# **Original Research Article**

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# A study on prevalence of rotavirus infection in children below 5 years, with acute gastroenteritis

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#### **ABSTRACT**

**Background:** Diarrheal diseases are among the most frequent causes of morbidity and mortality in children worldwide, especially in India. This cross sectional study was conducted to investigate the causative agent of diarrhea among children, below five years, treated at an urban tertiary care hospital, Telangana.

**Methods:** Stool specimens were collected from 240 children under 5 years of age visiting Apollo Medical College, General Hospital in Hyderabad, hospitals due to acute diarrhea. Rapid stool antigen immunochromatograhic testing was used to diagnose rota virus diarrhea. In this test the VP 6 antigen present on the middle layer of the virus capsule is detected.

**Results:** During the 2 months of the study period, 60 children visited the institute seeking treatment of diarrhoea. 11 (18%) of, the children were found to be positive for rota virus infection. It was also observed that, due to advent of rotavirus vaccine, incidence of diarrhea has come down.

**Conclusions:** In our study the prevalence of rotavirus infection in children suffering from acute gastroenteritis below five years of age is 18.3%. This study is performed using an ICT rapid card test in which group A of the rotavirus is detected, the drawback of this study is other groups of rotavirus like B and C which also cause human infections are not detected. Following the rapid ICT card test which is a diagnostic test, ELISA, confirmatory test for the detection of rotavirus should also be done.

**Keywords:** Childhood diarrhoea, Enteropathogens predominance, Antigen immunochromatograhic testing

#### INTRODUCTION

Diarrhea is the third leading cause of childhood mortality in India, and is responsible for 13% of all deaths/year in children under 5 years of age. Acute gastroenteritis is an inflammatory condition of the stomach and the small intestine; this can be caused due to many organisms like bacteria, viruses and parasite. Among the various organisms causing diarrhea, viruses are the most common causative agent. Rotavirus is a type of infection that's most common in children under the age of 5. It's highly

contagious and easily transmittable. While it occurs most often in young children, adults can also get the infection, although it's usually less severe. Rotavirus is the most common cause of diarrheal disease among infants and young children.<sup>2</sup>

66.7% of diarrheal deaths in children below five years of age are caused by rotavirus. Rotavirus was discovered in 1973 by Ruth Bishop and her colleagues by electron micrograph images and accounts for approximately one third of hospitalizations for severe diarrhea in infants and

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children; its importance has historically been underestimated within the public health community, particularly in developing countries. Rotavirus had often been associated with severe dehydration which is actually responsible for death associated with the infection.<sup>3</sup> In addition, children with dehydration had been found to be about two times more likely to have rotavirus diarrhea.<sup>4</sup> There is no specific treatment for rotavirus associated gastroenteritis but the complications can be prevented by fluid and electrolyte replenishment. This research would emphasize that the major causative organism causing gastroenteritis is rotavirus.

Rotavirus causes 78,500 deaths, 872,000 hospitalizations and over 3.2 million outpatient visits in children below five years of age all over India.<sup>5</sup> NICED in its report estimated that the total diarrheal death in India in 0-6 years of age is 1,58,2009 in the year 2012.<sup>6</sup>

With the advent of newer medical techniques and advent of rotavirus vaccine, the incidents of rotavirus infection have reduced. According to latest study reports, the prevalence of rotavirus infection is about 11.5% to 17.8%. The experimental design used in this research is detection of VP6 antigen present in the middle capsid layer. The technique of rapid ICT card test is used, being a rapid detection test for rotavirus; the results were obtained in 15 minutes. The age and name of the patient is noted and stool sample of the patient was collected, by using rapid ICT card test, the stool sample is tested for the presence of antigen, also, history of rotavirus vaccination was asked and noted down, if the guardians of the patient were unaware of the vaccine we informed them about the vaccine and the importance of the vaccine.

The rotavirus vaccine helps protect babies against diarrhoea and vomiting caused by rotavirus. It does not protect against diarrhoea and vomiting caused by other viruses. The vaccine contains a weakened form of rotavirus that does not cause disease.

# Economic burden of rotavirus diarrhoea in India

Every episode of rotavirus diarrhoea costs over 7% of average annual income of Indian families, enough to push low income families below poverty line. It is estimated that India spends over a staggering INR 1000 crores every year on management of rotavirus diarrhoea.

There are currently three licensed orally administered rotavirus vaccines available in markets and used in India. These vaccines have been shown to be safe and effective in large scale clinical trials.

#### Aims and objectives

• To estimate the burden of rotavirus associated gastroenteritis.

 Create awareness on environmental sanitation to break the chain of transmission of rotavirus diarrhea in the communit.

#### **METHODS**

The study is conducted by using the rapid immunochromatographic test. In this test the VP 6 antigen present on the middle layer of the virus capsule is detected

#### Study design

Cross-sectional study

#### Site

Apollo General Hospital, Hyderabad

# Study population

Children below five years of age attending Pediatrics OPD and IP of Apollo Institute of Medical Sciences and Research, (AIMSR General Hospital), Hyderabad.

#### Duration of study

2 months (July-2016 to August-2016)

#### Sample size

60

# Inclusion criterion

In this cross-sectional study children below 5 years of age came to the pediatric OPD and IP as well as in patients with symptoms of acute gastroenteritis- like nausea and vomiting, diarrhea and abdominal cramps (as per WHO) were included.

# Exclusion criterion

Critically ill children, those suffering from persistent or chronic diarrhea with mucus and blood, children already vaccinated against rotavirus, were excluded.

# **Procedure**

Rapid ICT card test which detects the VP 6 antigen present in the middle layer of the capsid of the rotavirus. This test is a rapid diagnostic method which detects only A group of rotavirus that infects man. The sensitivity of this test is 100% and specificity is 92.4%.

#### SD rotavirus BIO LINE

Samples were subjected to SD rotavirus BIO LINE, based on the result obtained, i.e. two bands on the card

which reads for test and control indicates that the test is positive, if only one band is present on the card which reads for control, indicates that the test is negative. For each day the total number of the positives and the negatives are noted. At the end the entire data is compiled and analyzed.

*Principle:* A nitrocellulose membrane is pre coated with rabbit monoclonal anti-rotavirus antibodies and mouse monoclonal anti-rotavirus antibodies which are used as detector materials. By using these detector materials, Group A rotavirus antigen can be directly detected from the faecal sample. The faecal sample will specifically react with rabbit anti-rabbit antibodies.

#### Preparation of extracted sample

Stool sample was collected in a sterile container. The device and sample were allowed to reach the room temperature. Assay diluents is taken up to the fill line, and then transferred into the given sample collection tube. These steps were repeated. 500 mg of the sample was taken with the sample collection swab. The swab was inserted into the sample collection tube containing assay diluents. The Swab was Swirled the swab at least 10 times until the sample has been dissolved into the assay diluents and discard the swab while squeezing the swab against the wall of tube.

# Test procedure

A control band will appear on the left section of the result window indicating that the test is working properly. The right side of the result window indicates the test result. If a band (test band) appears on the right side of the window along with the control band it indicates that the test is positive. If only control band is present on the result window, it indicates that the test is negative. If control band does not appear on the window, this indicates that the test is invalid.

Standard procedure for the collection and disposal of the sample and was followed and all precautions were taken while performing the test. The specimen was tested soon after the collection; in cases of delay they were stored in refrigerators at a temperature of 2 to 8°C. Samples which were stored for more than 72 hrs were not tested. The sample, collection swabs and the collection container were discarded in respective biohazard containers.

#### **RESULTS**

The diarrheal samples from children below five years of age suffering from acute gastroenteritis were collected and tested with rapid ICT card test. At the end the entire data was compiled and analyzed. The results were analyzed based on the results obtained from ICT card test. Out of 60 samples which were tested 11 isolates were positive when ICT was performed (Table 1). Out of 60 samples of acute gastroenteritis obtained from

children below five years of age 11 were positive for rotavirus infection and 49 were negative for rotavirus infection as shown in Table 1. The percentage of positivity is 18.3% shown in Figure 1. Rapid ICT card test is a reliable test for the detection of rotavirus in the diarrheal sample and shows 100% sensitivity and 92.4% specificity. It is a rapid detection test for Rotavirus in which VP 6 antigen present in the middle layer of the capsid which is responsible for infection, is detected.

**Table 1: Prevalence of rotavirus infection.** 

Total number of samples obtained	Positive for rotavirus infection	Negative for rotavirus infection
60	11	49

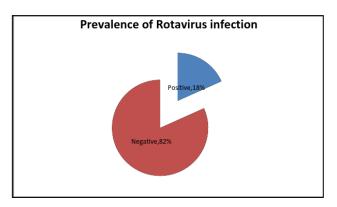


Figure 1: Prevalence of rotavirus infection.

# DISCUSSION

Immunochromatographic assay is an antigen antibody agglutination technique that allows the identification of group specific proteins, including the major inner capsid protein, present in group A of rotavirus. The test is based on the detection of VP 6 antigen present on the middle layer of the capsid, ICT test is a rapid test and the specificity of the test is 97%.

The antigen VP6 which is responsible for the pathogenesis and causes diarrhea in children is detected by ICT. The nitrocellulose based membrane pre-coated with rabbit monoclonal antibodies and the specially-selected monoclonal anti rotavirus antibodies are used as detector materials and used as detectors. These enable the test to identify Group A rotavirus antigen directly from the sample in human fecal specimens and then this mixture will react specifically with the rabbit anti rotavirus antibody on the membrane. The test is a rapid qualitative test for the detection of group A rotavirus in human fecal specimen. The ICT test was performed using the standard protocol and the all the rules of WHO were followed.

Out of the 60 samples we obtained from children suffering from acute gastroenteritis, 11 were positive. Our study on the prevalence of rotavirus in children

below five years which gave 18.3% positivity is showing good agreement with other studies conducted by Chakravarti et al (17.8%), Nag et al (15.6%), Anand et al (16.2%).

Though humans of all age groups are susceptible to rotavirus infection, the infection shows its severity in children below 5 years of age. The individual suffering from rotavirus infection excretes large amount of virus (rotavirus) which can be spread even through contaminated hands and fomites also. The ICT kit used in this study is an immunoassay to detect Group A rotaviruses in the fecal specimen.

#### **CONCLUSION**

In our study the prevalence of rotavirus infection in children suffering from acute gastroenteritis below five years of age is 18.3%. The test performed to estimate the prevalence of rotavirus infection is immune-chromatographic test which is a rapid diagnostic test.

ICT test is a reliable test for the detection of the presence of rotavirus in the children suffering from acute gastroenteritis, being a rapid test and sensitivity of the test being more than 97%, the results can be obtained within 15 to 20 minutes, this saves the time and helps in deciding the treatment regime and avoids the unnecessary prescription of antibiotics.

The approach of treatment of a diarrhea case in children below five years should be modified. By seeing the prevalence percentage of rotavirus we can say that the disease caused in children below five years of age is predominantly rotavirus infection and administration of antibacterial agents without a laboratory diagnosis is futile and contributes to antibiotic resistance.

This study is performed using an ICT rapid card test in which group A of the rotavirus is detected, the drawback of this study is other groups of rotavirus like B and C which also cause human infections are not detected. Following the rapid ICT card test which is a diagnostic test, ELISA for the detection of rotavirus should also be done as ELISA is a confirmatory test.

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