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Timing of Rotavirus Vaccine Doses and Severe Rotavirus Gastroenteritis among Vaccinated Infants in Low- and Middle-Income Countries

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Abstract

Background—Altering rotavirus vaccine schedules may improve vaccine performance in low- and middle-income countries. We analyzed data from clinical trials of the monovalent (RV1) and pentavalent (RV5) rotavirus vaccines in low- and middle-income countries to understand the association between vaccine dose timing and severe rotavirus gastroenteritis incidence.

Methods—We assessed the association between variations in rotavirus vaccine administration schedules and severe rotavirus gastroenteritis risk. We used the complement of the Kaplan–Meier survival estimator to estimate risk differences for different schedules. To adjust risk differences (RDs) for confounding, we calibrated estimates in the vaccinated arm using estimates from the placebo arm.

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Process to Replicate Results: Applications to access the GSK and Merck and Co, Inc. clinical trial data can be submitted to ClinicalStudyDatarequest.com and http://engagezone.msd.com/ds_documentation.php, respectively. Computing code can be made available by the first author.

Conflicts of Interest: Data for this manuscript were provided by GlaxoSmithKline (GSK), Rixensart, Belgium through a third party, ClinicalStudyDatarequest.com, and by Merck and Co, Inc., Kenilworth, NJ USA. Neither company had involvement in this study design and analysis. Both Merck and Co, Inc., Kenilworth, NJ USA and GSK, Rixensart, Belgium were given the opportunity to comment on this manuscript prior to publication. All decisions regarding the content of this manuscript were made by the authors. Dr. Gruber was employed by Merck and Co, Inc., Kenilworth, NJ USA as a graduate research assistant from January – December 2014 to work on research related to their rotavirus vaccine. Dr. Jonsson Funk is a member of the Scientific Steering Committee (SSC) for a post-approval safety study of an unrelated drug class funded by GSK. All compensation for services provided on the SSC is invoiced by and paid to UNC Chapel Hill. Dr. Jonsson Funk receives salary support from the Center for Pharmacoepidemiology in the Department of Epidemiology, Gillings School of Global Public Health (current members: GSK, UCB BioSciences, Merck and Co, Inc.). Dr. Jonsson Funk does not accept personal compensation of any kind from any pharmaceutical company. Dr. Brookhart has received investigator-initiated research funding from the NIH and through contracts with the AHRQ's DEcIDE program and the PCORI. Within the past three years, he has received research support from Amgen and AstraZeneca and has served as a scientific advisor for Amgen, Merck and Co, Inc., GSK, UCB BioSciences, and RxAnte. Within the past three years, Dr. Brookhart has received research support from Amgen and AstraZeneca and has served as a scientific advisor for Amgen, Merck, GSK, Genentech, TargetPharma, and RxAnte. Dr. Brookhart owns equity in NoviSci, LLC, a data sciences company.

Results—There were 3114 and 7341 children included from the RV1 and RV5 trials, respectively. The 18-month adjusted severe rotavirus gastroenteritis risk was 4.0% (95% confidence interval (CI): 1.1, 7.1) higher for those receiving their first RV5 dose at <6 versus 6 weeks. For RV1, there was a 4.0% (95% CI: 0.0, 8.2) increase in 12-month adjusted risk for a 4- versus 6-week interval between doses. Further analysis revealed those receiving their first RV5 dose at 3–4 and 5–7 weeks had 2.9% (95% CI: 0.8, 5.3) and 1.3% (95% CI: –0.3, 3.0), respectively, higher risk compared to those at 9–12 weeks. Those receiving their first dose at 8 weeks had the lowest risk [RD: –2.6% (95% CI: –5.4, –0.1)] compared to those at 9–12 weeks.

Conclusions—A modest delay in rotavirus vaccination start and increase in interval between doses may be associated with lower severe rotavirus gastroenteritis risk in low- and middle-income countries.

Keywords

rotavirus; vaccines; developing countries; vaccine schedules; infants; gastroenteritis

Introduction

Despite the success of rotavirus vaccines at reducing health care encounters including hospitalizations,^{1–3} rotavirus vaccine effectiveness remains lower in low- and middle-income countries than in high-income countries.^{4,5} Many factors may contribute this lower vaccine effectiveness including: concomitant vaccination with oral polio vaccine;^{6,7} malnutrition;^{8,9} interference by transplacental maternal antibodies;^{10,11} and environmental enteropathy and the infant microbiota.^{12–14}

Some factors shown to decrease rotavirus vaccine performance may be overcome by altering rotavirus vaccine schedules. A few studies have investigated the influence of vaccine schedules on vaccine performance,^{10,11,15} but these studies have generally been restricted to immunologic endpoints. Since there is no known correlate of protection for anti-rotavirus immunoglobulin A (IgA) levels,^{16,17} there is still uncertainty about the effect alternative schedules have on clinical endpoints. No data have been reported on the effect of timing of commonly used rotavirus vaccines as they are routinely administered (i.e., two doses of the monovalent [RV1, Rotarix™; GlaxoSmithKline Biologicals, Rixensart, Belgium] and three doses of the pentavalent [RV5, RotaTeq™; Merck & Co, Inc.; Kenilworth, NJ, USA] rotavirus vaccines) using clinical endpoints.

We analyzed data from two large rotavirus vaccine trials to understand the association of timing of rotavirus vaccine doses and incidence of severe rotavirus gastroenteritis among children in low- and middle-income countries.

Methods

Parent Study Data

This analysis used data from two randomized, Phase III, placebo-controlled, multicenter clinical trials of RV1 and RV5 (Clinical Trial Number: NCT00241644 and NCT00362648,

respectively) conducted in low- and middle-income countries. The trials have been described in depth elsewhere,^{18–20} but a brief summary of each trial is below.

The RV1 clinical trial was conducted in South Africa and Malawi from 2005 – 2009. Healthy infants aged 5 to 10 weeks were enrolled and randomized to receive three placebo doses, a placebo dose followed by two RV1 doses, or three RV1 doses at approximately 6, 10, and 14 weeks of age. Enrolled infants were actively followed for occurrence of gastroenteritis from enrollment until study conclusion at 1 year of age with a subset followed for up to 2 years of age. Study staff visited parents or guardians at the homes of participants weekly to collect diary cards and also visited health clinics serving the study population. Stool samples were collected and tested for rotavirus using an enzyme-linked immunosorbent assay (Rotaclone, Meridian Bioscience) followed by reverse transcriptase polymerase chain reaction (RT-PCR) confirmation. Gastroenteritis was defined as three or more, looser than normal stools within a 24-hour period. Rotavirus gastroenteritis severity was determined using the 20-point Vesikari score.²¹

The RV5 study was conducted in Ghana, Kenya, Mali, Bangladesh, and Vietnam from 2007 – 2009. Infants four to 12 weeks were enrolled and randomly assigned to receive three doses of vaccine or placebo at approximately six, 10, and 14 weeks of age. During the study, there was active surveillance for gastroenteritis at local clinics and hospitals. Any participant presenting with gastroenteritis provided a stool sample for testing of rotavirus using an enzyme immunoassay followed by RT-PCR confirmation. Gastroenteritis was defined as three or more watery or looser than normal stools within a 24-hour period or forceful vomiting. RVGE severity was classified using the 20-point modified Vesikari score.^{21–23}

Study Data

We analyzed data from the placebo and vaccinated arms of each trial as they are recommended for use (two doses of RV1 and three doses of RV5). In the RV1 trial, we included infants randomized to receive placebo or two doses of RV1 (precise schedules were three placebo doses, or one placebo dose and two RV1 doses). In the RV5 trial, we included infants randomized to receive three doses of placebo, or three doses of RV5. Each trial was analyzed separately, but the results are presented in parallel. This research was approved by the University of North Carolina at Chapel Hill Institutional Review Board (15–0401).

Study Design Overview

We used a modified version of the study design described by Hernán et al. 2006²⁴ to understand if timing of rotavirus vaccine doses, which was not randomized, was associated with risk of severe rotavirus gastroenteritis. We began by defining five different aspects of vaccine timing. For a specific aspect of timing (e.g., timing of first dose), we compared two or more predefined schedules related to that aspect of timing (e.g., first dose given at < 10 versus 10 weeks). We compared the schedules by assessing the cumulative risk of severe rotavirus gastroenteritis on an age-specific time scale to include severe episodes from 12 weeks of age. We did this by partitioning the follow-up time of infants such that infants could contribute person-time and severe events to more than one of the predefined schedules so schedules with delayed vaccine doses included severe episodes after 12 weeks of age in

their estimated associations. After partitioning follow-up time and events, we estimated the association between each schedule and incidence of severe rotavirus gastroenteritis. We used the estimated associations of schedules in the placebo arm, which should have a null effect, to calibrate the estimates within the rotavirus vaccinated arm to obtain adjusted estimates of the associations. This approach is similar to the negative control design described by Tchetgen Tchetgen (2013).²⁵

Defining Schedules

We created and compared simplified schedules to understand the aspects of timing that may be associated with risk of severe rotavirus gastroenteritis. Because of the numerous combinations of schedules received, we could not simply compare the schedules received in each trial. Instead, we classified the timing of doses *a priori* using five main aspects of dose timing: 1) timing of first-dose holding interval(s) between doses constant at 4 to 6 weeks; 2) timing of first dose; 3) timing of last dose; 4) length of interval(s) between doses; and 5) number of doses received at 10 weeks of age. For each aspect of timing, we defined and compared two or more schedules. All schedules were developed based on biologic plausibility, the potential for realistic interventions (e.g., alterations in rotavirus schedules that would fit at times routine vaccines are given as part of the Expanded Program on Immunization), and the nature of the data. The schedules for each aspect of timing are specified in Table 1 and a detailed description of each schedule can be found in eTable 1. Due to the number of associations estimated, we chose our primary aspect of timing *a priori* to be the timing of the first dose holding interval(s) between doses constant at 4 to 6 weeks. Completed weeks of age were used for all schedule definitions (e.g., 6 weeks and 5 days of age was categorized as 6 weeks of age)

Outcome

We classified the outcome, first episode of severe rotavirus gastroenteritis, as infants experiencing gastroenteritis with a Vesikari or modified-Vesikari score of 11. For all analyses, we analyzed data on an age-specific time scale with follow-up beginning at 12 weeks of age (the latest time any infant enrolled in either trial). As mentioned above, we partitioned the follow-up time of infants such that infants could contribute person-time and severe gastroenteritis events to more than one schedule until the timing of their actual doses deviated from the schedule(s). This approach allowed us to include any severe events occurring after 12 weeks of age. Figure 1 presents three hypothetical infants and how their person-time and severe rotavirus gastroenteritis episodes were partitioned for timing of last dose. In this example, all infants contributed person-time to both predefined schedule until the actual timing of their last doses deviated from the predefined schedule. For example, infant A experienced a severe episode at 14 weeks of age, and this event was included in both schedules, because at 14 weeks of age, the timing of infant A's last dose was consistent with both predefined schedules. Additional details of the approach are included in eTable 2.

Covariates

We categorized covariate data on demographic information; breastfeeding and growth status; and infection status, antibiotic use, and other vaccinations. We categorized growth status

using the World Health Organization (WHO) criteria.²⁶ We classified infection status, antibiotic use, and concomitant vaccination using medical histories.

Statistical Analysis

To estimate the association between timing of rotavirus vaccine doses and incidence of severe rotavirus gastroenteritis, we estimated risk differences (RDs) and ratios (RRs) between different schedules for each aspect of timing at 6, 12, and 18 months of age using cumulative risk estimates obtained from the complement of the Kaplan–Meier survival estimator at those time points.²⁷ We chose *a priori* to focus on 12 months of age as our primary time point of interest, because that provided adequate time for severe gastroenteritis events to occur while allowing a majority of participants to remain in the cohorts. We did not estimate RDs or RRs at specific time points if any schedule had fewer than five severe events at that time point. We also estimated hazard ratios using Cox proportional hazard models.

There was potential for bias in the association between dose timing and severe rotavirus gastroenteritis, because of confounding and administrative censoring in the study design. Data from the placebo arms was used as a negative control in order to adjust for both potential sources of bias. Since the timing of placebo doses should not influence the incidence of severe rotavirus gastroenteritis in the placebo arm, any association observed would be due to bias. This association provided an estimate of amount of bias expected from uncontrolled confounding within the vaccinated arms, assuming potential uncontrolled confounders influencing the timing of receipt of doses in the placebo arms were the same confounders as those in the rotavirus-vaccinated arms. Associations in the placebo arm were used to calibrate (i.e., adjust) the estimates among those in the vaccinated arm. The directed acyclic graphs (DAGs) in eFigure 1 provide a conceptual diagram of this approach.

Before calibrating estimates, we empirically verified that imbalances in measured covariates between schedules were similar in the placebo and vaccinated arms using standardized mean differences. Standardized mean differences were calculated as $(p_1 - p_2) / [\{p_1(1 - p_1) + p_2(1 - p_2)\} / 2]^{1/2}$, where p_1 was the proportion (or mean) of the binary covariate for a specific schedule (e.g., first dose at < 6 weeks) and p_2 was the proportion in a different schedule (e.g., first dose at ≥ 6 weeks). If the imbalance in covariates was similar (<10% difference) between the placebo and vaccinated arms, we assumed calibration of the estimates in the vaccinated arm would yield a sufficiently adjusted estimate of the associations.

To calibrate (i.e., adjust) the associations of rotavirus vaccine dose timing, we estimated RD and RR measures comparing schedules for each aspect of timing, as described above, for both the placebo and vaccinated arms of each trial. We then calibrated the estimates among those vaccinated with the estimates among those in the placebo arms by subtracting the difference measures and dividing the ratio measures (i.e., difference in differences and ratio of ratios).²⁸ A nonparametric bootstrap with 2000 sample draws with replacement was used to obtain the point estimates and 95% empirical confidence of intervals.²⁹ The median of the distribution of calibrated estimates was reported for the difference and ratio estimates, and the 2.5th and 97.5th percentiles of the distribution were reported for the lower and upper bounds of the 95% empirical confidence intervals.

We conducted additional *post hoc* analyses to assess the association between timing of first dose and incidence of severe rotavirus gastroenteritis. We estimated the 12-month RD of severe gastroenteritis by week of age at first dose, requiring 100 infants in each stratum. We then collapsed across ages with similar RDs to estimate the RDs more precisely. We also estimated the 12-month RD between a 6/10/14 and 8/12/16 week schedule for RV5. All analyses were performed using SAS Clinical Trial Data Transparency (Version 4.5.2; SAS Institute Inc., Cary, NC, USA).

Results

There were 3114 and 7341 children included in this analysis from the RV1 and RV5 trials, respectively (Table 2). Infants were followed for a median time of 286 and 301 days after 12 weeks of age in both the placebo and RV1 vaccinated arms, respectively. Median follow-up was 483 days after 12 weeks of age in both the placebo and RV5 vaccinated arms. A total of 154 (100 and 54 in placebo and RV1 arms, respectively) and 324 (205 and 119 in placebo and RV5 arms, respectively) severe rotavirus gastroenteritis events occurred in the RV1 and RV5 trials, respectively.

The distributions of measured covariates between schedules for each aspect of timing were similar for the placebo and rotavirus vaccinated arms (data available upon request). This provided empirical evidence for our assumptions for using calibration for adjustment. However, one comparison, first dose of RV1 with 4 – 6 weeks between doses, had a number of deviations, which may indicate presence of residual confounding.

First Dose with 4 – 6 Weeks between Doses

The adjusted risk of severe RVGE was about 2% higher for those infants receiving their first RV5 dose at 3 – 6 weeks of age compared to 10 – 12 weeks of age (holding intervals between all subsequent doses at 4 – 6 weeks) [12-month RD: 2.1% (95% confidence interval (CI): 0.1, 4.0); 12-month RR: 2.2 (95% CI: 0.7, 6.7)] (Figures 2 and 3).

First Dose

The 18-month adjusted risk of severe rotavirus gastroenteritis was 4.0% (95% CI: 1.1, 7.1) higher for those receiving their first RV5 dose at < 6 versus 6 weeks of age. The 18-month RR was 2.4 (95% CI: 1.3, 4.8).

Last Dose

Infants receiving their last RV5 doses at 15 versus > 15 weeks of age had almost a 2% increase in 12-month adjusted risk of severe rotavirus gastroenteritis [12-month RD: 1.9% (95% CI: 0.6, 3.2); 12-month RR: 1.7 (95% CI: 0.7, 4.2)]. This was slightly attenuated and less precise for RV1 [12-month RD: 1.4% (95% CI: -1.1, 4.1); 12-month RR: 0.7 (95% CI: 0.1, 2.2)].

Interval between Doses

For RV1, there was a 4.0% (95% CI: 0.0, 8.2) increase in 12-month adjusted risk for a 4- versus 6-week interval between doses [12-month RR: 3.6 (95% CI: 0.9, 31.6)]. This

association was diminished to about 2% when comparing a 5- to 6-week interval, but this estimate was imprecise [12-month RD: 1.9% (95% CI: -2.2, 5.9); 12-month RR: 4.5 (95% CI: 0.9, 46.5)].

Number of Doses 10 Weeks of Age

Those receiving only one dose of RV5 at 10 weeks of age had a higher adjusted risk of severe RVGE compared to those receiving three doses at 10 weeks [12-month RD: 2.6% (95% CI: 0.7, 4.7); 12-month RR: 4.8 (95% CI: 0.7, 75.3)]. When the analysis comparing one versus three doses of RV5 at 10 weeks of age excluded infants missing the second or third dose of vaccine, the effect seen was similar [12-month RD: 2.5% (95% CI: 0.5, 4.7); 12-month RR: 4.7 (95% CI: 0.7, 70.5)].

Additional Results

The estimated uncalibrated cumulative risk of severe rotavirus gastroenteritis stratified by schedule and treatment status (i.e., placebo or vaccinated) is shown for all comparisons in eFigures 2 – 10. Hazard ratios are presented in eFigure 11 and were similar to RRs. Calibrated, uncalibrated, and placebo RDs and RRs at 12 and 18 months are presented in eFigure 12 and 13, respectively. RDs and RRs were not estimated for any comparisons at 6 months of age and for first dose at < 6 versus 6 weeks of age schedule at 12 months of age, because there were fewer than five events for at least one schedule.

The *post hoc* assessment of the dose response relationship between timing of first dose and incidence difference of severe rotavirus gastroenteritis is presented in Figure 4. Those receiving their first RV5 dose at three to four and five to seven weeks had 2.9% (95% CI: 0.8, 5.3) and 1.3% (95% CI: -0.3, 3.0), respectively, higher severe gastroenteritis risk compared to those at 9–12 weeks. Those receiving their first dose at eight weeks had the lowest risk [RD: -2.6% (95% CI: -5.4, -0.1)] compared to those at 9–12 weeks. In addition, the estimated 12-month RD for a 6/10/14 compared to 8/12/14 week schedule of RV5 was 7.1% (95% CI: 2.9, 11.8).

Discussion

Comparisons from the RV5 trial indicated earlier vaccination of the first RV5 dose resulted in a higher risk of severe rotavirus gastroenteritis with this risk declining with later vaccination until approximately eight weeks of age. Importantly, these associations were present even when accounting for the occurrence of severe gastroenteritis episodes after 12 weeks of age. In a previous analysis conducted by the first author, there was heterogeneity of vaccine efficacy (comparing vaccinated infants to unvaccinated infants) for those with a first dose of RV5 received at < 8 weeks compared to 8 weeks of age.⁹ This analysis found a similar pattern when accounting for severe gastroenteritis episodes after 12 weeks of age and comparing different schedules among vaccinated infants. The results were consistent with trials investigating alternative rotavirus schedules reporting immunologic endpoints.³⁰ Vaccination with RV1 at 10/14 versus 6/10 weeks of age resulted in higher seroconversion percentages. At 18 weeks of age, the seroconversion percent difference comparing a 10/14

versus 6/10 week schedule was 12.8% (95% CI: 1.2, 23.2) in Ghana and 10.1% (95% CI: -0.4, 20.3) in Pakistan.^{10,11}

Lower incidence of severe rotavirus gastroenteritis with delays in first rotavirus vaccine dose could be due to a number of biologic factors including a decline in transplacentally-acquired anti-rotavirus immunoglobulin G (IgG) antibodies or changes in the microbiota that allow for a stronger immune response with slightly older ages at first dose. In Ghana and Pakistan, there was higher seroconversion among infants in the lowest compared to highest quartile of pre-vaccination IgG levels.^{10,11} In addition, a case control study of six week old Ghanaian infants found RV1 vaccine responders (post-vaccination IgA antibody levels ≥ 20 IU/mL) had microbiotas more closely resembling Dutch infants than Ghanaian non-responders (post-vaccination IgA antibody levels < 20 IU/mL).¹³ It may be possible such differences in microbiota between responders and non-responders can be overcome with age, leading to improved vaccine response when vaccinated at slightly later ages. However, a study in Vellore, India reported similar immunologic response to RV1 regardless of the presence of enteropathogens at the time of vaccination.³¹ Therefore, the intestinal microbiota may not be the mechanism by which the association between timing of rotavirus vaccine dose and severe gastroenteritis is mediated.

We also found an increase in risk of severe rotavirus gastroenteritis for those infants with a 4-week interval between RV1 doses compared to a six week interval between doses. For RV5, there was no association between length of interval between doses and severe gastroenteritis. To our knowledge, no studies have investigated the association between interval length and outcomes for rotavirus vaccines. A study of intervals between oral polio vaccine doses in Bangladesh reported similar seroconversion proportions among those vaccinated with a 2 versus 4-week interval between doses.³² However, seroconversion proportions for some poliovirus types were lower for the two versus four week intervals. A longer delay between doses may provide a greater booster effect for oral vaccines, including RV1. However, the underlying mechanism for the observed association was not investigated and remains unknown.

There were a number of limitations of this research. Since this was a re-analysis of previously collected data, there were restrictions on the comparisons we could assess. Also, we were unable to account for events that occurred before 12 weeks of age. In the RV5 cohort, no events occurred prior to 12 weeks of age, but three events did occur prior to 12 weeks of age in the RV1 trial (one and two in the placebo and RV1 arms, respectively). eTable 3 provides estimates from the placebo arms of each trial of the expected number of severe rotavirus gastroenteritis episodes that would occur by delaying vaccination start by two or four weeks. We were also unable to stratify by continent or country due to the relatively few number of severe gastroenteritis episodes. There may be important differences between low- and middle-income countries that we could not assess. In addition, there may have been residual confounding of some estimates that we were unable to account for in this analysis. Although the vast majority of schedules had similar covariate imbalances in the placebo and vaccinated arms, there were a few comparisons that did not deviate in a similar manner. This means that after calibration of estimates, these estimates may have had some residual confounding. In addition, because we were unsure of which aspect of timing could

potentially be associated with risk of severe rotavirus gastroenteritis, we defined *a priori* a number of schedules to compare for each aspect of timing. These multiple comparisons may have resulted in observing associations by random chance.

Despite these limitations, there were a number of strengths of this study. The data from these trials allowed us to assess how the timing of rotavirus doses relates to the occurrence of severe rotavirus gastroenteritis, which has not been previously reported. The study had four notable advantages: 1) our study used data from two trials of two rotavirus vaccines to understand the association between rotavirus vaccine timing; 2) the sample sizes of the trials were large enough to assess severe gastroenteritis as the outcome of interest; 3) we were able to use a novel study design to include rotavirus episodes after 12 weeks of age to ensure later schedules were penalized for any early episodes that occurred prior to receipt of the vaccine; and 4) data in the placebo arms were leveraged as a negative control to adjust estimates of the association between timing of rotavirus vaccine doses and incidence of severe gastroenteritis.

In this re-analysis of two clinical trials of rotavirus vaccines in low- and middle-income countries, we found there was an association between rotavirus vaccine dose timing and incidence of severe rotavirus gastroenteritis. These data suggest a modest delay in RV5 start and increase in interval between RV1 doses, when the series begins at approximately 10 weeks of age, may improve rotavirus vaccines performance in low- and middle-income countries. However, the decision to delay rotavirus vaccination needs to be carefully considered in conjunction with early rotavirus exposures and potential missed vaccination opportunities to fully evaluate the potential benefits of delaying rotavirus vaccination start in low- and middle-income countries.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Figure 1.
Example of the person-time and event categorization for three hypothetical infants.

^a Due to differences in the covariate patterns between the placebo and vaccine arms, the calibrated estimate should be interpreted with caution.

Figure 2.
Uncalibrated and calibrated risk differences (RDs) and 95% confidence intervals.

^a Due to differences in the covariate patterns between the placebo and vaccine arms, the calibrated estimate should be interpreted with caution.

Figure 3.
Uncalibrated and calibrated risk ratios (RRs) and 95% confidence intervals.

Figure 4.

Post hoc analysis of 12-month calibrated risk differences (RDs) and 95% confidence intervals for timing of first RV5 dose (referent 9 – 12 weeks).

Table 1.

Predefined rotavirus vaccine schedules for each aspect of dose timing.

Aspect of Timing	N ^a	N ^b	RV Type	Age at Dose 1 (weeks)	Timing of Dose 2	Timing of Dose 3
First dose with 4 – 6 weeks between doses	1167	1073	RV5	3 – 6	4 – 6 wks after 1 st dose	4 – 6 wks after 2 nd dose
	1669	1516	RV5	7 – 9	4 – 6 wks after 1 st dose	4 – 6 wks after 2 nd dose
	784	720	RV5	10 – 12	4 – 6 wks after 1 st dose	4 – 6 wks after 2 nd dose
	1299	1212	RV1	10 – 12	4 – 6 wks after 1 st dose	NA
First dose	207	166	RV1	13 – 16	4 – 6 wks after 1 st dose	NA
	519	506	RV5	< 6	10 wks after 1 st dose	10 wks after 2 nd dose
	3147	3063	RV5	6	10 wks after 1 st dose	10 wks after 2 nd dose
	2882	2805	RV5	< 10	10 wks after 1 st dose	10 wks after 2 nd dose
Last dose	784	764	RV5	10	10 wks after 1 st dose	10 wks after 2 nd dose
	1527	1216	RV5	7	11 wks of age	15 wks of age
	3520	2353	RV5	12	10 wks after 1 st dose	> 15 wks of age & 10 wks after 2 nd dose
	1048	420	RV1	11	15 wks of age	NA
Interval between doses	1559	1056	RV1	16	> 15 wks of age & 10 wks after 2 nd dose	NA
	2589	1310	RV5	12	4 wks after 1 st dose	4 wks after 2 nd dose
	3345	1453	RV5	12	4 or 5 wks after 1 st dose ^c	4 or 5 wks after 2 nd dose ^c
	3474	546	RV5	12	4, 5 or 6 wks after 1 st dose ^d	4, 5, or 6 wks after 2 nd dose ^d
Number of doses 10 weeks of age ^e	1559	337	RV1	16	4 wks after 1 st dose	NA
	1559	926	RV1	16	5 wks after 1 st dose	NA
	1559	167	RV1	16	6 wks after 1 st dose	NA
	1030	2	RV5	< 10	< 10 wks of age	< 10 wks of age
Number of doses 10 weeks of age ^e	3666	397	RV5	< 10	< 10 wks of age	10 wks of age & 32 wks of age
	3271	2432	RV5	< 10	10 wks of age & 32 wks of age	10 wks of age & 32 wks of age
	784	765	RV5	10	10 wks of age & 32 wks of age	10 wks of age & 32 wks of age

RV1, monovalent rotavirus vaccine; RV5, pentavalent rotavirus vaccine; Wks, weeks; NA, not applicable

- ^bNumber of infants beginning follow-up at 12 weeks of age in this schedule. Total sample size across schedules for an aspect of timing can sum to more than the total number of vaccinated infants in each trial, because infants can begin in > 1 schedule.
- ^cNumber of infants being followed for severe RVGE for a particular schedule at 6 months of age.
- ^dAt least one interval between the three doses must be 5 weeks.
- ^eAt least one interval between the three doses must be 6 weeks.
- ^fOther timing of doses resulting in the same number of doses received 10 are possible and are included in eTable 1.

Table 2.

Characteristics of the trial populations.

Infant Characteristics	Trial 1 (N = 3114)		Trial 2 (N = 7341)	
	RV1 (N = 1560)	Placebo (N = 1554)	RV5 (N = 3666)	Placebo (N = 3675)
Median Length of Follow-Up in Days from 12 Weeks of Age	301	286	483	483
First Severe Rotavirus Gastroenteritis Episode	54	100	119	205
Age in weeks at Vaccine or Placebo Receipt				
Dose 1 ^a , mean (SE)	11.2 (0.03)	11.3 (0.03)	7.6 (0.03)	7.5 (0.03)
Dose 2 ^b , mean (SE)	16.2 (0.04)	16.3 (0.04)	12.2 (0.04)	12.1 (0.04)
Dose 3 ^c , mean (SE)	--	--	16.7 (0.04)	16.7 (0.04)
Demographic				
Female Sex, %	48.8	48.6	48.6	49.7
African Race, %	97.2	96.8	72.3	72.4
Asian Race, %	--	--	27.6	27.6
Growth Status at Enrollment				
Stunted, %	22.6	21.7 ^d	10.1	10.4 ^e
Underweight, %	3.9	4.4	11.5	11.2
Wasting, %	3.7 ^d	4.3 ^d	23.1 ^d	20.9 ^{de}
Exclusively Breastfed				
At Dose 1, %	--	--	80.0	81.6
At Dose 2, %	--	--	75.2 ^d	74.7
At Dose 3, %	--	--	69.9	70.1 ^d
1 Infection(s)				
At Dose 1, %	0.5	0.9	5.5	5.6
At Dose 2, %	0.3	0.4	0.5	0.6
At Dose 3, %	--	--	1.1	1.3
1 Antibiotic Used ^f				
At Dose 1, %	12.9	14.1	2.2	2.8
At Dose 2, %	15.5	14.6	5.6	5.2
At Dose 3, %	--	--	6.2	6.1
Routine Vaccines				
1 BCG Vaccine				
At Dose 1, %	--	--	21.9	22.0
1 DTP-HB/HIB Vaccine ^g				
At Dose 1, %	99.3	99.3	45.7	47.0
At Dose 2, %	99.1	99.5	43.6	43.8

Infant Characteristics	Trial 1 (N = 3114)		Trial 2 (N = 7341)	
	RV1 (N = 1560)	Placebo (N = 1554)	RV5 (N = 3666)	Placebo (N = 3675)
At Dose 3, %	--	--	42.0	42.2
1 Oral Polio Vaccine				
At Dose 1, %	99.3	99.3	54.1	55.6
At Dose 2, %	99.2	99.5	51.1	51.0
At Dose 3, %	--	--	48.2	48.1

BCG, Bacillus Calmette–Guérin; DTP-HB/HIB, diphtheria-tetanus-pertussis-Hepatitis B and -Haemophilus influenza B; DTaP, diphtheria and tetanus toxoids and acellular pertussis

^aThere were 25 and 21 in the placebo and RV1 arms, respectively, missing dose one.

^bThere were 63 and 45 in the placebo and RV1 arms, respectively, missing dose two. There were 17 and 10 in the placebo and RV5 arms, respectively, missing dose two.

^cThere were 57 and 47 in the placebo and RV5 arms, respectively, missing dose three.

^dMissing 1 – 15 observations, excluding those missing doses of vaccine or placebo.

^eExcluding Bangladesh

^fExcluding topical antibiotics

^gOr DTaP & HB, which were the standard vaccines given in Asian countries