Supplementary Appendix

This appendix has been provided by the authors to give readers additional information about their work.

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WEB-ONLY SUPPLEMENT

The Burden of Human Metapneumovirus Infection in Young Children

Short title: HMPV infection in children

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The study was designed in collaboration between the CDC and the Principal Investigators at each site. Data were electronically transferred to a central database, where they were analyzed by the investigators. All the

authors vouch for the accuracy and completeness of the data. The manuscript was drafted by the first author and last author, with input from all of the other investigators listed as authors.

METHODS

Study design and methods

The Centers for Disease Control and Prevention (CDC) initiated the New Vaccine Surveillance Network (NVSN), a prospective, population-based inpatient and outpatient surveillance system enrolling children <5 years old with ARI or fever beginning in 2000 in Nashville and Rochester, and in 2003 in Cincinnati[1-4]. The NVSN surveillance study ended in 2009. Inpatient specimens collected in Cincinnati between 2005-2008 seasons were excluded due to protocol deviations but outpatient samples were retained; inpatient and outpatient samples from Rochester for the 2008-2009 year were not included due to inadequate sample amounts. Nasal and throat swabs were obtained for viral detection by the research team.

Children < 5 years old with ARI or fever were enrolled within 48 hours of hospital admission from Sunday through Thursday; outpatient surveillance was conducted concurrently. Outpatients were enrolled from 1 to 4 urban and suburban pediatric practices either 1 or 2 days per week. ED subjects were enrolled at Nashville and Rochester during daytime and nighttime shifts 3 or 4 days per week; and at Cincinnati every fourth day.

Each subject was enrolled and tested for HMPV in only one clinical setting. If subjects were enrolled in the clinic or ED but later were admitted on non-surveillance days (and thus not enrolled as inpatients), they were kept as clinic or ED visits. If children were enrolled in the clinic or ED but were admitted within 6 days later on a surveillance day, they were re-categorized as inpatients. If children sought care in the ED on a surveillance day within 4 days of the preceding clinic visit, they were considered ED patients.

There were 283 children who were enrolled more than once during the course of the study; of these, 244 children tested negative and 39 children tested positive for HMPV. No child tested HMPV+ more than once.

RESULTS

Other viruses tested among inpatients

The primary NVSN study was funded for influenza surveillance in pediatric populations, and thus other pathogens were not systematically tested. However, other viruses were tested in some years and some settings, and in some cases, population-based rates of hospitalization calculated. Influenza inpatient and outpatient rates were published for NVSN study years 2000-2004[3]. Additional testing and population-based rate estimates have been performed for RSV inpatient and outpatient study years 2000-2004[1]; parainfluenza viruses (PIV) inpatient study years 2000-2004[4]; coronaviruses (HCoV) inpatient study years 2001-2003[5]; and human rhinovirus (HRV) inpatient study years 2001-2003[6]. Thus, not all years tested for different viruses overlap between these different substudies. **Supplemental Table 1** shows the estimated population-based rates of hospitalization by age group for each of these viruses. For clarity of the Table, 95% confidence intervals are not shown.

Two additional NVSN studies reported HCoV inpatient and healthy controls in study years 2003-2005[7] and HRV inpatient and healthy controls in study years 2003-2005[8]. These studies did not calculate population-based rates of hospitalization, but reported the rate of viral detection in the two groups. HCoV was detected in 113 (7.6%) of 1481 hospitalized children and 53 (7.1%) of 742 controls, and thus the prevalence of HCoV was not significantly higher among hospitalized children than controls. Overall, HRV was detected in 226 (14.9%) of hospitalized children <5 years and 99 (12.5%) of control children <5 years and thus not significantly different. However, the HRV-A detection rate among children ≥24 months was 8.1% in hospitalized and 2.2% in control children (p=0.009), and the HRV-C detection rate in children ≥6 months were 8.2% and 3.9%, respectively (p=0.002). Therefore, while HCoV and HRV are respiratory pathogens, these viruses are also frequently detected in asymptomatic children, complicating the attribution of causation in some cases[9].

Supplemental Table 1. Rates of hospitalization for children <5 years old (per 1000 children) associated with all viruses tested during different years of the NVSN study. For clarity, 95% confidence intervals are not shown.

	Incidence of hospitalization per 1000 children <5 years old							
Study years	2003-2009	2000-2004	2000-2004	2000-2004	2000-2004	2000-2004	2001-2003	2001-2003
Age group (months)	**HMPV**	**RSV**	**Influenza#**	**PIV1**	**PIV2**	**PIV3**	**HCoV***	**HRV**
<6	3.0	16.9	4.5	1.0	0.4	1.6	1.0	7.2
6-11	2.0	5.1	0.9	0.6	0.2	1.0	0.4	2.6
12-23	1.2	2.7		0.6	0.2	0.7		
24-59	0.5	0.4	0.3	0.2	0.1	0.2	0	1.2
0-59	**1.0**	**3.0**	**0.9**	**0.4**	**0.1**	**0.5**	**2.3**	**2.2**
Reference	This study	[1]	[3]	[4]			[5]	[6]

Includes influenza A and B

* Includes HCoV HKU1, NL63, OC43, 229E

Other viruses tested among outpatients

Other viruses were tested in some years for the outpatient settings and population-based visit rates calculated. These include RSV inpatient and outpatient study years 2000-2004[1]; and influenza inpatient and outpatient rates for NVSN study years 2000-2004[3]. Supplemental Table 2 shows the estimated population-based rates of clinic and ED visits by age group for each of these viruses. For clarity, 95% confidence intervals are not shown.

Supplemental Table 2. Rates of outpatient visits associated with viruses tested during different years of the NVSN study.

	Rate of outpatient visits per 1000 children <5 years old			
Study years	2003-2009	2002-2004	2002-2003	2003-2004
Pediatric clinics				
Age group (months)	**HMPV**	**RSV**	**Influenza#**	
<6	58	132	28	59
6-11	102	177	52	125
12-23	75	66		
24-59	39	57	53	88
0-59	**55**	**80**	**50**	**95**
Emergency department				
<6	16	55	0	23
6-11	29	57	8	39
12-23	17	32		
24-59	9	13	7	23
0-59	**13**	**28**	**6**	**27**
Reference	This study	[1]	[3]	

Includes influenza A and B

Other viruses detected in children with HMPV infection

Other viruses were detected in 21 (10%) of HMPV-positive hospitalized children and 27 (6%) of HMPV-positive outpatient children. The severity of illness was not different in children with more than one virus, but the numbers were small (not shown).

Supplemental Table 3. Co-detection of other viruses in patients with HMPV.

HMPV+	RSV+	Influenza+	PIV+
Number (%)			
Inpatient			
21 (10)	13 (6)	6 (3)	2 (1)
Outpatient			
27 (6)	14 (4)	11 (2)	2 (1)

Supplemental Table 4. Discharge diagnoses for children with and those without HMPV infection.

Variable	Inpatients		Outpatients	
	HMPV positive (n=200)	HMPV negative (n=3290)	HMPV positive (n=446)	HMPV negative (n=5812)
Discharge diagnosis				
Pneumonia	99/200 (50)	731/3290 (22)	39/446 (9)	259/5812 (4)
Bronchiolitis	44/200 (22)	987/3290 (30)	46/446 (10)	544/5812 (9)
Asthma	29/200 (14)	419/3290 (13)	62/446 (14)	530/5812 (9)
Viral illness	11/200 (6)	391/3290 (12)	152/446 (34)	2058/5812 (35)
Fever	8/200 (4)	294/3290 (9)	19/446 (4)	314/5812 (5)
Influenza	0/200 (0)	59/3290 (2)	4/446 (1)	77/5812 (1)
Seizure	1/200 (0)	52/3290 (2)	2/446 (0)	15/5812 (0)
Bacteremia/septicemia	2/200 (1)	39/3290 (1)	0/446 (0)	3/5812 (0)
Pharyngitis	0/200 (0)	13/3290 (0)	13/446 (3)	206/5812 (4)
Sinusitis	1/200 (0)	4/3290 (0)	5/446 (1)	74/5812 (1)

Diagnosis at discharge was reported on the basis of up to 10 discharge diagnoses, ordered by pneumonia, bronchiolitis, asthma, viral illness (including nonspecific viral illness, croup, respiratory syncytial virus infection, and upper respiratory infection), fever, influenza, seizure, bacteremia or septicemia, pharyngitis, and sinusitis.

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