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Research Article

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Prevalence, Diagnosis, and Seasonal Patterns of Enteric Viruses Among Hospitalized Adults with Acute Gastroenteritis in the Central Region of Ukraine

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ABSTRACT

Acute infectious gastroenteritis (AIG) remains one of the leading causes of mortality in children under 5 years of age worldwide. Among the wide range of causative agents of this infection, rotavirus has been the most prevalent pathogen over the past decade; however, it is currently being progressively displaced by another agent, namely norovirus.

Our study analyzed stool samples from 265 hospitalized patients aged 18 to 90 years who presented with symptoms of acute gastrointestinal infection at the Saint Michael Clinical Hospital in Kyiv, Ukraine, between November 2024 and April 2026. Testing was performed using a real-time multiplex PCR test system. According to the results, viruses (noroviruses, rotaviruses, astroviruses, adenoviruses, and sapoviruses) were identified as a monoinfection in 44.9% of the samples. Norovirus was detected in the majority of positive cases (32.03%), followed by rotavirus (10.54%).

Norovirus activity was observed throughout the year, with peak incidence recorded in December and during the spring season. Rotavirus activity peaked in early summer, whereas adenovirus was detected exclusively in winter and spring. Sapovirus and astrovirus occurred rarely, predominantly during the autumn and winter months.

Keywords: Gastrointestinal Infection, Diarrhea, Acute Gastroenteritis, Polymerase Chain Reaction, Ukraine

Introduction

Acute gastrointestinal infections represent the second most common category of infectious diseases globally, surpassed only by respiratory tract infections, particularly in pediatric populations.

According to data from the World Health Organization (WHO), more than 1.7 billion cases of diarrheal diseases are recorded annually worldwide. Despite a reduction in mortality achieved through the implementation of rotavirus vaccination and the widespread use of oral rehydration solutions, diarrheal diseases rank third among the causes of death in children under five years of age globally, with the highest burden observed in Africa, South America, and Southeast Asia.

Increased accessibility and frequency of global travel have led to an unprecedented spread of infectious diseases worldwide, with gastrointestinal disorders being one of the most frequent health concerns among travelers and immigrants.

Acute infectious gastroenteritis (AIG) is primarily caused by viruses, bacteria, and parasites. These pathogens are transmitted to the host via various routes, including the fecal-oral route, person-to-person contact, and via fomites.

An etiological diagnosis is not always mandatory and should be established primarily when it directly influences patient management or carries implications for public health, such as during outbreaks or when isolation is required [1]. In such scenarios, early identification is essential to optimize clinical and epidemiological patient management.

Among the primary viral agents associated with childhood AIG, rotavirus A, norovirus, and human adenovirus represent the predominant etiological agents worldwide. However, the etiology of a significant proportion of AIG cases (up to 40%) remains unresolved, particularly in developing countries. It is

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estimated that this diagnostic gap accounts for one-third of all AIG cases, which may suggest that less common viral agents play a role in disease manifestation.

Materials and Methods

This study evaluated stool samples from 265 adult patients aged 18 to 90 years (mean age: 39.7 ± 19.3 years) who were admitted with symptoms of acute infectious gastroenteritis to the infectious disease department of the Saint Michael Clinical Hospital in Kyiv, Ukraine, between November 2024 and April 2026, Samples were collected within the clinical department and transferred to the DNA Diagnostics Laboratory. Viral nucleic acids (DNA and RNA) were purified in accordance with the diagnostic procedures of the Centers for Disease Control and Prevention (CDC).

Prior to amplification, nucleic acid extraction was performed using the "B-Prep-100" reagent kit (manufactured in Poland) designed for low-copy samples [2]. Nucleic acids were eluted into a final volume of 50 µL. PCR amplification was carried out on a CFX-96 Real-Time PCR Detection System (Bio-Rad, California, USA) according to the manufacturer's operational instructions.

Detection of viral nucleic acids was performed using the RevoDx Gastro Viral Pathogen Detection Kit (manufactured in Turkey), a multiplex real-time PCR test system. This assay utilizes TaqMan technology, featuring an internal oligonucleotide probe positioned between two PCR primers. The probe is modified with a fluorescent reporter dye at the 5' end and a quencher molecule at the 3' end. During the elongation step of PCR replication, the fluorescently labeled probe hybridizes to the target DNA template and is cleaved by the 5'→3' exonuclease activity of Thermus aquaticus (Taq) DNA polymerase. The probe is cleaved exclusively during target DNA replication, resulting in the physical separation of the reporter dye and the quencher. The resulting PCR products are monitored in real time via an exponential increase in fluorescent signal intensity with each successive amplification cycle [3,4]. The Ct (cycle threshold) parameter corresponds to the specific amplification cycle number at which the fluorescence signal exceeds a fixed baseline threshold. PCR amplification is performed directly on the DNA and RNA extracted from patient samples. Pathogen DNA/RNA detection is carried out via two distinct multiplex reactions, which simultaneously amplify an in vitro transcribed RNA internal control to verify extraction efficiency and monitor for PCR inhibition.

This multiplex PCR panel enables the concurrent detection of 5 major gastrointestinal pathogens: Adenovirus (F40/41), Astrovirus, Norovirus GI and GII, Rotavirus A, and Sapovirus (I, II, IV, V). The assay features high sensitivity and specificity combined with a short turnaround time, confirming its utility for the early diagnosis of acute hospital-acquired infections [5].

Results

Out of the 256 analyzed stool samples collected from adult patients presenting with acute diarrhea, 124 (44.9%) tested positive for at least one of the five target enteric viruses. All confirmed viral cases occurred exclusively as monoinfections; no cases of mixed viral coinfections were detected in this cohort. Norovirus genogroups I/GII was the most frequently identified pathogen, accounting for 32.03% (82/256) of cases, followed by Rotavirus A at 10.54% (27/256), Astrovirus at 4.29% (11/256), enteric Adenovirus (F40/41) at 1.17% (3/256), and Sapovirus (I, II, IV, V) at 1.17% (3/256) [6].

The correlation between patient age groups and the isolated enteric viral pathogens is detailed in Table 1. Patient age ranged from 18 to 90 years, with a mean age of 39.3 ± 19.7 years. Norovirus was confirmed as an etiological agent across all age brackets. This pathogen was most frequently detected in young adults aged 20–29 years (32.9%, 27 cases) and individuals of middle age spanning 30-39 years (24.4%, 20 cases). Patients under 20 years accounted for 7.3% (6 cases), while elderly patients aged 70 years and older comprised 12.2% (10 cases) of the total viral infections. Rotavirus demonstrated a lower overall prevalence but was present across all age cohorts, with higher frequencies observed in the population under 20 years (22.2%, 6 cases) and individuals aged 20–29 years (29.6%, 8 cases). Astrovirus infections were evenly distributed across the 20–29, 30–39, 40–49, and 70+ age brackets, each accounting for 25.0% (2 cases each) of positive astrovirus findings. The prevalence of enteric Adenovirus and Sapovirus remained low: Adenovirus was identified in young adults aged 20–39 years (33.3%, 1 case per decade), while Sapovirus was detected in the 20–29 age bracket (66.7%, 2 cases) and the 60–69 cohort (33.3%, 1 case).

Table 1: Age Distribution of Viral Pathogens in Patients with Acute Gastroenteritis

Age Category (years)	Astrovirus (n=11)	Norovirus (n=82)	Rotavirus (n=27)	Adenovirus (n=3)	Sapovirus (n=3)	Total (n=124)
Under 20	1 (9.1%)	6 (7.3%)	6 (22.2%)	2 (66,7%)	0 (0.0%)	15 (12.1%)
20–29	2 (18.2%)	27 (32.9%)	8 (29.6%)	0 (0.0%)	2 (66.7%)	39 (31.5%)
30–39	2 (18.2%)	20 (24.4%)	2 (7.4%)	1 (33.3%)	0 (0.0%)	25 (20.2%)
40–49	2 (18.2%)	11 (13.4%)	2 (7.4%)	0 (0.0%)	0 (0.0%)	15 (12.1%)
50–59	0 (0.0%)	5 (6.1%)	2 (7.4%)	0 (0.0%)	0 (0.0%)	7 (5.6%)
60–69	0 (0.0%)	3 (3.7%)	4 (14.8%)	0 (0.0%)	1 (33.3%)	8 (6.5%)
70 and older	2 (18.2%)	10 (12.2%)	3 (11.1%)	0 (0.0%)	0 (0.0%)	15 (12.1%)

According to the seasonal analysis, enteric viral infections were documented across all seasons (median number of cases per season: 30.5; $p = 0.005$), displaying a significantly higher frequency during the winter-spring period (Table 2). The peak prevalence was recorded in spring (36 cases, 29.0%) and winter (32 cases, 25.8%). Norovirus infections predominated during winter, spring, and

autumn, accounting for 28 (87.5%), 25 (69.4%), and 21 (72.4%) of positive viral samples within those seasons, respectively. In contrast, rotavirus infections demonstrated an elevated case count during the spring-summer transition, with 8 (22.2%) and 12 (44.4%) cases reported, respectively [7-10]. Astrovirus did not exhibit a pronounced seasonal pattern; a low number of sporadic cases were recorded throughout the year, with a minor increase during summer and autumn (3 cases each; 11.1% and 10.3%, respectively). Solitary episodes of adenovirus infection were restricted to the winter-spring period, while sapovirus cases occurred strictly during summer and autumn.

Table 2: Seasonal Distribution of Viral Pathogens in Patients with Acute Gastroenteritis

Season	Total Cases	Norovirus	Rotavirus	Adenovirus F40/41	Astrovirus	Sapovirus
Winter	32 (25.8%)	28 (87.5%)	1 (3.1%)	1 (3.1%)	2 (6.3%)	0 (0.0%)
Spring	36 (29.0%)	25 (69.4%)	8 (22.2%)	2 (5.6%)	1 (2.8%)	0 (0.0%)
Summer	27 (21.7%)	11 (40.7%)	12 (44.4%)	0 (0.0%)	3 (11.1%)	1 (3.7%)
Autumn	29 (23.3%)	21 (72.4%)	3 (10.3%)	0 (0.0%)	3 (10.3%)	2 (6.9%)

Figure 1 outlines the monthly distribution of viral agents in patients with acute diarrhea. Noroviruses were continuously detected throughout the entire calendar year ($p < 0.001$). The peak incidence of norovirus gastroenteritis was logged between November and December. Rotavirus cases were also recorded year-round, displaying significant peaks in March and June ($p = 0.0002$). Astrovirus infections were uniformly distributed across all twelve months without statistically significant seasonal fluctuations ($p = 0.895$). Adenovirus presentation was confined to the colder months, while sapovirus was detected in minimal amounts during the summer and autumn.

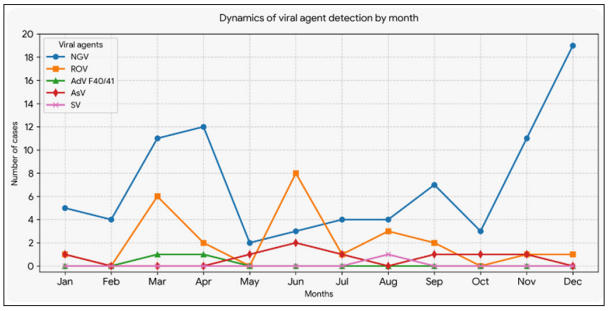


Figure 1: Monthly detection dynamics of viral agents

(Note: Refer to the graph embedded in the manuscript for the monthly distribution curve showing NGV, ROV, AdV F40/41, AsV, and SV case levels).

Consequently, statistically verified and significant monthly variations in incidence were established exclusively for Norovirus and Rotavirus ($p < 0.05$). The remaining three pathogens (Adenovirus, Astrovirus, Sapovirus) displayed a sporadic or completely uniform distribution pattern without direct correlation to specific months of the year.

Discussion

Pathogenic enteric viruses, characterized by a fecal-oral transmission route and intestinal replication, represent a primary source of global morbidity and mortality. Localized intestinal infections caused by enteric viruses such as Adenoviridae (adenoviruses A, F, and G), Caliciviridae (norovirus and sapovirus), Astroviridae (astrovirus), and Reoviridae (rotavirus)-induce acute gastroenteritis characterized by mucosal inflammation, disruption of the epithelial barrier, malabsorption, and severe diarrhea [11-13].

Traditional etiological diagnosis of gastrointestinal infections relies on stool culture, antibiotic susceptibility testing, microscopic evaluation for parasites, and antigen detection via enzyme-linked immunosorbent assays (ELISA).

While conventional bacterial culture remains the gold standard for enteric bacterial pathogens, classic methodologies feature distinct limitations: low diagnostic sensitivity and extended processing times mean that 60% to 70% of cases remain etiologically undiagnosed. The majority of cases with unresolved etiologies are highly likely to be viral in nature [14]. By implementing molecular diagnostic tools (nucleic acid amplification tests) characterized by superior sensitivity and specificity, this diagnostic gap in AIG can be substantially narrowed. Molecular approaches are exceptionally valuable for the swift detection of noroviruses and rotaviruses.

In recent years, multiplex molecular syndromic panels designed for the identification of gastrointestinal pathogens in stool samples from patients with acute diarrhea have emerged as invaluable clinical instruments [15]. These assays successfully complement and, in some settings, replace traditional diagnostic workflows, accelerating epidemiological tracking and outbreak containment. Gastrointestinal multiplex PCR serves as a state-of-the-art diagnostic kit helping clinicians manage diarrheal illnesses. It utilizes nucleic acid testing to rapidly identify unique genomic sequences of target microorganisms. Within emerging healthcare structures or specialized settings, such as military medicine or contained community environments, multiplex PCR diagnostics dramatically accelerate epidemiological investigations and outbreak management [16].

In this study, we evaluated the prevalence of the aforementioned enteric viruses in stool samples from 256 adult patients aged 18 to 90 years (mean age: 39.7 ± 19.3 years) hospitalized with symptoms of acute infectious gastroenteritis at the Saint Michael Clinical Hospital in Kyiv. The overall viral positivity rate among the stool samples was 44.9% (124 cases). Norovirus was confirmed as the most prevalent viral pathogen, identified in 23.03% of adult patients across all age categories [17]. This finding correlates strongly with global epidemiological data. A comprehensive meta-analysis of global acute gastroenteritis data indicates that norovirus is responsible for approximately 24% of all inpatient diarrheal infections [18].

Our findings regarding the monthly distribution of viral infectious agents partially diverge from classic literature, which typically describes a strict winter-spring seasonality for enteric viruses. Based on our data, while the highest case numbers were verified in spring and winter (36 cases, 29.0% and 32 cases, 25.8%, respectively), the prevalence of viral gastroenteritis during summer and autumn remained substantial, recording 27 (21.7%) and 29 (23.3%) cases, respectively [19].

A major non-linear factor responsible for the modification of classic epidemiological trends and the disruption of natural viral seasonality is the ongoing warfare in Ukraine and the corresponding state of martial law. Throughout the 2024–2026 period, the Central Region of Ukraine experienced massive socio-demographic shifts and critical infrastructure strain. Specifically, the anomalous June peak of Rotavirus documented in our study may be a direct consequence of water-sanitation infrastructure destabilization caused by summer power grid disruptions and impaired wastewater treatment facilities, which facilitated waterborne and foodborne transmission pathways [20].

Concurrently, the dual-peak activation of Norovirus (occurring in November-December and March-April) mirrors periods of maximum population density, migration waves, and forced prolonged stays of adult cohorts in enclosed, poorly ventilated spaces. Chronic psycho-emotional stress and secondary immunodeficient states arising within the adult population of Central Ukraine acted as supplementary pathogenic triggers, exacerbating infection severity, increasing hospitalization rates, and ultimately deforming the classic textbook epidemiological curves of incidence [21].

Conclusion

Acute gastrointestinal infections constitute a broad group of diseases with diverse etiologies unified by a fecal-oral transmission mechanism, resulting in gastrointestinal tract damage, systemic intoxication, and dehydration. The clinical challenge of acute gastroenteritis remains highly critical due to several factors: a wide array of potential pathogens, insufficient laboratory identification rates, high population prevalence, significant risks of severe complications, and high mortality rates, particularly in pediatric cohorts. These elements underscore the continuous need for comprehensive monitoring, optimized diagnostic workflows, and improved therapeutic and prophylactic interventions.

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