

AN EVALUATION OF ROTAVIRUS AND ADENOVIRUS ANTIGENS BY THE IMMUNOCHROMATOGRAPHIC METHOD IN SAMPLES WITH AN INITIAL DIAGNOSIS OF ACUTE GASTROENTERITIS

NEVZAT UNAL^{1*}, KERAMETTIN YANIK^{2*}, SEMA AYDOĞDU³, CAFER EROĞLU², MURAT GUNAYDIN⁴, MURAT HOKELEK⁴

¹Adana Numune Education and Research Hospital, Laboratory of Microbiology, Adana - ²Ondokuz Mayıs University, Faculty of Medicine, Department of Medical Microbiology, Samsun - ³Bafra State Hospital, Laboratory of Microbiology, Samsun, Turkey; ⁴Istanbul University, Cerrahpaşa Faculty of Medicine, Department of Medical Microbiology, İstanbul, Turkey

ABSTRACT

Aim: Infectious gastroenteritis is one of the most significant causes of morbidity and mortality, particularly in children. Rotavirus and enteric adenovirus are among the most common causes of viral diarrhoea among young children worldwide. The aim of this study of samples sent to the laboratory with an initial diagnosis of gastroenteritis was to define the prevalence of rotavirus and enteric adenovirus according to age and seasonal distribution.

Materials and methods: The presence of rotavirus and enteric adenovirus antigens was researched using a qualitative immunochromatographic method in stool samples from patients with gastroenteritis who were admitted to Ondokuz Mayıs University Hospital between January 2009 and April 2013. Chi-square tests were used for comparisons between groups.

Results: Of the 2185 stool samples examined, 343 were determined to be positive for rotavirus antigen and 157 for adenovirus antigen. The highest rate of rotavirus antigen positivity (212 cases, 62%) was seen in the 0-23 months age group. Seasonally, rotavirus gastroenteritis was seen mostly in winter (144 cases, 42%). The highest rate of adenovirus positive samples was determined in the 0-23 month's age group (86 cases, 55%). The distribution of the adenovirus cases in autumn, winter, spring and summer was 54 (34%), 31 (20%), 27 (17%) and 45 (29%) cases, respectively. Sixty-three specimens (3% of cases) were positive for both antigens.

Conclusion: As rotavirus and adenovirus were found to be significant factors in gastroenteritis in the 0-23 months age group, in the differential diagnosis of infectious gastroenteritis in this age group of patients, it is important to consider rotavirus as a factor, particularly in the winter months, while adenovirus should be considered in summer and autumn. Rotavirus and adenovirus can be diagnosed quickly by the immunochromatographic test from stool samples, making it possible to prevent the development of dehydration and the unnecessary use of antibiotics.

Key words: Viral gastroenteritis, rotavirus, adenovirus, immunochromatographic method.

DOI:10.19193/0393-6384_2016_1_12

Received May 30, 2015; Accepted November 02, 2015

Introduction

Infectious gastroenteritis is one of the most significant causes of morbidity and mortality, particularly in infants and young children, throughout the world. Diarrhoea is the second biggest cause of death in children under 5 years of age in the world, with a rate of 9%⁽¹⁾. Rotavirus is the leading cause of acute gastroenteritis requiring hospitalization among infants and young children around the world and the great majority of cases occur in the first 2

years of life^(2,3). Viruses are implicated in the majority of non-bacterial gastroenteritis. Rotavirus and enteric adenovirus (serotype 40, 41) are among the most common causes of viral diarrhoea in young children worldwide^(2,4). Each year, rotavirus causes an estimated 111 million episodes of diarrhea requiring only home-care, 25 million clinic visits, 2 million hospitalizations, and 352,000-592,000 deaths in children <5 years of age⁽⁵⁾.

Rotaviruses are primarily transmitted via the fecal-oral transmission due to their resistance to

dryness and the fact that they can live on objects such as hands, furniture and toys. They are the most common cause of gastroenteritis, which can lead to severe dehydration. Enteric adenoviruses are the second most common cause of acute and prolonged diarrhoea after rotaviruses, and are most frequent among children in the 0-3 year old age group. The incidence is about 5-15% and it can occur in all seasons (6). Symptoms of gastroenteritis begin with fever, diarrhea and vomiting at the end of an 8-10 day incubation period. Because of the long-term continued viral excretion even after the disappearance of symptoms, enteric adenoviruses are able to cause an epidemic in children.

Imaging with an electron microscope, cell culture, serological tests and the detection of antigens and nucleic acids can be used in diagnosis⁽⁷⁾. Tests selected in the laboratory for the routine diagnosis must be easy, fast and inexpensive, and have high-sensitivity. In the fight against viral gastroenteritis, the incidence of infectious pathogens and the factors affecting them should be followed by surveillance studies, since these infections are a major cause of mortality and effective treatment methods have not yet been developed. Epidemiological surveillance and the control of viral gastroenteritis is of prime importance due to the lack of effective treatments and unnecessary use of antibiotics.

In this study of samples sent to the laboratory with an initial diagnosis of gastroenteritis, we aimed to define the prevalence of rotavirus and enteric adenovirus according to age and seasonal distribution.

Materials and methods

Subjects

Data obtained from patients diagnosed with acute gastroenteritis and registered to several clinics and polyclinics of Ondokuz Mayıs University School of Medicine Hospital between January 2009 and April 2013 were retrospectively evaluated. Repeat samples, samples that indicated blood under macroscopic examination or indicated parasites under microscopy, as well as fecal specimens in which were positive for pathogen bacteria like *Salmonella* spp. or were positive for *Entamoeba histolytica* adhesin antigen, were excluded from the study.

Immunochromatographic method

The presence of rotavirus and enteric aden-

ovirus antigens was determined by the qualitative immunochromatographic method (RIDA Quick Rota-Adeno-Combi R-Biopharm AG, Germany) in fresh stool samples. The reported sensitivity of the test for rotavirus is 97.8 and the specificity is 94.4%; sensitivity for adenovirus is 72.7 and the specificity is 98.2%.

Statistical analysis

Chi-square tests were used for comparisons between groups and in statistical evaluation, $p < 0.05$ was accepted as the level of statistical significance.

Results

Of the 2185 stool samples examined, 343 (16%) were determined as positive for rotavirus antigen and 157 (7%) for adenovirus antigen. Of the patients that were positive for rotavirus, 196 (57%) were male and 147 (43%) were female. Ninety-nine of the patients with adenovirus were male (63%) and 58 (37%) were female (Table 1).

		Number of cases of rotavirus (%)	Number of cases of adenovirus (%)	Number of cases of rotavirus + adenovirus (%)	Total number of patients (%)
Gender	Male	196 (57)	99 (63)	42 (67)	1205 (55)
	Female	147 (43)	58 (37)	21 (33)	980 (45)
Total		343 (100)	157 (100)	63 (100)	2185 (100)

Table 1: The distribution of cases of rotavirus and adenovirus by antigen detection agent and gender.

Although the rate of adenovirus positivity was found to be significantly higher in men than in women ($p < 0.05$), there was not a significant difference between the two genders in terms of rotavirus positivity. The highest rate of rotavirus antigen positivity was seen in the 0-23 months age group (212 cases, 62%). The distribution of positive results for the rotavirus antigen in the other age groups was as follow: 83 cases (24%) aged 2-6 years, 26 cases (7%) aged 7-10 years, 19 cases (6%) aged 11-20 years, and 3 cases (1%) over the age of 40 (Table 2).

The rotavirus antigen was found to be statistically significantly more prevalent ($p < 0.05$) in the 0-2 year old age group in comparison to other age groups. Seasonally, rotavirus gastroenteritis was seen mostly in the winter months (December, January and February) (144 cases, 42%), with significantly more cases becoming in this period than

at any other time of year ($p < 0.05$). The distribution of positive cases of rotavirus and adenovirus in the different months of the year is presented in Figure 1.

	Number of cases of rotavirus (%)	Number of cases of adenovirus (%)	Number of cases of rotavirus + adenovirus (%)	Total number of patients (%)
0-23 months	212 (62)	86 (55)	33 (52)	1018 (47)
2-6 years	83 (24)	32 (21)	13 (21)	517 (24)
7-10 years	26 (7)	21 (13)	8 (13)	246 (11)
11-20 years	19 (6)	13 (8)	8 (13)	320 (15)
21 years >	3 (1)	5 (3)	1 (1)	84 (3)
Total	343 (100)	157 (100)	63 (100)	2185 (100)

Table 2: Distribution of ages of patients in whom rotavirus and adenovirus antigens were detected.

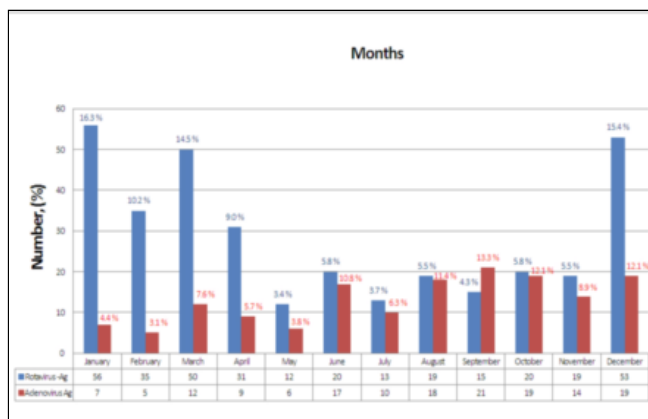


Fig 1: Distribution of positive cases of rotavirus and adenovirus in the different months of the year.

The highest rate of positive detection of adenovirus was found in the 0-23 months age group (86 cases, 55%). In the other age groups, rates of adenovirus positivity were as follows: 32 cases (21%) aged 2-6 years, 21 cases (13%) aged 7-10 years, 13 cases (8%) aged 11-20 years, and 5 cases aged 21 years and over (Table 2). There was no statistically significant difference among the age groups in terms of adenovirus positivity ($p > 0.05$). The distribution of the adenovirus cases according to the seasons (autumn, winter, spring and summer) was 54 (34%), 31 (20%), 27 (17%) and 45 (29%), respectively. Seasonally, cases of adenovirus gastroenteritis were seen mostly in the autumn and summer, and there were significantly more cases in these months.

Positive results for both adenovirus and rotavirus antigens in the same specimen were found in 63 cases (3%).

Rotavirus and adenovirus rates were assessed over five years. The rotavirus antigen positivity rate was 13% in 2009, 11% in 2010, 21% in 2011, 17% in 2012 and 22% in the first four months of 2013. The positivity rate for adenovirus was 8% in 2009, 7% in 2010, 12% in 2011, 5% in 2012 and 6% in the first four months of 2013.

Discussion

In the treatment of patients and prediction of prognosis, determination of gastroenteritis factors is most important during infancy when the disease can have severe consequences. The most common cause of viral gastroenteritis reported in the literature is rotavirus, with adenovirus in second place. Rotaviruses can lead to epidemics in places where people aggregate. Members of the community who stay in kindergarten or live in homes for the elderly or garrisons, as well as health-care workers and tourists, are affected by rotavirus more frequently than other people⁽⁸⁾. Several international studies showed that the incidence of rotavirus as a cause of gastroenteritis in children ranged from 16.5% to 53%^(3,9,10). The rates of positive observations of rotavirus antigens in domestic studies were reported as 9.7-39.8% in patients with gastroenteritis⁽¹¹⁻²⁰⁾. It is clear that the data given on the incidence and distribution of serotypes within these studies are different from each other. Rotavirus incidence was found to be 16% in our study. This difference may stem from differences in people’s awareness about health and sanitary issues and differences in health care services.

The incidence of rotavirus gastroenteritis gradually decreases with age⁽¹³⁾. In our study, incidences of rotavirus gastroenteritis were observed most frequently in the 0-23 month old age group and inversely proportional to the age of positivity rate has decreased.

It has been reported that gastroenteritis associated with rotavirus shows seasonal variations, with 70-95% of cases occurring in winter and 0-20% in summer^(3,6,21). However, unlike most of the other studies, Dağı et al. found that the rate of rotavirus antigen positivity was significantly higher in September and November whereas the highest rate of adenovirus positivity was in August. They thought that these season differences might be caused by the data being obtained in 2013⁽²²⁾. In our study, the high incidence of rotavirus gastroenteritis during the winter was similar to findings in most of

the studies in the literature, with significantly higher levels than in the other seasons.

Ahlywalia et al. reported that the incidence of adenovirus gastroenteritis during the period of infancy was 4.1%⁽²³⁾. Several other studies done in our country have been carried out, showing a range of incidence rates from 2.9 to 16.2%^(15-17, 4-25). The rate identified in this study (7%) is consistent with the information in the literature. Previous studies have reported that gastroenteritis which depends on adenovirus can be seen in all age groups, although it is most commonly seen in children under the age of 2 years^(16,26,27). Consistent with the results of other studies, in our study, more than half of the children (55%) with adenovirus were in the 0-23 months age group. Positive test results for adenovirus were seen in all seasons but most frequently in the autumn and summer months in our study.

Several studies have shown that concurrent positive results for rotavirus and adenovirus were found in 0.4-7.2% of cases^(17,22,28-32). Our study found that 3% of patients with gastroenteritis were positive for both rotavirus and adenovirus; variance among different studies may be due to the number of specimens examined.

The results of this study show that as rotavirus and adenovirus are significant factors in gastroenteritis in the 0-23 months age group, it is important to consider rotavirus as a factor in the differential diagnosis of infectious gastroenteritis in this age group. For rotavirus gastroenteritis this is particularly important in the winter months, while adenovirus is an important factor in summer and autumn. Rotavirus and adenovirus can be determined quickly by the immunochromatographic test from stool samples, preventing the patient from developing dehydration and limiting the unnecessary use of antibiotics.

References

- 1) World Health Organization (WHO). World health statistics; 2014. Available from: http://apps.who.int/iris/bitstream/10665/112738/1/9789240692671_eng.pdf
- 2) Parashar UD, Gibson CJ, Bresse JS, Glass RI. *Rotavirus and severe childhood diarrhea*. Emerg Infect Dis. 2006; 12: 304-306.
- 3) Patricia BV, Quach C, and Bonten M. *Nosocomial rotavirus infections: a meta-analysis*. Pediatrics 2012; 129: 4: 1011-1019.
- 4) Koletzko S, Osterrieder S. *Acute infectious diarrhea in children*. Dtsch Arztebl Int. 2009; 106: 33: 539-48.
- 5) Parashar UD, Hummelman EG, Bresee JS, Miller MA, Glass RI. *Global illness and deaths caused by rotavirus disease in children*. Emerg Infect Dis. 2003; 9: 5: 565-72.
- 6) Topçu AW, Söyletir G, Doğanay M. *Enfeksiyon Hastalıkları ve Mikrobiyolojisi. 3. Baskı*. İstanbul: Nobel Tıp Kitabevi. 2008.
- 7) Rhee EG, Barouch DH. *Adenoviruses*. in Mandell GL, Bennett JE, Dolin R, eds. 7th ed. *Mandell, Douglas and Bennett's Principles and Practice of Infectious Diseases*. Philadelphia: Elsevier, Churchill, Livingstone 2009: 143: 2027-33
- 8) Dormitzer PR. *Rotaviruses*. in Mandell GL, Bennett JE, Dolin R, eds. 7th ed. *Mandell, Douglas and Bennett's Principles and Practice of Infectious Diseases*. Philadelphia: Elsevier, Churchill, Livingstone 2009: 150: 2105-15.
- 9) Köksal AO, Köksal T. *Frequency of rotavirus children with acute gastroenteritis between 0-5 years of age in Ankara*. Yeni Tıp Dergisi 2013; 30.2: 121.
- 10) Gagneur A, Nowak E, Lemaitre T, Segura JF, Delaperrière N, et al. *Impact of rotavirus vaccination on hospitalizations for rotavirus diarrhea: the IVAN-HOE study*. Vaccine 2011; 29: 21: 3753-3759.
- 11) Iraz M, Ceylan A. *Frequency of Rotavirus in Children with Acute Gastroenteritis Between 0-5 Years of Age*. Ankem Derg. 2013; 27: 1: 2-6.
- 12) İlkaç M, Şahin A, Nazik H, Öngen B. *Investigation of Rotavirus Frequency and Following up the Rotavirus Season among Children with Acute Gastroenteritis: Evaluation of Five-year Results*. Ankem Derg, 2012; 26:1: 25-29.
- 13) Şimşek Y, Bostancı İ, Bozdayı G, Öner N, Ahmed A, Rota S ve ark. *Frequency and serotype features of rotavirus in 0-5 age children with acute gastroenteritis*. Türkiye Klinikleri J Pediatr. 2007; 16: 165-70.
- 14) Meral M, Bozdayı G, Ozkan S, Dalgıç B, Alp G, et al. *Rotavirus Prevalence in Children with Acute Gastroenteritis and the Distribution of Serotypes and Electropherotypes*. Mikrobiyol Bul. 2011; 45:1: 104-12.
- 15) Koçak M, Çalışkan E, Köksal AO. *Rotavirus Frequency in Children With Acute Gastroenteritis Who Were Hospitalized in Keçiören Education and Research Hospital Pediatric Clinic*. Ankem Derg. 2014; 28.4: 134-137.
- 17) Gül M, Garipardıç M, Çıragıl P, Aral M, Karabiber H, Güler. *Investigation of Rotavirus and Adenovirus types 40/41 in Children with Gastroenteritis between 0-5 Years of Age*. Ankem Derg. 2005; 19: 64-67.
- 18) Biçer S, Bezen D, Sezer S, Yavuzcan D, Akpınar Tekgündüz S, et al. *Rotavirus and Adenovirus Infections in Acute Gastroenteritis Cases in Pediatric Emergency Service*. Ankem Derg. 2006; 20: 206-209.
- 19) Karakus YT, Savran B, Dibeklioglu SE. *Incidence of rotavirus and adenovirus 40/41 in children and infants*. European Journal of Medical Sciences 2014; 1:1: 22-25.
- 20) İnan N, Ünsür EK, Demirel A, Mamçu D, Sönmez E, et al. *Investigation of Frequency of Rotavirus, Adenovirus and Norovirus in Patients with Acute Gastroenteritis*. Ankem Derg. 2014; 28:1: 14-19.
- 21) Tekin M, Topaloğlu N, Yıldırım Ş, Binnetoğlu K, Kaymaz N, et al. *Frequency Of Rotavirus in Children With Acute Gastroenteritis*. International Journal of Clinical Research 2014; 2: 1: 18-20.

- 22) Koopmans M, Brown D. *Seasonality and diversity of group A rotavirus in Europe*. Acta Paediatr Suppl. 1999; 88: 38-41.
- 24) Dađı HT, Fındık D. *Investigation of rotavirus and adenovirus antigens in patients with acute gastroenteritis*. Journal of Clinical & Experimental Investigations/Klinik ve Deneysel Arastirmalar Dergisi 2014; 5: 2: 256-260.
- 25) Ahluwalia GS, Scott-Taylor TH, Klisko B, Hammond GW. *Comparison of detection methods from enteric clinical specimens*. Diagn Microbiol Infect Dis. 1994; 18: 161-166.
- 26) Balkan ÇE, Çelebi D, Çelebi Ö, Altoparlak Ü. *Investigation of Rotavirus and Adenovirus Frequency Among 0-5 Years Old Children with Acute Gastroenteritis in Erzurum*. Türk Mikrobiyol Cem Derg. 2012, 42.2: 51-4.
- 26) Yazıcı V, Manzur Y, Akbulut A. *Investigation of Rotavirus and Enteric Adenovirus Infection Frequency in Cases with Acute Gastroenteritis*. Klimik Dergisi 2013; 26:1: 13-6.
- 27) Akıncı N, Erener Ercan T, Yalman N, Eren A, Sevege B, Ercan G. *Adenovirus and Rotavirus in Children with Acute Gastroenteritis*. Journal of Pediatric Infection 2007; 1: 98-101.
- 28) Bates PR, Bailey AS, Wood DJ, Morris DJ, Couriel JM. *Comparative epidemiology of rotavirus, subgenus F (types 40 and 41) adenovirus, and astrovirus gastroenteritis in children*. J Med Virol 1993; 39: 224-228.
- 29) Ozdemir S, Delialiođlu N, Emekdaş G. *Investigation of rotavirus, adenovirus and astrovirus frequencies in children with acute gastroenteritis and evaluation of epidemiological features*. Mikrobiyoloji Bulteni 2010; 44:4: 571.
- 30) Alicem T. *The frequency of rotavirus and enteric adenovirus in children with acute gastroenteritis in Mardin*. J Clin Exp Invest 2010; 1.1.
- 31) Altındıř M, Beřtepe G, Çeri A, Yavru S, Kalaycı R. *Frequency Of Rotavirüs And Enteric Adenovirüs Infection in Children With Acute Gastroenteritis*. SDÜ. Tıp Fak. Derg. 2008; 15: 2: 17-20.
- 32) Bayraktar B, Toksoy B, Bulut E. *Detection of Rotavirus and Adenovirus in Children with Acute Gastroenteritis*. Klinik Dergisi 2010;23, 15-7.
- 33) Balcı YI, Polat Y, Çövuıt İE, Canural R, Görüřen İ, et al. *Frequency of rotavirus and adenovirus types 40/41 in children with gastroenteritis between 0-5 years of age in Denizli*. Yeni Tıp Derg 2010; 27:1: 15-7.

Corresponding author

NEVZAT ÜNAL, MD

Adana Numune Education and Research Hospital

Laboratory of Microbiology

Adana

(Turkey)