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Effectiveness of pentavalent rotavirus vaccine against a diverse range of circulating rotavirus strains in Nicaragua

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Abstract

Background: Because over 60 rotavirus strains have been reported worldwide, concerns exist about strain replacement after the introduction of rotavirus vaccines particularly in developing country settings where strains are diverse and efficacy is lower.

Methods: We used the case-control design in 4 hospitals in Nicaragua to assess strain-specific vaccine effectiveness (VE) of a pentavalent rotavirus vaccine (RotaTeq) against rotavirus diarrhea. Cases were identified through prospective strain surveillance with reverse transcription-polymerase chain reaction for 3 years among children hospitalized for diarrhea, and controls were children negative for rotavirus.

Results: We enrolled 1178 case-patients, 1082 (92%) with G and P typing, and 4,927 controls. A different strain predominated each year with increasing age of the vaccine-eligible cohort during the study period: G2P[4] in 2008 (97%; mean age = 11.9 months), G1P[8] in 2009 (55%; mean age = 17.0 months), and G3P[8] in 2010 (78%; mean age = 17.3 months). Overall VE was 45% (95% CI: 25 to 59). Regardless of the strain, VE estimates were 12%-79% lower among children older than 12 months relative to those 6-11 months of age, consistent with waning immunity observed for RotaTeq in Africa. Thus, the lower VE for G3P[8] was related to the higher mean age of cases (17.3 months) compared to the G2P[4] strains (11.9 months), with a significant trend ($R^2=0.819$; $P<0.001$) of declining effectiveness with increasing mean age of the cases.

Conclusions: Introduction of RotaTeq did not result in sustained emergence of any particular strain in Nicaragua. Variation in strain specific effectiveness was due to an age-related decline in effectiveness rather than differences in protection against the observed strains.

Rotavirus is the leading cause of severe and fatal diarrhea among children <5 years of age. In 2009, the World Health Organization recommended two rotavirus vaccines (RotaTeq—

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Merck & Co; and Rotarix—GlaxoSmithKline).¹ Over the past 5 years, rotavirus vaccines have substantially reduced the number of children dying and being hospitalized from diarrhea in countries that have introduced a vaccine.²⁻¹²

Nicaragua, a lower-middle income GAVI eligible country in Central America, introduced RotaTeq in October 2006.¹⁰ RotaTeq is a pentavalent bovine human reassortant vaccine which expressed the 4 most common human rotavirus G-types (G1-G4) and the most common P-type, P1A[8].¹³ Because strains in the rotavirus vaccines might be different from those circulating in nature, the possibility exists that evolutionary pressure exerted by rotavirus vaccines might result in emergence of select strains after the introduction of vaccine.¹⁴

Secular strain variation before the introduction of RotaTeq was quite common in Nicaragua, changing from G2P[4] in 2001, to G1P[8] in 2002, and G3P[8] in 2003.¹⁵ During the 2006–2007 rotavirus season in Nicaragua, when few children had yet to be vaccinated, the partially heterotypic G2P[4] and the fully homotypic G4P[8] comprised approximately 40.5% and 53.2% of the circulating rotavirus strains.¹⁶ However, in 2007–2008, the first rotavirus season after one birth cohort was vaccinated with RotaTeq, G2P[4] became the predominant circulating strain in Nicaragua, suggesting natural fluctuation as the likely cause of the strain predominance.¹⁰ A case-control study demonstrated that effectiveness of RotaTeq in Nicaragua was ~50% against severe rotavirus disease from this strain, similar to the efficacy of the vaccine against diverse homotypic and heterotypic strains in low-income countries in Africa and Asia.^{17,18} In this article, we estimate strain specific effectiveness of RotaTeq in Nicaragua using surveillance data collected during 2008–2010.

MATERIALS AND METHODS

Setting and design

The Nicaragua Ministry of Health added RotaTeq to the routine childhood immunization program in October 2006, recommending 3 doses of vaccine at 2, 4, and 6 months of age. After the introduction of vaccine, active surveillance for acute gastroenteritis was conducted at 4 hospitals in different cities across Nicaragua among children who were age-eligible for vaccine (i.e., those born after August 2006) from July 2007–June 2010. We used a case-control approach to assess the effectiveness of RotaTeq against children hospitalized for rotavirus gastroenteritis. To identify case-patients, we conducted active hospital-based surveillance for children with acute gastroenteritis, defined as (defined as ≥ 3 loose stools in a 24-hour period and with onset <14 days prior to the hospital visit), 24 hours a day in the emergency department (ED) and inpatient wards. We enrolled all children with acute gastroenteritis who were admitted or treated in the ED with intravenous hydration and were born after August 26, 2006. Case-patients were children who tested positive for laboratory-confirmed rotavirus diarrhea. Controls were children who were enrolled during the gastroenteritis surveillance but tested negative for rotavirus (ie, test-negative controls).

We report the strain distribution and strain-specific vaccine effectiveness (VE) results from Nicaragua obtained by comparing vaccination rates among children with laboratory-confirmed rotavirus diarrhea against the background rates of vaccine among children with acute gastroenteritis who tested negative for rotavirus (i.e., test-negative controls).

This evaluation was approved by the Human Subjects Offices at the Program for Appropriate Technology in Health, Pan American Health Organization, and the Nicaragua Ministry of Health.

Specimen collection

Bulk stool specimens were collected from children with acute gastroenteritis within 48 hours of admission. Specimens were stored at 2–8 °C prior to transfer to the national laboratory on a weekly basis, where rotavirus testing was conducted using a commercially available enzyme immunoassay (EIA) [IDEIA™; Oxoid, Cambridge, United Kingdom]. At the national laboratory in Nicaragua, specimens were frozen at –70°C. Rotavirus positive samples were shipped to the Centers for Disease Control and Prevention where they were maintained at –70°C until further analysis for strain characterization using reverse transcription-polymerase chain reaction (RT-PCR), as previously described.

Data Collection

Parents of children with acute gastroenteritis were interviewed face-to-face during the hospital visit to obtain information on vaccination history, demographics, socioeconomic factors, birth weight, prematurity at birth, body mass index, history of breastfeeding, and medical history. Information on clinical characteristics, treatment, and course of illness was obtained by interviewing the medical provider and reviewing the medical chart. Vaccination history was obtained from the parent and considered confirmed if the parent presented a vaccination card or through review of the vaccine registry at the clinic where the child was vaccinated.

Viral RNA Extraction and RT-PCR Genotyping

For EIA-confirmed positive samples, rotavirus double-stranded RNA (dsRNA) or total nucleic acid was extracted by using 1 of 3 methods and corresponding standard protocols: the automated NucliSens Extractor (Biomerieux, Durham, NC); the automated KingFisher extraction system (Thermo Electron Corporation, Vantaa, Finland); or the semi-automated miniMAG magnetic extractor (Biomerieux, Durham, NC).

VP7 and VP4 genotyping were carried out following previously described 2-step amplification methods^{19,20} with modification on a GeneAMP PCR System 9700 thermocycler (Applied Biosystems, Foster City, CA) using the One-Step RT-PCR kit (Qiagen, Inc., Valencia, CA). In this procedure, the dsRNA was denatured at 97°C for 5 minutes and then the reaction conditions were set up as described in the kit instructions. Reverse transcription was performed at 42°C for 30 minutes, followed by 15 minutes at 95°C, and the PCR was performed for 30 cycles consisting of 30 seconds at 95°C, 30 seconds at 42°C, and 1 minute at 72°C. Reactions were then subjected to a final extension step of 72°C for 7 minutes then held at 4°C. For all 3 seasons, genotyping PCR was performed as described previously.^{19,20} G-typing used primer 9Con1-L in combination with primers 9T1–1, 9T1-Dg, 9T2, 9T3P, 9T-4, and 9T-9B.¹⁹⁻²¹ P-typing used primer con3 in combination with primers 1T-1, 1T1-V, 2T-1, 3T-1, 4T-1, 5T-1, and JRG237²⁰⁻²² and new primer 1T1-A (5'-CAC GTC GAT CCA GTA GA-3'). Genotyping reactions were analyzed by electrophoresis on a 3% agarose gel using a 2:1 ratio of NuSieve GTG: SeaPlaque (FMC

Bioproducts, Rockland, ME). The G- and P-types obtained were classified according to the system described by Estes and Cohen.²³

During these three surveillance years, all available rotavirus positive stool samples were genotyped, and nucleotide sequencing and sequence analysis were performed.

Nucleotide Sequencing and Sequence Analysis

Isolates that did not yield a visible PCR product by RT-PCR genotyping were characterized by nucleotide sequencing of the con3/ con2 and 9Con1-L/VP7-RDg RT-PCR amplicons. In cases when a con3/con2 product was not obtained, the VP4 gene was reverse transcribed and amplified with the primer pair primers VP4F and VP4R²⁴ using the one-step RT-PCR kit. Similarly, in cases where a 9Con1-L/VP7-RDg RT-PCR product was not obtained, primer pair Beg9/End9 was employed.²⁵ Analysis of RT-PCR reactions by gel electrophoresis, amplicon purification, and DNA sequencing was carried out as described previously.²⁶ Forward and reverse sequences were assembled using Sequencher software versions 4.7 or 4.8 (Gene Codes Corporation, Inc, Ann Arbor, MI). The consensus sequences obtained were compared with existing rotavirus sequences in the NR/NT database using the BLASTN program at the National Center for Biotechnology Information website (available at: <http://www.ncbi.nlm.gov/BLAST/>). Specimens that did not yield a visible product by RT-PCR were considered to be nontypeable.²⁷

Statistical Analysis

We used descriptive statistics to assess strain prevalence by year. We also conducted an analysis of vaccine effectiveness of 3 doses of RotaTeq against hospitalization due to each specific strain or pooled strain combinations (e.g., G1, G2, G3, and G4 combined) where sample size was >200. This exploratory analysis would provide us power of 80% to exclude VE of 55% or higher for the strains or strain combinations detected in Nicaragua. For this analysis, we constructed an unconditional logistic regression model to calculate the odds ratio of vaccination between rotavirus positive cases and test-negative controls, and 95% confidence intervals around the odds ratio. Vaccine effectiveness was computed for all children >6 months of age, 6-11 months of age, and those ≥12 months of age. Potential confounders were defined as any variable that changed the odds ratio by more than 10%. The final model included age at hospitalization (in months), month/year of birth, and hospital, in addition to vaccination status. Vaccine effectiveness was calculated as 1 minus the odds ratio x 100%. Because previous analysis has identified decline in effectiveness by age, we also fit linear regression models to examine the association between age at hospitalization and serotype specific vaccine effectiveness. Statistical significance was designated as a P value < 0.05. Analyses were done with SAS statistical software (version 9.3).

RESULTS

From July 2007–June 2010, 1,178 of 4,927 (24%) children hospitalized for diarrhea tested positive for rotavirus as part of sentinel surveillance established by the Ministry of Health at the four hospitals. Among the stools testing positive for rotavirus by EIA, 1,082 (92%)

samples had G- and P-genotypes detected by RT-PCR: 261 (24%) during 2008, 486 (45%) during 2009, and 335 (31%) during 2010 (Table 1). Of these children with strain typing, 938 (87%) were older than six months and eligible for rotavirus vaccination and met the case-definition of the vaccine effectiveness study.

During 2008, the first rotavirus season after a full birth cohort was eligible for RotaTeq vaccination in Nicaragua, G2 was the most common G-type strain, accounting for 90% of all circulating strains at the study 4 hospitals in Nicaragua. Of the 236 G2 strains, 230 (97%) were G2P[4] strains, a strain that was only seen in 26 of 486 (5%) of the specimens during 2009 and none of the specimens during 2010. Strain diversity was more pronounced during 2009 compared with 2008 and 2010. During this year, among the 486 specimens where strains were detected, G1P[8] was the most common strain (55%) followed by G1P[4] (13%), G9P[8] (9%), G1P[mixed] (7%), G2P[4] (5%), G9P[mixed] (4%), and G2P[8] (2%). During 2010, G3P[8] emerged as the predominant strain, being detected in 260 of the 335 (78%) specimens, followed by G1P[8] which was detected in 14% of the specimens.

The mean age of children was different for the different common strains reflecting the increasing age of the vaccine-eligible cohort during the study period: 11.9 months (G2P[4]) in 2008, 17.0 months (G1P[8]) in 2009, and 17.3 months (G3P[8]) in 2010.

During the 3-year study period, differences in vaccine effectiveness were noted among the commonly detected strains that could be explained by the phenomenon of declining effectiveness with increasing age. The overall VE was 45% (95% CI: 25 to 59). Strain specific VE was 63% (95% CI: 42 to 77) against partially heterotypic G2P[4] and 42% (95% CI: 6 to 64) and 23% (95% CI: -85 to 68) against the fully homotypic G1P[8] and G3P[8] strains, respectively (Table 2). VE was higher against non-vaccine P-types strains compared to those with fully homotypic vaccine type strains (53% vs. 35%, respectively) supporting the contention that differences in efficacy were not related to the strain type. Moreover, we observed an absolute decline in vaccine effectiveness with age that ranged from 8% to 51% (Table 2) and a significant trend ($R^2=0.819$; $P<0.001$) of declining effectiveness with increasing mean age of the cases (Figure 1). Thus, because mean age of G3P[8] and G1P[8] cases (17.1 to 17.3 months) was higher compared to the G2P[4] cases (11.9 months), the differences in vaccine effectiveness between the strains is largely a function of the difference in the mean age of the cases.

Estimates were consistently similar (63%-71%) for younger children 6-11 months of age for all strains except for G3P[8] (VE = 40%; -203 to 88); however, confidence limits were wide for this strain due to the small sample size (Table 2). Vaccine effectiveness estimates were 12%-79% lower among children older than 12 months relative to those 6-11 months of age, consistent with waning immunity observed for oral rotavirus vaccines in some developing country settings. Among children older than 12 months, VE was higher for G2P[4] (60%) compared to other single or pooled genotypes (13%-46%), but the mean age was also significantly lower (15 months) for G2P[4] compared to the other genotypes (18 months) ($p<0.001$).

Discussion

Rotavirus vaccines have been evaluated in a number of clinical trials that typically cover one or two rotavirus seasons.^{28,29} Because over 60 rotavirus strains have been documented to circulate naturally, with periodic emergence of novel strains in the prevaccine era, the full impact of these new vaccines on circulating strains will only be realized through postlicensure evaluations globally after a number of years of vaccine use.¹⁴ In particular, concerns have been raised in the literature and by decision makers on whether the current rotavirus vaccines will provide adequate protection against diverse strains and whether vaccine introduction will lead to changes in strain prevalence.³⁰⁻³² Our data from 3 years of intensive surveillance of over 6,000 hospitalized children with diarrhea in a developing country setting provide a strong evidence base on the complex interaction between the pentavalent rotavirus vaccine, rotavirus strains, and the age of the children.

We did not observe any sustained predominance of a particular strain during the three-year study period. The G2P[4] strain which is partially heterotypic to RotaTeq was predominant during the year 1 after vaccine introduction. In contrast, the fully heterotypic strains G1P[8] and G3P[8] strains became predominant during years 2 and 3, respectively, after vaccine introduction. Previous surveillance has shown that G2P[4] was present in the year before vaccine uptake in Nicaragua, supporting natural secular variation as the likely cause of the G2P[4] predominance in 2008.¹⁶ Similarly, year-to-year strain changes were common before the introduction of RotaTeq in Nicaragua.¹⁵ These data indicate that, at least for the observed study period, the pentavalent rotavirus vaccine did not induce any selective pressure on rotavirus strains or lead to emergence of novel or breakthrough strains.

For all genotypes and pooled combinations, we observed a consistent and significant decline in rotavirus VE among children older than 12 months compared to those 6-11 months of age. Similar findings of declining effectiveness with age have been observed in clinical trials from regions in Africa with the highest child mortality suggesting waning of vaccine induced protection from live, oral rotavirus vaccines.¹⁷ A previous study of rotavirus vaccine impact on nationwide hospitalizations from diarrhea also demonstrated epidemiological findings consistent with the declining vaccine effectiveness in older children in Nicaragua.¹¹ In our current study, effectiveness was lower during the second year with all strains suggesting that this was an effect related to waning immunity rather than a strain-specific phenomenon. This finding was different from a recently published vaccine effectiveness study of the monovalent rotavirus vaccine in Bolivia, a low-middle income setting. Both studies applied comparable WHO recommended case-definitions and severity scores thus suggesting that the differences are related to other factors such as host or vaccine differences. As these vaccines are introduced in diverse settings across Asia and Africa, further evaluation and monitoring is necessary to elucidate reasons for varying rotavirus vaccine performance in lower income settings.

Indeed, the mean age of children infected with the fully homotypic strains (G1P[8] and G3P[8]) was substantially higher (range 17-17.3 months) compared to those infected with the partially heterotypic strains (G2P[4], G1-P4 with non P[8] combinations) (range 11.9-12.9 months). Because vaccine effectiveness is expected to be higher against fully

homotypic G and P type combinations than partially or fully heterotypic combinations and because we observed a consistent decline in effectiveness with age for all strain types, we suspect an age effect was the likely explanation for the variation in effectiveness between the different strains.

Our study has limitations. We did not have any prevaccine data on strains or diarrhea surveillance in our study hospitals, but yearly changes in strains were reported from other sites in Nicaragua before the introduction of RotaTeq. We were also reassured by the absence of a persistent predominant strain. Moreover, national data on childhood hospitalizations for diarrhea have also shown a decline in disease burden consistent with the vaccine effectiveness. Ours was a case-control evaluation which assumes that the vaccinated and unvaccinated members of the community have a comparable risk of developing disease. Because we compared effectiveness against strains over 3 different rotavirus seasons, this assumption may have been violated particularly as the vaccination coverage increased during these early years after vaccine introduction. Thus, the strain specific effectiveness estimates may have different biases year to year. That said, the consistency in the age specific declines in effectiveness, with lower effectiveness among strain groups with higher mean ages is biologically consistent (with waning) and argues against bias as an explanation for the differences in effectiveness by strain.

In conclusion, in the largest strain-specific postlicensure evaluation of the pentavalent rotavirus vaccine to date, we demonstrated that vaccine has not led to a sustained predominance of a particular strain or emergence of a novel strain in a developing country setting. The decline in vaccine effectiveness was a consistent finding for all rotavirus strains in Nicaragua during the three-year study period and was related to age-specific decline in VE. Because of waning immunity with oral rotavirus vaccines in developing countries, careful attention is warranted to disentangle the effects of age and strain to accurately determine the relationship between vaccination and potential strain emergence after vaccine introduction.

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Disclaimer:

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REFERENCES

1. WHO. Rotavirus vaccines:an update. Wkly Epidemiol Rec. Dec 18 2009;84(50):533–40. [PubMed: 20034143]

2. do Carmo GM, Yen C, Cortes J, et al. Decline in diarrhea mortality and admissions after routine childhood rotavirus immunization in Brazil: a time-series analysis. *PLoS Med.* Apr 2011;8(4):e1001024. doi:10.1371/journal.pmed.1001024 [PubMed: 21526228]
3. Lanzieri TM, Linhares AC, Costa I, et al. Impact of rotavirus vaccination on childhood deaths from diarrhea in Brazil. *Int J Infect Dis.* Mar 2011;15(3):e206–10. doi:10.1016/j.ijid.2010.11.007 [PubMed: 21193339]
4. Richardson V, Hernandez-Pichardo J, Quintanar-Solares M, et al. Effect of rotavirus vaccination on death from childhood diarrhea in Mexico. *N Engl J Med.* Jan 28 2010;362(4):299–305. doi:10.1056/NEJMoa0905211 [PubMed: 20107215]
5. Richardson V, Parashar U, Patel M. Childhood diarrhea deaths after rotavirus vaccination in Mexico. *N Engl J Med.* Aug 25 2011;365(8):772–3. doi:10.1056/NEJMc1100062 [PubMed: 21864191]
6. Braeckman T, Van Herck K, Raes M, Vergison A, Sabbe M, Van Damme P. Rotavirus vaccines in Belgium: Policy and Impact *Pediatr Infect Dis.* 2011;(in this Supplement)
7. Buttery JP, Lambert SB, Grimwood K, et al. Impact of rotavirus vaccination on acute gastroenteritis hospitalizations among Australian children. *Pediatr Infect Dis.* 2011;(in this Supplement)
8. Cortes JE, Curns AT, Tate JE, et al. Rotavirus vaccine and health care utilization for diarrhea in U.S. children. *N Engl J Med.* Sep 22 2011;365(12):1108–17. doi:10.1056/NEJMoa1000446 [PubMed: 21992123]
9. de Palma O, Cruz L, Ramos H, et al. Effectiveness of rotavirus vaccination against childhood diarrhoea in El Salvador: case-control study. *BMJ.* 2010;340:c2825. [PubMed: 20551120]
10. Patel M, Pedreira C, De Oliveira LH, et al. Association between pentavalent rotavirus vaccine and severe rotavirus diarrhea among children in Nicaragua. *JAMA.* Jun 3 2009;301(21):2243–51. doi:10.1001/jama.2009.756 [PubMed: 19491186]
11. Patel M, Pedreira C, De Oliveira LH, et al. Duration of protection of pentavalent rotavirus vaccination in Nicaragua. *Pediatrics.* Aug 2012;130(2):e365–72. doi:10.1542/peds.2011-3478 [PubMed: 22753550]
12. Patel MM, Glass R, Desai R, Tate JE, Parashar UD. Fulfilling the promise of rotavirus vaccines: how far have we come since licensure? *Lancet Infect Dis.* Jul 2012;12(7):561–70. doi:10.1016/S1473-3099(12)70029-4 [PubMed: 22742639]
13. Clark HF, Offit PA, Ellis RW, et al. The development of multivalent bovine rotavirus (strain WC3) reassortant vaccine for infants. *J Infect Dis.* Sep 1996;174 Suppl 1:S73–80. doi:10.1093/infdis/174.supplement_1.s73 [PubMed: 8752294]
14. Gentsch JR, Laird AR, Bielfelt B, et al. Serotype diversity and reassortment between human and animal rotavirus strains: implications for rotavirus vaccine programs. *J Infect Dis.* Sep 1 2005;192 Suppl 1:S146–59. [PubMed: 16088798]
15. Espinoza F, Bucardo F, Paniagua M, Svensson L, Hallander HO, Bondeson K. Shifts of rotavirus g and p types in Nicaragua--2001-2003. *Pediatr Infect Dis J.* Nov 2006;25(11):1078–80. doi:10.1097/01.inf.0000242739.44424.0e [PubMed: 17072136]
16. Khawaja S, Cardellino A, Mast TC. Hospital-based surveillance and analysis of genotype variation in Nicaragua after the introduction of the pentavalent rotavirus vaccine. *Pediatr Infect Dis J.* Jan 2014;33(1):e25–8. doi:10.1097/INF.0000000000000074 [PubMed: 24042492]
17. Armah GE, Sow SO, Breiman RF, et al. Efficacy of pentavalent rotavirus vaccine against severe rotavirus gastroenteritis in infants in developing countries in sub-Saharan Africa: a randomised, double-blind, placebo-controlled trial. *Lancet.* Aug 21 2011;376(9741):606–14. doi:S0140-6736(10)60889-6 [pii] 10.1016/S0140-6736(10)60889-6
18. Zaman K, Dang DA, Victor JC, et al. Efficacy of pentavalent rotavirus vaccine against severe rotavirus gastroenteritis in infants in developing countries in Asia: a randomised, double-blind, placebo-controlled trial. *Lancet.* Aug 21 2011;376(9741):615–23. doi:S0140-6736(10)60755-6 [pii] 10.1016/S0140-6736(10)60755-6
19. Das BK, Gentsch JR, Cicirello HG, et al. Characterization of rotavirus strains from newborns in New Delhi, India. *J Clin Microbiol.* Jul 1994;32(7):1820–2. [PubMed: 7929782]
20. Gentsch JR, Glass RI, Woods P, et al. Identification of group A rotavirus gene 4 types by polymerase chain reaction. *J Clin Microbiol.* Jun 1992;30(6):1365–73. [PubMed: 1320625]

21. Freeman MM, Kerin T, Hull J, McCaustland K, Gentsch J. Enhancement of detection and quantification of rotavirus in stool using a modified real-time RT-PCR assay. *J Med Virol.* Aug 2008;80(8):1489–96. doi:10.1002/jmv.21228 [PubMed: 18551614]
22. Esona MD, Steele D, Kerin T, et al. Determination of the G and P types of previously nontypeable rotavirus strains from the African Rotavirus Network, 1996-2004: Identification of unusual G types. *J Infect Dis.* Sep 1 2010;202 Suppl:S49–54. doi:10.1086/653552 [PubMed: 20684717]
23. Estes MK, Cohen J. Rotavirus gene structure and function. *Microbiol Rev.* Dec 1989;53(4):410–49. doi:10.1128/mr.53.4.410-449.1989 [PubMed: 2556635]
24. Simmonds MK, Armah G, Asmah R, et al. New oligonucleotide primers for P-typing of rotavirus strains: Strategies for typing previously untypeable strains. *J Clin Virol.* Aug 2008;42(4):368–73. doi:10.1016/j.jcv.2008.02.011 [PubMed: 18378188]
25. Gouvea V, Glass RI, Woods P, et al. Polymerase chain reaction amplification and typing of rotavirus nucleic acid from stool specimens. *J Clin Microbiol.* Feb 1990;28(2):276–82. [PubMed: 2155916]
26. Esona MD, Geyer A, Page N, et al. Genomic characterization of human rotavirus G8 strains from the African rotavirus network: relationship to animal rotaviruses. *J Med Virol.* May 2009;81(5):937–51. doi:10.1002/jmv.21468 [PubMed: 19319943]
27. Griffin DD, Fletcher M, Levy ME, et al. Outbreaks of adult gastroenteritis traced to a single genotype of rotavirus. *J Infect Dis.* May 15 2002;185(10):1502–5. doi:10.1086/340218 [PubMed: 11992287]
28. Vesikari T, Karvonen A, Prymula R, et al. Efficacy of human rotavirus vaccine against rotavirus gastroenteritis during the first 2 years of life in European infants: randomised, double-blind controlled study. *Lancet.* Nov 24 2007;370(9601):1757–63. doi:10.1016/S0140-6736(07)61744-9 [PubMed: 18037080]
29. Vesikari T, Matson DO, Dennehy P, et al. Safety and efficacy of a pentavalent human-bovine (WC3) reassortant rotavirus vaccine. *N Engl J Med.* Jan 5 2006;354(1):23–33. [PubMed: 16394299]
30. Grimwood K, Kirkwood CD. Human rotavirus vaccines: too early for the strain to tell. *Lancet.* Apr 5 2008;371(9619):1144–5. [PubMed: 18395562]
31. Gurgel RQ, Cuevas LE, Vieira SC, et al. Predominance of rotavirus P[4]G2 in a vaccinated population, Brazil. *Emerging infectious diseases.* Oct 2007;13(10):1571–3. [PubMed: 18258011]
32. Matthijnssens J, Nakagomi O, Kirkwood CD, Ciarlet M, Desselberger U, Van Ranst M. Group A rotavirus universal mass vaccination: how and to what extent will selective pressure influence prevalence of rotavirus genotypes? Expert review of vaccines. Nov 2012;11(11):1347–54. doi:10.1586/erv.12.105 [PubMed: 23249234]

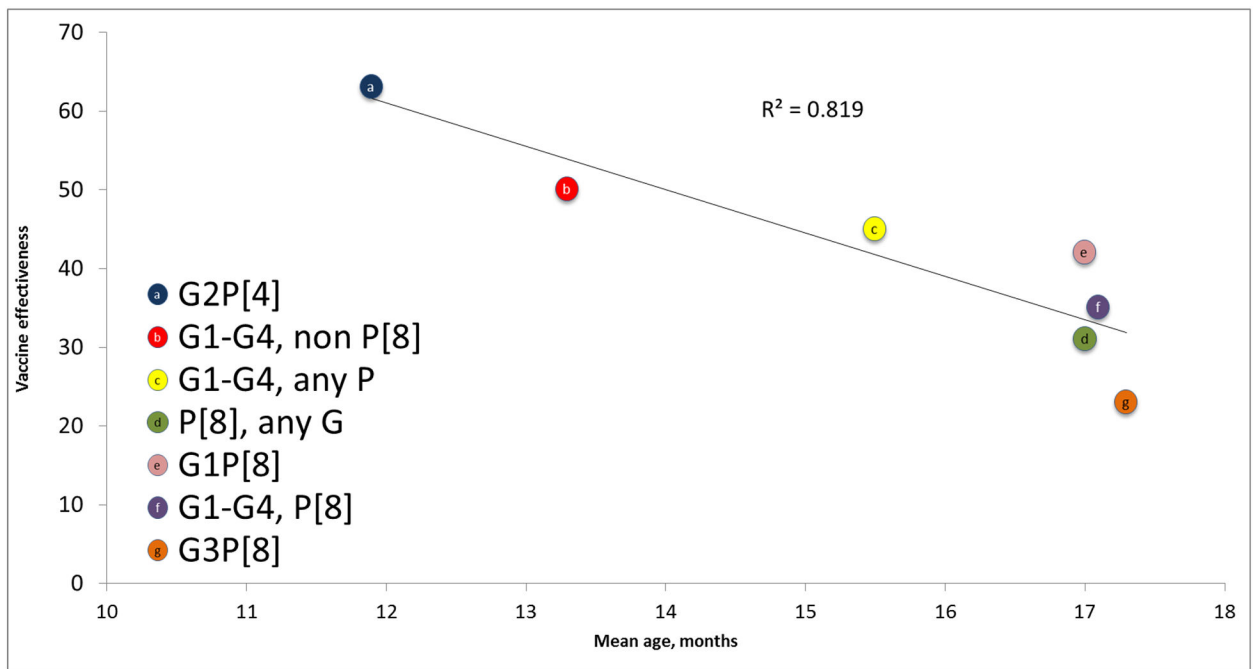


Figure 1.
Relationship between strain specific vaccine effectiveness and mean age at hospitalization.

Table 1.

Distribution of strains in Nicaragua after the introduction of RotaTeq, 2008-2010

	2008		2009		2010	
	N	%	N	%	N	%
TOTAL	261	100%	486	100%	335	100%
G1P[4]			63	13%		
G1P[8]	14	5%	269	55%	47	14%
G1P[M]	3	1%	34	7%		
G1P[NT]			1	0%	2	1%
G2P[4]	230	88%	26	5%		
G2P[8]	2	1%	9	2%		
G2P[M]	4	2%	1	0%		
G3P[6]					1	0%
G3P[8]			1	0%	260	78%
G3P[NT]					13	4%
G4P[6]	1	0%			1	0%
G4P[8]			4	1%	1	0%
G4P[NT]			1	0%		
G6P[1]	1	0%				
G9P[4]					2	1%
G9P[8]			43	9%		
G9P[M]			21	4%	2	1%
GM P[4]	2	1%	1	0%		
GM P[6]	1	0%				
GM P[8]	2	1%	4	1%		
GM P[M]	1	0%	1	0%		
GNT P[4]					1	0%
GNT P[6]					1	0%
GNT P[8]			3	1%	4	1%
GNT P[M]			2	0%		
GNT P[NT]			2	0%		

Table 2.

Effectiveness^a of RotaTeq against hospitalization related to select serotypes or serotype combinations, ordered by decreasing vaccine effectiveness among all ages

Strain	All Ages			Age 6-11 months			Age ≥ 12 months		
	Mean Age ^b	No.	VE (95% CI)	Mean Age ^b	No.	VE (95% CI)	Mean Age ^b	No.	VE (95% CI)
G2P[4]	11.9	214	63 (42 to 77)	8.7	104	68 (37 to 84)	15.0	110	60 (22 to 80)
Partially heterotypic ^c	13.3	374	50 (26 to 66)	8.8	167	63 (34 to 80)	17.0	207	37 (-8 to 64)
G1-G4, any P	15.5	852	45 (25 to 60)	8.8	292	63 (40 to 78)	19.0	560	34 (1 to 55)
P[8], any G	17.0	580	31 (-6 to 55)	8.7	161	62 (22 to 82)	20.2	419	13 (-46 to 48)
G1P[8]	17.0	289	42 (6 to 64)	8.9	76	71 (31 to 88)	19.9	213	25 (-34 to 58)
G1-G4, P[8]	17.1	529	35 (-1 to 58)	8.7	143	64 (21 to 83)	20.2	386	20 (-34 to 53)
G3P[8]	17.3	230	23 (-85 to 68)	8.6	66	40 (-203 to 88)	20.9	164	22 (-123 to 73)

^a: protection conferred against rotavirus hospitalization by 3 doses 14 days after vaccine receipt. Unconditional logistic regression used to calculate vaccine effectiveness using test negative controls. Adjusted for month and year of birth, age at hospitalization, and hospital.

^b: mean age among children >6 months of age who were included in this analysis

^c: G and P combinations that are partially heterotypic to RotaTeq: G1-G4, non P[8] and non-G1-G4, P[8]