DOI: 10.1111/tmi.13781

RESEARCH ARTICLE

Respiratory viruses in rural Zambia before and during the COVID-19 pandemic

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Funding information
National Institute of Allergy and Infectious Diseases, Grant/Award Number: HHSN272201400007C

Abstract

Objectives: With the emergence of the COVID-19 pandemic, restrictions were implemented globally to control the virus. Data on respiratory pathogens in sub-Saharan Africa during the COVID-19 pandemic are scarce. This analysis was conducted to evaluate patterns of respiratory pathogens in rural Zambia before and during the first year of the pandemic.

Methods: Surveillance was established in December 2018 at Macha Hospital in southern Zambia. Patients with respiratory symptoms in the outpatient and inpatient clinics were recruited. Nasopharyngeal samples were collected and tested for respiratory pathogens. The prevalence of respiratory symptoms and pathogens was evaluated and compared in the first (December 10, 2018–December 9, 2019) and second (December 10, 2019–November 30, 2020) years of surveillance.

Results: Outpatient visits and admissions for respiratory illness significantly decreased from the first to second year, especially among children. SARS-CoV-2 was not detected from any participants in Year 2. Among outpatients and inpatients with respiratory symptoms, the prevalence of respiratory syncytial virus and influenza viruses decreased from the first to second year. In contrast, the prevalence of rhinovirus/enterovirus, metapneumovirus and parainfluenza virus increased.

Conclusions: The epidemiology of respiratory viruses in rural Zambia changed during the first year of the COVID-19 pandemic, suggesting that public health interventions may have had an impact on the introduction and circulation of respiratory pathogens in this area.

KEYWORDS
Africa, COVID-19, influenza, respiratory syncytial virus, surveillance

INTRODUCTION

On March 11, 2020 the World Health Organisation designated coronavirus disease 2019 (COVID-19), the disease caused by the novel severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), as a global pandemic and urged countries to take public health action to prevent and control the virus.[1] This occurred as countries in the Southern Hemisphere were nearing their cold season when respiratory illness infections were expected to increase.[2] Initial concerns about a severe respiratory season with co-circulation of influenza viruses and SARS-CoV-2 did not materialise.

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Australia, for example, reported a dramatic decline in the prevalence of influenza-like illness (ILI), influenza cases and influenza admissions after implementing public health interventions designed to restrict COVID-19 transmission in March 2020.[3] Similar declines were observed for cases of respiratory syncytial virus (RSV) and human parainfluenza viruses among children in 2020.[4, 5]

Limited data are available from other regions in the Southern Hemisphere, particularly from sub-Saharan Africa. Surveillance from South Africa reported no respiratory specimens testing positive for influenza virus following implementation of public health interventions in 2020[6] and there has been little, if any, data on other respiratory pathogens.

In 2018, surveillance for respiratory infections was initiated in rural Zambia through the Johns Hopkins Center for Excellence in Influenza Research and Surveillance (JHCEIRS) to provide information on the burden and epidemiology of respiratory pathogens in a region that has been historically underrepresented in surveillance networks. In 2019, a significant burden of influenza virus, RSV and other pathogens was found.[7]

On March 18, 2020, Zambia recorded its first two cases of COVID-19 among individuals with a history of travel to Europe.[8] Within weeks, cases were detected among individuals without a history of travel. Zambia experienced a first wave of COVID-19 in July and August 2020 with cases occurring across a wide geographic area. A second wave began in December 2020. While 20,725 cases and 388 deaths had been reported nationwide by the end of 2020, only approximately 1900 cases were reported from Southern Province and 280 cases from Choma District, the region where JHCEIRS surveillance activities have been conducted.[8, 9] On March 26, 2020, public health interventions were imposed by the Zambian government, including screening and quarantine of travellers into Zambia, restrictions on travel for residents and public gatherings, closure of schools, indoor dining and recreational facilities and mandatory mask-wearing in public.[8] Easing of restrictions began in a phased manner on April 24, 2020, and continued until September 11, 2020 when most restrictions were fully lifted.

The JHCEIRS surveillance, which continued through 2020, provided the unique opportunity to increase our understanding of the epidemiology of respiratory pathogens during the COVID-19 pandemic in a rural area in sub-Saharan Africa. The objective of this analysis was to evaluate the burden and patterns of respiratory pathogens, including SARS-CoV-2, in rural Zambia in 2020 compared to previously reported data from 2019 prior to the emergence of the COVID-19 pandemic.[7]

METHODS

Study setting and participants

Surveillance for ILI and respiratory viruses was established in December 2018 at Macha Hospital, located in a rural area of Southern Province, Zambia. The hospital is a 208-bed facility that includes an outpatient department and seven inpatient wards. The hospital serves a catchment population of approximately 150,000 individuals, consisting primarily of subsistence farmers. The epidemiology of viral respiratory infections in Macha during the first year of surveillance, prior to the onset of the global COVID-19 pandemic, has been previously reported.[7] For the present analysis, Year 1 is defined as the period from December 10, 2018 to December 9, 2019 and Year 2 as the period from December 10, 2019 to November 30, 2020, coinciding closely with the emergence and spread of COVID-19 in sub-Saharan Africa.

The surveillance method has been previously described.[10] Briefly, all patients presenting to the outpatient department were screened for ILI. ILI was defined as documented (≥38°C) or reported fever together with cough or sore throat, with onset or clinical worsening in the past 7 days. Each week, an age-stratified sample of outpatients with ILI were approached for enrolment. In addition, inpatient surveillance was conducted. The medical records of all patients admitted to the male, female and paediatric medical wards were evaluated for respiratory illness. Those patients with a respiratory illness were further screened for ILI and approached for enrolment.

At enrolment, study staff administered a questionnaire detailing symptoms, exposures and medical history and collected a nasopharyngeal specimen. Participants were followed up 3–5 weeks later to ascertain vital status and assess the clinical course of their illness.

Testing and specimen selection

At enrolment, nasopharyngeal swabs were collected from all study participants, placed in 3 ml of viral transport media, and transported to the Clinical Research Laboratory at Macha Research Trust where they were tested using the GeneXpert Xpress Flu/RSV assay (Cepheid, Sunnyvale, CA). In addition, from May 1, 2020 onwards, all samples underwent testing for SARS-CoV-2 using various RT-qPCR kits (the Charité-Berlin protocol[11]; Da An Gene Co Ltd, Guangzhou, China[12]; and Maccura Biotechnology Co Ltd., Chengdu, China[13]) provided by the Zambian Ministry of Health through the Zambia National Public Health Institute. The remaining sample volume was stored at −80°C and shipped to Johns Hopkins University in Baltimore, Maryland for further testing.

After shipment, samples from all study participants ($n = 671$) enrolled during the first year of surveillance underwent testing using the BioFire® FilmArray Respiratory Panel EZ (BioFire Diagnostics, Salt Lake City, UT) to detect a range of respiratory viruses and atypical bacteria: adenovirus, human metapneumovirus, human rhinovirus/enterovirus, influenza A virus, influenza B virus, parainfluenza virus, RSV, coronaviruses (HKU1, 229E, OC43 and NL63 subtypes, but not SARS-CoV-2), *Bordetella pertussis*, *Chlamydia pneumoniae* and *Mycoplasma pneumoniae*. Samples from

the second year of surveillance underwent testing with the ePlex Respiratory Pathogen Panel 2 (GenMark Diagnostics, Inc., Carlsbad, CA). This panel identified the same pathogens as the FilmArray assay, with the exception of *Bordetella pertussis* and the addition of SARS-CoV-2. Given limited ePlex assay availability during Year 2, a subset of nasopharyngeal specimens was selected for testing ($n = 346$ of 476). This subset included specimens from all inpatient participants ($n = 31$), to enrich for those with severe disease, and a random sample of outpatient participants recruited each month ($n = 27$ or 28).

Statistical methods

Temporal trends in visits and ILI among outpatients and inpatients were summarised using descriptive statistics and graphical representations. Locally weighted scatterplot smoothing (LOWESS) methods were used to visualise temporal trends for age-stratified figures. To compare patient volume across years, we used two-sided paired t-tests, grouped by month. Age- and month-adjusted log-binomial regression was used to compare the prevalence of ILI across years.

Characteristics of the study population were summarised by year using descriptive statistics. Comparisons between years were performed using log-binomial regression for categorical characteristics and linear regression for continuous attributes.

The prevalence of respiratory pathogens was estimated for both the outpatient and inpatient population with ILI by month and surveillance year. As the ePlex Respiratory Panel did not include *Bordetella pertussis* and no cases of *Chlamydia pneumoniae* or *Mycoplasma pneumoniae* were detected, the analysis was limited to respiratory viruses. Due to the sampling of the outpatient population with ILI for enrolment and testing, survey sampling weights were used to estimate the prevalence of each viral respiratory pathogen among the outpatient population with ILI. In Year 1, weights were calculated based on the monthly age distributions of the outpatient population with ILI. In Year 2, additional weights were calculated based on the monthly distribution of samples selected for testing. The weighted prevalence of each pathogen was calculated by month and year. As all inpatients with ILI were recruited and tested, the monthly prevalence of each viral respiratory pathogen among the inpatient population with ILI was estimated as the proportion of cases among inpatient participants.

Co-infection was defined as the concurrent detection of two or more respiratory viruses among participants. Age-adjusted log-binomial models were employed to compare the prevalence of co-infections across participant populations (inpatient vs. outpatient) and age groups.

Statistical analyses were conducted using the Stata Statistical Software, version 14 (StataCorp LLC., College Station, TX) and SAS/STAT software, version 9.4 (SAS Institute Inc., Cary, NC).

Research ethics and consent

The surveillance was approved by the Johns Hopkins Institutional Review Board (IRB00168163), the Macha Research Trust Institutional Review Board (E.2018.02) and the Zambian National Health Research Authority. Adult participants and the parents or legal guardians of paediatric participants provided written informed consent. Children 12–15 years of age provided written assent.

RESULTS

ILI surveillance

Overall, 21,493 patients presented to the Macha Hospital Outpatient Department during Year 1 (December 10, 2018–December 9, 2019) and 20,846 patients during Year 2 (December 10, 2019–November 30, 2020). While there was no significant difference in the monthly patient volume across the 2 years (Y1 monthly mean: 1758.5 patients, Y2: 1769.5 patients, $p = 0.93$), the prevalence of ILI among outpatients was significantly lower in Year 2 (10.0%) than in Year 1 (17.1%; age-adjusted prevalence ratio [adjPR] 0.68, 95% confidence interval [CI] 0.65–0.71). The decline in ILI prevalence coincided with the introduction of wide-ranging national regulations aimed at combatting the spread of SARS-CoV-2 in early 2020 (Figure 1a). The decline in the prevalence of ILI was most substantial among outpatients in the youngest age groups (Figure 1b). There was a small but statistically significant difference in the age distribution of the outpatient population between Years 1 and 2, with fewer outpatients in the youngest of age groups presenting for care in Year 2 ($p < 0.001$; Figure S1).

Similar to outpatients, a decrease in the prevalence of acute respiratory illness was observed among inpatients admitted to the medical wards in Year 2 compared to Year 1 (Figure 1c). Despite an overall increase in the number of monthly inpatient admissions in Year 2 (Y1 monthly average: 144.8 patients, Y2: 161.4 patients), the proportion admitted with a respiratory diagnosis declined significantly from 15.6% (278/1783) in Year 1 to 9.0% (176/1950) in Year 2 (adjPR 0.57; 95% CI: 0.48–0.68), particularly among children (Figure 1D). The age distribution of inpatients changed from Year 1 to Year 2 with a significant decrease in the number of infants admitted (Figure S1).

Study population

The characteristics of the study population enrolled in Year 1 ($n = 671$) and Year 2 ($n = 476$) are presented in Table 1. Compared to the population recruited during Year 1, the population recruited in Year 2 had a significantly lower proportion of inpatients (14.2% vs. 6.5%; PR: 0.46, 95% CI 0.31–0.68; Table 1) and significantly lower prevalence of hypoxemia (15.1% vs. 5.8%; PR: 0.38, 95% CI 0.25–0.58).

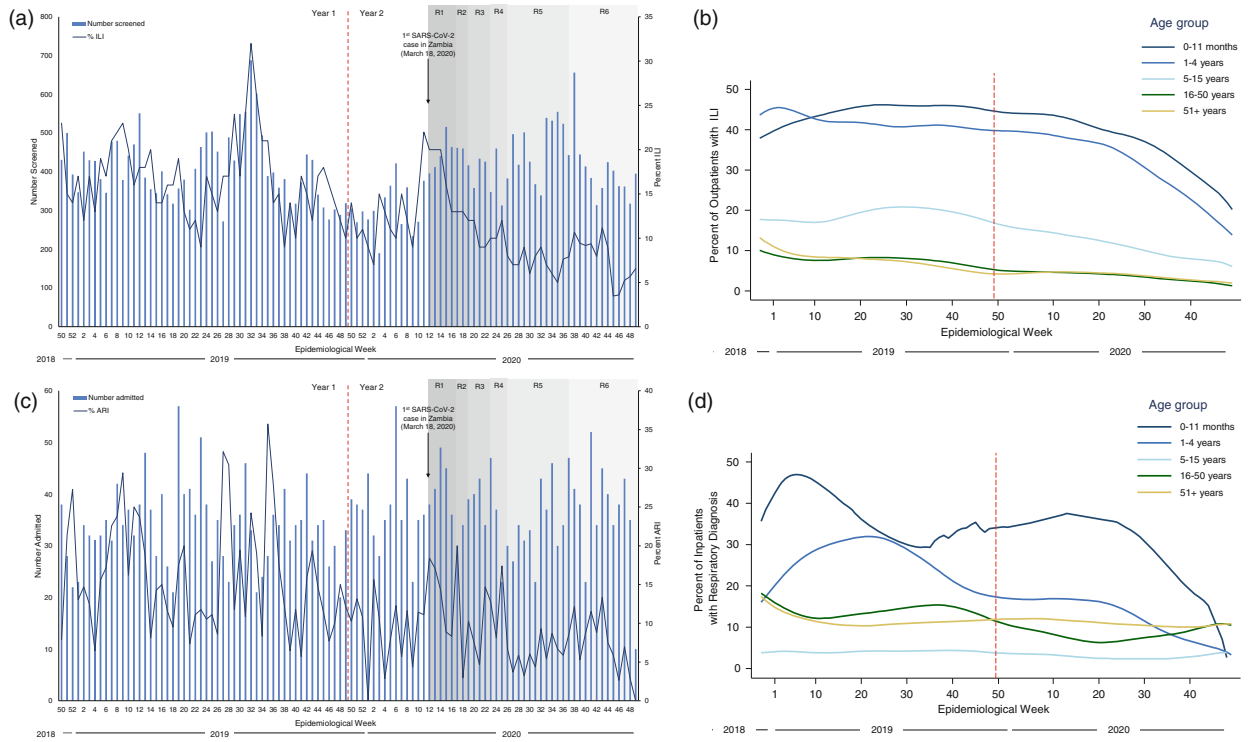

FIGURE 1 Respiratory illness among outpatients and inpatients in Macha, Zambia: December 2018–November 2020. (a) Weekly outpatient volume and proportion with influenza-like illness. Grey shaded backgrounds delineate the introduction, duration and easing of COVID restrictions. R1 (March 18, 2020): Schools closed, mandatory masking in public, screening and isolation of travellers into Zambia, all airports outside of Lusaka closed to international flights, suspension of non-essential travel outside of Zambia, restriction of public gathering, suspension of indoor dining, bars, nightclubs, cinemas, gyms, casinos closed. R2 (April 24, 2020): Resumption of congregation at places of worship, non-contact sports and operation of salons and barbershops. R3 (May 8, 2020): Restaurants, cinemas, gyms and casinos reopened. Hotels, lodges, tour operators and other businesses that closed voluntarily advised to consider reopening. R4 (June 1, 2020): Primary and secondary schools reopened for examination classes only. June 8, 2020: colleges and universities began a phased reopening for final year students only. R5 (June 25, 2020): All international airports reopened. R6 (September 11, 2020): Reopening of all non-examination classes in universities, colleges and schools. Partial reopening of bars, taverns and nightclubs. (b) Proportion of outpatients with influenza-like illness (ILI) stratified by age. (c) Number of inpatient medical admissions and proportion admitted for acute respiratory infections (ARI). (d) Proportion of inpatients with a diagnosed acute respiratory illness at admission stratified by age. The red dashed line in all figures separates Year 1 (December 10, 2018–December 9, 2019) and Year 2 (December 10, 2019–November 30, 2020) of surveillance.

The subset of participants from Year 2 selected for further microbiological testing ($n = 346$) was similar to the broader study population from that year except for a higher proportion of inpatients (9.0% vs. 6.5%), due to the preferential sampling of inpatient participants.

Respiratory virus prevalence

The proportion of specimens from which a respiratory virus was detected was similar in Year 1 (63.2%; 424/671) and Year 2 (65.9%; 228/346; $p = 0.42$). However, the observed epidemiology of respiratory infections was markedly different across the 2 years among both outpatient and inpatient participants. SARS-CoV-2 was not detected in any patient specimen from Year 2, either through PCR testing or the GenMark ePlex panel.

In contrast to Year 1, during which influenza viruses and RSV were commonly detected, these viruses did not appear to be major drivers of respiratory illness among outpatients with ILI in Year 2 (Figure 2). The estimated prevalence of RSV and influenza viruses among outpatients with ILI dramatically declined from 16.1% and 20.7% (13.5% influenza A, 7.2% influenza B) in Year 1 to 0.3% and 0% in Year 2, respectively. In contrast, the prevalence of rhinovirus/enterovirus (Y1: 23.5%; Y2: 42.4%), parainfluenza (Y1: 2.7%; Y2: 11.5%) and human metapneumovirus (Y1: 0.7%; Y2: 9.5%) all increased in Year 2, while adenovirus (Y1: 2.2%; Y2: 3.6%), and non-SARS-CoV-2 coronavirus (Y1: 5.1%; Y2: 7.6%) were similar across years.

In stratified analysis, the increased prevalence of rhinovirus/enterovirus was observed across outpatient age-groups while the increases in parainfluenza and metapneumovirus prevalence occurred predominantly among younger participants (Figure S2).

In Year 2, as in Year 1, we observed substantial year-round prevalence of rhinovirus/enterovirus among outpatients. A low prevalence of adenovirus was also observed throughout the year. In contrast, parainfluenza virus (early 2020), metapneumovirus (early 2020) and non-SARS-CoV-2 coronavirus (late 2020) all presented with a single annual prevalence peak in Year 2.

TABLE 1 Characteristics of the study population by year of enrolment

		Year 1	Year 2	Year 2
		(*n* = 671)	All participants (*n* = 476)	Sampled participants (*n* = 346)
Inpatient, *n* (%)[a]		95 (14.2%)	31 (6.5%)	31 (9.0%)
Age, years	Median (IQR)	3.2 (0.8–19)	2.8 (0.8–16)	2.7 (0.8–13)
	0–11 months, *n* (%)	187 (27.9%)	130 (27.3%)	93 (26.9%)
	1–4 years, *n* (%)	212 (31.6%)	151 (31.7%)	118 (34.1%)
	5–15 years, *n* (%)	86 (12.8%)	74 (15.5%)	55 (15.9%)
	16–50 years, *n* (%)	109 (16.2%)	78 (16.4%)	50 (14.5%)
	≥51 years, *n* (%)	77 (11.5%)	43 (9.0%)	30 (8.7%)
Female, *n* (%)		360 (53.7%)	247 (51.9%)	173 (50.0%)
Number of individuals in household, median (IQR)		7 (5–9)	7 (5–9)	7 (5–9)
Number of individuals sharing sleeping space, median (IQR)		3 (2–3)	3 (2–3.5)	3 (2–3)
Underweight, *n* (%)		68 (10.2%)	33 (7.0%)	23 (6.7%)
HIV-infected, *n* (%)		29 (4.3%)	12 (2.5%)	10 (2.9%)
History of tuberculosis, *n* (%)[a]		22 (3.3%)	5 (1.1%)	4 (1.2%)
Hypoxemic, *n* (%)[a,b]		81 (15.1%)	27 (5.8%)	23 (6.7%)
Died during follow-up, *n* (%)[a]		10 (1.8%)	0 (0.0%)	0 (0.0%)

Abbreviation: IQR, interquartile range.
[a]Statistically significant difference in prevalence/incidence (p <0.05) between Year 1 (December 10, 2018 to December 9, 2019) and Year 2 (December 10, 2019 to November 30, 2020) overall study populations. Statistical testing involving zero values performed using Fisher's exact test.
[b]Hypoxemia defined as oxygen saturation (SpO_2) ≤92% at enrolment.

Among inpatients with ILI, similar trends were observed. Influenza A virus (20.1%), influenza B virus (2.1%) and RSV (15.8%) were frequently identified in Year 1 but not at all in Year 2 (Table 2; Figure S3). In contrast, the prevalence of parainfluenza virus (Y1: 3.2%; Y2: 22.6%) and human metapneumovirus (Y1: 0.0%; Y2: 12.9%) increased in Year 2, while adenovirus (Y1: 4.2%; Y2: 6.5%) and rhinovirus/enterovirus (Y1: 17.9%; Y2: 19.4%) were similar across years. Notably, despite an increase in non-SARS-CoV-2 coronavirus prevalence among outpatients with ILI in Year 2, non-SARS-CoV-2 coronaviruses were not detected among any inpatients with ILI in Year 2.

Viral co-infections

The prevalence of viral co-infections among all participants was similar across years (Y1: 6.4%, Y2: 6.4%; Table 3) and consistently higher among children than adults. Rhinovirus/enterovirus was the most common co-infecting species across both years (Table S2). In a pooled analysis across both years, the likelihood of viral co-infection was not significantly different between inpatient and outpatient participants (age-adjusted risk ratio [adjRR]: 0.51, 95% CI 0.19–1.35).

DISCUSSION

Our surveillance infrastructure provided a unique opportunity to evaluate changes in the epidemiology of respiratory infections during the early months of the pandemic in a rural area of Zambia, a region that has historically been under-surveilled. Perhaps most notably, we did not detect a single SARS-CoV-2 infection in our study population through the end of November 2020 and no cases were reported through government testing at the hospital. During this time, over 17,000 cases of COVID-19 were reported across Zambia, including approximately 1900 in Southern Province.[9, 14] The reported number of cases is likely to be a substantial underestimate given limited surveillance and testing infrastructure, particularly outside urban centres. In July 2020, a cross-sectional prevalence survey across six high burden districts in Zambia estimated the ratio of reported to actual cases to be 1–92.[15] A sizeable prevalence of SARS-CoV-2 infection was found across all districts (range: 6.0%–14.4%). Furthermore, published findings from Lusaka suggest a substantial prevalence of SARS-CoV-2 infection among deceased patients from June to September 2020.[16] The findings from our surveillance program therefore suggest that the virus may not have penetrated this rural area in the context of public health interventions and that important sub-national differences exist. Differences in the course of the pandemic in rural and urban areas were initially projected for several African countries[17] and have been reported anecdotally.[18]

Among non-SARS-CoV-2 viruses, we observed marked differences in the diversity of respiratory viruses during the first and second years of surveillance in southern Zambia. In particular, influenza A and B viruses and RSV were major disease drivers in Year 1 but were scarcely detected in Year 2. Similar declines in influenza virus and RSV activity

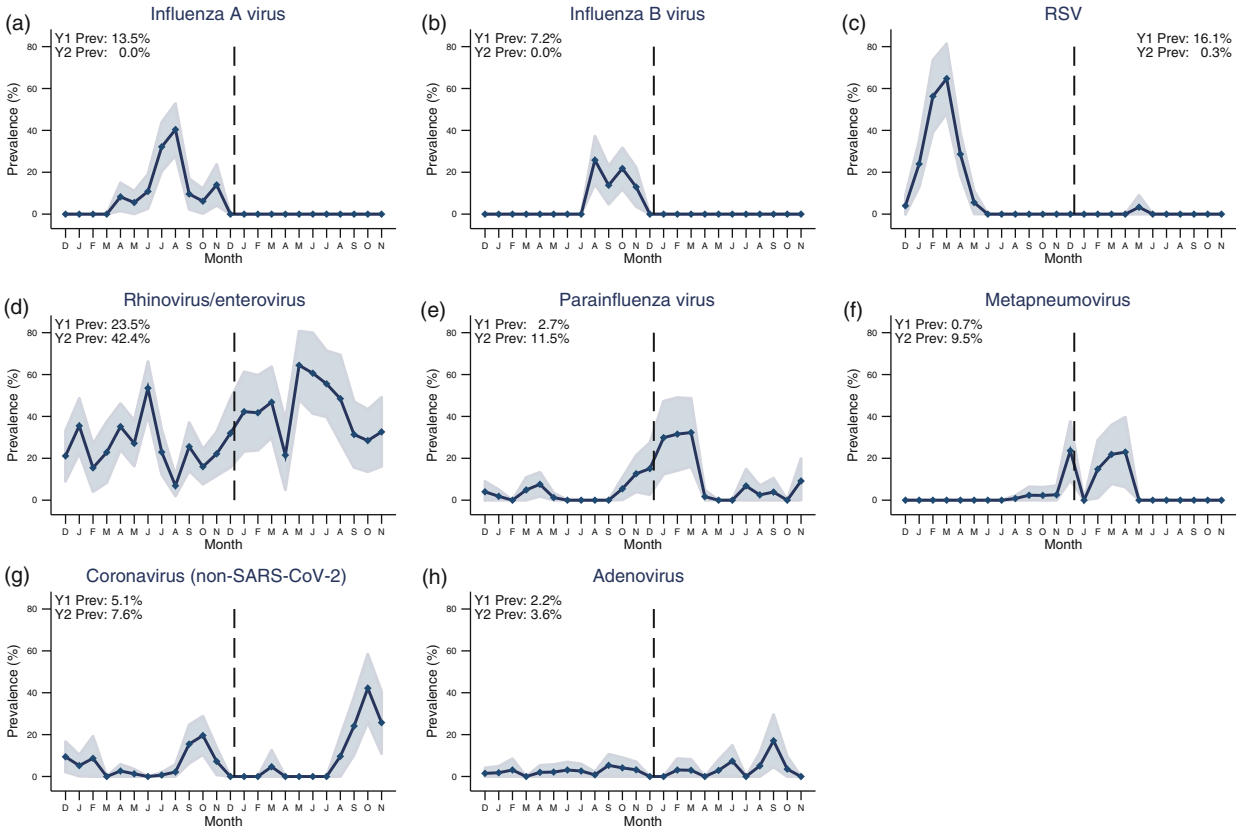

FIGURE 2 Prevalence of respiratory viruses among outpatients with ILI in Macha, Zambia from December 2018 to November 2020. Estimated prevalence of pathogens among outpatients with influenza-like illness (ILI) from December 2018 to November 2020 for (a) influenza A virus, (b) influenza B virus, (c) respiratory syncytial virus (RSV), (d) rhinovirus/enterovirus, (e) parainfluenza virus, (f) human metapneumovirus, (g) non-SARS-CoV-2 coronavirus, (h) adenovirus. Percentages for Year 1 (December 10, 2018–December 9, 2019) and Year 2 (December 10, 2019–November 30, 2020) represent estimated yearly pathogen prevalence overall among all outpatients with ILI. Lines represent estimated monthly prevalence, grey bands represent 95% confidence intervals. The vertical dashed line in all figures separates Year 1 and Year 2. The overall pathogen prevalence estimates for Year 1 differ from those previously published[7] due to the different methodology used to account for sampling at recruitment and testing in this analysis.

TABLE 2 Prevalence of respiratory viruses among inpatients with influenza-like illness in Macha, Zambia from December 2018 to November 2020

Pathogen	Year 1 ($n = 95$)	Year 2 ($n = 31$)
Influenza A virus	20 (20.1%)	0 (0.0%)
Influenza B virus	2 (2.1%)	0 (0.0%)
Respiratory syncytial virus	15 (15.8%)	0 (0.0%)
Adenovirus	4 (4.2%)	2 (6.5%)
Coronavirus (non-SARS-CoV-2)	2 (2.1%)	0 (0.0%)
Parainfluenza virus	3 (3.2%)	7 (22.6%)
Rhinovirus/enterovirus	17 (17.9%)	6 (19.4%)
Human metapneumovirus	0 (0.0%)	4 (12.9%)

Note: Year 1 is defined as the period from December 10, 2018 to December 9, 2019 and Year 2 as the period from December 10, 2019 to November 30, 2020.

during the COVID-19 pandemic have been described in a number of Southern[4, 6, 19] and Northern Hemisphere settings,[20–23] with only one report from sub-Saharan Africa to date.[6] The dramatic decline in prevalence suggests a large impact of preventive public health measures in this and other settings on the transmission of these viruses. In particular, non-pharmaceutical interventions that restrict movement and gatherings have had a large impact on viral transmission.[24] While reported travel outside of the district was rare among our study population both before and during the pandemic, reductions in movement into the area and in the size and frequency of gatherings during the pandemic may have limited introduction and spread of these viruses into this rural area. Further investigation will be needed to understand the relative importance of local, national and international interventions in altering the epidemiology of seasonal and pandemic respiratory infections and the long-term impact of these disruptions on immunity and health.

In contrast to influenza virus and RSV, other respiratory viruses continued to circulate and to be detected among both inpatients and outpatients despite implementation of public health interventions. Other surveillance efforts have reported on respiratory virus diversity during the pandemic but none, to our knowledge, from sub-Saharan Africa. Australia, New Zealand, Japan, Germany, England and

TABLE 3 Prevalence of co-infections among participants with influenza-like illness in Macha, Zambia from December 2018 to November 2020

		Prevalence of co-infections		
		Year 1	Year 2	Overall
Type of participant, % (*n*/*N*)	Inpatients	3.2% (3/95)	3.2% (1/31)	3.2%
	Outpatients	6.9% (40/576)	6.7% (21/315)	6.9%
Age group, % (*n*/*N*)	0–11 months	12.8% (24/187)	14.0% (13/93)	13.2%
	1–4 years	3.8% (8/212)	6.8% (8/118)	4.9%
	5–15 years	5.8% (5/86)	1.8% (1/55)	4.3%
	16–50 years	3.7% (4/109)	0.0% (0/50)	2.5%
	≥51 years	2.6% (2/77)	0.0% (0/30)	1.9%
Overall, % (*n*/*N*)		6.4% (43/671)	6.4% (22/346)	6.4%

Note: Year 1 is defined as the period from December 10, 2018 to December 9, 2019 and Year 2 as the period from December 10, 2019 to November 30, 2020.

Austria continued to experience entero-rhinovirus infections through 2020, particularly once restrictions were eased,[3, 25–29] but very low prevalence of human metapneumovirus, adenovirus, parainfluenza virus and coxsackievirus A and B was observed.[25, 28, 29] Structural and transmission-related properties of these viruses may be driving the observed epidemiological differences. For entero-rhinovirus, potential reasons for continued circulation that have been noted include the year-round circulation of the virus in non-pandemic years,[25] the non-enveloped structure of the virus rendering it more stable in the environment and resistant to disinfectants,[25, 27, 28] a relatively increased contribution of fomite transmission for this virus which is less effectively prevented by masking,[25, 27] and the potentially lower efficacy of masks in preventing transmission.[30] In rural settings, such as in Macha, the impact of these factors may be enhanced, as hygiene-related interventions may be more difficult to implement and sustain and travel restrictions may be more effective in preventing introductions of seasonal viruses into remote areas.

This study has some limitations. Firstly, hospital-based virus surveillance is less sensitive than community-based approaches, particularly in the detection of asymptomatic or pauci-symptomatic respiratory disease. This may be particularly true if patients avoid health facilities for fear of contracting SARS-CoV-2 infection, leading to an underestimation of disease burden. However, we saw little evidence of hospital avoidance, with similar numbers of visits and admissions before and during the pandemic period. Furthermore, no cases of SARS-CoV-2, influenza virus or RSV were detected even among hospitalised inpatients, suggesting a true absence of severe respiratory infections with these viruses. Secondly, the use of a varied set of PCR primers for detecting SARS-CoV-2 may have decreased the sensitivity of the initial test. However, all specimens underwent subsequent testing with the ePlex Respiratory Panel 2, with a reported percent positive agreement for SARS-CoV-2 of 100% with known positive clinical samples,[31] and inclusivity of 99.99% for the B.1.351 variant[32] which was detected in Zambia shortly after the study period.[33] No discrepant results were observed between the assays.

During the first year of the global COVID-19 pandemic, rural southern Zambia experienced a marked change in the epidemiology of respiratory viruses. While many areas in Zambia, particularly urban centres and transit hubs, experienced a large wave of SARS-CoV-2 infections, rural southern Zambia was spared. These results and the declining prevalence of key viral drivers of disease raise questions about the impact of non-pharmaceutical public health interventions and strongly support the need for broad-based, geographically diverse viral surveillance.

ACKNOWLEDGEMENTS

We thank the study team and participants for their contributions to the study. We also thank the staff at Macha Hospital for their support and assistance in conducting the study.

FUNDING INFORMATION

This work was supported by the National Institute of Allergy and Infectious Diseases (contract HHSN 272201400007C) awarded to the Johns Hopkins Center of Excellence in Influenza Research and Surveillance (JHCEIRS). Any opinions, findings, conclusions or recommendations expressed are those of the authors and do not represent the policy or position of NIAID or NIH. The funding agency had no role in the analysis of the data, writing of the manuscript or the decision to submit the manuscript for publication.

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SUPPORTING INFORMATION

Additional supporting information may be found in the online version of the article at the publisher's website.

How to cite this article: Loevinsohn G, Hamahuwa M, Hardick J, Sinywimaanzi P, Fenstermacher KZJ, Munachoonga P, et al. Respiratory viruses in rural Zambia before and during the COVID-19 pandemic. Trop Med Int Health. 2022;27(7):647–54. https://doi.org/10.1111/tmi.13781