Microbial etiology of acute gastroenteritis in pediatric patients in Western India

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Abstract

Introduction: Acute gastroenteritis is a major cause of illness and death among infants and young children worldwide. According to the World Health Organization (WHO), an estimated 2.5 billion cases of diarrhea occur among children under 5 years of age each year. Nearly one in five pediatric deaths (about 1.5 billion each year) is due to diarrhea. Present study was thus drawn to identify microbial etiology of acute gastroenteritis in pediatric patients in Western India.

Materials and Method: This was Prospective laboratory based study approved by Institutional Ethical committee. 100 stool samples were collected from children admitted with acute diarrhea of 72 hours or less duration in 1- 60 months of age over a period of one year. Bacteria were identified by standard Microbiological methods and serotyping of isolated *Escherichia coli (E. coli)* was done. *Rotavirus* antigen was detected by Enzyme Linked Immunosorbant Assay (ELISA) followed by genotyping by Reverse Transcriptase Polymerase chain Reaction (RT-PCR) and Multiplex PCR. Parasitic identification was done by microscopy.

Results: 74% of children with diarrhea were in the age group of 7 to 12 months. Watery diarrhea (94%) was the commonest clinical presentation, followed by vomiting (78%), fever (78%), and dehydration (74%). Pathogenic bacteria were isolated in 51% of samples. *Escherichia coli* was most common (48%) followed by *Shigella flexneri* (2%) and *Vibrio cholerae* (1%). The most prevalent *E. coli* type was *Enterotoxigenic Escherichia coli (ETEC)* (20.8%) followed by *Enteropathogenic Escherichia coli (EPEC)*(16.7%), *Enterohaemorrhagic Escherichia coli (EHEC)* (4.1%), and *STEC* (2.1%). The most prevalent serotypes of *ETEC* were O27, O23, and O169. Among *EPEC* most prevalent serotypes were O90, O26. The most prevalent *EHEC* strain found in this study was O71. *Rotavirus* was detected in 35% of patients. Most prevalent *Rotavirus* genotype was G9P[4] (28.6%) followed by G2P[4] (21.4%), G1P[8] (21.4%), G12P[6] (14.3%), G9P[8] (7.1%). Parasitic etiology was detected in 5% of cases. Coinfection of *E.coli* and *Rotavirus* was detected in 23% of children. *Rotavirus* was most commonly associated with *EPEC* (25.7%) followed by *ETEC* (17.1%).

Conclusion: In the present study, *E. coli* was the commonest microorganism followed by *Rotavirus*. Thus, the importance of safe water and food hygiene would be most important intervention to prevent acute gastroenteritis in children along with *Rotavirus* vaccine.

Keywords: *Rotavirus, E. coli, WHO, NIV, ETEC, EPEC, EHEC, STEC*

Introduction

Acute gastroenteritis is a major public health concern for both developed and developing countries. According to the World Health Organization (WHO), an estimated 2.5 billion cases of diarrhea occur among children under 5 years of age each year. Nearly one in five pediatric deaths (about 1.5 billion each year) is due to diarrhea.[1]

Diarrhea is caused by different etiologic agents in both developed and developing countries.[2] These organisms included bacteria, viruses, and parasites. Most of diarrheal episodes that had been reported in developing countries were found to be associated with bacterial infections, while *Rotavirus* was reported as the most causes of diarrhea in developed countries.[3] The most predominant bacterial infections that cause diarrhea are *Salmonella spp, Shigella spp, Campylobacter jejuni, Yersinia enterocolitica, and Diarrheagenic Escherichia coli (DEC)*. *DEC* is recognized to be the most importance cause of diarrhea among children in the world and reported as one of the most common and important causes of endemic and epidemic diarrhea worldwide.[4]

Repeated and persistent diarrhea in young children contributes to significant cognitive and growth impairment that can impact school performance and development.[5-7] Diarrhea makes a greater contribution to malnutrition and growth impairment than other common infections and *Escherichia coli* diarrheas may be even more detrimental than *Rotavirus* infections in this regard.[5,8,9]

Thus the Present study was drawn to identify microbial etiology of acute gastroenteritis in pediatric patients in Western India.

Materials and Method

This was a Prospective laboratory based study approved by Institutional Ethical committee.

Specimens: 100 stool samples were collected from children admitted in pediatric ward with acute diarrhea of 72 hours or less duration in 1- 60 months of age over a period of one year.

Epidemiologic data were recorded for all patients.

Bacteria were identified by standard Microbiological methods. Identified *E. coli* were

transported to National Institute of *Salmonella* and *Escherichia coli* center, Kasauli for further Serotyping.

Testing for *Rotavirus* antigen was done by *Rotavirus* antigen capture Enzyme Linked Immunosorbant Assay (ELISA) by IVD ELISA kit (Carlsbad, CA, USA). Ten percent (w/v) suspensions of all *Rotavirus* positive stool specimens prepared in 0.01 M phosphate buffered saline (PBS) (pH 7.2) were transported to National Institute of Virology (NIV), Pune for G and P Genotyping by Reverse Transcriptase Polymerase chain Reaction (RT-PCR) and Multiplex PCR.

Parasitic identification was done by standard methods (Formal ether method, saline and iodine wet mount and Modified ZN staining.

Gel electrophoresis pictures after multiplex PCR
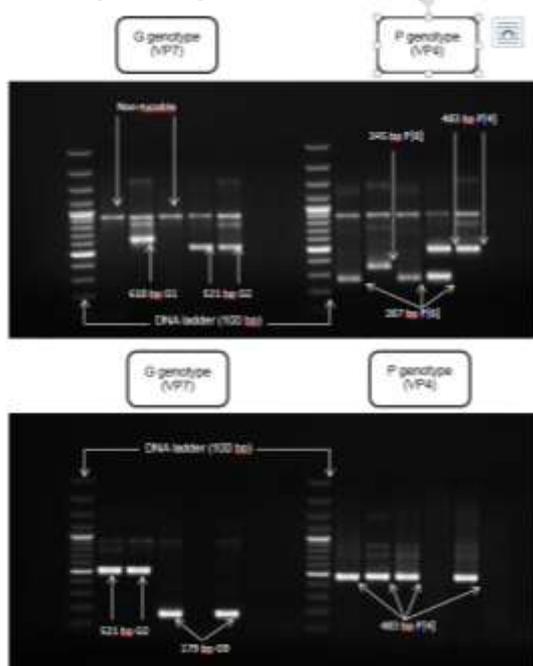
Results

74% of children with diarrhea were in the age group of 7 to 12 months. Watery diarrhea (94%) was the commonest clinical presentation, followed by vomiting (78%), fever (78%), and dehydration (74%).

Pathogenic bacteria were isolated in 51% of samples. *Escherichia coli* was most common (48%) followed by *Shigella flexneri* (2%) and *Vibrio cholerae* (1%).

The most prevalent *E. coli* Serotype was *ETEC* (20.8%) followed by *EPEC* (16.7%), *EHEC* (4.1%), and *STEC* (2.1%).

The most prevalent serotypes of *ETEC* were O27, O23, and O169. Among *EPEC* most prevalent serotypes were O90, O26. The most prevalent *EHEC* strain found in this study was O71.

Rotavirus was detected in 35% of patients. Most prevalent *Rotavirus* genotype was G9P[4] (28.6%)

followed by G2P[4] (21.4%), G1P[8] (21.4%) , G12P[6] (14.3%), G9P[8] (7.1%) .

Parasitic etiology was detected in 5% of cases. *Entamoeba histolytica* and *Cryptosporidium parvum* were detected in 2% each followed by *Giardia lamblia* (1%).

Coinfection of bacteria and *Rotavirus* was detected in 23% of children. *Rotavirus* was most commonly associated with *EPEC* (25.7%) followed by *ETEC* (17.1%), *EHEC* (2.9%), *STEC* (2.9%) and *Shigella flexneri* (2.8%)

Discussion

In the present study a total of 100 children aged less than 5 years of age suffering from acute diarrhea, admitted in pediatric ward, in tertiary care hospital were included during the period of 1 year (September 2012 to August 2013). The youngest child was 1 month old and the oldest child was 48 month old. The majority of children who presented with acute gastroenteritis were in the age group of 7 to 12 months of age (74%).

The probable reason for this age distribution could be that the most common etiological agents of acute gastroenteritis in children < 5 years of age viz. *E. coli* and *Rotavirus* cause maximum morbidity less than 18 months of age and in the 7-24 months of age group, respectively.

In the present study bacteria isolated were *E. coli* in 48%, *Shigella flexneri* in 2% and *Vibrio cholera* in 1%. *Rotavirus* was positive in 35% by ELISA and RT-PCR. Parasites identified were *Entamoeba histolytica* (2%), *Cryptosporidium parvum* (2%) and *Giardia lamblia* in 1% of children < 5 years of age.

Escherichia coli were serotyped at National *Salmonella* and *Escherichia* Center, Kasauli and *ETEC* (20.8%) was most common type followed by *EPEC* (16.7%), *EHEC* (4.1%), and *STEC* (2.1%).

Similarly Osman MM al[10] (2012) reported that 19% of the cases had diarrheal disease caused by bacterial isolates and *Enteropathogenic E.coli (EPEC)* was the predominant isolate (42.11%), followed by *Salmonella paratyphi B* (31.58%), *Salmonella typhi* (10.53%), *V. cholerae* (10.53%), and *Shigella flexneri* (5.26%). Panikar et al[11] isolated pathogenic bacteria from 12.5% of cases *Enteropathogenic E. coli* was most frequent (7.3%), followed by *Shigella spp.* (2.9%) and *Aeromonas hydrophila* (1.6%).

In studies discussed above *EPEC* was most commonly isolated but in the present study *ETEC* was the most common type of *Escherichia coli*.

Out of 100 samples collected over the period of one year, 35 samples (35%) were positive by *Rotavirus* antigen detection ELISA. Thus the prevalence of *Rotavirus* causing acute gastroenteritis in the present study was found to be 35%. Indian studies[12-17] have shown a wide range of *Rotavirus* prevalence ranging from 4 to 89.9%.

In the present study *Escherichia coli* was the most common pathogen followed by *Rotavirus*. *Rotavirus* is only diarrheogenic pathogen that can be prevented by vaccine.

Escherichia coli is transmitted by feco-oral rout mostly due to poor hygiene and contaminated water and food. Thus for prevention of *E. coli* diarrhea mere hand hygiene and use of safe water and food is very important. As *E. coli* diarrhea is more common in children's < 5 years of age to prevent diarrhea related complications education of mother is important.

As of 2006, for *Rotavirus* there are two live oral vaccines which have undergone large scale clinical trials and are being introduced globally. In countries considering a *Rotavirus* vaccination program, public health officials need to collect data on the *Rotavirus* disease burden to assess the need for a vaccine.

Conclusion

In the present study, *Escherichia coli* was the commonest microorganism followed by *Rotavirus*. Thus, the importance of safe water, hand hygiene and food hygiene would be most important intervention to prevent acute gastroenteritis in children along with *Rotavirus* vaccine.

Acknowledgement

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