Brief Report

Predominance of Rotavirus G1[P8] Genotype among Under-Five Children with Gastroenteritis in Mwanza, Tanzania

by Adolfine Hokororo,[1,*] Benson R. Kidenya,[2,*] Jeremiah Seni,[3,*] Seheri Mapaseka,[4] Jeffrey Mphahlele,[4] *and* Stephen E. Mshana[3]

[1]*Department of Pediatrics and Child Health Bugando Medical Centre, P.O.Box 1370, Mwanza, Tanzania*

[2]*Department of Biochemistry and Molecular Biology Catholic University of Health and Allied Sciences, P.O.Box 1464, Mwanza, Tanzania*

[3]*Department of Microbiology and Immunology Catholic University of Health and Allied Sciences, P.O.Box 1464, Mwanza, Tanzania*

[4]*Department of Virology, University of Limpopo, College of Health and Allied Sciences, P.O.Box 173, Limpopo, South Africa*

*Correspondence: Stephen E. Mshana, Department of Microbiology/Immunology Catholic University of Health and Allied Sciences, BOX 1464, Mwanza, Tanzania. Tel: +255 282 500 881; Fax: +255 282 502 678; E-mail <mshana72@yahoo.com>.*These authors contributed equally to this work.*

Summary

We analyzed stool samples from underfives with gastroenteritis for rotavirus infection between January 2010 and June 2011. A total of 393 stool specimens were examined for rotavirus infection using enzyme-linked immunosorbent assay (ELISA). Hundred selected positive specimens were genotyped using multiplex polymerase chain reaction. Out of 393 underfives, 194 (49.4%) had rotavirus infection, with 96.9% of infected underfives being <2 years. Underfives infected with rotavirus had prolonged hospital stay than those without rotavirus infection ($P = 0.0001$). G1 was the most predominant G type (59%) followed by G8 (13%) while P[8] was the most predominant P type (25%). In single-type infection, common G–P combinations were G1P[8] (24%) and G1P[6] (17%). Common mixed infections were G1/G8 (16%) and P4/P8 (13%). G1 genotype is common among underfives with gastroenteritis in Mwanza. Diversity of genotypes causing gastroenteritis in Mwanza necessitates a continuous surveillance after the introduction of RotaRix® vaccine.

Key words: rotavirus, genotypes, Tanzania.

Rotavirus infection is the leading cause of diarrhea among underfives worldwide and is usually associated with prolonged hospitalization, severe dehydration and deaths notably in children from developing countries [1–3]. Different studies have shown that the prevalence could be as high as 45%, with higher rates being reported in countries where there is no routine rotavirus immunization [3–6].

Rotavirus serogroup A is the most predominant group and clinically important in causing endemic gastroenteritis in children. A structural viral proteins (VP)-based classification of rotaviruses is composed of 12 different VP7 antigens (G-types) and 15 different VP4 antigens (P-types). Five G–P combinations G1P[8], G2P[4], G3P[8], G4P[8] and G9P[8] contribute to ~90% of all rotavirus infections in human worldwide. Of these, G1P[8] is the most predominant genotype [1, 7–10]. Of G-types, G1 has been found to predominate in many studies [11–13], with exception of two studies from Kenya and Dar es Salaam, Tanzania, that showed the predominance of G4 and G9, respectively [4, 14]. The monovalent vaccine

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TABLE 1
Demographic and clinical characteristics of 393 children with gastroenteritis

Variable	Results
Age	
Median	10 months
IQR	7–14 months
Sex	
Female	169 (43%)
Male	224 (57%)
Dehydration status	
No dehydration	140 (35.6%)
Mild dehydration	60 (15.3%)
Moderate dehydration	100 (25.5%)
Severe dehydration	13 (3.3%)
Shock	80 (20.4%)
Temperature	
Median	37.4°C
IQR	36.9–38°C
Hospital stay (days)	
Median	4
IQR	2–5
ELISA result	
Positive	194 (49.4%)
Negative	199 (50.6%)

TABLE 2
Association of demographic, clinical characteristics and rotavirus results of 393 children with gastroenteritis

Variable	Positive rotavirus	Negative rotavirus	*P*
Age			
Median	9 months	10 months	
IQR	7–11months	7–12 months	0.0024
Age category			
≤ 12 months	150 (56%)	118 (44%)	
>12 months	44 (35.2%)	81 (65.8%)	0.00001
Sex			
Female	88 (52.1%)	81 (47.9%)	
Male	106 (47.3%)	118 (52.7%)	0.351
Dehydration status			
No dehydration	69 (49.3%)	71 (50.7%)	
Any degree of dehydration	125 (49.4%)	128 (50.6%)	0.982
Season			
Dry	25 (44.6%)	31 (55.4%)	
Rainy	169 (50.2%)	168 (49.8%)	0.445
Temperature (°C)			
Median	37.2°C	37.4°C	
IQR	36.7–38°C	37–38°C	0.2568
Hospital stay (days)			
Median	4	3	
IQR	3–6	2–4	<0.00001

based on an attenuated human rotavirus strain of P1 A[8] G1, RIX 4414 (RotaRix®, GSK Biologicals, Belgium), which has been introduced in Tanzania recently, is effective against G1P[8], G3P[8], G4P[8] and G9P[8] [15, 16]. Because of possible variations of genotypes, this vaccine should be evaluated not only across countries but also within different regions in each country for its suitability in the respective populations.

A prospective cross-sectional study was conducted in Mwanza, Tanzania, between January 2010 and June 2012. During the study, 543 children were admitted due to gastroenteritis. Gastroenteritis was defined as acute occurrence of at least three motions of loose or watery stools in a 24 h period and/or two or more episodes of vomiting unexplained by other reasons. Stool specimens were collected and transferred to the laboratory on the same day for analysis using commercially available DAKO IDEIA rotavirus EIA detection kit (Dako Ltd, Ely, United Kingdom) [17]. Hundred randomly selected positive specimens were stored at −20°C before being transported to South Africa for genotyping at MRC/UL Diarrhoeal Pathogens Research Unit, Department of Virology, University of Limpopo [18].

Data were managed using Epi Data 3.1 (CDC Atlanta, USA) and analyzed using STATA version 12 (College Station, TX, USA). Categorical variables were summarized as proportions, and significance of their difference in distribution with the outcome was assessed using Pearson's chi-square test and probability plots and Shapiro–Wilk normality test to assess the normality of continuous variables. In all analyses, the difference with p-value <0.05 was considered as significant.

Of 543 children with gastroenteritis, 393 (72%) had valid rotavirus results with complete information and were included in the analysis. The median age was 10 months with interquartile range (IQR) of 7–14 months. Out of 393 children, 253 (64.3%) had degree of dehydration. The median temperature was 37.4°C with IQR of 36.9–38.0°C (Table 1). Regarding hospital stay, the median duration was 4 days with IQR of 2–5 days. Of 393 underfives, 194 (49.4%) were found to be infected with rotavirus infection. Infants were significantly more infected than older children [150 of 268 (56.0%) vs. 44 of 124 (35.2%), $P < 0.001$]. Children with rotavirus infections were found to have prolonged hospital stay than those without rotavirus infections (4 vs. 3 median days, $P < 0.0001$). More rotavirus infections occurred during rainy season (February, March, April, November and December) than dry season, although the difference was not statistically significant ($P = 0.445$) (Table 2).

Of 100 randomly selected positive stool specimens for genotyping, G1 was detected in 79 specimens followed by G8 in 29 specimens (Table 3). The common P-type detected was P[8], which was detected in 53

TABLE 3
Distribution of various genotypes in 100 stool specimens of children with positive rotavirus infections

Genotypes/serotypes	*N*
VP7 (G-type) (100)	
G1	59
G8	13
G2	4
G9	2
G1/G8	16
G1/G2	4
VP4 (P-type) (100)	
P[8]	25
P[6]	25
P[4]	12
P4/P8	13
P8/P6	11
P4/P6	10
P4/P6/P8	4
Genotypic combinations (100)	
G1 P[8]	24
G1 P[6]	17
G8P[4]	7
G8P[6]	4
G2P[4]	2
G8P[8]	6
G2/P8/P6	1
G1/P4/P6	9
G2/P8/P4	1
G1/P4/P8	1
G9/P8/P6	2
G1/G8/P[8]	5
G1/G8/[P6]	2
G1/G8/[P4]	3
G8/G1/[P6]	2
G8/P4/P6/P8	2
G1/G2/P4/P8	4
G1/G8/P4/P8	3
G1/G8/P6/P4	1
G1/G8/P8/P6	2

specimens. In single-type infections, the predominant G–P combinations were G1P[8], G1P[6] and G1[P4], whereas the least combination was G2P[4], which was detected in two specimens only.

Mixed infections were common in this study; the common mixed infection observed was G1 G8 (16%) followed by G1 G2 (4%). The G1 G2 mixed infection was also observed previously in Spain and Italy [19, 20]. Spain and Italy had low prevalence of G8 type; this could explain why they had no G1 G8 mixed infection. In the current study and studies in Italy and Spain, the common P-type combination was P[4] P[8]. In contrast to the study in Spain and Kenya, [21] the commonest mixed infection combination in this study was G1P[4] [6], whereas in the studies in Spain and Kenya, it was G1P[4] [8] and G8 P[4] [6].

Overall, in this study, G1 was the most predominant G-type followed by G8, whereas P[8] and P[6] constituted over 50% of the P-types, and the G1[8], G1P[6] and G1P[4] formed over three quarter of the G–P combinations. These findings are similar to those found elsewhere in African countries [11–13, 22] and in other countries outside Africa [7, 9, 10, 23]. The finding of predominance of G1 is in contrary to the study in Dar es Salaam, Tanzania [4], whereby the predominant G-type was G9 (81.6%) and G9 P[8] combination. This shows the diversity of circulating genotypes in Tanzania, which emphasizes the need of ongoing country-wide surveillance.

As demonstrated previously [3–5], infants were significantly more infected with rotavirus infection than older children. In this study, more cases of rotavirus infections were detected during rainy season than in dry season, although the difference was not statistically significant. As in previous studies [24, 25], there was a prolonged duration of hospitalization among children infected with rotavirus compared with those without rotavirus infection.

Rotavirus infection is a leading cause of gastroenteritis in our setting and is associated with prolonged hospitalization. The diversity of genotypes and variation of genotypes in the same country emphasizes the continuous surveillance of rotavirus strains. RotaRix® vaccine is expected to reduce infection in Mwanza; however, continued surveillance is warranted, especially regarding the long-term effects of the vaccine.

References

1. Mandell GL, Bennett JE, Dolin R. Principles and practice of infectious diseases. In: Dormitzer PR (ed.). Rotaviruses. 6th edn., Vol. 2. Churchill Livingstone: Philadelphia, PA, 2005.
2. WHO. Rotavirus vaccines. Weekly epidemiological record. Geneva: World Health Organization, 2013. 88:49–64.
3. Nakawesi JS, Wobudeya E, Ndeezi G, *et al*. Prevalence and factors associated with rotavirus infection among children admitted with acute diarrhea in Uganda. BMC Pediatr 2010;10:69.
4. Moyo SJ, Gro N, Kirsti V, *et al*. Prevalence of enteropathogenic viruses and molecular characterization of group A rotavirus among children with diarrhea in Dar es Salaam Tanzania. BMC Public Health 2007; 7:359.
5. Junaid SA, Umeh C, Olabode AO, *et al*. Incidence of rotavirus infection in children with gastroenteritis attending Jos university teaching hospital, Nigeria. Virol J 2011;8:233.
6. Temu A, Kamugisha E, Mwizamholya DL, *et al*. Prevalence and factors associated with Group A rotavirus infection among children with acute diarrhea in Mwanza, Tanzania. J Infect Dev Ctries 2012;6:508–15.
7. 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;15:29–56.
8. WHO. Manual of rotavirus detection and characterization methods (WHO/IVB/08.17). Geneva: World Health Organization, 2009.
9. Kirkwood CD, Boniface K, Bogdanovic-Sakran N, *et al*. Rotavirus strain surveillance—an Australian perspective of strains causing disease in hospitalised children from 1997 to 2007. Vaccine 2009;27:F102–7.
10. Hu L, Crawford SE, Hyser JM, *et al*. Rotavirus nonstructural proteins: structure and function. Curr Opin Virol 2012;2:380–8.
11. Todd S, Page NA, Duncan Steele A, *et al*. Rotavirus strain types circulating in Africa: review of studies published during 1997-2006. J Infect Dis 2010;202:S34–42.
12. Esona MD, Armah GE, Steele AD. Rotavirus VP4 and VP7 genotypes circulating in Cameroon: identification of unusual types. J Infect Dis 2010;202:S205–11.
13. Bonkoungou IJ, Damanka S, Sanou I, *et al*. Genotype diversity of group A rotavirus strains in children with acute diarrhea in urban Burkina Faso, 2008-2010. J Med Virol 2011;83:1485–90.
14. Nakata S, Gatheru Z, Ukae S, *et al*. Epidemiological study of the G serotype distribution of group A rotaviruses in Kenya from 1991 to 1994. J Med Virol 1999; 58:296–303.
15. Parashar UD, Alexander JP, Glass RI. Prevention of rotavirus gastroenteritis among infants and children. Recommendations of the Advisory Committee on Immunization Practices (ACIP). MMWR Recomm Rep 2006;55:1–13.
16. Salinas B, Pérez Schael I, Linhares AC, *et al*. Evaluation of safety, immunogenicity and efficacy of an attenuated rotavirus vaccine, RIX4414: a randomized, placebo-controlled trial in Latin American infants. Pediatr Infect Dis J 2005;24:807–16.
17. Coulson BS, Holmes IH. An improved enzyme-linked immunosorbent assay for the detection of rotavirus in faeces of neonates. J Virol Methods 1984;8:165–79.
18. Akran V, Peenze I, Akoua-Koffi C, *et al*. Molecular characterization and genotyping of human rotavirus strains in Abidjan, Cote d'Ivoire. J Infect Dis 2010; 202:S220–4.
19. De Donno A, Grassi T, Bagordo F, *et al*. Emergence of unusual human rotavirus strains in Salento, Italy, during 2006–2007. BMC Infect Dis 2009;9:43.
20. Cilla G, Montes M, Gomariz M, *et al*. Rotavirus genotypes in children in the Basque Country (North of Spain): rapid and intense emergence of the G12 [P8] genotype. Epidemiol Infect 2013;141:868–74.
21. Nokes DJ, Peenze I, Netshifhefhe L, *et al*. Rotavirus genetic diversity, disease association, and temporal change in hospitalized rural Kenyan children. J Infect Dis 2010;202:S180–6.
22. Enweronu-Laryea CC, Sagoe KW, Glover-Addy H, *et al*. Prevalence of severe acute rotavirus gastroenteritis and intussusceptions in Ghanaian children under 5 years of age. J Infect Dev Ctries 2011;6:148–55.
23. Yen C, Tate JE, Patel MM, *et al*. Rotavirus vaccines: update on global impact and future priorities. Hum Vaccin 2011;7:1282–90.
24. Dennehy PH. Acute diarrheal disease in children: epidemiology, prevention, and treatment. Infect Dis Clin North Am 2005;19:585–602.
25. Perl S, Goldman M, Berkovitch M, *et al*. Characteristics of rotavirus gastroenteritis in hospitalized children in Israel. Isr Med Assoc J 2011;13:274–7.