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Prevalence and Genetic Diversity of Adenovirus 40/41, Astrovirus, and Sapovirus in Children With Acute Gastroenteritis in Kansas City, 2011–2016

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Abstract

Background.—Most acute gastroenteritis (AGE) episodes in children in the United States are attributed to norovirus, whereas very little information is available on adenovirus 40/41 (AdV40/41), astrovirus, or sapovirus. The New Vaccine Surveillance Network (NVSN) conducted prospective, active, population-based AGE surveillance in young children.

Methods.—We tested and typed stool specimens collected between December 2011 and June 2016 from 1 NVSN site in Kansas City for the 3 viruses, and calculated hospitalization and emergency department (ED) detection rates.

Results.—Of 3205 collected specimens, 2453 (76.5%) were from AGE patients (339 inpatients and 2114 ED patients) and 752 (23.5%) were from healthy controls. In patients with AGE, astrovirus was detected in 94 (3.8%), sapovirus in 252 (10.3%), and AdV40/41 in 101 (4.5%) of 2249 patients. In healthy controls, astrovirus was detected in 13 (1.7%) and sapovirus in 15 (2.0%) specimens. Astrovirus type 1 (37.7%) and genogroup I sapoviruses (59.3%) were most prevalent. Hospitalization rates were 5 (AdV40/41), 4 (astrovirus), and 8 (sapovirus) per 100 000 children <11 years old, whereas ED rates were 2.4 (AdV40/41), 1.9 (astrovirus), and 5.3 (sapovirus) per 1000 children <5 years old.

Conclusions.—Overall, AdV40/41, astrovirus, and sapovirus were detected in 18.6% of AGE in a large pediatric hospital in Kansas City.

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Disclaimer. The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the Centers for Disease Control and Prevention.

Keywords

acute gastroenteritis; adenovirus 40/41; astrovirus; sapovirus; genetic diversity

Despite introduction of preventive strategies, acute gastroenteritis (AGE) remains a major global public health problem and is associated with an estimated 525 000 deaths annually in children <5 years of age including >400 children in the United States (US) [1, 2]. Most AGE episodes in children are attributed to viruses that are primarily transmitted by the fecal-oral route, principally rotavirus, norovirus, sapovirus, astrovirus, and adenovirus 40/41 (AdV40/41) [3]. Since the successful introduction of rotavirus vaccines, followed by the subsequent decline of these viruses in young children, norovirus has become the most common cause of AGE in most regions in the world [4–6]. In a recent study, norovirus was the most frequently detected pathogen of medically attended acute gastroenteritis cases in the US, followed by sapovirus and astrovirus, although the specimens were not tested for AdV40/41 [7]. In an 8-country multiyear community-based surveillance study in low- and middle-income countries, sapovirus, rotavirus, AdV40/41, and astrovirus had the highest attributable diarrheal incidence after *Shigella* in children 12–24 months of age [8].

Human adenoviruses in the genus *Mastadenovirus* are classified into 9 species (A to G) and at least 90 genotypes have been identified [9], of which species F (genotypes 40 and 41 [AdV40/41]) have been associated with AGE. AdV40/41 are responsible for an estimated 5%–20% of AGE in young children [10, 11]. Astroviruses, belonging to the family *Astroviridae*, are nonenveloped single-stranded RNA viruses [12]. Of the 19 species of genus *Mamastrovirus*, MAstV-1 includes the classic human astrovirus (HAstV) genotypes 1–8 that cause AGE [13]. Of these, HAstV type 1 is the most frequently detected type, accounting for >50% of AGE cases worldwide, whereas HAstV type 7 and HAstV type 8 viruses are rare [14]. In recent years, 2 novel groups of highly divergent astroviruses, named HAstV-MLB and HAstV-VA/HMO, have been identified in human stools of individuals with diarrhea [15, 16], but also in asymptomatic healthy controls (HCs) [17].

Sapoviruses are nonenveloped single-stranded RNA viruses belonging to a separate genus *Sapovirus* in the *Caliciviridae* family and are classified into 19 genogroups (GI to GXIX) [18]. Of these, viruses from GI, GII, GIV, and GV infect humans and cause AGE [19], and worldwide most infections are caused by GI viruses [20].

Because AGE continues to be a substantial healthcare burden in the US, and AGE etiology frequently remains undetermined during standard of care management [7], data from active surveillance systems like the New Vaccine Surveillance Network (NVSN) [21] can be used to estimate the prevalence and incidence of medically attended viral AGE, particularly for viruses for which routine clinical testing is not readily available. The NVSN sites include 7 pediatric hospitals across the US conducting prospective, active, population-based, laboratory-confirmed surveillance for AGE to determine the cause and burden of gastrointestinal hospitalizations and emergency department (ED) visits in children as well as evaluate the effectiveness of pediatric vaccines [4]. Unique for NVSN is that age- and time-matched HCs are also enrolled to assess baseline population levels of AGE pathogens.

In this study, we determined the prevalence and genetic diversity of AdV40/41, astrovirus, and sapovirus in stool specimens collected from children with AGE and HCs from 2011 to 2016.

MATERIALS AND METHODS

Recruitment and Enrollment

Between December 2011 and June 2016, children between 14 days and 11 years old were enrolled if they had symptoms of AGE for ≤ 10 days regardless of whether they were hospitalized or seen in the ED at Children's Mercy hospital in Kansas City, Missouri. The Children's Mercy hospital has the primary market share for inpatient ($>90\%$), and thus children seeking medical care are most likely cared for in Children's Mercy ED and hospital. ED patients admitted within 6 days of enrollment were classified as inpatients. AGE symptoms were defined as ≥ 3 diarrhea episodes within 24 hours, ≥ 1 vomiting episode within 24 hours, or both, and symptoms lasting for <10 days prior to enrollment. Stool specimens were collected within 10 days of illness onset. Surveillance was conducted in the ED for 8 hours/day and >4 days each week and enrollment in inpatient was conducted during Monday–Friday of each week.

Healthy controls were enrolled during well-child visits of children between >14 days and 11 years of age at Children's Mercy primary care clinic as described previously [3]. HC were enrolled each week to match the seasonal distribution of the AGE enrollees. HC stools were collected at enrollment or requested to be brought back/couriered within 5 days after enrollment.

Parent/guardian interviews were conducted for each enrolled child with AGE and each HC. Demographic, epidemiologic, and clinical data, including age, sex, race, ethnicity, previous medical history, clinical signs and symptoms, date of onset of symptoms, and date of specimen collection, were collected. All hospitalized and ED AGE patients from the predefined surveillance area (6-county area within the Kansas City metropolitan region) were considered eligible for the study. Approval for the study was obtained from the Institutional Review Board of Children's Mercy Hospital–Kansas City and from the Centers for Disease Control and Prevention.

Specimen Collection and Nucleic Acid Extraction

Raw stool specimens were collected directly from whole stool or diapers and several aliquots were made and stored at -80°C for later use. Clarified stool suspensions were made by adding 100 μL of liquid stool to 400 μL of nuclease-free distilled water and nucleic acid was extracted by mixing 180 μL of clarified stool suspension with 20 μL of bacteriophage MS2 (internal control) by using the NucliSENS easyMag automated extraction system according to manufacturer instructions (bioMérieux, Durham, North Carolina).

Testing and Typing of Astrovirus and Sapovirus

Nucleic acids were tested for sapoviruses and classical astroviruses by real-time reverse-transcription polymerase chain reaction (RT-qPCR) using previously published assays

[22, 23]. Specimens with cycle threshold (Ct) values of ≤ 35 were considered positive. RT-qPCR-positive specimens were typed by amplification of small regions of the genome by conventional RT-PCR (astrovirus: 449 bp, sapovirus: 420 bp) [24, 25]. PCR products were visualized on a 2% agarose gel (Seakem-ME, Lonza, Allendale, New Jersey) containing Gel Red (Biotium, Fremont, California) and purified by ExoSAP-IT (Affymetrix, USB, Cleveland, Ohio) or gel purified by the QIAquick PCR purification kit (Qiagen, Germantown, Maryland). Purified PCR products were sent to Eurofins MWG Operon (Louisville, Kentucky) for Sanger sequencing. Sapovirus sequences were typed by using an online human calicivirus typing tool at <https://calicivirustypingtool.cdc.gov> [26]. For the classical human astrovirus sequences, alignments with prototype reference sequences (HAstV 1–8) were generated using MUSCLE [27]. Phylogenetic trees were constructed using the maximum-likelihood method with 100 bootstraps replications to assess phylogenetic robustness using MEGA version 11 [28]. Additionally, all specimens were also tested for nonclassical astroviruses using a pan MAstV conventional RT-PCR assay [29]. PCR-positive specimens were then further tested by conventional RT-PCR using specific oligonucleotide primers for nonclassical astroviruses [29].

Testing by xTAG Gastrointestinal Pathogens Panel

A subset of 2249 AGE specimens, but no HC specimens, was available for additional testing using the xTAG Gastrointestinal Pathogens Panel (GPP, Luminex Molecular Diagnostics, Austin, Texas) kit that includes targets for 9 bacteria (*Campylobacter*, *Clostridioides difficile* toxin A/B, *Escherichia coli* 0157, enterotoxigenic *E coli* (ETEC) LT/ST, Shiga-like toxin-producing *E coli* (STEC) stx1/stx2, *Salmonella*, *Shigella*, *Vibrio cholerae*, *Yersinia enterocolitica*), 3 viruses (AdV40/41, norovirus GI/GII, rotavirus A), and 3 parasites (*Cryptosporidium*, *Entamoeba histolytica*, *Giardia*). AdV40/41-positive specimens were not further typed.

Calculation of Incidence Rates

Population-based AdV40/41, astrovirus, and sapovirus hospitalization rates per 100 000 per year were calculated from 1 January through 30 June for 5 consecutive seasons: December 2011–November 2012, December 2012–November 2013, December 2013–November 2014, December 2014–November 2015, and December 2015–November 2016. Hospitalization rates were calculated using the weighted number of laboratory-confirmed AdV40/41, astrovirus, or sapovirus hospitalizations divided by the number of children within the age cohort study site's catchment area (Johnson County, Leavenworth County, and Wyandotte County, Kansas; Clay County, Jackson County, and Platte County, Missouri), as determined by the 2020 US bridged-race population estimates (National Center for Health Statistics, https://www.cdc.gov/nchs/nvss/bridged_race.htm). Weighting was performed to account for the number of surveillance days, the proportion of the eligible children enrolled, and the percentage of stool specimens collected and tested. The 95% confidence intervals (CIs) were determined by conducting 1000 bootstraps for each rate and using the resulting 2.5th and 97.5th percentiles as the lower and upper confidence limits, respectively [30–33].

The ED visit rates per 10 000 per year were calculated by multiplying the percentage of children with AGE that were AdV40/41, astrovirus, or sapovirus positive by the total

number of AGE-related ED visits during the full 24-hour surveillance periods and then divided by the number children in the site's catchment area. The numerator data were weighted to account for the estimated market share of the Children's Mercy Hospital–Kansas City ED among residents <11 years of age in the 6-county catchment area and the number of surveillance days conducted each week. This approach precluded calculation of 95% CIs.

Statistical Analyses

A 2-tailed Fisher exact test (GraphPad Prism version 9.2.0, San Diego, California) was used to compare the proportion of virus-positive specimens from AGE children versus HCs, and between the proportion of AGE-positive specimens in children 12–59 months old versus all other age groups (younger and older were pooled in a single group). A Mann-Whitney test was used to compare the differences of Ct values between the different groups. *P* values of <.05 were considered statistically significant.

RESULTS

A total of 3205 fecal specimens were collected from children <11 years of age (average, 34 months; median, 22 months) at Children's Mercy–Kansas City between December 2011 and June 2016 (55 months). Of those, 2453 (76.5%) specimens were from AGE patients (339 inpatients, 2114 ED visit patients) and 752 (23.5%) from age-/time-matched HCs (Table 1).

From the 2453 AGE specimens, astrovirus was detected in 94 (3.8%), sapovirus in 252 (10.3%), and AdV40/41 in 101 (4.5%) of the 2249 tested specimens as 204 specimens were not available for GPP testing. A combination of sapovirus/astrovirus, sapovirus/adenovirus, or astrovirus/adenovirus co-detections were detected in 12 (0.5%) specimens. No AdV40/41, astrovirus, or sapovirus was detected in the remaining 1994 (81.3%) specimens. Among the 752 HC specimens, 15 (2.0%) tested positive for sapovirus, 13 (1.7%) tested positive for astrovirus, and neither virus was detected in 724 (96.3%) specimens (Figure 1). HC specimens were not tested by GPP and therefore no AdV40/41 data were obtained. The majority (91.7% AdV40/41, 82.6% astrovirus, and 86.2% sapovirus) of detections were in AGE or HC children <5 years of age. For sapovirus, hospitalized inpatients and ED patients (mean Ct value 25) had significantly ($P = .021$) higher viral loads than the HCs (mean Ct value 29), whereas for astrovirus there was no difference between the Ct values of hospitalized children and HCs (Figure 2).

Among the 339 specimens from hospitalized children with AGE, 13 (3.8%) tested positive for astrovirus, 28 (8.3%) tested positive for sapovirus, and 11 (5.6% of the 197 AGE-Inpatient) specimens were positive for AdV40/41 (Table 1). Among the 2114 ED specimens, 89 (4.2%) tested positive for astrovirus, 233 (11.0%) tested positive for sapovirus, and 97 (4.7% of the 2052 specimens tested) were positive for AdV40/41 (Table 1).

Of the 108 AdV40/41-positive specimens, 61 (56.5%) were from children 12–59 months old. Of these, 3 (4.9%) specimens were from inpatients and 58 (95.1%) from ED patients. Astrovirus was detected in 115 specimens, including 68 (59.1%) from children 12–59 months old. Seven (10.3%) were from hospitalized children, 54 (79.4%) from ED patients,

and 7 (10.3%) from HCs (Table 1). Of the 276 sapovirus-positive specimens, 168 (60.9%) were from children 12–59 months old. Twelve of these (7.1%) specimens were from inpatients, 146 (86.9%) from ED patients, and 15 (6.0%) from HCs (Table 1). Overall, AdV40/41, astrovirus, and sapovirus prevalence was higher in the age group 12–59 months compared to 0–11 months ($P < .05$) and >60 months ($P < .05$).

In a subset of 2249 AGE specimens that were tested by xTAG GPP, coinfections with other enteric viruses and/or bacteria were present in 33% of AdV40/41-positive specimens, 37% of astrovirus-positive specimens, and 32% of sapovirus-positive specimens (Table 2). Twelve children were coinfecting including astrovirus and sapovirus ($n = 5$), AdV40/41 and sapovirus ($n = 4$), or AdV40/41 and astrovirus ($n = 3$), while 6 of these 12 specimens had a third detection. Coinfections with norovirus were detected in 8 (7.4%) AdV40/41-positive specimens (1 was a child with 5 coinfections), 15 (14.7%) astrovirus-positive specimens (3 included a third detection and 1 child was infected with 5 pathogens), and 20 (7.7%) sapovirus-positive specimens (2 included a third detection). *Clostridioides difficile* was the most often detected bacterial coinfection, while ETEC, *Salmonella*, *Shigella*, and STEC were also detected (Table 2). Fourteen of the 15 (93.3%) astrovirus coinfections with *C. difficile* were in children <2 years old. Similarly, 19 of the 21 (90.5%) AdV40/41 infections and 35 of the 36 (97.2%) sapovirus coinfections with *C. difficile* were among the <2 years age group. AdV40/41, astrovirus, and sapovirus were detected throughout the year, with no clear seasonal trend for AdV40/41. In contrast, astrovirus and sapovirus prevalence was highest in the cooler months, November to April, with some annual fluctuations (Figure 3).

For adenovirus-positive specimens ($n = 108$), 46 (42.6%) were from Black children, 30 (27.8%) from White children, 16.7% from children for whom race information was not available, and 11.1% from children identified as multiracial (Table 3). Among the 115 astrovirus-positive specimens, 34 (29.6%) were from Black children, 42 (36.5%) from White children, 25.2% from children for whom race information was not available, and 8.4% from children identified as multiracial (Table 3). Sapovirus prevalence was higher in males than in females (Table 3); however, the differences were not statistically significant ($P > .05$). Among 276 children with sapovirus-positive specimens, 119 (43.1%) were from Black children and 88 (31.9%) from White children; children who identified as multiracial, Asian, or another race, or who refused to disclose their race, accounted for the remaining 25%. Children who were identified as Hispanic accounted for approximately one-fourth of the AdV40/41- and sapovirus-positive specimens and one-third of the astrovirus-positive specimens compared to people of non-Hispanic ethnicity. Overall, positivity rate for each of the viruses was not significantly different in Hispanic children compared to the overall enrollment population (Table 3).

The hospitalization rates were 5 (95% CI, 3–8) for AdV40/41, 4 (95% CI, 2–4) for astrovirus, and 8 (95% CI, 5–10) for sapovirus per 100 000 children <11 years old. The AdV40/41-, astrovirus-, and sapovirus-associated ED rates were 2.4, 1.9, and 5.3 per 1000 children <5 years old and 0.3 and 0.8, and 1.5 per 1000 children 5–10 years old, respectively.

Of the 115 astrovirus-positive specimens, 106 (92.2%) were successfully typed. HAsV type 1 was the most prevalent (40 [37.7%]), followed by HAsV type 5 (22 [20.8%]), HAsV type 4 (18 [17.0%]), HAsV type 2 (16 [15.1%]), HAsV type 3 (9 [8.5%]), and HAsV type 8 (1 [0.9%]). Of the remaining 9 specimens that could not be typed, 7 had Ct values between 30 and 35. Additionally, 13 specimens tested positive for nonclassical astroviruses including MLB1 (n = 10), MLB2 (n = 1), VA1 (n = 1), and VA2 (n = 1) (Figure 4B).

Of the 276 sapovirus-positive specimens, 182 (66%) were successfully genotyped including 108 GI (GI.1, n = 74; GI.2, n = 25; GI.3, n = 2; and GI.6, n = 7), 46 GII (GII.1, n = 19; GII.2, n = 7; GII.3, n = 7; GII.4, n = 5; and GII.5, n = 8), 11 GIV, and 17 GV (Figure 4A). Of the 58 specimens that had Ct values between 30 and 35, 19 (32.7%) could be typed.

DISCUSSION

Since the introduction of rotavirus vaccines, norovirus has become the leading cause of medically attended AGE in US children younger than 5 years of age [4]. Very few US reports are available on other established gastroenteritis viruses such as AdV40/41, astrovirus, and sapovirus. We aimed to reduce that data gap by testing specimens from children with AGE enrolled in Kansas City, which is 1 of the 7 NVSN sites, which conducts prospective, population-based surveillance on viral causes of AGE and respiratory infections [4, 21]. Among 2453 enrolled children with AGE seeking medical care at the Kansas City site during 2011–2016, the overall prevalence was 4.5% for AdV40/41, 3.8% for astrovirus, and 10.3% for sapovirus, whereas the prevalence in HCs was 1.7% for astrovirus and 2% for sapovirus (no samples were tested for AdV40/41). In addition, we also detected nonclassical MLB and VA astroviruses, albeit at a low rate (0.4%) suggesting that these viruses do not play a major role in pediatric gastroenteritis in the US. Genogroup I sapoviruses were most frequently detected (59.3%) followed by GII (25.3%), GIV (6.0%), and GV (9.3%).

Until recently, AdV40/41 was an underappreciated cause of diarrhea in children [34]. We found an overall prevalence of AdV40/41 of 4.5% and 6.3% among children <2 years of age, which is on the lower end of the detection rates previously reported across 5 NVSN sites [3, 32]. There was no apparent outbreak detected and no discernable seasonality for AdV40/41.

Globally, the overall prevalence of astrovirus is 4.2% [35], ranging from 0% to almost 30% [14], with rates in the US between 3% and 5% [36, 37]. All 3 clades of HAsV infecting humans were detected in this study, including the classic HAsV (types 1–5, 8), as well as the novel HAsV-MLB and HAsV-VA. HAsV type 1 viruses were the most common genotype (37.7%) in our 5-year study, in line with global data [35]. Serological studies indicate that most children are infected with HAsV and develop antibodies to the virus early in life, and recent data from an adult population in the US showed that 73% have immunoglobulin G antibodies against HAsV-1 [38]. Astrovirus clearly showed a winter seasonality, with the vast majority of detections between November and May each year comparable with previous studies in the US [36, 37].

Globally the pooled prevalence of sapovirus associated with AGE is 3.4% [20] with prevalence in children younger than 5 years ranging between 2% and 17% [39, 40]. Higher

sapovirus prevalence rates are frequently reported in studies that, like ours, used RT-qPCR for detection [36, 41]. Rates in HCs, 4% in this study, are typically significantly lower than in cases [36, 41]. We detected all 4 sapovirus genogroups that infect humans, with GI viruses comprising >50% of all sapovirus detections, consistent with other studies [41–43]. They were detected throughout the year with peak detection during the winter months [36].

Coinfections with combinations of AdV40/41, astrovirus, and sapovirus were found in 0.5% of the specimens, which is significantly lower than reported in studies from Thailand, Nigeria, or Germany [44]. When expanded to also include norovirus, rotavirus, and bacterial coinfections, norovirus and *C difficile* were more frequently co-detected, ranging from 8% to 19% of AdV40/41-, astrovirus-, and sapovirus-positive specimens. However, the majority of *C difficile* co-detections were in children <2 years of age, suggesting *C difficile* colonization rather than infections [45] and increasing confidence that the AGE symptoms were due to AdV40/41, astrovirus, or sapovirus. Detection of multiple viruses/pathogens in pediatric AGE cases has become more common, including the increasing use of gastroenteritis multiplex molecular assays [46]. Our co-detection rates in pediatric AGE patients are slightly higher than the range (4%–13%) previously reported [3, 35]. Viral loads, using Ct values as a surrogate, for sapovirus appeared slightly higher (ie, lower Ct values) in children with AGE (ED visits and hospital) compared to HCs, whereas for astrovirus, the highest viral loads were found in children visiting the ED with AGE. Previous studies for norovirus have shown that viral load, as measured by RT-qPCR testing of stools from AGE patients, does not clearly distinguish nonpathogenic detections from disease-associated detections [47]; in addition, many molecular diagnostic gastrointestinal panels such as the GPP used in this study do not provide information on viral loads. Extraction of stool was done for <20% of the specimens; further elution may not affect results in these sample because viruses are generally shed at high titers in stool samples and molecular assays are highly sensitive to detect viruses even if extraction could have diluted the sample.

In summary, AdV40/41, astrovirus, and sapovirus contributed to 18.6% of medically attended AGE during a 5-year prospective surveillance period in a large pediatric hospital in Kansas City, whereas the rate in HCs was 3.7% for sapovirus and astrovirus. Together with rotavirus and norovirus [48], 49.1% of the children with AGE in this cohort had a virus detected. Our study provides a better understanding of the importance of viral causes of pediatric AGE and to consider including AdV40/41, astrovirus, and sapovirus in ongoing viral AGE surveillance as well as valuable data on which astrovirus and sapovirus genotypes are responsible for the disease burden in young children. Future studies providing data on the prevalence of these viruses from more recent NVSN years will be important to further confirm the importance of these viruses as the cause of AGE and implications for current norovirus vaccine development [49].

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Potential conflicts of interest.

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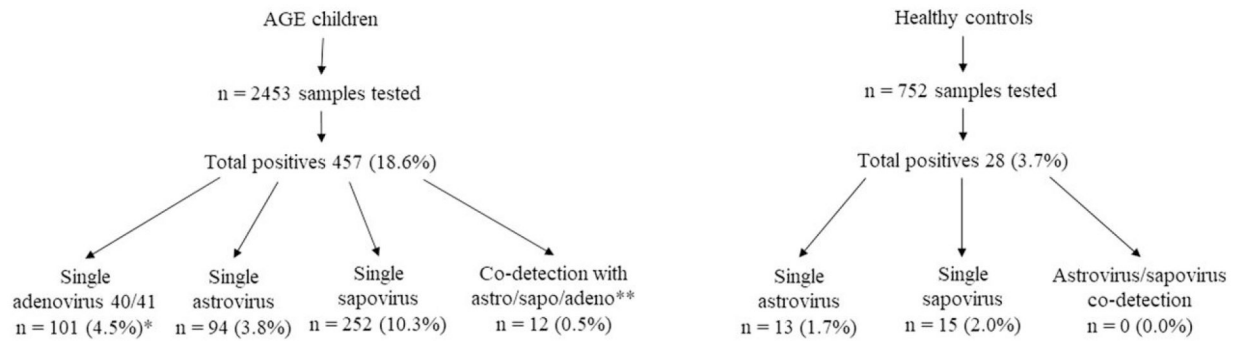


Figure 1.

Study enrollment (n = 3205) of children with acute gastroenteritis (AGE) (n = 2453) and healthy controls (n = 752) and results of adenovirus 40/41 (AdV40/41), astrovirus, and sapovirus testing. *Of the 2453 AGE specimens, 2249 were tested for AdV40/41. **Five astrovirus/sapovirus, 3 adenovirus/astrovirus, and 4 adenovirus/sapovirus co-detections.

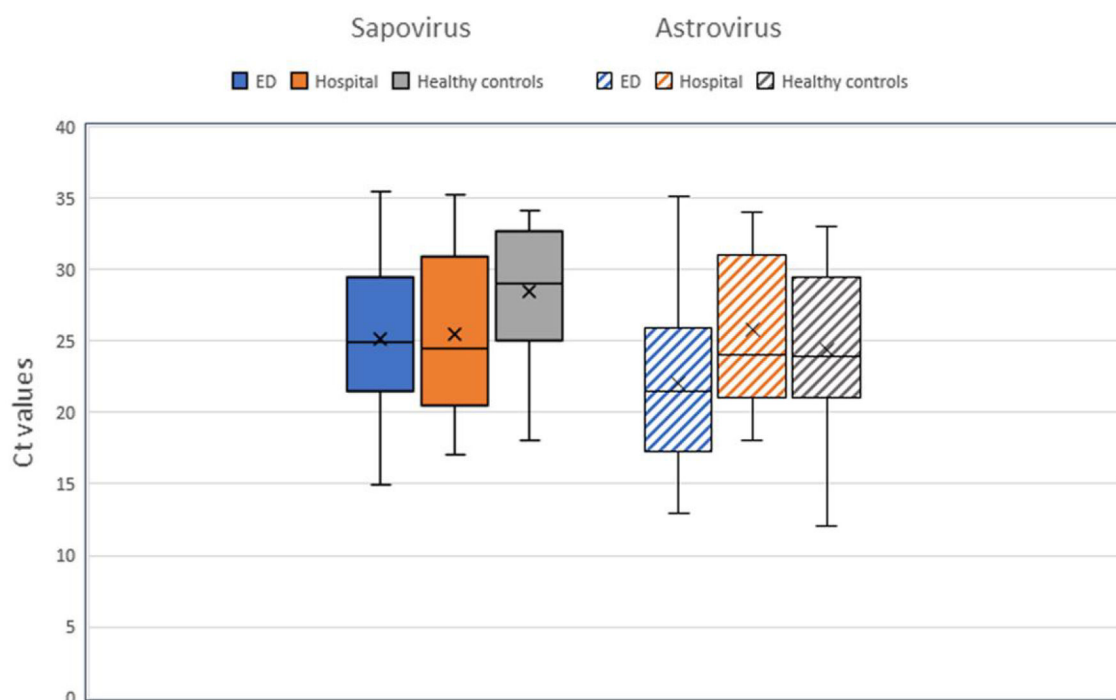


Figure 2. Cycle threshold (Ct) value distribution among astrovirus and sapovirus real-time reverse-transcription polymerase chain reaction–positive samples from children with acute gastroenteritis (seen in emergency department [ED] or hospital) and healthy controls. The bars represent the range of Ct values (min and max).

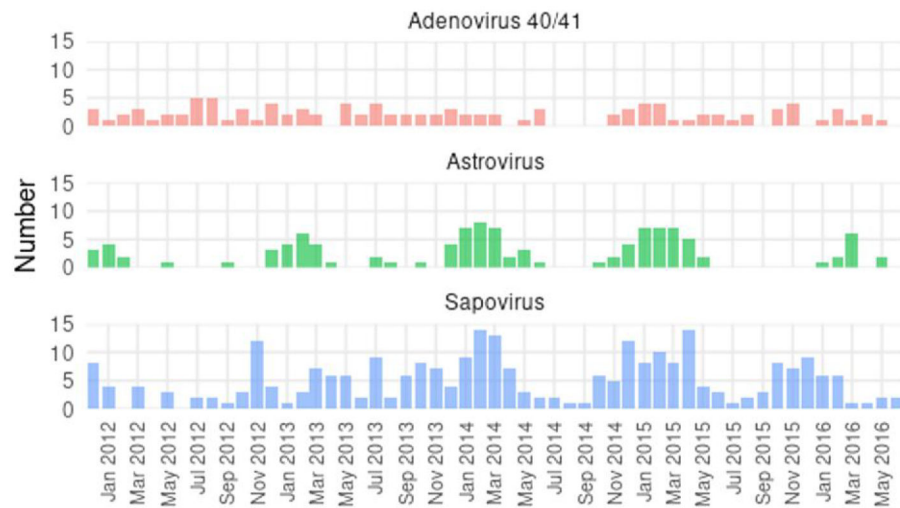


Figure 3. Seasonal distribution of adenovirus 40/41 (red), astrovirus (green), and sapovirus (blue) infections between December 2011 and June 2016 in Children’s Mercy–Kansas City.

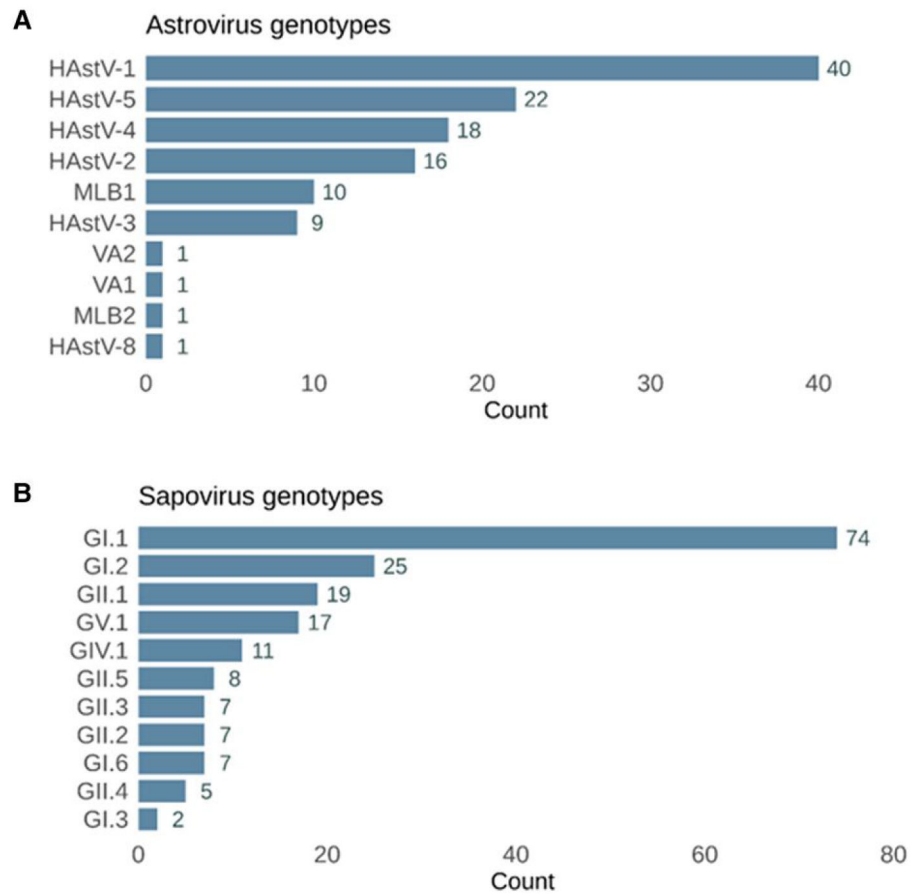


Figure 4.

Genotype distribution of astrovirus (*A*) and sapovirus (*B*) in children aged <5 y enrolled in an acute gastroenteritis study in Children's Mercy–Kansas City, December 2011 to June 2016.

Table 1.

Adenovirus 40/41, Astrovirus, and Sapovirus Test Results for Children With Acute Gastroenteritis Seen in the Emergency Department or as Inpatients (n = 2453^a) and Healthy Controls (n = 752) by Age Group and Setting

Characteristic	Total Tested	Sapovirus Detected	Astrovirus Detected	Adenovirus 40/41 Detected
AGE-inpatient				
0–11 mo	132	13	4	7
12–59 mo	147	12	7	3
≥60 mo	60	3	2	1
Subtotal	339 ^b	28 (8.3%)	13 (3.8%)	11 (5.6%) ^a
AGE-ED				
0–11 mo	478	53	17	31
12–59 mo	1134	146	54	58
≥60 mo	502	34	18	8
Subtotal	2114 ^b	233 (11.0%) ^a	89 (4.2%) ^a	97 (4.7%) ^a
Healthy controls				
0–11 mo	287	4	6	NT
12–59 mo	341	10	7	NT
≥60 mo	124	1	0	NT
Subtotal	752	15 (2.0%)	13 (1.7%)	NT
Total	3205	276 (8.6%)	115 (3.6%)	108 (4.8%) ^a

Abbreviations: AGE, acute gastroenteritis; ED, emergency department; NT, not tested.

^a A subset of 2249 AGE specimens was additionally tested for adenovirus 40/41: 197 AGE-inpatient, 2052 AGE-ED, and 0 healthy control specimens.

^b Totals include specimens with co-detections; 5 astrovirus/sapovirus, 3 astrovirus/adenovirus, 4 sapovirus/adenovirus.

Table 2.

Co-detections With Other Viral, Bacterial, and Parasitic Enteric Pathogens

AGE Pathogen	Total ^a	Co-detection With:							
		Single	Astrovirus	Adenovirus	Norovirus	Rotavirus	Sapovirus	<i>C. difficile</i>	Other Bacteria ^b <i>Giardia</i> ≥ 3 Pathogens ^c
Astrovirus	102	64 (62.7)	...	1 (1.0)	12 (11.8)	3 (2.9)	3 (2.9)	8 (7.8)	3 (2.9) 0 8 (7.8)
Sapovirus	260	176 (67.7)	3 (1.2)	2 (0.8)	18 (6.9)	14 (5.4)	...	30 (11.5)	9 (3.5) 1 (0.4) 7 (2.7)
AdV40/41	108	72 (66.7)	1 (0.9)	...	7 (6.5)	4 (3.7)	2 (1.9)	16 (14.8)	1 (0.9) 0 5 (4.6)

Data are presented as No. (%).

Abbreviations: AdV40/41, adenovirus 40/41; AGE, acute gastroenteritis; *C. difficile*, *Clostridioides difficile*.

^aOne sapovirus-positive specimen was not tested by xTAG Gastrointestinal Pathogens Panel assay.

^b*Shigella*, *Salmonella*, enterotoxigenic *Escherichia coli* (ETEC), Shiga-like toxin–producing *E. coli* (STEC).

^cAstrovirus/*C. difficile*/STEC (n = 1), astrovirus/*C. difficile*/adenovirus (n = 2), astrovirus/sapovirus/*C. difficile* (n = 1), astrovirus/norovirus/*C. difficile* (n = 2), astrovirus/sapovirus/rotavirus (n = 1), sapovirus/*C. difficile*/ETEC (n = 1), sapovirus/*C. difficile*/norovirus (n = 2), sapovirus/adenovirus/*C. difficile* (n = 2), astrovirus/rotavirus/norovirus/*C. difficile* (n = 1), adenovirus/norovirus/rotavirus/*C. difficile*/cryptosporidium (n = 1).

Table 3.

Demographic Characteristics and Adenovirus 40/41, Astrovirus and Sapovirus Test Results of 3205 Children Enrolled in a Medically Attended Acute Gastroenteritis Study in Children’s Mercy–Kansas City, 2011–2016

Characteristic	Enrolled	Sapovirus	Astrovirus	Adenovirus	Negative
Sex					
Male	1724 (53.8)	165 (59.8)	60 (52.2)	58 (53.7)	1446 (53.2)
Female	1481 (46.2)	111 (40.2)	55 (47.8)	50 (46.3)	1272 (46.8)
Total	3205 ^a	276	115	108	2718
Ethnicity					
Non-Hispanic	2373 (74.0)	210 (76.1)	78 (67.8)	83 (76.9)	2011 (74.0)
Hispanic	831 (25.9)	66 (23.9)	37 (32.2)	25 (23.1)	706 (26.0)
Unknown	1 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.0)
Total	3205 ^a	276	115	108	2718
Race					
Black	1348 (42.1)	119 (43.1)	34 (29.6)	46 (42.6)	1156 (42.5)
White	947 (29.5)	88 (31.9)	42 (36.5)	30 (27.8)	788 (29.0)
Unknown/Refused	578 (18.0)	43 (15.6)	29 (25.2)	18 (16.7)	491 (18.1)
Multiracial	269 (8.4)	21 (7.6)	10 (8.7)	12 (11.1)	227 (8.3)
Other race	37 (1.2)	2 (0.7)	0 (0.0)	1 (0.9)	34 (1.3)
Asian	26 (0.8)	3 (1.1)	0 (0.0)	1 (0.9)	22 (0.8)
Total	3205 ^a	276	115	108	2718

Data are presented as No. (%).

^aTotals include specimens with co-detections: 5 astrovirus/sapovirus, 3 astrovirus/adenovirus, 4 sapovirus/adenovirus. A subset of 2249 AGE specimens was tested for adenovirus 40/41.