Respiratory Viral Infection: An Underappreciated Cause of Acute Febrile Illness Admissions in Southern Sri Lanka

L. Gayani Tillekeratne,^{1,2,3*} Champica K. Bodinayake,^{3,4} Ryan Simmons,² Ajith Nagahawatte,^{3,4} Vasantha Devasiri,^{3,4}
Wasantha Kodikara Arachchi,^{3,5} Bradly P. Nicholson,^{1,3} Lawrence P. Park,^{1,2} Sky Vanderburg,^{1,3} Ruveni Kurukulasooriya,³
Aruna Dharshan De Silva,^{3,6} Truls Østbye,^{1,2,3} Megan E. Reller,^{1,2,3†} and Christopher W. Woods^{1,2,3†}

¹Duke University, Durham, North Carolina; ²Duke Global Health Institute, Durham, North Carolina; ³Duke-Ruhuna Collaborative Research Centre, Galle, Sri Lanka; ⁴Faculty of Medicine, University of Ruhuna, Galle, Sri Lanka; ⁵Teaching Hospital Karapitiya, Galle, Sri Lanka; ⁶General Sir Kotelawala Defence University, Ratmalana, Sri Lanka

Abstract. The contribution of respiratory viruses to acute febrile illness (AFI) burden is poorly characterized. We describe the prevalence, seasonality, and clinical features of respiratory viral infection among AFI admissions in Sri Lanka. We enrolled AFI patients ≥ 1 year of age admitted to a tertiary care hospital in southern Sri Lanka, June 2012–October 2014. We collected epidemiologic/clinical data and a nasal or nasopharyngeal sample that was tested using polymerase chain reaction (Luminex NxTAG, Austin, TX). We determined associations between weather data and respiratory viral activity using the Spearman correlation and assessed respiratory virus seasonality using a Program for Appropriate Technology definition. Bivariable and multivariable regression analyses were conducted to identify features associated with respiratory virus detection. Among 964 patients, median age was 26.2 years (interquartile range 14.6–39.9) and 646 (67.0%) were male. One-fifth (203, 21.1%) had respiratory virus detected: 13.9% influenza, 1.4% human enterovirus/rhinovirus, 1.4% parainfluenza virus, 1.1% respiratory syncytial virus, and 1.1% human metapneumovirus. Patients with respiratory virus identified were younger (median 9.8 versus 27.7 years, $P < 0.001$) and more likely to have respiratory signs and symptoms. Influenza A and respiratory viral activity peaked in February–June each year. Maximum daily temperature was associated with influenza and respiratory viral activity ($P = 0.03$ each). Patients with respiratory virus were as likely as others to be prescribed antibiotics (55.2% versus 52.6%, $P = 0.51$), and none reported prior influenza vaccination. Respiratory viral infection was a common cause of AFI. Improved access to vaccines and respiratory diagnostics may help reduce disease burden and inappropriate antibiotic use.

INTRODUCTION

Acute febrile illness (AFI) or fever without localizing signs is a common reason for hospitalization in tropical and subtropical settings.¹ Acute febrile illness etiology varies widely by geographic region, season, and climate, and is challenging to identify based on clinical features alone.² Confirmation of diagnoses with laboratory testing is essential, but many settings lack the necessary infrastructure.³ Acute febrile illness etiologies such as malaria and dengue, which carry high morbidity and mortality and are of considerable public health interest, thus become the focus of many diagnostic clinical laboratories.⁴ Other diseases implicated in AFI may consequently be underdiagnosed and inappropriately managed.²

The contribution of respiratory viruses to global AFI burden is not well described in the literature.¹ Influenza affects 10–20% of the world's population annually, and other viruses such as respiratory syncytial virus (RSV), parainfluenza virus (PIV), and human rhinovirus/enterovirus (HRV/HEV) as a group account for even greater annual morbidity and mortality.⁵ Respiratory illnesses account for most of the acute infections at all ages, but few AFI studies have incorporated respiratory viral testing into the diagnostic panel.⁶

In 2007, we determined the burden of AFI due to dengue, rickettsial infections, leptospirosis, and chikungunya among patients presenting to a tertiary care hospital in southern Sri Lanka.^{7–10} Respiratory viral testing was not performed and more than 50% of patients did not receive a laboratory-

confirmed diagnosis. In the present study, we repeated surveillance for AFI in southern Sri Lanka from 2012 to 2014. We determined the prevalence, seasonality, and clinical features of respiratory viral infection among patients admitted with AFI.

MATERIALS AND METHODS

Study population. We prospectively enrolled patients with AFI admitted to the pediatric and adult wards of the largest (1,500 bed) tertiary care hospital in the southern province of Sri Lanka from June 2012 to October 2014. Consecutive patients ≥ 1 year of age with documented fever ($> 38^{\circ}\text{C}$) at presentation or within 48 hours of hospital admission were eligible. We obtained epidemiological and clinical data and a nasopharyngeal sample at enrollment. Patients who did not tolerate nasopharyngeal sampling had a nasal sample collected instead. Details regarding patients' clinical management and outcomes during hospitalization were subsequently extracted from the medical record.

Respiratory pathogen molecular testing. The upper respiratory sample collected from each patient was placed in viral transport media and frozen at -80°C . The nasal or nasopharyngeal sample of alternate (i.e., every other) enrolled patient was selected for testing by real-time reverse transcription–polymerase chain reaction (PCR) with the Luminex Integrated System NxTAG Respiratory Pathogen Panel platform. If a patient did not have a nasal or nasopharyngeal specimen collected, the next alternate patient was selected for testing. The Luminex platform detects 19 respiratory viruses (RSV A and B; nonspecific influenza A; influenza A subtypes H1, H3, and 2009 H1N1; influenza B; parainfluenza 1–4; human metapneumovirus (hMPV); adenovirus; HRV/HEV; coronavirus types NL63, HKU1, 229E, and OC43; and human bocavirus)

* Address correspondence to L. Gayani Tillekeratne, Duke Global Health Institute, 310 Trent Dr., Durham, NC 27705. E-mail: gayani.tillekeratne@dm.duke.edu

† Please note that Megan E. Reller and Christopher W. Woods are co-senior authors.

and three bacteria (*Chlamydophila pneumoniae*, *Legionella pneumophila*, and *Mycoplasma pneumoniae*).¹¹

Statistical analysis. Seasonality of influenza and all respiratory viruses was determined using a definition from the Program for Appropriate Technology (PATH).¹² The monthly proportion of cases of influenza out of all positive cases of influenza for a given calendar year was calculated. A “peak” in activity was defined as the monthly proportion being $\geq 10\%$ during two consecutive years. If circulation varied from year to year (i.e., proportion $< 10\%$ per month), then a peak was not identified according to the definition. The PATH methodology was also used to determine seasonality of all respiratory viruses as a group.

Monthly weather data were obtained for the Galle region from the Sri Lanka Department of Meteorology.¹³ Correlation between weather data (monthly rainfall, monthly minimum temperature, monthly maximum temperature, daytime humidity, and nighttime humidity) and the monthly proportion of subjects with influenza or respiratory viruses was determined using the Spearman correlation.¹⁴

Sociodemographic and clinical characteristics of patients with a positive compared with a negative respiratory viral test were compared using the Fisher exact test for categorical variables and the Kruskal–Wallis test for continuous variables. Bivariable and multivariable logistic regressions were carried out to determine the association (odds ratios [OR] with 95% confidence intervals [CI]) between patients’ sociodemographic and clinical characteristics, and having a positive respiratory viral PCR test result. Tachypnea was defined as > 40 breaths/minute for children aged 1 to < 5 years, and > 20 breaths/minute for children aged ≥ 5 years and adults.¹⁵ Tachycardia was defined as > 180 beats/minute for children aged 1 to < 2 years, > 140 beats/minute for children aged 2 to < 6 years, > 130 beats/minute for children aged 6 to < 13 years, > 110 beats/minute for children aged 13 to < 18 years, and > 100 beats/minute for adults ≥ 18 years.¹⁶

For the multivariable analysis, an adjusted model was constructed to determine features associated with respiratory

viral infection. Any sociodemographic characteristic, clinical symptom, or examination finding that was associated with the dependent variable at a P -value < 0.25 on bivariable analysis was a candidate for inclusion in the multivariable model. Variables were checked for collinearity and nonlinear relationships before inclusion in the model. To create a more parsimonious model, Bayesian model averaging was performed.¹⁷ The multivariable model was constructed by adding the variables with the highest posterior inclusion probabilities until an effect was no longer significant. To check for confounding and moderation, excluded variables and interaction effects were then added into the final model sequentially.

All analyses were performed using STATA, version 11 (STATACorp, College Station, TX). Bayesian model averaging was performed using the BMA package in R, version 3.3.3 (Vienna, Austria).

Written informed consent was obtained from all patients and from parents or guardians of minors. The Institutional Review Boards of the University of Ruhuna Faculty of Medicine (Sri Lanka), Duke University Medical Center (Durham, NC), and Johns Hopkins University (Baltimore, MD) approved this study.

RESULTS

Study cohort and respiratory testing results. Of 1,954 patients enrolled from June 2012 to October 2014, 968 patients had upper respiratory samples collected and tested. Of these, four had an invalid respiratory test result and were excluded from further analyses. For the remaining 964 patients, the median age was 26.2 years (IQR 14.6–39.9 years) and 135 patients (14.0%) were aged < 5 years. The majority (646, 67.0%) were male. A total of 205 (21.3%) patients had an upper respiratory sample that tested positive for a respiratory organism, with all but two of the detected organisms being viruses (Table 1). The most commonly detected pathogen was influenza for both adults and children (134, 13.9% of all tested patients), with approximately two-thirds being influenza A and

TABLE 1
Organisms identified from the upper respiratory tract in patients admitted with acute febrile illness, southern Sri Lanka, 2012–2014

Virus type	Number (%) in all patients	Number (%) in patients aged < 5 years	Number (%) in patients aged ≥ 5 years	P -value
Influenza A	88 (9.1)	14 (10.8)	74 (8.9)	0.49
H1N1p2009	37 (3.8)	8 (6.2)	29 (3.5)	0.14
H3	44 (4.6)	6 (4.6)	38 (4.6)	0.98
Untypeable	7 (0.7)	–	7 (0.8)	0.29
Influenza B	46 (4.8)	7 (5.4)	39 (4.7)	0.73
Parainfluenza virus	13 (1.4)	11 (8.5)	2 (0.2)	< 0.001
Parainfluenza 1	3 (0.3)	2 (1.5)	1 (0.1)	0.007
Parainfluenza 2	2 (0.2)	2 (1.5)	–	< 0.001
Parainfluenza 3	8 (0.8)	7 (5.4)	1 (0.1)	< 0.001
Human rhinovirus/enterovirus	13 (1.4)	8 (6.2)	5 (0.6)	< 0.001
Human metapneumovirus	11 (1.1)	6 (4.6)	5 (0.6)	< 0.001
RSV	11 (1.1)	10 (7.7)	1 (0.1)	< 0.001
RSV A	6 (0.6)	6 (4.6)	–	< 0.001
RSV B	5 (0.5)	4 (3.1)	1 (0.1)	< 0.001
Bocavirus	7 (0.7)	4 (3.1)	3 (0.4)	< 0.001
Adenovirus	5 (0.5)	4 (3.1)	1 (0.1)	< 0.001
Coronavirus	2 (0.2)	–	1 (0.1)	0.69
<i>Mycoplasma pneumoniae</i>	1 (0.1)	–	1 (0.1)	0.69
<i>Chlamydophila pneumoniae</i>	1 (0.1)	–	1 (0.1)	0.69
Co-infections	7 (0.7)	4 (3.1)	3 (0.4)	< 0.001
Negative	759 (78.7)	62 (47.7)	696 (83.6)	< 0.001
Total	964	130	833	–

RSV = respiratory syncytial virus. $P < 0.15$ listed in bold.

the remainder being influenza B. Other viruses identified included HRV/HEV (1.4% of tested patients), PIV (1.4%), RSV (1.1%), hMPV (1.1%), and bocavirus (0.7%). Respiratory coinfections were identified in 0.7% of patients. Children aged < 5 years were more likely to have respiratory virus identified and to have a respiratory coinfection compared with patients aged ≥ 5 years (Table 1, Figure 1). Severity of disease in the tested patients was low, with five (0.5%) patients requiring intensive unit care, two (0.2%) requiring vasopressors, two (0.2%) requiring intubation, and seven (0.7%) patients dying. None of the patients with severe illness had a respiratory pathogen detected.

Seasonality of respiratory viruses and associated weather parameters. Respiratory viral activity peaked in February–June of both 2013 and 2014 (excluding the month of March; Figure 2A). Influenza A activity also peaked during these same months (Figure 2B), whereas influenza B activity peaked in July 2012 and 2013 (Figure 2C). Of weather parameters, only maximum daily temperature was correlated with either respiratory virus or influenza detection: $r = 0.42$ (95% CI: 0.05–0.68, $P = 0.03$) for all respiratory viruses and $r = 0.42$ (95% CI: 0.05–0.68, $P = 0.03$) for influenza. Rainfall ($r = -0.24$, 95% CI: -0.56 to 0.15 , $P = 0.22$), minimum daily temperature ($r = 0.29$, 95% CI: -0.10 to 0.60 , $P = 0.14$), day-time humidity ($r = -0.11$, 95% CI: -0.46 to 0.28 , $P = 0.59$), and nighttime humidity ($r = -0.14$, 95% CI: -0.49 to 0.25 , $P = 0.49$) were not correlated with respiratory virus activity. Rainfall ($r = -0.29$, 95% CI: -0.60 to 0.09 , $P = 0.13$), minimum daily temperature ($r = 0.23$, 95% CI: -0.16 to 0.55 , $P = 0.25$), day-time humidity percentage ($r = -0.21$, 95% CI: -0.54 to 0.17 , $P = 0.28$), and nighttime humidity percentage ($r = -0.19$, 95% CI: -0.53 to 0.20 , $P = 0.34$) were also not correlated with influenza activity.

Sociodemographic and clinical features associated with respiratory viral infection. Results of the bivariable

associations of sociodemographic characteristics, exposures, and clinical features with respiratory viral infection are shown in Table 2. The two patients with bacterial respiratory pathogens on testing were excluded from the analysis. Patients with respiratory virus identified were younger than patients with no respiratory virus (median age 9.8 versus 27.7 years, $P < 0.001$) and were more likely to report a sick contact within the prior 4 weeks (OR 1.51, 95% CI: 1.08–2.10). Clinical symptoms were different in patients with and without respiratory virus identified. Patients with respiratory virus had a shorter duration of fever (median 4 versus 5 days, $P < 0.001$) and were more likely to report respiratory symptoms such as rhinitis/congestion (OR 6.40, 95% CI: 4.54–9.03), cough (OR 4.80, 95% CI: 3.36–6.85), and shortness of breath (OR 1.91, 95% CI: 1.24–2.95). However, the duration of cough was shorter in patients who had respiratory virus identified than in those who did not. Patients with respiratory virus identified were also less likely to report nonspecific symptoms such as fatigue, joint pain, muscle pain, and headache. On physical examination, patients with respiratory virus were more likely to have crepitations (OR 3.15, 95% CI: 2.18–4.54) and rhonchi/wheezing (OR 5.63, 95% CI: 3.36–9.42) than patients with no respiratory virus identified. Other examination findings, such as hepatomegaly, tender liver, and rash, were significantly less common in patients with respiratory virus than those without respiratory virus.

After adjusting for other sociodemographic and clinical factors in the multivariable regression model, age < 5 years was associated with higher odds of respiratory viral infection (OR 3.62, 95% CI: 2.05–6.43) compared with age 5–65 years (Table 3). Increasing white blood cell count, diarrhea, and the presence of rash on examination were also negatively associated with respiratory viral infection. By contrast, respiratory signs and symptoms including cough, rhinitis/congestion,

FIGURE 1. Proportion with respiratory viruses identified, by age category, among patients hospitalized with acute febrile illness, southern Sri Lanka, 2012–2014.

FIGURE 2. Respiratory virus (A), influenza A (B), and influenza B (C) seasonality using the program for appropriate technology definition among patients hospitalized with acute febrile illness in southern Sri Lanka, 2012–2014.

and rhonchi/wheezing on examination were associated with having respiratory virus. Interestingly, among patients who had cough, increasing duration of cough was negatively associated with having respiratory virus (OR 0.83, 95% CI: 0.75–0.92).

Clinical management of patients with respiratory viral infection. Patients with respiratory virus identified were more likely to receive a clinical diagnosis of upper respiratory tract infection (OR 1.95, 95% CI: 1.05–3.62), lower respiratory tract infection (OR 3.60, 95% CI: 2.41–5.37), or unspecified viral

TABLE 2

Bivariable analysis of sociodemographic characteristics, exposures, and clinical features associated with respiratory virus detection among patients admitted with acute febrile illness, southern Sri Lanka, 2012–2014

Characteristic	Number (%) or median (IQR) in RVP+ (n = 203)	Number (%) or median (IQR) in RVP- (n = 761)	Odds ratio (95% confidence interval)	P-value
Sociodemographic characteristics and exposures				
Age (years)	9.8 (3.9–30.5)	27.7 (18.4–41.1)	0.97 (0.96, 0.98)	< 0.001
Male	125 (61.6)	521 (68.5)	0.74 (0.54–1.02)	0.06
Occupation (adults)				
Unemployed/retired	24 (27.0)	133 (22.8)	Reference	0.79
Laborer	32 (36.0)	221 (37.9)	0.80 (0.45, 1.42)	
Merchant/office	12 (13.5)	94 (16.1)	0.71 (0.34, 1.49)	
Other	16 (18.0)	98 (16.8)	0.91 (0.46, 1.79)	
Education ≥ 12th grade (adults)	30 (33.7)	209 (35.8)	0.91 (0.57–1.47)	0.71
Common hometown				
Galle	45 (22.2)	142 (18.7)	Reference	0.71
Akuressa	5 (2.5)	29 (3.8)	0.54 (0.20, 1.49)	
Ahangama	7 (3.5)	24 (3.2)	0.92 (0.37, 2.28)	
Wanduramba	8 (3.9)	37 (4.9)	0.68 (0.30, 1.57)	
Poddala	6 (3.0)	26 (3.4)	0.73 (0.28, 1.88)	
Sick contact in past 4 weeks	72 (35.1)	199 (26.3)	1.52 (1.08–2.14)	0.01
Travel in past 4 weeks	21 (10.3)	144 (18.9)	0.49 (0.30–0.80)	0.004
Rural residence	46 (22.7)	143 (18.8)	1.27 (0.87–1.84)	0.22
Passive or active smoking	40 (19.7)	222 (29.2)	0.60 (0.41–0.87)	0.008
Prior antibiotic use	60 (29.6)	194 (25.5)	0.93 (0.64, 1.33)	0.68
Clinical features				
Days of fever	4 (3–5)	5 (3–6)	0.88 (0.82, 0.94)	< 0.001
Rhinitis/congestion	103 (50.7)	106 (13.9)	6.40 (4.54–9.03)	< 0.001
Cough	153 (75.4)	299 (39.3)	4.80 (3.36–6.85)	< 0.001
Days of cough	4 (3–5)	4 (3–6)	1.13 (1.08, 1.18)	< 0.001
Sore throat	36 (17.7)	112 (14.7)	1.27 (0.84–1.91)	0.26
Shortness of breath	35 (17.2)	75 (9.9)	1.91 (1.24–2.95)	0.004
Pain with breathing	17 (8.4)	69 (9.1)	0.93 (0.53–1.62)	0.79
Decreased appetite	146 (71.9)	568 (74.6)	0.88 (0.62–1.24)	0.44
Vomiting	82 (40.4)	354 (46.5)	0.76 (0.56–1.05)	0.09
Diarrhea	18 (8.9)	133 (17.5)	0.46 (0.27–0.77)	0.003
Abdominal pain	31 (15.3)	156 (20.5)	0.71 (0.47–1.08)	0.11
Decreased urine output	12 (5.9)	60 (7.9)	0.73 (0.39–1.39)	0.34
Dysuria	7 (3.4)	78 (10.2)	0.31 (0.14–0.69)	0.004
Headache	112 (55.2)	548 (72.0)	0.48 (0.35–0.66)	< 0.001
Fatigue	100 (49.3)	470 (61.8)	0.60 (0.44–0.82)	0.002
Joint pain	68 (33.5)	437 (57.4)	0.38 (0.27–0.52)	< 0.001
Muscle pain	79 (38.9)	451 (59.3)	0.44 (0.32–0.61)	< 0.001
Admission temperature	102.0 (100.4–102.6)	102.0 (100.2–102.4)	1.01 (0.39–2.65)	0.99
Tachycardia on exam	60 (29.6%)	265 (34.9%)	0.80 (0.57–1.11)	0.15
Tachypnea on exam	77 (37.9%)	298 (39.2%)	0.95 (0.67, 1.34)	0.95
Oxygen saturation on exam	99 (98–100)	99 (98–100)	1.02 (0.94–1.10)	0.60
Conjunctival injection	18 (8.9)	135 (17.7)	0.45 (0.27–0.76)	0.003
Throat erythema	10 (4.9)	41 (5.4)	0.91 (0.45–1.85)	0.79
Enlarged lymph nodes	26 (12.8)	81 (10.6)	1.24 (0.77–1.99)	0.37
Crepitations on exam	64 (31.5)	97 (12.7)	3.15 (2.18–4.54)	< 0.001
Rhonchi/wheezing on exam	37 (18.2)	29 (3.8)	5.63 (3.36–9.42)	< 0.001
Enlarged liver on exam	5 (2.5)	47 (6.2)	0.38 (0.15–0.98)	0.04
Tender liver	2 (1.0)	50 (6.6)	0.14 (0.03–0.59)	0.007
Enlarged spleen	1 (0.5)	20 (2.6)	0.18 (0.02–1.37)	0.10
Rash on exam	9 (4.4)	102 (13.4)	0.30 (0.15–0.60)	< 0.001

RVP = respiratory virus positive. *P* < 0.15 listed in bold.

fever on admission (OR 1.42, 95% CI: 1.04–1.94; Table 4). However, patients with respiratory virus detected were equally likely as other patients to receive antibiotic therapy at enrollment (55.2% versus 52.6%), with the most common antibiotics prescribed being erythromycin, second-generation cephalosporins, and penicillins. Patients with respiratory virus detected had a higher leukocyte count within 48 hours of admission (7.7 versus 5.9×10^3 cells/ μ L) and were as likely as patients without respiratory virus to have a chest X-ray obtained during hospitalization (25.1% versus 24.8%) or to have an abnormality on chest X-ray (8.4% versus 7.6%). Patients with respiratory virus detected were hospitalized for a shorter duration (4 versus 5 days, *P* < 0.001) than patients

without respiratory virus. Two patients in the entire cohort reported a prior history of influenza vaccination, with both these patients testing negative for respiratory virus.

DISCUSSION

In this study, one-fifth of patients hospitalized with AFI in southern Sri Lanka had a respiratory virus identified. Definite seasonal variation in respiratory viral and influenza A activity was seen, with peak activity in February through June. Patients with respiratory virus isolated had less severe illness than patients with other causes of AFI, with fewer days of fever and shorter duration of hospitalization, and were more likely to

TABLE 3

Multivariable analysis of sociodemographic features, exposures, and clinical characteristics associated with respiratory virus detection among patients admitted with acute febrile illness, southern Sri Lanka, 2012–2014

Variable	Odds ratio (95% confidence interval)	P-values
Age < 5 years*	3.62 (2.05–6.43)	< 0.0001
Age > 65 years*	1.25 (0.47–3.03)	0.6430
White blood cell count (cells/uL)†	0.96 (0.92–0.99)	0.0165
Cough	5.88 (3.20–10.80)	< 0.0001
Cough × days of cough‡	0.84 (0.76–0.93)	0.0014
Rhinitis/congestion	3.16 (2.02–4.95)	< 0.0001
Diarrhea	0.51 (0.27–0.92)	0.0319
Joint pain	0.58 (0.38–0.87)	0.0083
Rhonchi/wheezing on exam	3.16 (1.63–6.16)	0.0007
Rash on exam	0.27 (0.11–0.59)	0.0025

* Reference category: age 5–65 years.

† Within 48 hours of admission; per 1-unit increase.

‡ Per 1-unit increase.

report respiratory signs and symptoms. More than half of the patients with respiratory viral illnesses were treated with antibiotics at enrollment, suggesting an inappropriate use of

antibiotics. In addition, no patients with respiratory virus reported a history of influenza vaccination, indicating a possible target for future public health intervention.

Few other studies have described the contribution of respiratory viruses to AFI, and most of these studies have focused on influenza alone or on outpatients, who generally have less severe disease. In the Asian-Pacific region, influenza A accounted for 11.8% of outpatients with AFI.¹⁸ In Vietnam, respiratory viruses accounted for 10% of AFI among outpatients, with PIV and influenza B being most common.¹⁹ In Ghana, Jones et al.²⁰ showed that influenza accounted for 6% of AFI among patients hospitalized at three hospitals, with most of the cases being in patients aged > 5 years. In a recent study of both outpatients and inpatients ≥ 1 year of age with AFI in Tanzania, influenza A was detected in 24% and HRV was detected in 25%.²¹ In our study, the overall prevalence of respiratory viral infection at 20% was generally higher than that previously reported, which may be secondary to a substantial proportion of our cohort consisting of children. More than half of the children aged < 5 years in our cohort had respiratory virus identified, although up to 20% of patients in other age groups also tested positive for respiratory viruses.

TABLE 4

Clinical management and outcomes of patients with and without respiratory virus isolated among a cohort with acute febrile illness, southern Sri Lanka, 2012–2014

Characteristic	Number (%) or median (IQR) in RVP+ (n = 203)	Number (%) or median (IQR) in RVP- (n = 761)	Odds ratio (95% confidence interval)	P-value
Clinical diagnosis at admission				
Dengue	21 (10.3)	226 (29.7)	0.27 (0.17–0.44)	< 0.001
Viral fever	98 (48.3)	301 (39.6)	1.42 (1.04–1.94)	0.03
Leptospirosis	8 (3.9)	103 (13.5)	0.26 (0.13–0.55)	< 0.001
Atypical pneumonia	4 (2.0)	6 (0.8)	2.53 (0.71–9.04)	0.15
Upper resp infection	16 (7.9)	32 (4.2)	1.95 (1.05–3.62)	0.04
Lower resp infection	53 (26.1)	68 (8.9)	3.60 (2.41–5.37)	< 0.001
Antibiotic therapy at enrollment	112 (55.2)	400 (52.6)	1.11 (0.81–1.52)	0.51
Penicillin	21 (10.3)	107 (14.1)	0.73 (0.45–1.21)	0.22
2nd generation cephalosporin	27 (13.3)	38 (5.0)	2.92 (1.74–4.91)	< 0.001
3rd generation cephalosporin	17 (8.4)	86 (11.3)	0.72 (0.42–1.24)	0.23
Erythromycin	37 (18.2)	86 (11.3)	1.75 (1.15, 2.67)	0.01
Doxycycline	2 (1.0)	59 (7.8)	0.12 (0.03–0.49)	0.003
Chest X-ray obtained	51 (25.1)	189 (24.8)	1.02 (0.71–1.45)	0.93
Chest X-ray abnormality	17 (8.4)	58 (7.6)	1.13 (0.58–2.18)	0.72
Alveolar	15 (7.4)	56 (7.4)	1.03 (0.52–2.04)	0.97
Interstitial	2 (1.0)	1 (0.1)	7.71 (0.68–87.52)	0.94
Multilobular involvement	5 (2.5)	5 (0.7)	4.00 (1.11, 14.40)	0.03
Effusion	1 (0.5)	8 (1.1)	0.45 (0.06–3.70)	0.46
White blood cell count (×10 ³ cells/μL)*	7.7 (5.9–10.5)	5.9 (3.6–10.2)	1.02 (0.99, 1.04)	0.09
Leukocytosis*	55 (27.1)	201 (26.4)	1.16 (0.81–1.65)	0.42
C-reactive protein performed*	51 (25.1)	150 (19.7)	1.37 (0.95–1.97)	0.09
High CRP (> 8 mg/L)*	34 (16.8)	114 (15.0)	1.14 (0.75–1.74)	0.53
Hospitalization duration	4 (3–5)	5 (4–6)	0.86 (0.79, 0.93)	< 0.001
Intensive unit care	0 (0)	5 (0.7)	n/a	0.98
Intubation	0 (0)	2 (0.3)	n/a	0.99
Supplemental oxygen use	1 (0.5)	6 (0.8)	0.62 (0.08–5.20)	0.66
Use of vasopressors	0 (0)	2 (0.3)	n/a	0.99
Clinical diagnosis at discharge				
Dengue	10 (4.9)	229 (30.1)	0.12 (0.06–0.23)	< 0.001
Viral fever	83 (40.9)	225 (29.6)	1.66 (1.21–2.29)	0.002
Leptospirosis	9 (4.4)	86 (11.3)	0.37 (0.18–0.74)	0.005
Atypical pneumonia	4 (2.0)	7 (0.9)	2.18 (0.63–7.51)	0.22
Upper resp infection	18 (8.9)	26 (3.4)	2.76 (1.48–5.14)	0.001
Lower resp infection	60 (29.6)	67 (8.8)	4.38 (2.96–6.48)	< 0.001
Antibiotics given at discharge				
Yes	47 (23.2)	155 (20.4)	1.13 (0.76–1.67)	0.68
Unsure	25 (12.3)	87 (11.4)	1.07 (0.65–1.75)	0.98
Death	0 (0)	7 (0.9)	n/a	0.98

n/a = not applicable; RVP = respiratory virus positivity. P-values in cells with 0 are calculated from Chi-square tests. P < 0.15 listed in bold.

* Within 48 hours of admission.

The burden of respiratory viral disease may be underappreciated in settings where testing is limited because clinical judgment alone may not suffice for differentiating between illnesses. For example, influenza A has previously been shown to present with hemorrhagic manifestations and thrombocytopenia, and may be confused with other illnesses such as dengue or leptospirosis.^{22,23}

In Sri Lanka, limited prior studies have identified the epidemiology of respiratory viral infection, and all of these studies have focused on pediatric populations. Among children aged ≤ 5 years hospitalized with respiratory illness in the Central and North Central provinces, $> 40\%$ had respiratory viruses detected, with RSV being most common and PIV, adenovirus, influenza, and hMPV also being present.²⁴ Among children aged ≤ 3 years hospitalized with respiratory illness in the Sabaragamuwa Province, 32% had respiratory virus, with 90% being RSV and the remainder being PIV and influenza.²⁵ Interestingly, in our study in the southern province, influenza was the most common respiratory virus identified, even among children aged < 5 years. This finding is likely because of our cohort including patients with AFI rather than solely respiratory illness. By performing surveillance over a 2-year period and including a large cohort of both children and adults, we were able to obtain a relatively comprehensive picture of the epidemiology of respiratory viral disease in this region. The World Health Organization (WHO) has highlighted the need for greater knowledge and interventions on viral respiratory diseases to reduce morbidity and mortality by initiating the Battle against Respiratory Viruses agenda.²⁶ Among other measures, this initiative recommends improving global surveillance for respiratory viruses and improving understanding of the geographical patterns, seasonality, and distribution of respiratory viruses.²⁶

In our study, both influenza A and respiratory viral activity peaked in February–June of 2013–2014. We have previously shown that among outpatients presenting with influenza-like illness in the Southern Province, influenza and respiratory viral activity peaked in March–June of 2013–2014, substantiating our present findings from the inpatient setting.²⁷ In tropical and subtropical countries such as Sri Lanka, studies have shown that the seasonality of respiratory viruses varies widely and may be impacted by local variations in temperature, humidity, and rainfall.^{28,29} Thus, even within a country, respiratory viral seasonality may fluctuate, and understanding local epidemiology is important for targeting preventative health measures.³⁰ Presently, less than 1% of the Sri Lankan population receives influenza vaccination and there is no national vaccination policy.³¹ Using data available through the WHO's FluNet from 2010 to 2015 and national surveillance data available from 2000 to 2014 (excluding 2009–2010), Hirve et al.¹² found that the primary influenza peak in Sri Lanka occurred in October–December, with a lesser peak in January–March. Hirve et al. recommended that the Northern Hemisphere vaccine formulation (available annually in October) be used in Sri Lanka, with targeted vaccination from October to December. However, our findings suggest that in the Southern Province of Sri Lanka, peak influenza activity is in February–June, and that the Southern Hemisphere formulation (available annually in April) may be important in providing protection. Enhanced local surveillance, typing of influenza strains, and correlation with vaccine formulation may help determine the best strategy for vaccine deployment in the country in the future.

Respiratory viral activity was associated with warmer temperatures in our study, which is contrary to the pattern generally seen in temperate regions.³² Associations of respiratory viral activity with temperature are often not observed in tropical and subtropical settings, and of meteorological parameters, relative humidity is most often correlated with respiratory viral activity in the literature.^{33–35} As with seasonality, associations between meteorological parameters and respiratory viral activity may vary by subregion within a country in tropical and subtropical settings.

Patients with respiratory viruses generally had lower severity of illness than other patients in our AFI cohort, with shorter durations of fever and hospitalization. Diagnoses such as dengue, leptospirosis, and enteric fever in the remainder of the cohort may account for such differences because these illnesses are traditionally associated with longer illness. The symptom of cough with non-respiratory illnesses such as dengue has been documented in other studies, and was present in more than a third of our patients who did not have a respiratory viral infection.³⁶ The duration of cough may be an important feature in helping a clinician to distinguish between respiratory viral versus other etiologies of AFI because patients with respiratory viruses generally had 7 days of cough or less in our study. This finding is in keeping with surveillance definitions for influenza-like illness, which limit cough duration to 7–10 days.^{37,38}

Patients with respiratory viral infection were as likely as other patients to receive chest X-rays and to have abnormalities on chest X-ray. However, more than 50% of patients who had respiratory virus identified were treated with antibiotics at enrollment. This group represents an important population that should be targeted for improved antimicrobial stewardship efforts. We have previously demonstrated that almost 80% of outpatients with respiratory viral illnesses in Sri Lanka receive antibiotic prescriptions.³⁹ Given the current global crisis in antimicrobial resistance, rapid and multiplex diagnostics that identify respiratory viruses may help reduce antimicrobial overuse in this and other settings.^{40,41} However, the cost and availability of such diagnostics may continue to be a barrier to access, and cost-effectiveness studies remain scarce.⁴²

Some limitations should be noted. The identification of respiratory virus from the nares or nasopharynx may represent passive colonization rather than infection, and we did not have an age-matched cohort without infectious symptoms as a control group. However, most patients with respiratory virus identified ($> 80\%$) had respiratory symptoms, and prior studies suggest that the isolation of influenza, which was the most commonly identified virus in our study, correlates with infection rather than colonization.^{21,43} We only recorded antibiotic use at enrollment and at hospital discharge, and did not track antiviral use during hospitalization. Assessment of antimicrobial use throughout the course of hospitalization may help better identify targets for future antimicrobial stewardship efforts. Finally, our cohort consisted of patients with acute undifferentiated febrile illness; patients who did not develop a fever or patients who had a focal pneumonia identified on examination or chest X-ray would thus have been excluded from this analysis.

In conclusion, respiratory viruses were a common and under-recognized cause of illness among patients hospitalized with AFI in southern Sri Lanka. An improved understanding

of the burden and seasonality of respiratory viruses may help inform preventative and clinical decisions such as vaccination and the use of antimicrobials.

Received August 26, 2018. Accepted for publication November 9, 2018.

Published online December 26, 2018.

Acknowledgments: We thank the research and laboratory staff who helped conduct this study.

Financial support: This work was supported by a grant from the National Institutes of Allergy and Infectious Diseases (K23AI125677 to LGT); Fogarty International Center and the National Institute of Mental Health (#R25 TW009337), the U.S. Department of Defense (BA150703 to CWW); the Office of Naval Research to the Emerging Infectious Diseases Programme, Duke-NUS Graduate Medical School, Singapore; a Johns Hopkins Center for Global Health Junior Faculty Grant (MER); and the Hubert-Young Center for Global Health.

Conflicts of Interest: Dr. Woods reports other relationships with Predigen; personal fees from IDbyDNA, Becton Dickinson, and Giner; grants and personal fees from bioMerieux; and grants from Pfizer, Biofire, Openbiome, Janus, BioMeme, MRI Global, and RTI, outside the submitted work.

Authors' addresses: L. Gayani Tillekeratne, Truls Østbye, Megan E. Reller, and Christopher W. Woods, Duke University, Durham, NC, Duke Global Health Institute, Durham, NC, and Duke-Ruhuna Collaborative Research Centre, Galle, Sri Lanka, E-mails: gayani.tillekeratne@dm.duke.edu, truls.ostbye@duke.edu, megan.reller@duke.edu, and chris.woods@duke.edu. Champica K. Bodinayake, Ajith Nagahawatte, and Vasantha Devasiri, Duke-Ruhuna Collaborative Research Centre, Galle, Sri Lanka, and Faculty of Medicine, University of Ruhuna, Galle, Sri Lanka, E-mails: bodinayake@gmail.com, ajithnagahawatte@yahoo.co.uk, and vdevasiri@gmail.com. Ryan Simmons, Duke Global Health Institute, Durham, NC, E-mail: ryan.simmons@duke.edu. Wasantha Kodikara Arachchi, Duke-Ruhuna Collaborative Research Centre, Galle, Sri Lanka, and Teaching Hospital Karapitiya, Galle, Sri Lanka, E-mail: kody@sltnet.lk. Bradley P. Nicholson and Sky Vanderburg, Duke University, Durham, NC, and Duke-Ruhuna Collaborative Research Centre, Galle, Sri Lanka, E-mails: brad.nicholson@duke.edu and sky.vanderburg@duke.edu. Larry P. Park, Duke University, Durham, NC, and Duke Global Health Institute, Durham, NC, E-mail: larry.park@duke.edu. Ruvini Kurukulasooriya, Duke-Ruhuna Collaborative Research Centre, Galle, Sri Lanka, E-mail: ruhunasearch@gmail.com. Aruna Dharshan De Silva, Duke-Ruhuna Collaborative Research Centre, Galle, Sri Lanka, and General Sir Kotelawala Defence University, Ratmalana, Sri Lanka, E-mail: dslv90@yahoo.com.

REFERENCES

1. Prasad N, Murdoch DR, Reyburn H, Crump JA, 2015. Etiology of severe febrile illness in low- and middle-income countries: a systematic review. *PLoS One* 10: e0127962.
2. Crump JA et al., 2013. Etiology of severe non-malaria febrile illness in northern Tanzania: a prospective cohort study. *PLoS Negl Trop Dis* 7: e2324.
3. Petti CA, Polage CR, Quinn TC, Ronald AR, Sande MA, 2006. Laboratory medicine in Africa: a barrier to effective health care. *Clin Infect Dis* 42: 377–382.
4. Moreira J, Bressan CS, Brasil P, Siqueira AM, 2018. Epidemiology of acute febrile illness in Latin America. *Clin Microbiol Infect* 24: 827–835.
5. Tang JW, Lam TT, Zaraket H, Lipkin WI, Drews SJ, Hatchette TF, Heraud JM, Koopmans MP, 2017. Global epidemiology of non-influenza RNA respiratory viruses: data gaps and a growing need for surveillance. *Lancet Infect Dis* 17: e320–e326.
6. Shek LP, Lee BW, 2003. Epidemiology and seasonality of respiratory tract virus infections in the tropics. *Paediatr Respir Rev* 4: 105–111.
7. Reller ME et al., 2013. Chikungunya as a cause of acute febrile illness in southern Sri Lanka. *PLoS One* 8: e82259.
8. Reller ME et al., 2012. Unsuspected dengue and acute febrile illness in rural and semi-urban southern Sri Lanka. *Emerg Infect Dis* 18: 256–263.
9. Reller ME, Bodinayake C, Nagahawatte A, Devasiri V, Kodikara Arachchi W, Strouse JJ, Flom JE, Dumler JS, Woods CW, 2011. Leptospirosis as frequent cause of acute febrile illness in southern Sri Lanka. *Emerg Infect Dis* 17: 1678–1684.
10. Reller ME et al., 2012. Unsuspected rickettsioses among patients with acute febrile illness, Sri Lanka, 2007. *Emerg Infect Dis* 18: 825–829.
11. Chen JH, Lam HY, Yip CC, Wong SC, Chan JF, Ma ES, Cheng VC, Tang BS, Yuen KY, 2016. Clinical evaluation of the new high-throughput luminex NxTAG respiratory pathogen panel assay for multiplex respiratory pathogen detection. *J Clin Microbiol* 54: 1820–1825.
12. Hirve S, Newman LP, Paget J, Azziz-Baumgartner E, Fitzner J, Bhat N, Vandemaale K, Zhang W, 2016. Influenza seasonality in the tropics and subtropics—when to vaccinate? *PLoS One* 11: e0153003.
13. Department of Meteorology SL, 2016. *Meteorological Data*. Colombo, Sri Lanka: Department of Meteorology.
14. Minh AN DT, Ngoc NT, Nilsson M, 2014. Influenza-like illness in a Vietnamese province: epidemiology in correlation with weather factors and determinants from the surveillance system. *Glob Health Action* 7: 23073.
15. Jain S et al.; CDC EPIC Study Team, 2015. Community-acquired pneumonia requiring hospitalization among U.S. children. *N Engl J Med* 372: 835–845.
16. Goldstein B, Giroir B, Randolph A, 2005. International pediatric sepsis consensus conference: definitions for sepsis and organ dysfunction in pediatrics. *Pediatr Crit Care Med* 6: 2–8.
17. Viallefont V, Raftery AE, Richardson S, 2001. Variable selection and Bayesian model averaging in case-control studies. *Stat Med* 20: 3215–3230.
18. Capeding MR et al., 2013. Dengue and other common causes of acute febrile illness in Asia: an active surveillance study in children. *PLoS Negl Trop Dis* 7: e2331.
19. Phuong HL et al., 2010. Viral respiratory tract infections among patients with acute undifferentiated fever in Vietnam. *Southeast Asian J Trop Med Public Health* 41: 1116–1126.
20. Jones AH et al., 2016. Sentinel surveillance for influenza among severe acute respiratory infection and acute febrile illness inpatients at three hospitals in Ghana. *Influenza Other Respir Viruses* 10: 367–374.
21. Hercik C et al., 2017. A diagnostic and epidemiologic investigation of acute febrile illness (AFI) in Kilombero, Tanzania. *PLoS One* 12: e0189712.
22. Kuberski TT, 1977. Haemorrhage associated with influenza. *Lancet* 1: 709.
23. Lorenzi OD et al., 2013. Acute febrile illness surveillance in a tertiary hospital emergency department: comparison of influenza and dengue virus infections. *Am J Trop Med Hyg* 88: 472–480.
24. Jayaweera J, Noordeen F, Morel A, Pitchai N, Kothalawala S, Abeykoon A, Peiris J, 2016. Viral burden in acute respiratory tract infections in hospitalized children in the wet and dry zones of Sri Lanka. *Int J Infect Dis* 45S: 1–477.
25. Muthulingam A, Noordeen F, Morel A, 2014. Viral etiology in hospitalized children with acute respiratory tract infection in the Kegalle area of Sri Lanka. *J Pediatr Infect Dis* 9: 167–170.
26. World Health Organization, 2013. *Research Needs for the Battle against Respiratory Viruses (BRaVe)*. Available at: http://www.who.int/influenza/patient_care/clinical/BRaVe_Research_Agenda_2013.pdf?ua=1. Accessed March 5, 2018.
27. Shapiro D et al., 2017. Burden and seasonality of viral acute respiratory tract infections among outpatients in southern Sri Lanka. *Am J Trop Med Hyg* 97: 88–96.
28. Breiman RF, Van Beneden CA, Farnon EC, 2013. Surveillance for respiratory infections in low- and middle-income countries: experience from the Centers for Disease Control and Prevention's Global Disease Detection International Emerging Infections Program. *J Infect Dis* 208 (Suppl 3): S167–S172.
29. Peasah SK, Azziz-Baumgartner E, Breese J, Meltzer MI, Widdowson MA, 2013. Influenza cost and cost-effectiveness studies globally—a review. *Vaccine* 31: 5339–5348.

30. Newman LP, Bhat N, Fleming JA, Neuzil KM, 2018. Global influenza seasonality to inform country-level vaccine programs: an analysis of WHO FluNet influenza surveillance data between 2011 and 2016. *PLoS One* 13: e0193263.
31. Hirve S, Lambach P, Paget J, Vandemaële K, Fitzner J, Zhang W, 2016. Seasonal influenza vaccine policy, use and effectiveness in the tropics and subtropics—a systematic literature review. *Influenza Other Respir Viruses* 10: 254–267.
32. Azziz Baumgartner E et al., 2012. Seasonality, timing, and climate drivers of influenza activity worldwide. *J Infect Dis* 206: 838–846.
33. Soebiyanto RP et al., 2015. Associations between seasonal influenza and meteorological parameters in Costa Rica, Honduras and Nicaragua. *Geospat Health* 10: 372.
34. Munshili Njifon HL et al., 2018. Influence of meteorological parameters in the seasonality of influenza viruses circulating in northern Cameroon. *Influenza Other Respir Viruses*, doi:10.1111/irv.12612.
35. Soebiyanto RP et al., 2014. The role of temperature and humidity on seasonal influenza in tropical areas: Guatemala, El Salvador and Panama, 2008–2013. *PLoS One* 9: e100659.
36. Dietz V, Gubler DJ, Ortiz S, Kuno G, Casta-Velez A, Sather GE, Gomez I, Vergne E, 1996. The 1986 dengue and dengue hemorrhagic fever epidemic in Puerto Rico: epidemiologic and clinical observations. *P R Health Sci J* 15: 201–210.
37. World Health Organization, 2012. *WHO Interim Global Epidemiological Surveillance Standards for Influenza*. Geneva, Switzerland: World Health Organization.
38. Fitzner J et al., 2018. Revision of clinical case definitions: influenza-like illness and severe acute respiratory infection. *Bull World Health Organ* 96: 122–128.
39. Tillekeratne L et al., 2015. An under-recognized influenza epidemic identified by rapid influenza testing, southern Sri Lanka, 2013. *Am J Trop Med Hyg* 92: 1023–1029.
40. World Health Organization, 2014. *Antimicrobial Resistance—Global Report on Surveillance*. Geneva, Switzerland: World Health Organization.
41. Rappo U, Schuetz AN, Jenkins SG, Calfee DP, Walsh TJ, Wells MT, Hollenberg JP, Glesby MJ, 2016. Impact of early detection of respiratory viruses by multiplex PCR assay on clinical outcomes in adult patients. *J Clin Microbiol* 54: 2096–2103.
42. Vallieres E, Renaud C, 2013. Clinical and economical impact of multiplex respiratory virus assays. *Diagn Microbiol Infect Dis* 76: 255–261.
43. Robinson J, 2004. Colonization and infection of the respiratory tract: what do we know? *Paediatr Child Health* 9: 21–24.