ORIGINAL ARTICLE

Detection of Rotavirus in Fecal samples from Children under Five Years among pediatric patients with diarrhea in Central Sudan

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Abstract

Background: Rotavirus is severe diarrhea in young children, resulting in over 200,000 deaths worldwide. Various methods, including ELISA and RT-PCR, are used to detect rotavirus in stool samples. Methods: The study screened 98 stool samples from hospitalized children under five with diarrhea in Gezira State, Sudan, using ELISA. Results: Rotavirus was significantly detected (P-value = 0.04), with a higher incidence in males (73%) than in females (27%). The infection was most prevalent in children aged 2-3 years. Discussion: The findings indicate a significant prevalence of rotavirus, consistent with previous studies in Egypt and Sudan. The high infection rate in children aged 2-3 may relate to improved hygiene in older children. A notable 87% prevalence was observed in children from low socioeconomic backgrounds, likely due to poor sanitation. The male predominance in infection rates aligns with similar findings from India. Conclusions: ELISA proves effective for screening rotavirus in fecal specimens. The study's results support the potential introduction of a rotavirus vaccine.

Keywords: Rotavirus, Under Five Years among Children, diarrhea, Central Sud

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INTRODUCTION

Rotavirus is a contagious virus that causes inflammation of the stomach and intestines, leading to severe diarrhea and vomiting, especially in infants and young children (1). Rotavirus is a leading cause of severe gastroenteritis and diarrhea among children under five years of age, contributing significantly to childhood morbidity and mortality, particularly in developing countries (2). Rotavirus is the predominant enteric pathogen linked to severe gastroenteritis in young children, accounting for more than 200,000 fatalities worldwide (3). Acute diarrhea represents a major public health concern in both low- and high-income countries, leading to considerable morbidity and mortality. Diarrhea is a leading cause of pediatric hospitalization globally, particularly among children under the age of five.

Despite the availability of vaccines, rotavirus remains a major health concern, especially in regions with limited healthcare infrastructure, inadequate sanitation, and low vaccination coverage. In Sudan, as in many other low-income countries, diarrheal diseases are a substantial burden on public health, accounting for a large proportion of pediatric hospital admissions and deaths (4). Among the various pathogens responsible, rotavirus stands out as the predominant viral cause, particularly in young children (5).

The introduction of rotavirus vaccines into national immunization programs resulted in a reduction of severe rotavirus-associated diarrhea by approximately 40% in low- and middle-income countries (LMICs) between 2008 and 2016, a period during which one in six children died due to diarrhea (6). More than a quarter (26.93%) of diarrhea-related deaths occurred in children under the age of five, with nearly 90% (89.37%) of these fatalities occurring in South Asia and sub-Saharan Africa (7).

In Central Sudan, a region characterized by a mix of urban and rural populations, rotavirus continues to pose a significant health threat. The challenges in accurately diagnosing rotavirus infections in this setting are exacerbated by the scarcity of diagnostic tools, limited laboratory facilities, and a shortage of trained healthcare professionals. As a result, there is often a delay in diagnosis, leading to inadequate management of the disease, which can result in dehydration, malnutrition, and, in severe cases, death (8). This underscores the importance of accessible, reliable, and cost-effective diagnostic methods. Serological detection of rotavirus in fecal samples has emerged as a valuable tool for confirming rotavirus infections. Immunoassays, such as enzyme-linked immunosorbent assays (ELISA) and rapid antigen detection tests (RDTs), are widely used for their ability to quickly and accurately identify the virus in stool samples, even in resource-limited settings (9). These tests offer several advantages, including their simplicity, cost-effectiveness, and the ability to be used in both clinical and field settings. However, the successful implementation of these diagnostic methods in Central Sudan faces several challenges, including financial constraints, inadequate infrastructure, and logistical barriers.

Latex agglutination assays, commonly employed for rotavirus diagnosis, offer rapid, cost-effective screening for specimens (10). Other diagnostic methods such as latex agglutination (LA), enzyme-linked immunosorbent assay (ELISA), reverse transcription polymerase chain reaction (RT-PCR), immunochromatographic test (ICT), and electron microscopy (EM) are frequently used to detect rotavirus in stool samples (11). Group A rotaviruses are routinely identified using ELISA, which detects the group-specific antigen, as is commonly practiced in the United Kingdom (12).

Despite the progress made in understanding the role of rotavirus in pediatric diarrhea, there remains a lack of comprehensive data on its prevalence and the effectiveness of diagnostic

methods in Central Sudan. This gap in knowledge makes it difficult to formulate evidence-based public health strategies, including vaccination campaigns and targeted interventions. Furthermore, without accurate surveillance, the true burden of rotavirus-related diarrhea may be underestimated, hindering efforts to reduce its impact.

This original article aims to explore the role of serological detection in diagnosing rotavirus among children under five years of age with diarrhea in Central Sudan. By examining the advantages and challenges of these diagnostic methods, we seek to highlight the importance of timely diagnosis in improving clinical outcomes and shaping effective public health responses to this preventable infection.

MATERIALS AND METHODS

Study Area

This cross-sectional study performed in the Department of Microbiology at Wad Medani Pediatric Hospital, Sudan, between June and August 2022.

Ethical consideration

The study was approved by the Wad Medani Pediatric Hospital and the Sudanese Ministry of Health. Before collecting specimens and obtaining consents for participation, all enrolled patients' parents were told about the study's goal.

Data Analysis

The Statistical Package for the Social Sciences (SPSS) version 20 was used to analyze the data.

Sample Collection

Stool samples were collected in containers and labeled with the corresponding patient numbers. All samples were transferred to the virology lab at Wad Madani Pediatric Hospital for ELISA processing.

Sample Storage

Until processed, stool samples were labeled and stored at -20°C.

Assay Procedure

Stool samples were mixed with buffer and used as the antigen in an ELISA assay (DIAGNOSTIC AUTOMATION, INC, Cat. No. 8306-3). Control wells received negative and positive controls, while test wells were filled with stool supernatant. After incubating and washing, Reagent 1 (anti-rotavirus monoclonal antibodies) was added, followed by a second incubation and washing. Reagent 2 (anti-mouse antibodies conjugated to horseradish peroxidase) was then added, followed by another incubation and wash. A chromogen solution was added, and the samples were incubated before adding a stop solution. Results were either visually assessed or measured at 620-650 nm.

Interpretation of ELISA Results

ELISA results were interpreted as follows: a reactive result had an absorbance of 0.15 OD or higher, indicating the presence of rotavirus antigen; a non-reactive result had an absorbance below 0.15 OD, indicating no detectable antigen; and an equivocal result had an absorbance exactly at 0.15 OD.

RESULTS

In this study, the mean age of the subjects was 3.1 ± 3.3 years, with a minimum age of 9 months and a maximum age of 4 years. The highest incidence of the disease was found among the age group of 2-3 years. Among the children, 64 out of 98 (73%) were males and 34 out of 98 (27%) were females. The majority of the subjects (86 out of 98, or 87%) were from low socioeconomic backgrounds, while 12 out of 98 (13%) were from moderate socioeconomic backgrounds. According to locality, 69 out of 98 (71%) were from rural areas, and 29 out of 98 (29%) were from urban areas.

A total of 98 children suffering from watery diarrhea admitted to Wad Medani Pediatric Hospital were diagnosed by ELISA for rotavirus (Fig. 1). Of these, 79 (80.6%) tested positive for rotavirus by ELISA, while 19 (19.4%) tested negative, with $\chi^2 = 3.951$, df = 1, and P-value = 0.04 (Table 1).

Discussion

The results of this study show a significant prevalence of rotavirus among children with suspected diarrhea (P value = 0.04). This finding is consistent with research conducted in Egypt by Ibrahim B. Salwa and colleagues, which reported a 67.7% positivity rate for rotavirus by ELISA and identified a significant association between the severity of disease and rotavirus infection (13).

Our results align with a study conducted in Sudan by Magzoub Abbas Magzoub and others, which concluded that among children under five years old, rotavirus A is the most frequent cause of acute gastroenteritis in developing countries, including Sudan (14). The high incidence of rotavirus among children aged 2-3 years in our study may be attributed to improve mental and health habits in children older than five, which may reduce rotavirus transmission through better hygiene practices. Additionally, the results indicated a significantly higher prevalence of rotavirus in children from low socioeconomic backgrounds (87%). This could be due to inadequate sanitation practices, such as the lack of standardized water cycles or open defecation, which facilitates the transmission of rotavirus. The incidence of rotavirus was also higher in males (73%) compared to females (27%).

This finding aligns with a 2022 study conducted in India by Manaji Pawar and Geeta Karande (14), which reported a higher prevalence of rotavirus infection in male children (52.6%) compared to females (47.4%). The cause of this male predominance remains unclear, but it has been proposed that parents may prioritize male children when seeking healthcare. Additionally, the presence of two X chromosomes in females may provide greater resistance to infections (15).

A historical study conducted over a five-month period (September 1979 to January 1980) identified rotavirus in the feces of 16 out of 116 infants and young children (13.8%) diagnosed with acute, sporadic gastroenteritis (16). The highest incidence of rotavirus infection was observed in the 7- to 12-month age group (18.9%), with males representing 61% of the sample. Additionally, males excreted rotavirus at a significantly higher rate than females (P < 0.01) (17).

The study was conducted between June and August, a period that may coincide with a high transmission rate of the virus. This is consistent with previous findings that indicated higher rates of rotavirus infection during the drier months (November–January, 19.3%) compared to the rainy months (September and October, 8.5%) (18).

Conclusions

This study highlights the substantial health burden of rotavirus among children under five years of age. Furthermore, it demonstrates that ELISA is a useful method for screening rotavirus in fecal specimens due to its sensitivity, specificity, ease of use, and minimal equipment requirements.

Recommendations

This study provides valuable data that can be used to assess the potential benefits of introducing a rotavirus vaccine, particularly in terms of its impact on reducing rotavirus-associated gastroenteritis in young children.

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TABLES AND FIGURES

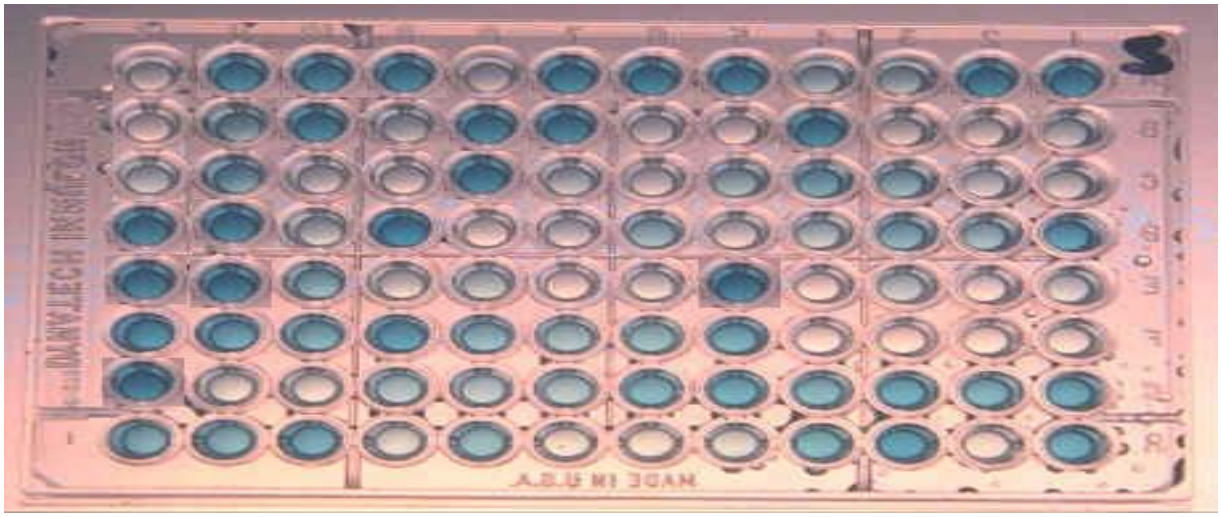

Figure 1: The ELISA results, positive result with blue color and negative result for colorless and showed the positive and negative control.

Table 1: The incidence of Rota results among the study group:

	frequency	Percent%
ELISA Rota positive	79	80.6
ELISA Rota negative	19	19.4
	98	100

($\chi^2 = 3.951$, df = 1, *P-value* = 0.04)

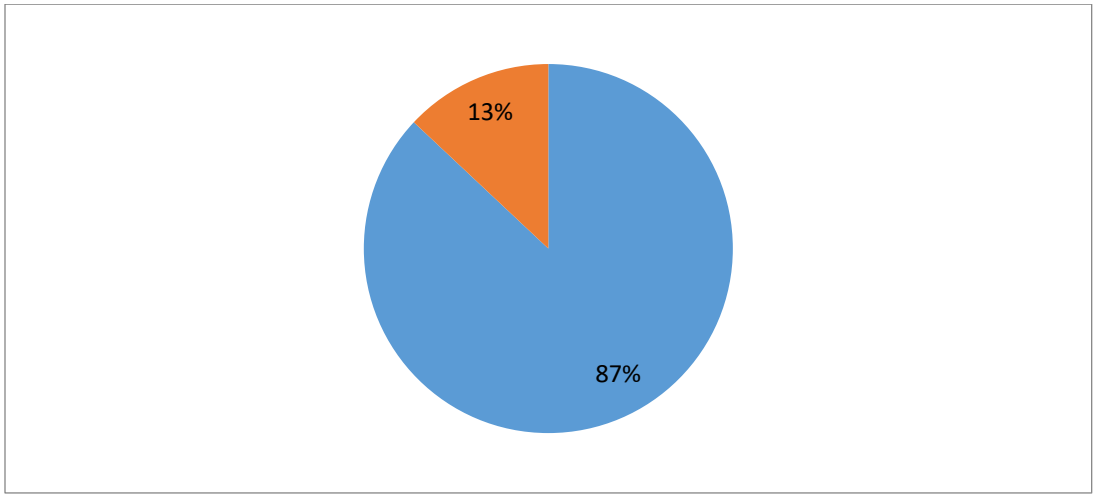

Figure 2: The results show the socioeconomic backgrounds of study cases, were (87%) low socioeconomic and (13%) of moderate socioeconomic status.

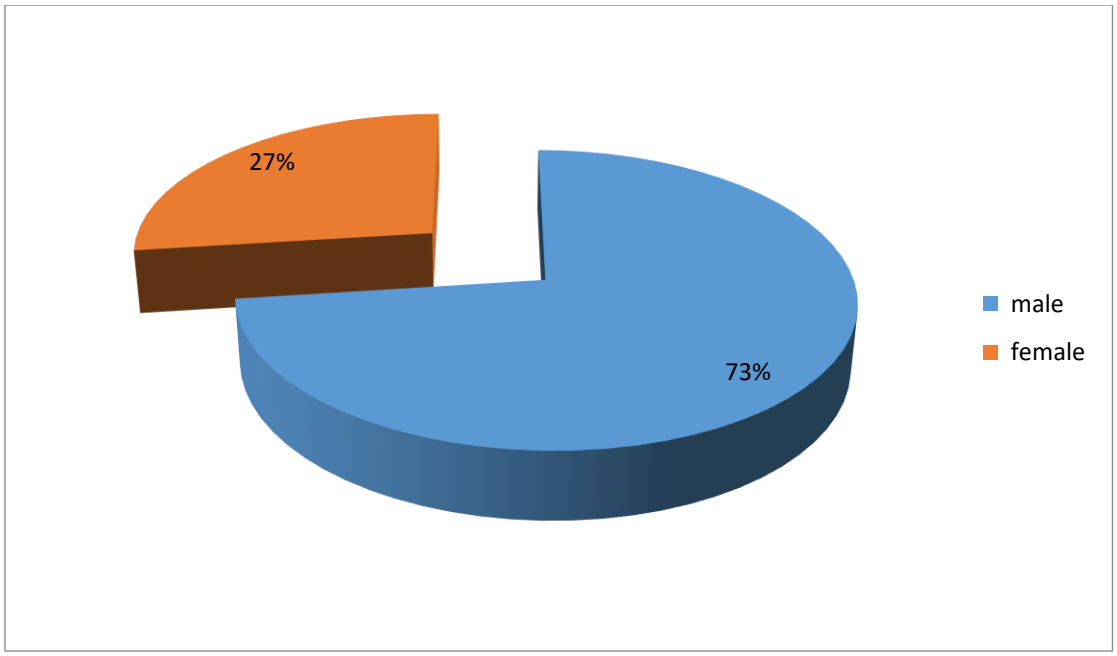

Figure 3: Distribution of gender in the children patients 64/98 (73%) of them were males and 34 /98 (27%) females