigations are required to clearly understand the underlying drivers of MPXV transmission in Cameroon.

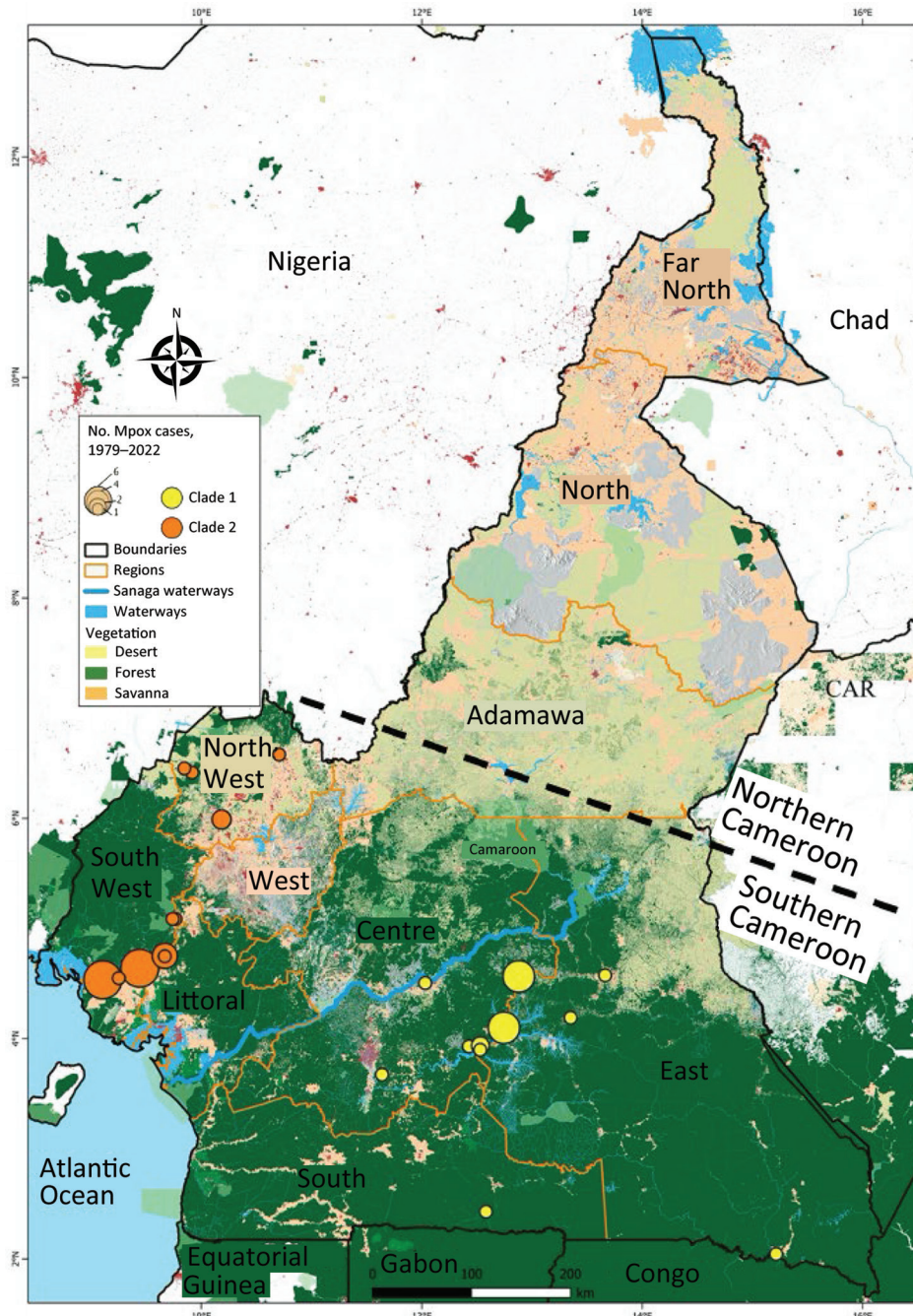


Figure 3. Geographic distribution of confirmed mpox cases and clades in a study of concurrent clade I and clade II monkeypox virus circulation, Cameroon, 1979–2022. A total of 137 suspected mpox cases were reported in the framework of the mpox surveillance system, among which 37 were PCR-confirmed for monkeypox virus infection. Clade I (12 cases) and clade II (18 cases) viral strains were identified circulating in the country. We noted a clear geographic segregation between the Centre, South, and East regions where only clade I (yellow dots) was reported, and the Northwest, and Southwest regions where only clade II (orange dots) was found. The size of each dot is proportional to the number of confirmed cases on the map. The map was designed by using Quantum GIS version 3.30.1 (QGIS, <https://qgis.org>). CAR, Central African Republic.

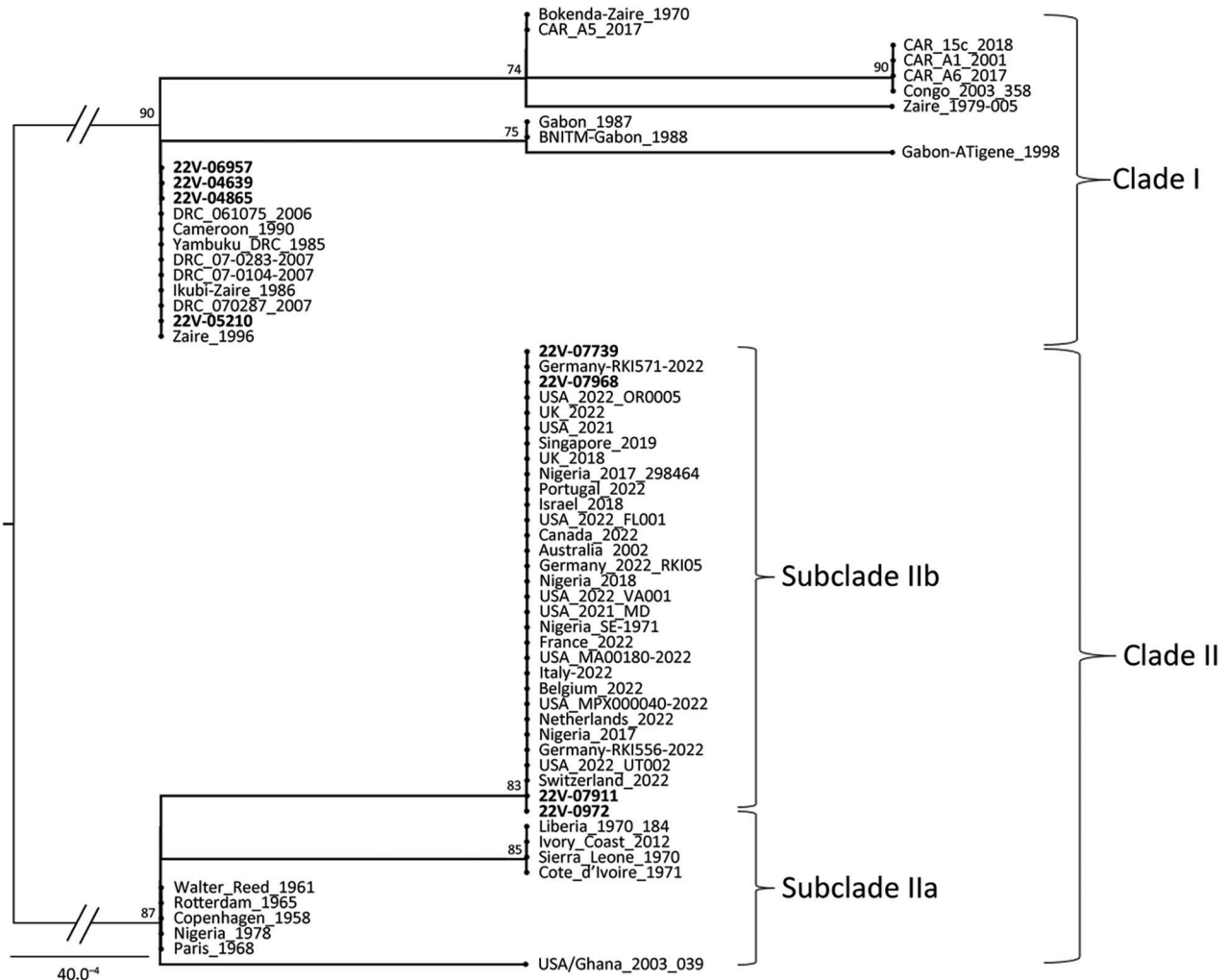


Figure 4. Maximum-likelihood phylogenetic tree of sequences in a study of concurrent clade I and clade II monkeypox virus circulation, Cameroon, 1979–2022. The tree is based on the Hasegawa-Kishino-Yano model inferred from a 942-bp fragment of the *AT1* gene, including 8 virus sequences from Cameroon generated in this work (bold text) and 55 reference sequences from GenBank. The tree with the highest log likelihood (–1,340.35) is shown. To test the robustness of the tree topology, 1,000 bootstrap replicates were performed. For a better display of the tree, the size of the 2 main midpoint rooted branches (represented in gray) that support the differentiation of the 2 monkeypox virus clades have been divided by half. Mpox strains from Cameroon are closely related to clades I and II, especially clade IIb for which a highlighted link to the ongoing global mpox epidemic is noted. Scale bar indicates number of substitutions per site. CAR, Central African Republic; DRC, Democratic Republic of the Congo; USA, United States.

A limitation of this study is our inability to perform more precise analyses to determine the characteristics independently describing the mpox epidemiology in Cameroon. Because the current surveillance system is still handwritten and forms are often incompletely filled, data are missing, as is common in paper-based data collection systems (48).

Since 1979, MPXV infections in Cameroon have occurred in 6 of the 10 administrative divisions of the country: Centre, South, East, Littoral, Northwest, and Southwest. All those administrative divisions are in the southern part of the country, which is a forested area encompassed by the lower mon-

tane forest of Guinea and the tropical rainforest of the Congo Basin, a favorable ecosystem for potential wildlife hosts. In contrast, northern Cameroon, a dry Sahelian and savannah zone, seems unlikely to be conducive to MPXV transmission because no cases have been confirmed in this region. That ecosystem is probably not suitable for MPXV reservoirs due to the dry environment. In most endemic countries, including Sierra Leone, Nigeria, Liberia, Central African Republic, and the Democratic Republic of the Congo, mpox cases mainly have been reported from forested areas (24,25,46). Most MPXV-confirmed cases in our study originated from the Centre (34 [38%])

and Southwest (31 [25%]) regions, which are the 2 most affected areas in the country. The Northwest region was the third (18 [75%] cases) most affected region. The Northwest and Southwest regions have been most seriously affected by civil unrest since 2017. That civil unrest has increased the number of IDPs in the country, and IDPs often move to different regions and neighboring countries. Furthermore, that situation has greatly increased human contact with wildlife as IDPs seek refuge in makeshift camps in the forest. By living in overlapping natural habitats of wild animals and potential MPXV reservoirs, populations of the Southwest and Northwest regions are under increased threat of zoonotic MPXV acquisition. Indeed, in Africa, civil unrest often leads to increases in mpox cases, and risk for any zoonotic disease is common (4,49). In several endemic countries, mpox outbreaks in the context of armed conflicts or massive population movements are a typical epidemiologic feature, and those conditions are usually associated with inefficient disease surveillance and control (4,49).

Genotypic and phylogenetic analyses revealed that both clade I and clade II are concurrently circulating in Cameroon and that a clear geographic segregation appears between the 2 clades. Circulation of both MPXV clades in Cameroon was previously reported in 2 published MPXV sequences from Cameroon (6,29). However, this study builds on those findings and provides more samples to further confirm that clades I and II concurrently circulate in a single country, a unique feature in MPXV epidemiology.

The geographic segregation of the clades is more perceptible in clade II case 21V-04877 in the Centre region. An epidemiologic investigation revealed that the case-patient was an IDP originating from the Northwest region, where MPXV clade II is endemic. The geographic segregation observed between MPXV strains circulating in Cameroon can be attributed to the natural barriers that potential animal reservoirs might not be able to cross between the Centre, East, and South regions, covered by the Congo Basin tropical forest, and the Northwest and Southwest regions, covered by lower montane moist forest of Guinea (38,40). Indeed, the Sanaga River, which is the largest river in the country, and the Cameroon highlands region sharply separate the 2 geographic areas into tropical moist forest ecoregions. The Cross-Sanaga-Bioko coastal forests lie to the north between the Sanaga River and the Cross River of Nigeria, and the Atlantic Equatorial coastal forests extends south of the river through

southwestern Cameroon and other neighboring countries of central Africa (38,39). Alternatively, the 2 ecologic environments potentially host different reservoirs. Several studies aimed to identify presumed MPXV reservoirs (31,33,47), but none have emphasized the potential of 2 distinct reservoirs that could be specific to a given ecosystem. Furthermore, MPXV circulation in humans in Cameroon after decades of absence might have resulted from movements of human populations, reservoir hosts, or both from endemic reservoirs in neighboring countries as armed conflicts intensified cross-border movements since 2017. That hypothesis is supported by the clustering of newly sequenced MPXV strains with counterparts originating from neighboring countries that have no physical barrier with the eastern and western parts of Cameroon but have long terrestrial borders.

In summary, this study provides detailed insight into the mpox epidemic in Cameroon during a 44-year period. The epidemiology of mpox in Cameroon involves both primary and secondary transmission. Segregated clade I and II virus strains concurrently circulate, suggesting potential existence of distinct viral reservoirs and cross-border circulation of MPXV. This study can inform the design, optimization, and evaluation of public health interventions for monitoring and controlling mpox in Cameroon and other countries in Africa with similar epidemiologic settings.

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Concurrent Clade I and Clade II Monkeypox Virus Circulation, Cameroon, 1979–2022

Appendix

Appendix Table 1. Public monkeypox virus genomes used for phylogenetic analysis*

No.	Country	Year	Original name	Name on tree	Host	Accession no.	Clade	Sub-clade	Lineage
1	Cameroon	1989	Cameroon-1990	Cameroon_1990	Human	KJ642618	I	I	1
2	Gabon	1987	Gabon 1988	Gabon_1988	Human	KJ642619	I	I	1
3	Gabon	1988	BNITM-Gabon1988	BNITM-Gabon_1988	Human	OP498046	I	I	1
4	DRC	1970	Congo_8	Bokenda-Zaire_1970	Human	KJ642613	I	I	2
5	DRC	2003	Congo_2003_358	Congo_2003_358	Human	DQ011154	I	I	2
6	CAR	2018	015c_contig_SPADES	CAR_15c_2018	Human	MN702448	I	I	2
7	CAR	2001	A1_contig_SPADES	CAR_A1_2001	Human	MN702453	I	I	2
8	CAR	2017	A6_contig_SPADES	CAR_A6_2017	Human	MN702451	I	I	2
9	CAR	2018	A5_contig_SPADES	CAR_A5_2017	Human	MN702444	I	I	2
10	DRC	1985	Yambuku_DRC_1985	Yambuku_DRC_1985	Squirrel	KP849471	I	I	3
11	DRC	2006	DRC_06–1074	DRC_06–1075–2006	Human	JX878411	I	I	3
12	DRC	2007	DRC_07–0286	DRC_07–0287–2007	Human	JX878422	I	I	3
13	DRC	2007	DRC_07–0283	DRC_07–0283–2007	Human	JX878420	I	I	3
14	DRC	1986	Ikubi	Ikubi-Zaire_1986	Human	KJ642612	I	I	4
15	DRC	1996	Monkeypox virus	Zaire_1996	Human	NC_003310	I	I	4
16	DRC	1979	Zaire 1979–005	Zaire_1979–005	Human	HM172544	I	I	
17	Gabon	1998	A-type inclusion body protein gene	Gabon-AT1gene_1998	Human	U84504	I	I	
18	DRC	2007	DRC_07–0104	DRC_07–0104–2007	Human	JX878417	I	I	5
19	USA	2022	hMPXV/USA/CA-LACPHL-MA00154/2022	USA_2022_MA00180	Human	OP580587	II	IIa	
20	USA	1971	Nigeria-SE-1970	Nigeria_SE_1971	Human	KJ642617	II	IIa	
21	Nigeria	2017	MPXV_Nig_2017_298464	Nigeria_2017_298464	Human	MG693724	II	IIa	
22	USA	2022	MPXV/Human/USA/CA-CDPH-MPX000040/2022	USA_2022_MPX000040	Human	OP587264	II	IIa	
23	Liberia	1970	Liberia_1970_184	Liberia_1970_185	Human	DQ011156	II	IIa	
24	Cote d'Ivoire	1970	Cote d'Ivoire_1971	Ivory Coast_1971	Human	KP849470	II	IIa	
25	Sierra Leone	1970	Sierra Leone	Sierra Leone_1970	Human	AY741551	II	IIa	
26	USA	1961	MPXV-WRAIR7–61	Walter Reed_1961	Human	AY603973	II	IIa	
27	USA	2003	USA_2003_039	USA/Ghana_2003	Human	DQ011157	II	IIa	
28	France	1968	PCH	Paris_1968	Human	KJ642616	II	IIa	
29	Netherlands	1965	UTC	Rotterdam_1965	Human	KJ642614	II	IIa	
30	Denmark	1958	COP-58	Copenhagen_1958	Human	AY753185	II	IIa	
31	Nigeria	1978	W-Nigeria	Nigeria_1978	Human	KJ642615	II	IIa	
32	Cote d'Ivoire	2012	Ivory Coast 2012	Ivory Coast_2012	Human	KJ136820	II	IIa	
33	Germany	2022	MPXV/Germany/2022/RKI571	Germany-RKI571_2022	Human	OP764629	II	IIa	
34	Germany	2022	MPXV/Germany/2022/RKI556	Germany-RKI556_2022	Human	OP764614	II	IIa	
35	USA	2022	MPXV_USA_2022_OR0005	USA_2022_OR0005	Human	OP752116	II	IIa	
36	Nigeria	2017	MPXV-M2957_Lagos	Nigeria_2017	Human	MT903338.1	II	IIb	A
37	Nigeria	2018	MPXV-M3021_Delta	Nigeria_2018	Human	MT903339.1	II	IIb	A
38	Singapore	2019	MPXV-Singapore	Singapore_2019	Human	MT903342.1	II	IIb	A.1
39	UK	2018	MPXV-UK_P2	UK_2018	Human	MT903344.1	II	IIb	A.1

No.	Country	Year	Original name	Name on tree	Host	Accession no.	Clade	Sub-clade	Lineage
40	Switzerland	2022	MPXV-CH-38134631/2022	Switzerland_2022	Human	ON595760.2	II	IIB	B.1
41	Italy	2022	INMI-Pt1	Italy-2022	Human	ON614676.1	II	IIB	B.1
42	Netherlands	2022	MPXV_2022_NL001	Netherlands_2022	Human	ON615424.1	II	IIB	B.1
43	UK	2022	MPXV_UK_2022_3	UK_2022	Human	ON619837.2	II	IIB	B.1
44	Belgium	2022	MPX/UZ_REGA_1/Belgium/2022	Belgium_2022	Human	ON622712.1	II	IIB	B.1
45	France	2022	MPXV_FR_HCL0001_2022	France_2022	Human	ON622722.2	II	IIB	B.1
46	Australia	2022	MPxV/VIDRL01/2022	Australia_2022	Human	ON631963.1	II	IIB	B.1
47	Portugal	2022	Monkeypox/PT0027/2022	Portugal_2022	Human	ON649710.1	II	IIB	B.1
48	USA	2022	MPXV_USA_2022_FL001	USA_2022_FL001	Human	ON674051.1	II	IIB	A.2
49	USA	2022	MPXV_USA_2022_VA001	USA_2022_VA001	Human	ON675438.1	II	IIB	A.2
50	USA	2022	MPXV_USA_2022_UT002	USA_2022_UT002	Human	ON676706.1	II	IIB	B.1
51	USA	2021	MPXV_USA_2021_TX	USA_2021_TX	Human	ON676707.1	II	IIB	A.2
52	USA	2021	MPXV_USA_2021_MD	USA_2021_MD	Human	ON676708.1	II	IIB	A.1.1
53	Germany	2022	MPXV/Germany/2022/RK105	Germany_2022_RK105	Human	ON682264.4	II	IIB	B.1
54	Canada	2022	MPX/2022/Canada/AB1	Canada_2022	Human	ON736420.1	II	IIB	B.1
55	Israel	2018	Israel_2018	Israel_2018	Human	MN648051.1	II	IIB	A.1

*Available in GenBank (<https://www.ncbi.nlm.nih.gov/genbank>). CAR, Central African Republic; DRC, Democratic Republic of the Congo; UK, United Kingdom; USA, United States of America.

Appendix Table 2. Epidemiologic and molecular features of monkeypox virus sequences obtained from Cameroon

No.	Country	Age, y/sex	Collection year	Original SEQ_ID	SEQ_ID on tree	Host	Isolation source region/health district	MPXV clade	Accession no.
1	Cameroon	36/F	2022	22V-0972	22V-0972	Human	South-West/Benakuma	II	OR038717
2	Cameroon	10/M	2022	22V-06957	22V-06957	Human	South/Djoum	I	OR038718
3	Cameroon	6/F	2022	22V-04639	22V-04639	Human	Centre/Ayos	I	OR038719
4	Cameroon	20/M	2022	22V-04865	22V-04865	Human	Centre/Ayos	I	OR038720
5	Cameroon	11/F	2022	22V-07739	22V-07739	Human	South-West/Kumba South	II	OR038721
6	Cameroon	52/M	2022	22V-07911	22V-07911	Human	South-West/Deido-Tombel	II	OR038722
7	Cameroon	40/M	2022	22V-05210	22V-05210	Human	Centre/Ayos	I	OR038723
8	Cameroon	43/M	2022	22V-07968	22V-07968	Human	South-West/Tombel	II	OR038724

Appendix Table 3. Epidemiologic information of mpox confirmed cases in Cameroon, 1979–2022

No.	CPC code	Country	Region	Age, y/sex	Patient progress	MPXV PCR	MPXV clade	VZV PCR	Date	
									Disease onset	Sampling
1	UNK	Cameroon	Centre	4/F	Unk	+	UNK	UNK	UNK	1979 Jan 1
2	UNK	Cameroon	East	3/M	Unk	+	UNK	UNK	UNK	1980 Jan 1
3	UNK	Cameroon	Centre	7/M	Alive	+	I	UNK	1989 Dec 4	1989 Dec 11
4	18V-04552	Cameroon	Northwest	20/M	Alive	+	II	UNK	UNK	2018 May 4
5	19V-06906	Cameroon	Southwest	14/M	Alive	+	II	UNK	2019 Sep 7	2019 Sep 19
6	20V-00071	Cameroon	Centre	27/F	Alive	+	I	–	2020 Jan 3	2020 Jan 4
7	20V-02998	Cameroon	East	36/M	Alive	+	I	–	2020 Mar 14	2020 Mar 24
8	20V-14349	Cameroon	East	31/F	Alive	+	I	–	2020 Aug 26	2020 Sep 6
9	20V-14644	Cameroon	Centre	46/M	Alive	+	I	–	2020 Sep 16	2020 Sep 23
10	20V-14645	Cameroon	Centre	43/F	Alive	+	I	–	2020 Sep 18	2020 Sep 23
11	21V-04877	Cameroon	Centre	30/F	Alive	+	II	UNK	UNK	2021 Jul 22
12	21V-09233	Cameroon	Centre	11/M	Alive	+	I	–	2021 Dec 3	2021 Dec 13
13	21V-09234	Cameroon	Centre	8/M	Alive	+	I	–	2021 Nov 27	2021 Dec 15

No.	CPC code	Country	Region	Age, y/sex	Patient progress	MPXV PCR	MPXV clade	VZV PCR	Date	
									Disease onset	Sampling
14	21V-09565	Cameroon	Southwest	0/M	Alive	+	II	-	2021 Dec 22	2021 Dec 29
15	21V-09567	Cameroon	Southwest	31/M	Dead	+	II	-	2021 Dec 23	2021 Dec 29
16	22V-00302	Cameroon	Southwest	27/F	Alive	+	II	-	2021 Dec 27	2022 Jan 27
17	22V-00972	Cameroon	Northwest	36/F	Alive	+	II	-	UNK	2022 Feb 16
18	22V-00973	Cameroon	Northwest	1/F	Alive	+	II	-	UNK	2022 Feb 16
19	22V-04639	Cameroon	Centre	10/M	Alive	+	I	-	2022 Jun 9	2022 Jun 13
20	22V-04865	Cameroon	Centre	6/F	Alive	+	I	-	2022 Jun 20	2022 Jun 21
21	22V-05210	Cameroon	Centre	20/M	Alive	+	I	-	2022 Jun 28	2022 Jul 4
22	22V-06957	Cameroon	South	11/F	Alive	+	I	-	2022 Sep 5	2022 Sep 7
23	22V-07739	Cameroon	Southwest	52/M	Alive	+	II	-	2022 Sep 19	2022 Oct 6
24	22V-07911	Cameroon	Littoral	40/M	Alive	+	II	-	2022 Sep 25	2022 Oct 11
25	22V-07968	Cameroon	Southwest	43/M	Alive	+	II	-	UNK	2022 Oct 13
26	22V-08077	Cameroon	Southwest	23/F	Alive	+	II	-	2022 Oct 12	2022 Oct 14
27	22V-08300	Cameroon	Northwest	28/M	Alive	+	II	-	2022 Oct 14	2022 Oct 22
28	22V-08501	Cameroon	Southwest	23/M	Alive	+	II	-	UNK	2022 Oct 27
29	22V-08502	Cameroon	Southwest	18/M	Alive	+	II	-	UNK	2022 Oct 27
30	22V-08503	Cameroon	Southwest	0/M	Alive	+	II	-	UNK	2022 Oct 26
31	22V-09327	Cameroon	Northwest	1/M	Alive	+	II	-	2022 Nov 3	2022 Nov 8
32	22V-09403	Cameroon	Northwest	43/M	Alive	+	II	-	2022 Nov 2	2022 Nov 11

*CPC, Centre Pasteur du Cameroun; MPXV, monkeypox virus; UNK, unknown; VZV, +, positive; -, negative

Appendix Table 4. Severity of monkeypox virus clades in 30 confirmed cases in Cameroon from 1979 to 2022

Clinical Characteristics	MPXV RT-PCR, no. (%)		Crude OR (95% CI)	p value
	Clade I, n = 12 (40.00)	Clade II, n = 18 (60.00)		
Active Skin Lesions				
Y	12 (100.00)	17 (94.44)	1 (Referent)	0.995
N	0 (0.00)	1 (5.56)	0 (0.00–58.5)	
Missing	0	0	NA	
Lesion progress				
Diffuse	1 (14.29)	4 (33.33)	1 (Referent)	0.066
Head to limbs	0 (00.00)	4 (33.33)	0.00 (0 to ∞)	
Limbs to head	3 (42.86)	3 (25.00)	4.00 (0.27–60.32)	
Others	3 (42.86)	1 (8.33)	12.00 (0.51–280.09)	
Missing	5	6	NA	
Lesions at same stage				
Y	7 (70.00)	6 (46.15)	1 (Referent)	0.249
N	3 (30.00)	7 (53.85)	2.72 (0.48–15.47)	
Missing	2	5	NA	
Lesions of same size				
Y	8 (80.00)	5 (38.46)	1 (Referent)	0.057
N	2 (20.00)	8 (61.54)	6.4 (0.95–43.23)	
Missing	2	5	NA	
Lesions deep				
Y	4 (40.00)	7 (53.85)	1 (Referent)	0.511
N	6 (60.00)	6 (46.15)	0.57 (0.11–3.04)	
Missing	2	5	NA	
Fever before rash				
Y	12 (100.00)	10 (71.43)	1 (Referent)	0.995
N	0 (0.00)	4 (28.57)	0.00 (0.00–1.61)	
Missing	0	4	NA	
Headache				
Y	9 (75.00)	6 (53.85)	1 (Referent)	0.149
N	3 (25.00)	7 (46.15)	3.50 (0.64–19.19)	
Missing	0	5	NA	
Cough				
Y	7 (58.33)	6 (40.00)	1 (Referent)	0.346
N	5 (41.67)	9 (60.00)	2.1 (0.45–9.84)	

Clinical Characteristics	MPXV RT-PCR, no. (%)		Crude OR (95% CI)	p value
	Clade I, n = 12 (40.00)	Clade II, n = 18 (60.00)		
Missing	0	3	NA	
Vomiting, nausea				
Y	2 (16.67)	2 (14.29)	1 (Referent)	0.867
N	10 (83.33)	12 (85.71)	1.20 (0.14–10.12)	
Missing	0	4	NA	
Chills, sweat				
Y	8 (66.67)	10 (66.67)	1 (Referent)	1.000
N	4 (33.33)	5 (33.33)	1.00 (0.20–5.00)	
Missing	0	3	NA	
Lymphadenopathy				
Y	8 (66.67)	10 (71.43)	1 (Referent)	0.059
N	4 (33.33)	4 (28.57)	5.00 (0.94–26.53)	
Missing	0	4	NA	
Sore throat when swallowing				
Y	9 (75.00)	7 (50.00)	1 (Referent)	0.199
N	3 (25.00)	7 (50.00)	3.00 (0.56–16.01)	
Missing	0	4	NA	
Oral ulcer				
Y	8 (66.67)	3 (21.43)	1 (Referent)	0.026
N	4 (33.33)	11 (78.57)	7.33 (1.27–42.29)	
Missing	0	4	NA	
Itchy lesions				
Y	11 (91.67)	8 (53.33)	1 (Referent)	0.052
N	1 (8.33)	7 (46.67)	9.62 (0.98–94.54)	
Missing	0	3	NA	
General fatigue				
Y	10 (83.33)	10 (66.67)	1 (Referent)	0.334
N	2 (16.67)	5 (33.33)	2.5 (0.39–16.05)	
Missing	0	3	NA	
Myalgia				
Y	5 (41.67)	4 (28.57)	1 (Referent)	0.486
N	7 (58.33)	10 (71.43)	1.79 (0.35–9.13)	
Missing	0	4	NA	
Conjunctivitis				
Y	1 (8.33)	1 (7.14)	1 (Referent)	0.910
N	11 (91.67)	13 (92.86)	1.18 (0.07–21.17)	
Missing	0	4	NA	
Contact with human case				
Y	7 (58.33)	10 (62.50)	1 (Referent)	0.510
N	5 (41.67)	5 (31.25)	1.42 (0.30–6.88)	
Unknown	0 (0.00)	1 (6.25)	0.00 (0 to ∞)	
Missing	0	2	NA	
Contact with animal				
Y	7 (58.33)	5 (38.46)	1 (Referent)	0.570
N	4 (33.33)	7 (53.85)	0.40 (0.08–2.19)	
Unknown	1 (8.33)	1 (7.69)	0.71 (0.04–14.35)	
Missing	0	5	NA	
Contact with wild or domestic animal				
Domestic animal	2 (20.00)	1 (10.00)	1 (Referent)	0.397
Wild animal	4 (40.00)	2 (20.00)	1.00 (0.05–18.91)	
No contact	4 (40.00)	7 (70.00)	0.29 (0.02–4.24)	
Missing	2	8	NA	

*Bold text indicates statistical significance. MPXV, monkeypox virus; NA, not applicable; OR, odds ratio; RT-PCR, reverse transcription PCR.