

Percentage of Rotavirus infection in stool of pediatric patients at Children Welfare Teaching Hospital in Baghdad

Ban A. Abdul Sattar*

BSc (microbiology), MSc (medical microbiology)

Khalida K. Al-Kareemi**

BSc (medical technology, pathological analysis), MSc (medical microbiology)

Ateka A. Jassim***

BSc (microbiology)

Summary:

Background: Gastroenteritis is a common illness in pediatric age group; the causes could be bacterial, fungal or viral infection. Rotavirus is the most common cause of infectious diarrhea in children. Children between 3 months to 2 years old have the most severe symptoms. Rotavirus is extremely easy to catch and is transmitted mainly by the oral- fecal route. For diagnosis; the specimen of choice is the stool which has been collected during the first 3 to 5 days of illness. If collected 8 or more days after onset of symptoms the specimen will rarely contain the virus.

Objective: The objective of this study includes a survey for Rotavirus that cause diarrhea in children admitted to Children Welfare Teaching Hospital in Baghdad between 13th. of January-2010 to 14th. of October-2010 using a rapid test method for detection of Rota virus to children from one day to 6 years.

Materials and Methods: Latex agglutination test for the stool of 61 children suspected with Rotavirus infection was done in addition to general and bacteriological test of these stool samples at Children Welfare Teaching Hospital from January 2010 to October 2010.

Results: There were 61 children from one day-6 years old, only 11(18.03%) had positive Rotavirus infection with high percentage in males (seven cases) and (four cases) in females and only one case detected with positive Rotavirus infection at the age of 42 month. No pathogenic bacteria or fungus were isolated.

Conclusion: The data indicate that male has highly incidence than female and children up to 78 months had diarrhea that not caused by Rotavirus.

Key words: Rotavirus, Latex agglutination, viral gastroenteritis.

J Fac Med Baghdad
2012; Vol.54, No. 4
Received June.2012
Accepted Oct.2012

Introduction:

Rotavirus is the most common cause of severe diarrhea among infants and young children (1), and is one of several viruses that cause infections often called stomach flu having no relation to influenza. Rota virus is a genus of double-stranded RNA virus in the family Reoviridae. By the age of five, nearly every child in the world has been infected with rotavirus at least once (2). Rotavirus is transmitted from person to person through the fecal-oral route. This occurs when the viruses found in stool of infected child is swallowed by another child, in other words children become infected if they put their finger in their mouth after touching something such as toys, books, clothing etc., that has been contaminated by stool of an infected person, this usually happen when children forget to wash their hands after using the toilet or before eating (3). Adults are rarely affected (4). There are five species of rotavirus, referred to as A, B, C, D and E. Humans are primarily infected by species

A, B and C, most commonly by species A. All five species cause disease in other animals (5). Within rotavirus A there are different strains, called serotypes (6) As with influenza virus, a dual classification system is used, which is based on two structural proteins on the surface of the virion. The glycoprotein VP7 defines G-types and the protease-sensitive protein VP4 defines P-types. Strains are generally designated by their G serotype specificities (e.g., serotypes G1 to G4 and G9), and the P-type is indicated by a number and a letter for the P-serotype and by a number in square brackets for the corresponding P-genotype. (P-serotypes are difficult to characterize; therefore, molecular methods based on sequence analysis are often used to define the corresponding P-genotype instead. These genotypes correlate well with known P-serotypes (7). Because the two genes that determine G-types and P-types can be passed on separately to off springs, various combinations occur in any one strain. The virus is transmitted by the feco-oral route. The virus infects and damages the cells that line the small intestine and causes gastroenteritis. Although rotavirus was discovered in 1973(8) and accounts for up to 50% of hospitalizations for severe diarrhea in infants and children (9), its importance is still not widely known within the public

* Assistant Lecturer of microbiology- Department of Biology / College of Science / Al-Mustansiriya University.

** Assistant Lecturer of microbiology /Department Microbiology/ College of Medicine/ University of Baghdad.

***Bsc. Bacteriologist, Children Welfare Teaching Hospital Laboratory.

health community, particularly in developing countries(10). In addition to its impact on human health, rotavirus also infects animals, and is a pathogen of livestock(11).

Diagnosis of infection with rotavirus normally follows diagnosis of gastroenteritis as the cause of severe diarrhea. Most children admitted to hospital with gastroenteritis are tested for rotavirus A (12, 13). Specific diagnosis for infection with rotavirus A is made by finding the virus in the child's stool by enzyme immunoassay. There are several licensed test kits on the market which are sensitive (14). Various other techniques have been developed to readily detect rotavirus in stool, including electron microscopy and polyacrylamide gel electrophoresis (PAGE) of viral nucleic acid and antibody-based assays such as enzyme immunoassay (EIA), immunofluorescence, radioimmunoassay, and solid-phase aggregation of coated erythrocytes(15). Reverse transcription-polymerase chain reaction (RT-PCR) can detect and identify all species and serotypes of human rotavirus (16).

The purpose of this study was to assess the rate of rotavirus infection among children less than six years of age with acute gastroenteritis at Children Welfare Teaching Hospital in Baghdad.

Materials and Methods:

All stool samples were tested bacteriologically by culturing on XLD agar, MacConkey agar and Sabuaroud agar. For Rotavirus a rapid test(Latex agglutination) were applied by taking 2 gram of feces and adding 2 ml diluents in test tube, vortex test tube and let stand for 10-15 min. at room temperature, then centrifuged for 10 min.50µL supernatant mixed with one drop of reagent(antigen) and (clumping) agglutination indicate positive Rotavirus.(code 3000-8010) (Biokit,U.S.A).

Results:

A total of 61 suspected Iraqi children cases with Rotavirus infection distributed as 39 (63.93%) male and 22(36.06%) female, eleven cases(18.03%) recorded with positive Rota virus infection distributed as 7 (63.64%)male and 4(36.36%) female as shown in (Figure 1) with high percentage in male infection than female. No pathogenic bacteria or Candida albicans was isolated, while (Figure 2) indicate that children under 18 months old have high risk for infection with Rota virus and only one male case with the age 42 months old had positive Rota virus infection.

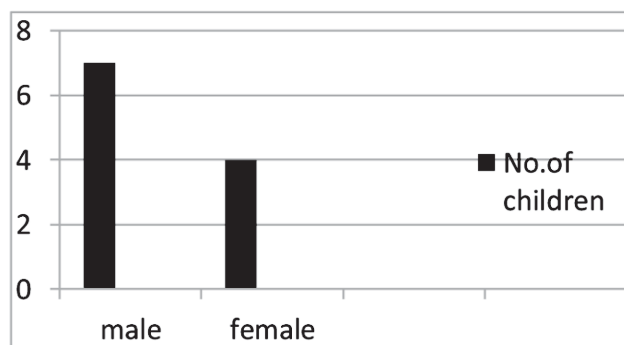


Figure (1) :(Distribution of Rotavirus infection in relation to gender of the patients)

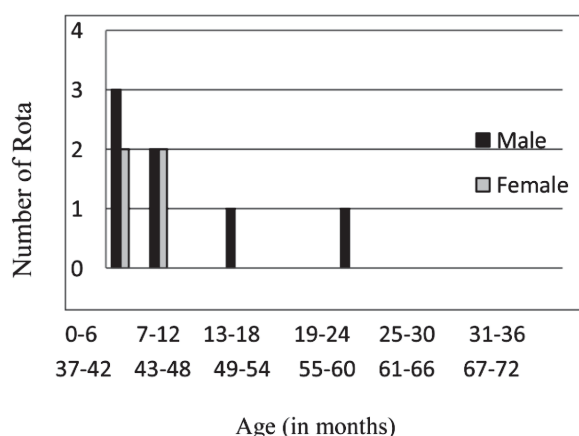


Figure (2): (Rotavirus infection among different age groups)

Discussion:

Rotavirus is recognized as a major cause of non bacterial gastroenteritis (infection of the stomach and intestinal tract leading to diarrhea and vomiting) especially in infants and young children worldwide (17, 18). A child may have rotavirus gastroenteritis more than once because there are many different rotavirus types but repeat infection tends to be less severe than the original infection. Asymptomatic infections occur by either subsequent infection of individuals whose immune system confers protection with rotavirus infection with seroconversion (19).

Diagnosis of rotavirus infection is based on the identification of rotavirus in feces or suspension of rectal swab collected early in the illness. Rapid serological test involve the use of latex agglutination kits while confirmatory test with Enzyme Linked Immunosorbent Assay (ELISA) test for rotavirus specific antigens is also should be used (20-21).Detection of rotavirus with higher numbers in males than in females was noted in our study with ratio of female to male was 1:1.75.This agree with the finding of (19), that boys have

been found to be twice susceptible and likely to be admitted to hospitals than girls. Whether this difference is due to sex susceptibility or by chance is however questionable and needs further investigation. The reason(s) for the difference in ratio occurrence within different geographical locations is not well understood, too. No pathogenic bacteria or *Candida albicans* were isolated, the cause may be viral agent but the test was negative due to sample duration i.e more than 8 days after symptoms onset.

In developing countries, 65%-80% of children have rotavirus antibodies by the age of 12 months. Thus, the incidence of symptomatic illness declines rapidly after 24 months of age, and repeated infections may be asymptomatic or accompanied by mild symptoms and in temperate climates, infections occur predominantly in winter (22). On the other hand there were high incidences in Rota virus infection among children under 13 months age and this may come from natural immune state to rotavirus does not exist. Through primary infection by the virus induces production of rotavirus-specific memory B and T cells, these are not normally sufficient to prevent re-infection by the virus. However, they do serve to reduce the severity of secondary infections. It has shown that serum IgA antibody titers correlate with protection against re-infection (23). It has been shown also in mice that in the absence of IgA, IgG is also sufficient to protect mice (24). However, in humans, high titers of IgG do not seem to be as protective as IgA against moderate to severe illness, so serum IgA is seen as the primary indicator of protective immunity to rotavirus (23). Intestinal immunoglobulin A (IgA) to rotavirus has been shown to be the most-sensitive marker of rotavirus infection (25). but titers of serotype-specific, heterotypic, and neutralizing serum antibodies and isotype-specific antibodies in serum and intestine or stools cannot be used reliably as markers of protection against subsequent illness (26). Further investigations are needed to provide a more accurate picture of epidemiology of rotavirus disease and also its dominated serotype in Iraq by using advanced technology and tests for the detection of Rota virus in all Iraqi hospitals and if there are any other factors like (seasons, type of feeding, education background,..etc) effect rates of infection should also be reported and documented.

References:

- 1-Dennehy PH. "Transmission of rotavirus and other enteric pathogens in the home". *Pediatr. Infect. Dis. J.* 2000, 19 (10 Suppl): S103-105.
- 2-Velázquez FR, Matson DO, Calva JJ, Guerrero L, Morrow AL, Carter-Campbell S, Glass RI, Estes MK, Pickering LK, Ruiz-Palacios GM. "Rotavirus infections in infants as protection against subsequent infections". *N. Engl. J. Med.* 1996, 335 (14): 1022-1028.
- 3-Chin J. *Control of Communicable Disease Manual*. Wash. DC: American Public Health Association. *J Clin Microbiol* 2000, 125(17):910-7.
- 4-Bishop RF. "Natural history of human rotavirus infection". *Arch. Virol. Suppl.* 1996, 12: 119-128.
- 5-Kirkwood CD. "Genetic and antigenic diversity of human rotaviruses: potential impact on vaccination programs". *The Journal of Infectious Diseases.* 2010, 202 Suppl: S43-48.
- 6- Santos N, Hoshino Y. "Global distribution of rotavirus serotypes/genotypes and its implication for the development and implementation of an effective rotavirus vaccine". *Rev. Med. Virol.* 2005, 15 (1): 29-56.
- 7-Dennehy PH. "Rotavirus vaccines: an overview" *Clin. Microbiol. Rev.* 2008, 21 (1): 198-208.
- 8-Bishop RF, Davidson GP, Holmes IH, Ruck BJ. "Virus particles in epithelial cells of duodenal mucosa from children with acute non-bacterial gastroenteritis". *Lancet* 2 (7841): 1281-1283.
- 9-Rheingans RD, Heylen J, Giaquinto C. "Economics of rotavirus gastroenteritis and vaccination in Europe: what makes sense?". *Pediatr. Infect. Dis. J.* 2006, 25 (1 Suppl): S48-55.
- 10-Simpson E, Wittet S, Bonilla J, Gamazina K, Cooley L, Winkler JL. "Use of formative research in developing a knowledge translation approach to rotavirus vaccine introduction in developing countries". *BMC Public Health* .2007, 7: 281.
- 11-Holland RE. "Some infectious causes of diarrhea in young farm animals" (PDF). *Clin. Microbiol. Rev.* 1990, 3 (4): 345-375.
- 12-Patel MM, Tate JE, Selvarangan R, et al. "Routine laboratory testing data for surveillance of rotavirus hospitalizations to evaluate the impact of vaccination". *Pediatr. Infect. Dis. J.* 2007, 26 (10): 914-919
- 13-The Pediatric Rotavirus European Committee (PROTECT). "The pediatric burden of rotavirus disease in Europe". *Epidemiol. Infect.* 2006, 134 (5): 908-916.
- 14-Smith TF, Wold AD, Espy MJ, Marshall WF. "New developments in the diagnosis of viral diseases". *Infect. Dis. Clin. North Am.* 1993, 7 (2): 183-201.
- 15-Bon F, Fascia P, Dauvergne M, et al. Prevalence of group A rotavirus, human calicivirus, astrovirus, and adenovirus type 40 and 41 infections among children with acute gastroenteritis in Dijon, France. *J Clin Microbiol.* 1999; 37: 3055-3058
- 16-Fischer TK, Gentsch JR. "Rotavirus typing methods and algorithms". *Rev. Med. Virology.* 2004, 14 (2): 71-82.
- 17-Parashar UD, Gibson CJ, Bresse JS, Glass RI. Rotavirus and severe childhood diarrhea. *Emerging Infect Dis.* 2006,

12(2):304-306.

18-Rheingan RD, Heylen J, Giaquinto C: Economics of rotaviruses gastro-enteritis and Vaccines in Europe. *Pediatr Infect Dis J.* 2006, 25:48-55.

19-Bass CW, Dorsey KN: Rotavirus and other agents of viral gastroenteritis. In *Nelson Textbook of Pediatrics*. Edited by Richard E and Behrman F. Raven Press, Philadelphia; 2004:107-110.

20-Jawetz E, Malnick JL, Adelberg EA: Reoviruses. In *Medical Microbiology*. 24th.ed.. Edited by Janet F, Jim R, Harriet L, Barbara R. Lange Medical Pub. Los Angeles, California; 2007:434-438.

21-Fischer TK, Bresee JS, Glass RI: Rotavirus vaccines and the prevention of hospital-acquired diarrhea in children. *Med Virology.* 2004, 22(5):49-54.

22- PAN, American Health Organization, regional office of(WHO). 2010. Scientific and technical publication, No. 623

23-Velazquez, F.R., et al., Serum antibody as a marker of protection against natural rotavirus infection and disease. *J Infect Dis.* 2000. 82(6): p. 1602-1609

24-O'Neal, C.M., G.R. Harriman, and M.E. Conner; Protection of the villus epithelial cells of the small intestine from rotavirus infection does not require immunoglobulin. *A. J Virology.* 2000. 4(9): p. 4102-4109.

25- Coulson B. S., Grimwood K., Masendycz P. J., Lund J. S., Mermelstein N., Bishop R. F., Barnes G. L. Comparison of rotavirus immunoglobulin A coproconversion with other indices of rotavirus infection in a longitudinal study in childhood. *J. Clin. Microbiol.* 1990. 28:1367-1374. Coulson B. S.,

26- Ward, R. L. Mechanisms of protection against rotavirus in humans and mice. *J. Infect. Dis.* 1996. 174(Suppl. 1):S51-S58.