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Prevalence of rotavirus infection in children under five years, referring to Indira Gandhi Children's Health Hospital, Kabul, Afghanistan

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Abstract

Background: About 5% of children's deaths in Afghanistan are related to severe rotavirus infections. The aim of this research is to investigate the status of rotavirus gastroenteritis among Afghan children referred to Indira Gandhi Children's Health Hospital from May 2018 to May 2019.

Methods: A cross-sectional study has conducted at Indira Gandhi Children's Health Hospital. 422 stool samples were examined for rotavirus and questionnaires were filled

Results: Out of 422 children (244 (57.8%) male and 178 (42.2%) female) examined, 215 children (131 (60.9%) male and 84 (39.1%) female) were positive for rotavirus infection. Significant differences were observed between positive and negative rotavirus cases from the point of view of diarrhea duration time, frequency of diarrhea/24hours and frequency of vomiting/24 hours.

Conclusion: Still more than 50% of diarrhea cases in children are related to rotavirus. The findings of this research emphasize the fact that the Ministry of Health of Afghanistan should seriously follow control and preventive strategies, especially in warm seasons.

Key words: Rotavirus, Afghanistan, Children, Diarrhea.

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Introduction

Diarrhea is one of the leading cause of childhood deaths in the world(1) and viral diarrhea located among the top causative factor of diarrhea(2). Rotavirus a detectable infectious disease is a common reason of diarrhea in children(3, 4). Rotavirus accounts for 29% of all diarrhea and in 2008 estimated 527,000 mortality among the children under 5 years' age worldwide per year (5-7). Vaccination is the most favored prevention of rotavirus which after vaccination in 2015 the global deaths caused rotavirus declined into 215,000 deaths(8). Key factors related with rotavirus diarrhea are the age less than 2 years, nutritional status, dehydration and earlier complementary feeding (9-11). The rotavirus infection ranges from asymptomatic to symptoms including malaise, vomiting, fever and life threatening diarrhea (6, 12). Prolonged diarrhea results to sever dehydration and leading to death of patients if not treated. Most of rotavirus infected cases occurred in developing countries such as sub-Saharan Africa and South-East Asia(6). Spread of rotavirus firstly occur via fecal-oral. In addition, its transmission can occur by fomites, contaminated hands, rarely by water and food (13). Accessibility to clean water is declined the predominance of most gut infectious disease while the rotavirus cases remain almost unchanged(13).

The association between feeding types and seasonality with rotavirus diarrhea remained unclear. The positive rotavirus cases percentage increased in wet season (14) while Kabul weather is not wet in summer season as well. Furthermore, although breast feeding for under 2 years' age children shows temporary delayed the rotavirus diarrhea but not totally prevented them.

Henceforth, in this study we aimed to perform a cross sectional observational study on rotavirus prevalence under 5years old children in Kabul city.

Materials and Methods

The current study is a cross-sectional study, which conducted from May 2018 to May 2019 at Indira Gandhi Children's Health Hospital. The study was conducted among children aged 1 to 60 months presenting with acute diarrhea, which was defined as passage of three or more loose stools per day lasting less than 14 days.

The children who had acute diarrhea and vomiting for less than two weeks were included in the program, and the collected stool samples were transported within 5 min of sample collection to a centrally placed refrigerator found in the pediatric sample room and stored at 2-8 °C. Thereafter, the stool samples were collected by a well-trained research assistant and transported several times daily to the laboratory. At the laboratory, the stool samples were frozen at − 20 °C prior to testing. They were tested for rotavirus antigen using a commercially available Enzyme-linked immunosorbent assay (EIA) kit ProSpect Rotavirus Micro plate Assay (Oxoid Ltd., Basingstoke Hants, UK) which is based on detection of group specific antigen. Rotavirus testing was performed by a well-trained laboratory technologist. The results were released and placed in the patient's medical records.After that, the collected data was analyzed in the SPSS program.

Results

Out of 422 children (244 (57.8%) male and 178 (42.2%) female) examined, 215 children (131 (60.9%) male and 84 (39.1%) female) were positive for rotavirus infection (Fig. 1). Maximum number of positive cases (42.3 %) were in the age group of 6 months to 10 months and Minimum number of them (6.5 %) were in the age group of 6 months to more than 20 months (Fig. 2). The most positive cases of rotavirus occurred in July and September of 2018 and the lowest cases occurred in March and February of 2019 (Fig. 3).

Results of Chi-squire test showed that there was a significant association between having diarrhea and positivity of rotavirus by EIA (p-value=0.001). The Chi-squire test also suggest that, on comparison of rotavirus with non-rotavirus diarrhea, it was found that there was no statistically significant association between age (in months) and rotavirus (p-value=0.065) or with gender and rotavirus (p-value=0.201). Table 1. Based on Mann-Whitney U test, there are significant differences between positive and negative rotavirus cases from the point of view of diarrhea duration time (p-value=0.007), frequency of diarrhea/24hours (p-value=0.000) and Frequency of vomiting/24 hours (p-value=0.001). So, the rotavirus positive children suffered from longer diarrhea duration time, more frequent diarrhea/24hours and Frequent of vomiting\24 hours.

Table 1. Association of EIA results (positive or negative) with age, gender and having diarrhoea

		EIA Result		p-value a
		Positive	Negative	
Age Group	=<5	27	45	.065
	6-10	91	74	
	11-15	58	46	
	16-20	26	22	
	>20	14	20	
Gender	Male	131	113	.201
	Female	84	94	
Have Diarrhea	No	20	43	.001*
	Yes	196	164	

A total of 19 premature and full-term neonates were included in the study during the year it lasted, 15 of whom had sepsis (6 early onset, 9 late onset and 2 both) (Table 2) and only 6 were positive in haemoculture (31%). Four neonates from hospital 1 did not have sepsis and one of the four presenting sepsis had both early and late onset neonatal sepsis. One of the three neonates from hospital 3 had early and late onset neonatal sepsis. Five neonates were classified as being stage 3 (i.e. advanced enterocolitis) and the remaining 14 as stage 2 or confirmed enterocolitis (Table: 3).

Table 3 summarises the neonates' main clinical and diagnostic characteristics. Twelve were female and 8 male; eleven of them developed NNE during the first week of life (57%). Regarding how they were fed, 17 neonates (89%) received enteral feeding, 7 (36%) parenteral and 6 (31%) both types of feeding. Regarding gestational age, 8 neonates were full-term (i.e. 37 or more weeks' gestation), 8 were preterm (having less than 34 weeks' gestation) and 4 were late-stage pre-term (34 to 36 weeks' gestation). Regarding birth-weight, 7 weighed less than 1,500 g and the remaining 13 (68%) weighed 1,630 to 3,170 g (Table: 4).

The present study's results showed that presentation of NNE was mainly characterised by symptoms such as intolerance concerning the oral route in 14 cases (73%), blood in faeces in 13 cases (68%), abdominal distension in 17 cases (89%), distended bowel loops and intestinal pneumatosis in all cases.

Conventional and molecular identification of bacterial isolates. The bacteria identified by the respective hospitals' clinical laboratories were *Staphylococcus epidermidis* (hospitals 1, 2, 4 and 5), *Escherichia coli* (hospital 1) and *Pantoea agglomerans* (hospital 1). The last mentioned isolate could not be identified at species level using 16 S rRNA sequencing due to the high identity of species forming the *P. ananatis*, *P. eucrina* and *P. dispersa* complex. The other isolates proved to be the same using both methods (Table 5). All *S. epidermidis* isolates were methicillin-resistant and the *Pantoea* spp isolate was sensitive to all antibiotics used in the susceptibility tests (Table 4). *S. epidermidis* ST81 was identified in the 2 neonates who died due to advanced enterocolitis (stage I3) and sepsis following molecular characterisation by MLST (Table 7); the first neonate was hospitalised in hospital 5 and the second in hospital 1

MLST analysis of *S. epidermidis* and *E.coli*. Figure 2 shows PCR amplification of *S. epidermidis* (the same for the 4 isolates) and *E. coli* constitutive genes. The DNA from these amplified fragments was sequenced by PCR and the results compared to MLST databases Table 6 shows the resulting allele profiles and sequence types. Two *S. epidermidis* isolates belonged to the same sequence type (ST81) and, coincidentally, were isolated from the neonates who died. The other *S. epidermidis* isolates corresponded to ST126 and ST2 and that from *E.coli* was identified as ST394.

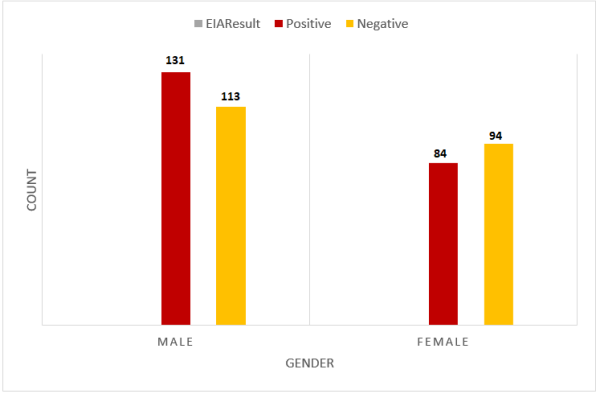

Fig. 1. Clustered bar count of gender by EIA result

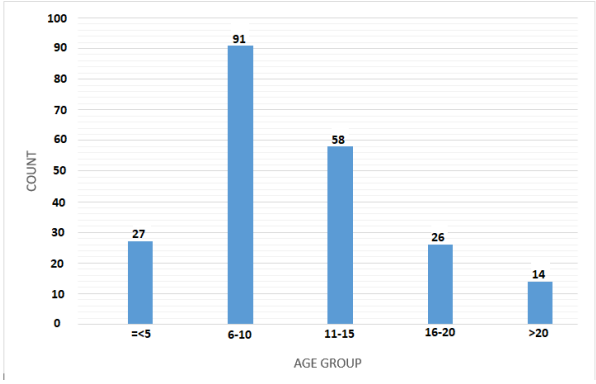

Fig. 2. Clustered Bar Count of Age Group by month in EIA positive results

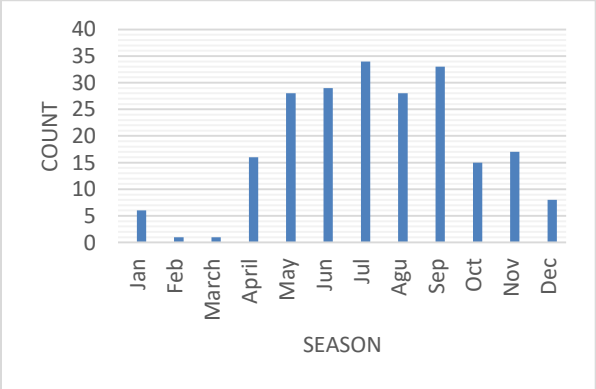

Fig. 3. Frequency of in EIA positive cases for rotavirus, by month

Discussion

Rotavirus is one of the causes of death of children in developing countries, which causes severe diarrhea (15). Many studies have been done on rotavirus in different countries of the world.

About 5% of children's deaths in Afghanistan are related to rotavirus, which shows the necessity of continuous research on this disease (16).

The findings of this study showed that more than 50% of children with diarrhea referred to Indira Gandhi Children's Health Hospital were related to rotavirus

and most of the cases were children under 20 months. These findings are in complete agreement with the study previously conducted by Anwari et al. and indicate that the situation of rotavirus in the country has not yet improved and requires special attention from the authorities (17).

By studying the seasons of the year, we came to the conclusion that the lowest rate of rotavirus diarrhea cases was related to cold seasons and the highest rate was related to warm seasons, which is not in accordance with the findings of previous studies (18). This shows that the activity of the virus is decreasing in Afghanistan due to the severe cold weather.

The longer diarrhea duration time, more frequent diarrhea/24hours and Frequent of vomiting\24 hours of the rotavirus emphasizes on modernizing, equipping and providing health services as much as possible in hospitals of under-developed countries like Afghanistan (19).

Although a significant difference has been observed between gender and rotavirus diarrhea in various studies, this significant difference was not observed in the present study (20-22). However, in our work the positivity of rotavirus was higher in male children compared with their female counterparts.

One of the most important gaps in relation to rotavirus in Afghanistan is the lack of a high-tech diagnostic tools and genotyping system, despite the existence of numerous equipment such as real time PCR machines and sequencing platform. The use of these tools not only helps to know the nature of the virus in the country, but also can examine the impact of vaccination in the future.

Conclusions

Our findings showed that still more than 50% of diarrhea cases in children are related to rotavirus. The findings of this research emphasize the fact that the Ministry of Health of Afghanistan should seriously follow control and preventive strategies, especially in warm seasons. Due to the worse condition of children suffering from rotavirus diarrhea and due to its high prevalence in Afghanistan, health services should be increased. Using high-tech tools is another solution that can be helpful in managing this disease.

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Ethics approval and consent to participate

Study protocol was reviewed and approved by Medical Ethic Committee of Ghalib University, Kabul, Afghanistan (AF.GKU.REC.1401.002).

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