MAJOR ARTICLE

Human Rhinovirus Species Associated With Hospitalizations for Acute Respiratory Illness in Young US Children

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Background. The contribution of human rhinovirus (HRV) to severe acute respiratory illness (ARI) is unclear.

Objective. To assess the association between HRV species detection and ARI hospitalizations.

Methods. Children <5 years old hospitalized for ARI were prospectively enrolled between December 2003 and April 2005 in 3 US counties. Asymptomatic controls were enrolled between December 2003 and March 2004 and between October 2004 and April 2005 in clinics. Nasal and throat swab samples were tested for HRV and other viruses (ie, respiratory syncytial virus, human metapneumovirus, parainfluenza virus, and influenza virus) by reverse-transcription–polymerase chain reaction, and genetic sequencing identified HRV species and types. HRV species detection was compared between controls and patients hospitalized during months in which controls were enrolled.

Results. A total of 1867 children with 1947 ARI hospitalizations and 784 controls with 790 clinic visits were enrolled and tested for HRV. The HRV-A detection rate among participants ≥24 months old was 8.1% in the hospitalized group and 2.2% in the control group ($P = .009$), and the HRV-C detection rates among those ≥6 months old were 8.2% and 3.9%, respectively ($P = .002$); among younger children, the detection rates for both species were similar between groups. The HRV-B detection rate was ≤1%. A broad diversity of HRV types was observed in both groups. Clinical presentations were similar among HRV species. Compared with children infected with other viruses, children with HRV detected were similar for severe hospital outcomes and more commonly had histories or diagnoses of asthma or wheezing.

Conclusions. HRV-A and HRV-C were associated with ARI hospitalization and serious illness outcomes.

Human rhinoviruses (HRVs), which are prime causes of the common cold, have been more frequently detected in upper and lower respiratory tract infections with the use of modern molecular methods. A causal role of HRV in lower respiratory tract infection is supported by isolation of HRV from the lower respiratory tract of infants and older persons [1–4]. Recent studies suggest differences in illness severity among HRV species [5, 6] and a more evident association between asthma exacerbation and HRV species C (HRV-C), compared with other HRV species (ie, HRV-A and HRV-B) [6–9].

Although reverse-transcription polymerase chain reaction (RT-PCR) enables detection of HRV RNA, including HRV-C, which has not been grown in standard tissue culture, HRV RNA has been detected in children for prolonged periods, even after symptoms have resolved [10–13], and is possibly more prolonged for asthma patients [14, 15]. Thus, detection may not reflect a current illness. To assess the contribution of HRV to severe respiratory illness, several studies compared HRV

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detection among hospitalized patients [5, 12, 16, 17] or outpatients with lower respiratory tract illness [6, 18–20] to HRV detection among controls without respiratory symptoms; most but not all studies noted higher HRV detection among participants with illness. The study populations, severity of cases, age, comorbidities, and choice of controls varied by study, with HRV detection rates for controls ranging widely, from 3% to 32%. Only 1 US study, involving Alaskan children, compared HRV detection between hospitalized cases and asymptomatic controls and did not find significant differences [17].

Our study aimed to delineate the association of HRV-A, HRV-B, and HRV-C with serious illness and to compare detection rates between hospitalized children with severe acute respiratory illness (ARI) and asymptomatic control children <5 years old in 3 large geographic areas of the United States.

METHODS

Study Design

Overview

The goal of this study was to determine whether HRV was associated with hospitalizations for ARI or fever (collectively referred to hereafter as ARI) among a large number of children <5 years of age who resided in 3 geographic areas in the United States. The primary objective was to compare the HRV species detection rates between 2 groups: hospitalized ARI cases and asymptomatic controls. The secondary objective was to compare the clinical severity of hospitalized ARI cases with only HRV to those only with viruses (ie, respiratory syncytial virus [RSV], parainfluenza virus types 1-3 [PIV], human metapneumovirus [HMPV], and influenza A and B viruses [influenza]) known to be associated with serious illness requiring hospitalization.

The design and methods of the New Vaccine Surveillance Network for ARI hospitalizations have been described elsewhere [21, 22]. The study was approved by the institutional review boards of each site and the Centers for Disease Control and Prevention (CDC). Informed consent was obtained from each parent or guardian prior to enrollment.

Hospitalized Group

Study staff prospectively enrolled children <5 years of age admitted to study hospitals in 3 counties (encompassing Rochester, NY; Nashville, TN; and Cincinnati, OH) from October 2003 to April 2005 who had an ARI admission diagnosis. Hospital surveillance was conducted 4 days/week, except during the 2004–2005 influenza season, when it was conducted 7 days/week. Surveillance was population based, with study hospitals accounting for >95% of the pediatric hospitalizations in each county. Children were excluded if they had been previously hospitalized during the prior 4 days, were newborns hospitalized since birth, had neutropenia from chemotherapy, or had been ill for >14 days before hospitalization.

Control Group

Children <5 years of age who resided in one of the 3 study counties were systematically enrolled as controls at well-child visits in 8–10 primary care practices if they did not have current ARI symptoms determined by questioning the family. Enrollment was conducted 1–2 days/week during December 2003–April 2004 and October 2004–April 2005, as described elsewhere [23]. Enrolled control children were excluded from analysis if they subsequently had ARI signs noted in the medical record for that visit.

Specimen and Data Collection

After obtaining informed consent, study staff collected a nasal swab specimen (both nares were sampled to the level of nasal turbinates) and throat (ie, tonsillopharyngeal) swab specimen and combined them in transport media. Study staff also conducted interviews with parents or guardians and performed medical record reviews to obtain demographic and clinical data.

A preexisting high-risk medical condition was considered present if noted in the medical record or if the parent or guardian responded that a healthcare professional told them the child had the condition. High-risk medical conditions corresponded to those included in the Advisory Committee on Immunization Practices recommendations for influenza vaccine, including history of cancer; diabetes mellitus; sickle cell disease; immunodeficiency; disease of the heart, kidney, or lung (including asthma); and neurologic/neuromuscular conditions, such as seizures, cerebral palsy, or muscular dystrophy [24]. History of asthma or wheezing included a diagnosis of asthma, reactive airway disease, or recurrent or chronic wheezing. We defined 3 clinical variables related to wheezing: (1) acute wheezing, which includes an admission or discharge diagnosis of asthma, bronchiolitis, or wheezing, consistent with our previous study [9]; (2) a discharge diagnosis of asthma (defined as *International Classification of Diseases, Ninth Revision,* code 493.xx); and (3) a first-listed discharge diagnosis of asthma (the likely reason for hospitalization).

Laboratory Methods

Aliquots of combined nasal and throat swab specimens were stored at −70°C and tested at the CDC for HRV by RT-PCR, using primers and probes that targeted the highly conserved HRV 5′ noncoding region (5′NCR) that can detect all 100 prototype HRVs and the novel species C strains [25]. To determine HRV species (HRV-A, HRV-B, and HRV-C) and type, RT-PCR was performed on all HRV-positive specimens, using other primers to amplify and sequence a partial region of the VP1 gene (method available on request). Specimens that were VP1 negative were further tested using primers that amplified partial VP4/VP2 or 5′NCR regions [26], resulting in identification of 74% of strains by partial VP1 sequencing, 18% by VP4/2, and 3% by 5′NCR. Specimens were also tested by RT-PCR for RSV, PIV, HMPV, and influenza A and B viruses.

Table 1. Study Enrollment and Human Rhinovirus (HRV) Detection Among Hospitalizations With Acute Respiratory Infection (ARI) and Asymptomatic Controls, by Age and Study Period

HRV Species[b]	Detected October 2003–April 2005, no. (%)	Detected during 12 mo in which controls were enrolled, no. (%)[a]							
		Age 0–59 mo		Age 0–5 mo		Age 6–23 mo		24–59 mo	
	ARI hospitalizations (n = 1947)	ARI hospitalizations (n = 1515)	Controls (n = 790)	ARI hospitalizations (n = 725)	Controls (n = 272)	ARI hospitalizations (n = 493)	Controls (n = 339)	ARI hospitalizations (n = 297)	Controls (n = 179)
Any	339 (17.4)	226 (14.9)	99 (12.5)	…	…	…	…	…	…
HRV-A	167 (8.6)	113 (7.5)	52 (6.6)	48 (6.6)	19 (7.0)	41 (8.3)	29 (8.6)	24 (8.1)[c]	4 (2.2)
HRV-B	29 (1.5)	15 (1.0)	4 (0.5)	6 (0.8)	0	6 (1.2)	2 (0.6)	3 (1.0)	2 (1.1)
HRV-C	136 (7.0)	93 (6.1)	32 (4.1)	28 (3.9)	12 (4.4)	37 (7.5)[c]	13 (3.8)	28 (9.4)[c]	7 (3.9)

[a] Data are for December 2003–April 2004 and October 2004–April 2005.

[b] Among HRV-positive hospitalizations, 4 had 2 HRV species codetected, 1 had 2 HRV-A types codetected, species was indeterminate in 11, and HRV type was indeterminate in 15. Among HRV-positive controls, 1 had 2 HRV species codetected, 1 had 2 HRV-A types codetected, species was indeterminate in 12, and HRV type was indeterminate in 5.

[c] $P < .05$ for comparison between hospitalized and control children.

Data Analysis

Hospitalizations and control visits occurring <30 days after a child's previous enrollment were excluded to remove multiple enrollments during what might have been the same illness episode and to be consistent with our previous study [9]. The entire 19-month study period was included to describe the seasonality, diversity of HRV types, and clinical course of HRV. However, comparisons of hospitalized and control children were restricted to the 12 months during which controls were enrolled.

A 2-sided .05 α level was applied for all statistical tests. SAS (SAS Institute) and SUDAAN software was used for all analyses [27]. Medians, interquartile ranges, and Wilcoxon rank sum tests were used for continuous data. Pearson χ^2 or Fisher exact tests were used for categorical data.

Comparisons of HRV Detection Between Hospitalized and Control Children (Restricted to Months With Controls) Crude unadjusted proportions of hospitalized children and controls who were positive for HRV-A, HRV-B, and HRV-C were calculated separately by age group (0–5, 6–23, and 24-59 months, specified a priori); exact tests were used to test bivariate associations between HRV species and study group. Multivariable logistic regression models were constructed for each of the following outcomes to estimate their association with study group: (1) HRV-A–positive versus HRV-A–negative children; (2) HRV-C–positive versus HRV-C–negative children; and (3) any HRV-positive versus HRV-negative children. Models were not fit for HRV-B because of small numbers. Models included terms for study group and age group, conditional on county and month of hospitalization or visit, to obtain adjusted odds ratios (ORs) and 95% confidence intervals (CIs) as estimates of relative risk. Candidate covariates in each model for HRV species were insurance status (ie, private vs other), race/ethnicity (ie, non-Hispanic white vs other), exposure to another child (ie, lives with another child or attends day care vs does neither), and living with a smoker, if they were statistically significant in bivariate analyses with the outcome.

We evaluated the effect of concurrent detections of other viruses that might have caused the hospitalization instead of HRV, by refitting HRV-A and HRV-C models and excluding the codetected cases and retaining the codetected cases by classifying the latter as HRV negative. Multiple enrollments ≥30 days apart for a single child were included in final analyses because the numbers were small and did not change the results or conclusions appreciably. This was determined by refitting final models and retaining only the first enrollment for each child, as well as by incorporating a correlation structure between multiple enrollments, which was less parsimonious and more complex (data not shown).

RESULTS

Study Population

The overall study enrollment included patients associated with 2082 ARI hospitalizations and asymptomatic controls associated with 861 clinic visits. The enrollment rate was 83% for hospitalized children and 90% for control children. The main reason for nonenrollment among hospitalized children (75%) and controls (87%) was parental refusal or unavailability. Of those enrolled, sufficient specimens were obtained from 95% of hospitalizations and 96% of controls. Among the enrolled children with available specimens, excluded were 1% of hospitalizations and 0.1% of control visits that were <30 days after a prior enrollment and 4% of control visits with signs of ARI noted in the medical record. Thus, the study included 1867 children with 1947 ARI hospitalizations during the 19-month study period, and 1473 children with 1515 ARI hospitalizations and 784

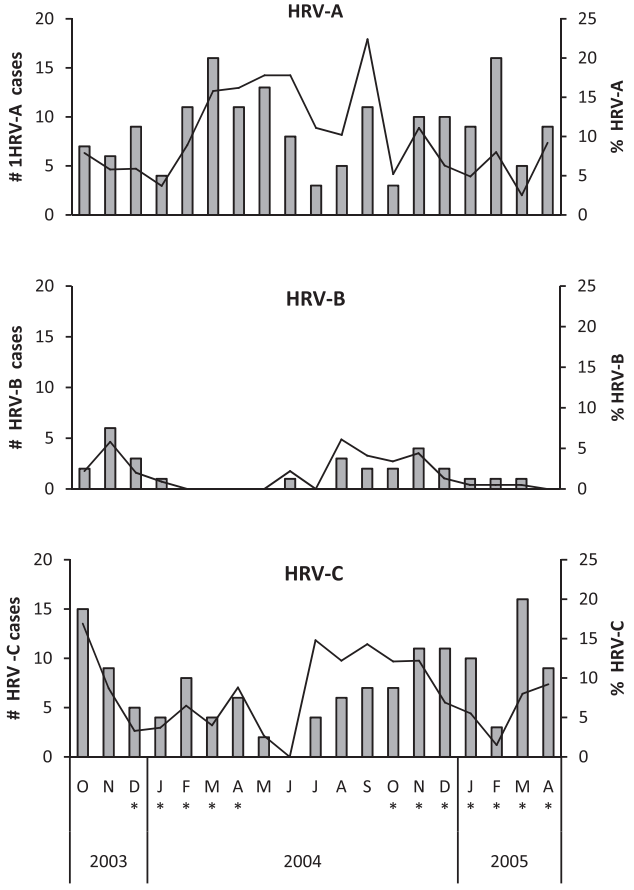

Figure 1. Number and percentage of ARI hospitalizations with human rhinovirus (HRV) A, HRV-B, and HRV-C, by month. Bars denote number of enrolled HRV cases; lines, percentage of hospitalizations that are HRV positive hospitalizations; asterisks, months in which controls were enrolled.

control children with 790 visits during the 12 months when controls were enrolled. Comparisons between hospitalized and control children were restricted to these 12 months.

Frequency of HRV Detections

During the 19-month study period, 339 (17.4%) of 1947 ARI hospitalizations were HRV positive; the number and proportions by HRV species are shown in Table 1, with HRV hospitalizations occurring in most study months (Figure 1). Multiple HRVs were codetected in 5 hospitalized children and 2 controls.

During the 12 months in which controls were enrolled, 226 (14.9%) of 1515 ARI hospitalizations and 99 (12.5%) of 790 controls were HRV positive (Table 1). Two percent of hospitalized children had multiple enrollments, including 5 with HRV (ie, 2 children with 2 HRV-C infections >6 months apart and 3 children with different HRV species or types). Among controls, 0.6% had multiple enrollments, and none had >1 visit during which HRV was detected.

Viruses other than HRV were codetected in a higher proportion of HRV-positive hospitalizations, compared with HRV-positive controls (18% [40/226] vs 6% [6/99]; $P = .006$). Among HRV-positive hospitalizations, RSV was codetected in 10%, PIV in 2%, HMPV in 4%, and influenza virus in 3%, whereas among HRV-positive controls, RSV was codetected in 3%, PIV in 1%, HMPV in 2%, and influenza virus in none.

Demographic Characteristics and Clinical History

Age and clinical characteristics of participants during the 12 months in which controls were enrolled are shown in Table 2. Hospitalized children with HRV-A were significantly older than controls with HRV-A ($P = .02$). History of high-risk conditions was significantly more common among hospitalized children with HRV-A or HRV-C, compared with control children with HRV-A or HRV-C ($P \leq .005$). More than 60% of hospitalized children ≥12 months old had a history of asthma or wheezing for all HRV species. Premature birth was significantly more common among hospitalized children with HRV-A, compared with control children with HRV-A ($P = .01$).

Compared with controls with HRV, hospitalized children with HRV were more likely to be privately insured (33% vs 22%; $P = .047$) or exposed to another child at home or day care (87% vs 71%; $P < .001$), but the groups did not significantly differ by sex, race/ethnicity, or living with a smoker.

Comparison of HRV Species Detection Between Hospitalized and Control Children

HRV detection rates differed between the hospitalized and control groups by age and species, as shown in Table 1.

HRV-A

Crude rates of HRV-A detection were similar between the 2 study groups among children <24 months old (7.3% in the hospitalized group vs 7.9% in the control group; $P = .67$), but among children ≥24 months old, crude rates were significantly higher among hospitalized children, compared with controls (8.1% vs 2.2%; $P = .009$). This trend was observed in each county and was unchanged after control for several covariates by logistic regression: adjusted ORs were 1.2 (95% CI, 0.8–1.8; $P = .39$) for children <24 months old and 4.3 (95% CI, 1.5–12.7; $P = .009$) for children ≥24 months old.

HRV-C

Crude rates of HRV-C detection were similar between the 2 study groups among participants aged <6 months (3.9% vs 4.4%; $P = .69$), but among children ≥6 months old, crude rates were higher among hospitalized children, compared with controls (8.2% vs 3.9%; $P = .002$). The logistic regression results supported these findings: adjusted ORs were 1.0 (95% CI, 0.6–2.1; $P = .96$) for children <6 months old and 2.0 (95% CI, 1.2–3.4; $P = .01$) for children ≥6 months old. Insurance status, race/ethnicity, and exposure to another child or smoker were not significantly associated with HRV-A or HRV-C detection and were not included in the final logistic regression models. Results were similar if children with codetection of other viruses were excluded or counted as HRV negative.

Table 2. Age and Medical History of Patients Hospitalized With Acute Respiratory Infection (ARI) and Asymptomatic Controls, Overall and by Human Rhinovirus (HRV) Species, During the 12 Months in Which Controls Were Enrolled[a]

Characteristic	ARI hospitalizations, %				Controls, %			
	HRV-A (n = 113)	HRV-B (n = 15)	HRV-C (n = 93)	Overall (n = 1515)	HRV-A (n = 52)	HRV-B (n = 4)	HRV-C (n = 32)	Overall (n = 790)
Age, mo								
0–5	42[c]	40	30	48[d]	37	0	38	34
6–23	36	40	40	32	56	50	41	43
24–59	21	20	30	20	8	50	22	23
High-risk condition[b]	37[c]	47	52[c]	30[d]	15	25	19	11
Asthma or wheezing[e]	30[c]	47	42[c]	21[d]	8	0	13	7
Age 0–11 mo	7	0	13	7[d]	3	0	0	2
Age 12–59 mo	63[c]	88	64[c]	45[d]	17	0	24	13
Chronic lung	7	7	5	5[d]	2	0	0	0.4
Neurologic	10	0	3	6[d]	4	25	3	1
Premature birth (>1 mo early)	21[c]	13	17	16[d]	6	0	9	9

[a] Controls were enrolled during December 2003–April 2004 and October 2004–April 2005. A total of 40 ARI hospitalizations and 6 controls with HRV detections had respiratory syncytial virus, parainfluenza virus, human metapneumovirus, or influenza virus codetected (among ARI hospitalizations, codetection occurred in 25 with HRV-A, 4 with HRV-B, and 11 with HRV-C; among controls, codetection occurred in 2 with HRV-A, 0 with HRV-B, and 2 with HRV-C). Species were indeterminate for 7 ARI hospitalizations and 12 controls .

[b] Includes history of cancer; diabetes mellitus; sickle cell disease; immunodeficiency; disease of the heart, kidney, or lung (including asthma); and neuromuscular conditions, such as seizures, cerebral palsy, or muscular dystrophy.

[c] $P < .05$ for comparison between hospitalized patients with ARI and controls with separate comparisons for HRV-A and HRV-C. No comparisons were made for HRV-B because of small numbers.

[d] $P < .05$ for comparison between hospitalized patients with ARI and controls.

[e] Includes history of asthma, reactive airway disease, or chronic or recurrent wheezing.

Results for children positive for any HRV were consistent with results for children with HRV-A and for those with HRV-C described above; the overall adjusted OR averaged across age groups was 1.46 (95% CI, 1.1–1.9), and the age-specific adjusted OR was 2.6 (95% CI, 1.4–4.9) for children aged 24–59 months and was not significantly different for younger children.

Association Between HRV and History of Asthma or Wheezing

HRV was significantly more common in hospitalized children >12 months old with a history of asthma or wheezing and in controls >24 months old ($P < .01$) (Table 4). Similar trends were seen for HRV-A and HRV-C.

Clinical Severity of Hospitalizations Among Children With HRV and Children With Other Respiratory Viruses

Clinical characteristics were compared between children with HRV only and those with other viruses only (Table 3). With regard to clinical indicators of severity, these 2 groups had similar rates of intensive care unit (ICU) admission, mechanical ventilation use, and supplemental oxygen use. A hospital stay >3 days was less common among children with HRV only, compared with those with other viruses only ($P < .001$), but the median duration of stay was not different. Acute wheezing was less frequently diagnosed in infants <6 months old with HRV only but more commonly diagnosed in older children with HRV only, compared with children in whom other viruses only were detected ($P < .002$ for both comparisons). A discharge diagnosis of asthma was more common among children with HRV only, compared with children with other viruses only ($P < .001$); a similar trend was seen for asthma as the first-listed discharge diagnosis (data not shown). Discharge diagnoses of croup and otitis media were less common among children with HRV only ($P \leq .01$ for both comparisons).

Codetections

Fifty-two children had both HRV and another respiratory virus (26 had RSV codetected, 12 had PIV, 9 had HMPV, and 7 had influenza virus), with codetections more common for HRV-A than for HRV-C among children aged 6–23 months. Children with codetections and those with other viruses only had similar clinical courses.

Clinical Severity of Hospitalizations Among HRV Species

The clinical characteristics of children hospitalized with HRV during the 19-month study period are shown for each HRV species (Table 3). Overall and for each HRV species, >75% had a diagnosis of acute wheezing, pneumonia, bronchiolitis, or asthma. Children with HRV-A were more likely to have a discharge diagnosis of pneumonia or bronchiolitis, compared with children with HRV-C (23% vs 13%; $P = .04$), but the 2 groups were not significantly different with respect to other clinical characteristics (Table 3), including indicators of severity (ie, ICU admission, receipt of mechanical ventilation, and length of hospital stay). Acute

Table 3. Clinical Course and Codetected Viruses Among Hospitalized Children During 19-Month Study Period[a]

Characteristic	Virus detected				
	HRV-A (n = 167)	HRV-B (n = 29)	HRV-C (n = 136)	HRV only[b] (n = 287)	Other viruses only[c] (n = 588)
Duration of illness before admission, median (IQR), days	2 (2–4)	2 (1–3)	2 (1–4)	2 (1–4)	3 (2–5)[d]
Duration of illness before swab sample date, median (IQR), days	3 (2–5)	2 (2–4)	3 (2–4)	3 (2–4)	4 (3–5)[d]
ICU admission	10 (6)	1 (3)	7 (5)	16 (6)	31 (5)
Mechanical ventilation	3 (2)	1 (3)	2 (1)	5 (2)	18 (3)
Supplemental oxygen	66 (40)	10 (34)	54 (40)	102 (36)	248 (42)
Length of hospital stay, median (IQR), days	2 (1–3)	2 (1–2)	1 (1–3)	1 (1–2)	2 (1–3)
Hospital stay >3 days	23 (14)	3 (10)	14 (10)	27 (9)	117 (20)[d]
Acute wheezing, by age[e]					
0–5 mo	32 (49)	8 (67)	16 (44)	38 (40)	179 (64)[d]
6–23 mo	45 (75)	7 (58)	46 (82)	85 (79)	135 (62)[d]
24–59 mo	35 (83)	4 (80)	42 (95)	77 (90)	50 (54)[d]
Discharge diagnosis					
Pneumonia or bronchiolitis	75 (45)[f]	12 (41)	43 (32)	95 (33)	369 (63)[d]
Any asthma, by age					
0–23 mo	37 (30)	5 (21)	34 (37)	69 (34)	95 (19)[d]
24–59 mo	33 (79)	4 (80)	41 (93)	74 (86)	44 (48)[d]
Croup	5 (3)	3 (10)	5 (4)	9 (3)	44 (7)[d]
Otitis media	15 (9)	1 (3)	17 (13)	27 (9)	111 (19)[d]
Codetected viruses, by age[g]					
All ages	34 (20)	6 (21)	12 (9)	…	…
0–5 mo	14 (22)	4 (33)	6 (17)	…	…
6–23 mo	16 (27)[f]	1 (8)	4 (7)	…	…
24–59 mo	4 (10)	1 (20)	2 (5)	…	…

Abbreviations: ICU, intensive care unit; IQR, interquartile range.

[a] Data represent No. (%) of ARI hospitalizations with the denoted viruses detected. The study period was October 2003–April 2005. Species were indeterminate in 11 (3%) of 339 human rhinovirus (HRV)–positive hospitalizations for acute respiratory infection.

[b] Data are for children in whom only HRV was detected.

[c] Data are for children in whom only the following viruses were detected: respiratory syncytial virus (n = 332), parainfluenza virus (n = 99), human metapneumovirus (n = 84), and influenza virus (n = 155). The sum is greater than the total of 588 because some children had multiple infections.

[d] $P < .05$ for comparison between patients with HRV only and those with other viruses only.

[e] Acute wheezing includes an admission or discharge diagnosis of wheezing, bronchiolitis, or asthma.

[f] $P < .05$ for comparison between HRV-A and HRV-C. (No comparisons were made for HRV-B because of small numbers.)

[g] The following viruses were codetected: respiratory syncytial virus, parainfluenza virus, human metapneumovirus, and influenza virus.

wheezing at admission or discharge was the most common diagnosis (66%–76% of children) among the HRV species, and for those with HRV-A and those with HRV-C, acute wheezing was significantly more common with increasing age ($P < .001$ for both comparisons). Although HRV-B was detected infrequently, clinical characteristics of children with this species appeared to be similar to those of children with HRV-A or HRV-C.

Table 4. Detection of Human Rhinovirus (HRV) Among Children With and Those Without a History of Asthma or Wheezing, by Age[a]

Study group	HRV Detection, %					
	Age 0–11 mo		Age 12–23 mo		Age 24–59 mo	
	Asthma	No asthma	Asthma	No asthma	Asthma	No asthma
ARI hospitalizations	18	12	29[b]	12	25[b]	11
Controls	11	13	10	17	22[b]	5

[a] Asthma is defined as a history of asthma or wheezing, which includes asthma, reactive airway disease, or chronic or recurrent wheezing. Among the group of patients hospitalized for acute respiratory infection (ARI), asthma was detected in 61 aged 0–11 mo, 97 aged 12–23 mo, and 165 aged 24–59 mo. Among controls, asthma was detected in 9 aged 0–11 mo, 20 aged 12–23 mo, and 27 aged 24–59 mo.

[b] $P < .05$ for comparison of HRV detection rates between the group with asthma and the group without asthma.

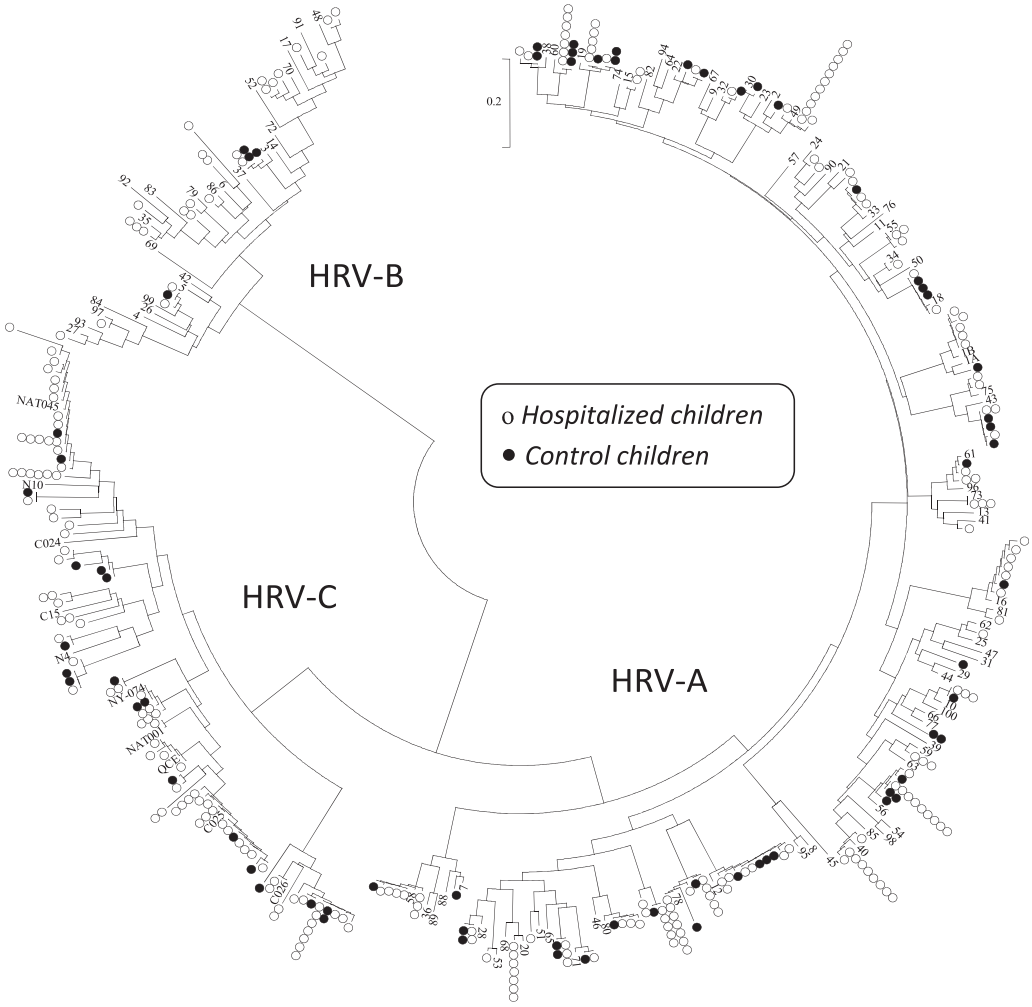

Figure 2. Phylogenetic tree of selected human rhinoviruses (HRVs) detected in hospitalized children (*white circles*) and asymptomatic controls (*black circles*) during the study period. Published HRV-A and -B prototype strains (numbers) and selected HRV-C strains (strain names) were included for context. The estimated neighbor-joining tree was constructed from alignments of partial predicted amino acid (aa) sequences of the VP1 protein (aa 625–716 of HRV1A [GenBank accession no. ACK37367]) using MEGA software, version 4.0.2 (Center for Evolutionary Medicine and Informatics. The Biodesign Institute, Tempe AZ). All positions containing alignment gaps and missing data were eliminated in pairwise sequence comparisons (pairwise deletion option). Bootstrap support values (2000 replicates) were plotted at selected internal branch nodes. Scale bar corresponds to 0.2 aa change per site.

Diversity of HRV Genetic Types Among Hospitalized and Control Children

The phylogenetic tree in Figure 2 depicts the wide distribution of HRV types among hospitalized children and controls. No discernible differences in the distribution of HRV-A and HRV-C species or types between hospitalized and control children were noted. Diversity of HRV-B viruses appeared more limited among controls than among hospitalized children, but the number of cases was small.

DISCUSSION

In children <5 years of age hospitalized with ARI, significantly higher rates of HRV-A and HRV-C detection were noted when compared with rates in asymptomatic control children, particularly among older children. However, clinical indicators of severity, such as ICU admission, were not significantly different between HRV species or between HRV and other respiratory viruses acknowledged to cause severe illness.

There are some similar comparisons to controls in the literature. One US study [17], 2 European studies [5, 12], and a study from Thailand [16] compared a group of children hospitalized with ARI with an asymptomatic control group. The US study, involving Alaskan Native children <3 years old, detected HRV in 32% of both hospitalized (n = 208) and control (n = 381) children, with similar detection rates regardless of whether controls were symptomatic in the prior 14 days [17]. A Finnish study of hospitalized children 3 months to 15 years of age with acute wheezing (n = 161) versus surgical controls (n = 79) found HRV detection rates

of 16% and 8%, respectively [12]. The controls were asymptomatic during the prior 4 weeks, and 5 of 13 HRV/HRV-enterovirus positive controls developed symptoms in the following week. A Spanish study of hospitalized children <14 years old (n = 1555) and elective surgery or food challenge controls who were asymptomatic for 14 days before and 10 days after enrollment (n = 211) reported a significant difference in HRV detection between the groups (27.2% and 12%, respectively) [5]. The Thailand study compared detection rates between hospitalized children and asymptomatic clinic controls and found significantly higher rates of HRV-A and HRV-C detection in hospitalized children 1–19 years old but not in those <1 year of age [16]. Additional studies of ARI cases seen in emergency or urgent care settings reported significantly higher detection of HRV [6, 18, 19] or HRV-C [20], compared with asymptomatic controls.

Our study noted a significant association between HRV and hospitalization among older children (ie, those aged 24–59 months for HRV-A and those aged 6–59 months for HRV-C) but not among younger children. Similar findings were reported in another US study comparing wheezing children treated in an emergency department with asymptomatic controls and found differences in HRV detection rates by RT-PCR for older children 2–16 years old but not for children <2 years old [28]. Differences in age distribution may be secondary to older children being at higher risk for asthma exacerbations or wheezing, possibly because of risk factors such as prior viral infections or atopy [29, 30] or because of the difficulty in determining whether younger children are asymptomatic. Finally, our study only required control children to be asymptomatic on the day of their visit, and children may have been shedding HRV from prior infections or incubating new infections. This is also true for the hospitalized children. The difference by age might also be explained if younger children have more frequent HRV infections, symptomatic or asymptomatic, or longer durations of HRV shedding than older children. Follow-up studies provide limited support for this explanation. One study of 329 children followed up from 2 to 24 months of age reported that 91% were HRV seropositive and 79% were HRV-positive by RT-PCR or isolation, with the bulk of infections occurring between 6 and 18 months of age [10]. More-recent cohort studies performed HRV typing over time and found the same type typically persisting <2 weeks in infants with multiple types circulating concurrently and sequentially in their families [11] and in children <7 years of age [13]. Other earlier studies without HRV typing and without age group comparisons among young children detected HRV for up to 5–6 weeks among children hospitalized for wheezing (78% were <3 years of age) [12] and among children 4–12 years of age with asthma exacerbation [14] and for up to 21 days in 20% of specimens from families with young children [31].

Our data support an association between HRV hospitalization and asthma or wheezing. This was also observed for HRV species, consistent with our previous study [9] and another study [32]. We did not find a history of asthma or asthma discharge diagnoses to be significantly more frequent in children with HRV-C than in those with HRV-A, as reported in our previous study, but the trends were in the same direction. Our previous study included 2 instead of 3 counties, did not include controls, and had fewer HRV cases, emphasizing the importance of studies of multiple locations and years and of controls.

Our study has several limitations. First, detection of HRV RNA by RT-PCR may not reflect an infection that caused the ARI hospitalization. Second, asymptomatic controls were not available for all months. However, we compared hospitalized and control groups during months with controls, which is a valid comparison. Third, our controls may not have been representative of the community or as closely matched as surgical controls for factors related to hospitalization. One study with a surgical control group noted the potential for negative selection bias against children with respiratory illness, which could have lowered their HRV detection rate [12]. Fourth, asthma is difficult to define clinically, and the diagnosis is generally assigned to older children. To account for these factors, we used different definitions for asthma and wheezing, as was done in a previous study. Fifth, although some HRV cases may have been undetected because of specimen storage for several years and repeated freezing and thawing of some specimens, most of the specimens were previously unthawed aliquots, and hospital and control specimens were concurrently tested. Finally, we did not do quantitative RT-PCR or longitudinal sampling, which precludes a more rigorous assessment of disease associations with viral load.

In conclusion, our study provides evidence that HRV-A and HRV-C are associated with hospitalization for ARI, particularly among older children. This could possibly be due to the association with asthma, a diagnosis more likely to be assigned to older children, but it may also be due in part to a longer duration or higher rate of asymptomatic HRV shedding in younger children. The association between HRV and wheezing is complex. Longitudinal studies over multiple years with healthy controls are needed to understand the causal associations in young children who have a high burden of severe ARI, HRV, and history of wheezing and to clarify whether particular HRV species or types cause more severe illness.

Notes

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