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Evaluation of non-specific effects of human rotavirus vaccination in medical risk infants

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ABSTRACT

Background: The WHO recommends research into non-specific effects of vaccination. For rotavirus vaccines, these have not yet been well established. We studied non-specific effects up to 18 months of age using data from a quasi-experimental before-after study comparing cohorts of rotavirus vaccinated and unvaccinated infants with medical risk conditions.

Methods: Infants were enrolled at six weeks of age before and after a stepped-wedge implementation of a hospital-based risk-group rotavirus vaccination program. Other infant vaccinations were administered according to the Dutch National Immunization Program and similar in both cohorts. Non-specific effect outcomes were prospectively collected using monthly questionnaires and included acute hospitalization (excluding for acute gastroenteritis), monthly incidence of acute respiratory illness and eczema. We used time-to-event analysis and negative binomial regression to assess the effect of at least one dose of rotavirus vaccination for each of these outcomes.

Findings

The analysis included 496 rotavirus unvaccinated and 719 vaccinated medical risk infants. In total, 1067 (88%) were premature, 373 (31%) small for gestational age and 201 (17%) had a congenital pathology. The adjusted hazard ratio for first acute hospitalization was 0·91 (95 %CI 0·76;1·16) for rotavirus vaccinated versus unvaccinated infants. Adjusted incidence rate ratio for acute respiratory illness was 1·05 (95 %CI 0·96;1·15) and for eczema 0·89 (95 %CI 0·69;1·15).

Conclusion: The results suggest no, or minimal non-specific effects from rotavirus vaccination on acute hospitalization, acute respiratory illness or eczema in medical risk infants.

Trial registration: as NTR5361 in the Dutch trial registry, www.trialregister.nl.

Abbreviations: HRV, human rotavirus vaccination; NIP, national immunization program; ARI, acute respiratory illness; BCG, bacillus Calmette Guerin.

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1. Introduction

Beneficial non-specific effects have been attributed to (live attenuated) vaccines in both adults and children.[1] Non-specific effects are defined as effects of vaccines on morbidity and mortality that are not explained by the prevention of the target diseases, [2] or as resistance towards unrelated pathogens in vaccine recipients.[3] For instance, the introduction of the first human vaccine, Vaccinia, led to reductions in measles and scarlet fever besides the effect on smallpox.[3] The proposed mechanism for non-specific effects on non-target infections and hospitalizations is trained immunity.[1] Trained immunity is induced by epigenetic reprogramming that results in enhanced innate immune responses to reinfection and to non-target pathogens.[4] Immunological studies in infants and adults established that vaccines can induce activation of innate immune cells against other than target diseases.[5–8] The duration of these beneficial non-specific effects is thought to last between three months and one year or until the next (non-live) vaccine is given.[1,9]

Epidemiologic studies suggest that bacillus Calmette-Guérin (BCG) vaccination can offer protection against respiratory infections and (recurrent) bladder cancer.[10] A recent randomized controlled trial showed that neonatal BCG vaccination prevented non-tuberculosis infectious diseases in the first six weeks of life. [11] For the Measles Mumps Rubella (MMR) vaccine, a reduction in childhood mortality was observed, mediated by prevention of respiratory infections.[12] For Oral Polio vaccine (OPV) research indicated lower hospitalization rates and protection against otitis media.[1] However, conflicting reports on reduction in all-cause case fatality after OPV have been published.[13–15]

Another observation from epidemiological studies is a non-specific effect on atopy. There are two reports of reduced atopy after BCG vaccination,[16,17] however a randomized controlled trial found no effect but a potential trend towards less (severe) eczema.[18] The mechanism resulting in atopy prevention is less well studied, the Th1 stimulating property of live-vaccines may prevent allergic sensitization and reduce atopy (which is Th2 mediated).[17,19]

Yet, two systematic reviews on routine childhood vaccinations and non-specific effects concluded that low quality studies and heterogeneity of the available evidence should raise caution in interpretation.[20,21] The World Health Organization recommends more research towards non-specific effects of vaccines. [22,23]

Rotavirus vaccines are live-attenuated and orally administered, like OPV. It is hypothesized that rotavirus vaccination can therefore induce beneficial non-specific effects through similar mechanisms, but this has been little studied thus far. One study reported a decrease of 31% in non-rotavirus hospitalization rates in the 60 days following rotavirus vaccination, but misclassification of rotavirus infections could not be completely ruled out.[24] To our knowledge, no literature is available on non-specific effects of rotavirus vaccination on respiratory infections or atopy.

We explored potential non-specific effects in a prospective cohort of medical risk infants in the Netherlands who did or did not receive rotavirus vaccination in a quasi-experimental setting.

2. Methods

For a detailed description of the study we refer to the study protocol. In short, thirteen Dutch hospitals with a neonatal intensive or high care ward participated in the project that combined the implementation of rotavirus vaccination program for medical risk infants with a prospective before-after cohort study. During both periods with and without rotavirus vaccination, infants were recruited at six weeks of age if they had at least one medical risk condition; 1) prematurity (gestational age < 36 weeks), 2) low birthweight (<2500 g) and/or 3) congenital pathology (list in suplementary material), and received prolonged pediatric care between six and 14 weeks of postnatal age at the participating site. Hospitals entered the study in a stepped-wedge approach and each site implemented rotavirus vaccination in routine care for medical risk infants between months 12–18 of the project. The schedule for other routine childhood immunizations according to the National Immunization Program (NIP) was left unchanged (Fig. 1). Post-implementation, the human rotavirus vaccine (HRV, Rotarix, GSK, Belgium) was used at all sites. HRV was given in a two-dose schedule, with first dose preferably between six and nine weeks of age and the second dose with a minimum interval of four weeks and no later than 24 weeks of age. The first dose was administered by a physician in participating hospitals, the second dose was given by parents at home after detailed instruction.

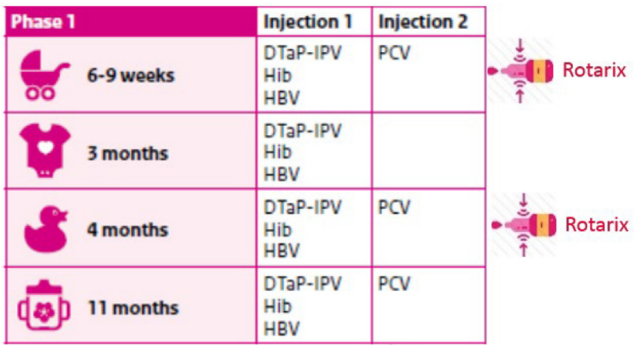

Fig. 1. Vaccination schedule RIVM@2021. Legend: = human rotavirus vaccination. Abbreviations: DTaP-IPV = Diphteria Tetanus accelular Pertussis-Inactivated Polio vaccine, Hib = Haemophilus influenza type B, HBV = Hepatitis B vaccine, and PCV = Pneumococcal conjugate vaccine.

2.1. Data collection

Data collection for cohort participants included monthly parental questionnaires until 18 months of age. Parents answered yes/no questions concerning acute respiratory illness (ARI) symptoms, presence of eczema symptoms and hospital admission in the past month. ARI symptoms included fever with or without nasal congestion/runny nose, cough and earache. In addition the number of days with ARI symptoms in each month was recorded. For hospitalization, the reason and number of hospital days was also collected.

2.2. Non-specific effect outcomes

We defined non-specific effect outcomes of rotavirus vaccination as the relative change in incidence of parent-reported acute hospitalization, ARI or eczema between rotavirus vaccinated and unvaccinated infants.

Acute hospitalization was defined as any hospital admission following initial post-natal discharge, excluding hospital admissions for acute gastroenteritis. Hospital admissions for scheduled medical or surgical interventions were also excluded.

2.3. Analyses

Descriptive statistics were used to compare patient characteristics and outcomes between vaccinated and unvaccinated infants.

We calculated the cumulative incidence and incidence rate for each non-specific effect outcome by group status.

Next, we used Cox regression to model the effect of at least one dose of rotavirus vaccination on time to first hospital admission, with age (between two and 18 months) as time axis. Infants were censored when lost to follow up, dropped-out, deceased or hospitalized, whichever came first. The proportional hazard assumption was visualized graphically and tested with Schoenfelds residuals. The final model with covariates was derived using log-likelihood ratio test, hazard ratios (HR) and their 95% confidence intervals (CI) were provided. Infants were categorized as rotavirus vaccinated from 28 days post-dose one onwards.

For ARI, we used a negative binomial model with offset for persontime of observation to compare the number of months with ARI between rotavirus vaccinated and unvaccinated groups. Incidence rate ratios (IRR) were obtained with their 95% CIs. We used Akaike Information Criterion to select the final model. The analysis was repeated for the outcome months with eczema.

As secondary outcomes, we calculated the cumulative acute hospitalization days and days with ARI symptoms for rotavirus vaccinated and unvaccinated infants and used a Poisson model. Because of overdispersion for hospitalization days we used a negative binomial model.

In sensitivity analyses, we separately analysed effects up to six months of age, covering the initial three months post-rotavirus vaccination, and up to eleven months of age, covering the period up to the booster vaccinations of the NIP. We also analysed hospitalizations for infectious diseases separately, using Cox modelling as described above. For all models, we considered the following covariates: sex, type of medical risk condition, gestational age, breastfeeding, daycare attendance, type of hospital care (i.e. academic versus general), family educational level, parental origin, NIP vaccination status, presence of siblings in the household and season with high rates of respiratory infections (from October until April).

All analyses were performed on according to protocol cohorts, where all medical risk infants whose parents indicated willingness to vaccinate their child against rotavirus in the 'before' cohort (i.e. pre-implementation) are compared to infants that received at least one HRV dose in the 'after' cohort (i.e. post-implementation of HRV). This information was based on a parental questionnaire at start of the study.[25]

We performed complete case analyses, missing information is documented. As statistical software SPSS version 25 and RStudio version 5.0 were used, with packages *MASS, fmsb, survival and survminer*.

3. Results

A total of 1482 high-risk infants with one or more medical risk conditions were enrolled in the study. The population for analysis included 719 rotavirus vaccinated infants and 496 infants whose parents indicated willing to vaccinate against rotavirus, but who were enrolled before rotavirus vaccination was implemented. In total, 1067 infants (87·8%) were born premature, 373 infants (30·7%) were small for gestational age and 201 infants (16·5%) had at least one congenital condition. Baseline characteristics were comparable between the rotavirus vaccinated and unvaccinated group (table 1), with the exception of follow up, which was complete for 450 vaccinated infants (62·6%) and 380 (76·6%) of unvaccinated infants. The proportion of observation months during seasons with or without high rates of respiratory infections was not significantly different between the groups, ARIs occurring in-season was also not different.

Non-specific effect outcomes by vaccination status are described in Table 2. Twenty-one hospitalizations for acute gastroenteritis were excluded from the acute hospitalizations (eleven among unvaccinated and ten among vaccinated infants, respectively). Thirty-nine percent of infants were hospitalized at least once or reported one month with eczema. Reasons for hospitalization are provided in Table S1 in supplementary material. For 82% of infants at least one ARI episode was reported. Incidence of any of the non-specific effect outcomes or the mean cumulative number of days hospitalized or with ARI symptoms show no significant differences (Table 2).

Based on the adjusted Cox model at least one dose of HRV was not significantly protective against first acute hospitalization (HR

Table 1
Baseline table with characteristics of vaccinated and unvaccinated infants.

	According to protocol groups protocol (ATP)		
	Pre-implementation: willing to vaccinate N = 496	Post-implementation: vaccinated N = 719	p-value
Male sex	286 (57·7%)	381 (53·0%)	0·11
Multiple births	115 (23·8%)	193 (26·8%)	0·15
Premature birth (gestation < 37 weeks)	432 (87·8%)	635 (88·3%)	0·79
Small for gestational age£	135 (27·2%)	238 (33·1%)	0·03
Presence of severe congenital disorder*	95 (19·2%)	106 (14·7%)	0·04
Vaccinated according to NIP	484 (97·6%)	652 (90·7%)	<0·001
Daycare attendance	289 (58·6%)	398 (59·6%)	0·74
Breastfed	312 (63·3%)	403 (60·2%)	0·29
Socioeconomic status^			0·36
Higher	352 (71·5%)	492 (74·1%)	
Moderate	119 (24·2%)	153 (23·0%)	
Lower	21 (4·3%)	19 (2·9%)	
Ethnicity#			0·75
European parent(s)	417 (84·8%)	557 (83·4%)	
Non-European parent(s)	28 (5·7%)	38 (5·7%)	
Mixed	47 (9·6%)	73 (10·9%)	
Mean number of months with completed follow-up (SD)	16·4 (5·5)	14·9 (6·5)	<0·001
Number with complete follow-up until 18 months of age	380 (76·6%)	450 (62·6%)	<0·001
Mean proportion of months during ARI season (SD)$	0·50 (0·18)	0·50 (0·18)	0·75

Percentages are computed excluding subjects with missing data on the variable. Statistical significance (p-value < 0·05) is highlighted in bold. *For a list of qualifying congenital disorders, see appendix Fig. 1 of (van Dongen et al., 2020). £ Based on the 10th percentile of Dutch perinatal growth curves ^Based on highest parental education level, i.e. high (higher vocational level or university degree), moderate (other educational levels) or low (primary school or no education). #Based on parental origin by World population by country. (*World Population by Country 2020*, n.d.) $ARI season in the Netherlands is defined from October until April.(Reukers et al., 2019) Abbreviations: GA = gestational age, SD = standard deviation, IQR = interquartile range, yrs = years, NIP = National Immunization Program and, N = number in group.

Table 2
Non-specific effect outcomes for vaccinated and unvaccinated infants.

	Pre-implementation: willing to vaccinate N = 496	Post-implementation: vaccinated N = 719
At least one acute hospitalization	212 (43·0%)	262 (39·2%)
Cumulative number of hospitalizations	367	433
Incidence of hospitalization* (95 % CI)	0·54 (0·48;0·59)	0·48 (0·43;0·52)
Mean cumulative number of hospitalization days	8.3	8.2
At least one ARI	411 (83·4%)	584 (87·3%)
Cumulative number of months with ARI	1888	2660
Incidence of months with ARI* (95 %CI)	2·77 (2·64;2·89)	2·92 (2·81;3·04)
Mean cumulative number of days with ARI	23·2	24·5
At least one month with eczema	204 (41·4%)	270 (40·4%)
Cumulative number of reported eczema	995	1215
Incidence of months with eczema* (95 %CI)	1·46 (1·37;1·55)	1·33 (1·26;1·41)

Percentages are derived excluding cases with missing information (n = 53). *Incidence per person year of observation. Abbreviations: N = number in group, CI = confidence interval and, ARI = acute respiratory illness.

for time to first acute hospitalization 0·91; 95 %CI 0·71;1·18, Table 3). The final model included gestational age, presence of a congenital disorder, type of hospital care and seasonality as covariates.

In addition, no effect was observed on the occurrence of ARI (adjusted incidence rate ratio (IRR) 1·05; 95 %CI 0·96;1·15), or eczema (adjusted IRR: 0·89; 95 %CI 0·69;1·15, Table 3). Comparing the secondary outcomes of cumulative or hospitalization days showed no statistically significant difference. The cumulative number of days with ARI was increased for vaccinated infants (Table 3).

In sensitivity analyses we observed that the non-specific effects estimates were closer to one and non-significant when restricting to the six or eleven months of age for all non-specific effect outcomes, Table 3. Restricting to acute hospitalizations for infectious diseases resulted in an adjusted HR of 0·96 (95 %CI 0·58;1·62).

4. Discussion

This quasi-experimental prospective study assessed the potential non-specific effects of rotavirus vaccination among 1215 infants with medical risk conditions. Our results suggest that HRV does not offer significant protection against non-target diseases leading to acute hospitalization, or result in reduced incidence of ARI or atopy up to 18 months of age.

Non-specific effects of (live attenuated) vaccines are increasingly being studied. Currently, 22 trials are being performed or completed on the potential protective effect of BCG vaccination against COVID-19 disease.[22] For rotavirus vaccines however, there has been very little research into non-specific effects despite their availability and widespread use for more than a decade. While some promising results of beneficial non-specific effects, in particular for BCG vaccine are available,[1,4,9,10] other studies report contradictory and less convincing results.[13–15,26,27] We were unable to detect any beneficial, nor any non-beneficial effects of rotavirus vaccination on the studied outcomes. The results of our study add to the growing body of evidence hinting towards absence of non-specific effects, at least for rotavirus vaccines.

Opposite to our result, a reduction in acute hospitalization due to non-target diseases after rotavirus vaccination was reported in a previous study from the United States.[24] As a secondary analysis, this study compared hospitalization rates in the 60-day post-vaccination window for rotavirus vaccinated and unvaccinated infants, excluding hospitalizations coded as rotavirus gastroenteritis. A reduction of 31% (95 %CI 27–35%) was reported. However, coding for rotavirus hospitalizations is known to be incomplete.[28] By including hospitalizations for gastroenteritis without the rotavirus specific code, misclassification may have occurred and could explain the observation mediated by direct, rather than indirect vaccine effects.

Most likely, a reduction in acute hospitalizations resulting from non-specific effects would be mediated by lowering infection incidence. This is however not supported by our findings that show a similar rate of ARI in vaccinated and unvaccinated infants. Furthermore, when restricting the analysis to hospitalizations for infectious diseases only, an even smaller effect was estimated. Alternatively, one could argue that non-specific effects following rotavirus vaccination reduce severity of ARI, rather than ARI incidence, thereby reducing the risk of hospitalization. However, no reduction was observed in days with ARI symptoms, which could be considered a proxy for severity. Another hypothesis mentions

Table 3
Effect estimates of non-specific effects.

	Univariate	95% CI	Multivariate estimate	Lower 95% CI	Upper 95% CI
HR for acute hospitalization*	0·85	0·66-1·09	0·91	0·71	1·18
IRR for ARI#	1·07	0·99-1·17	1·05	0·96	1·15
IRR for eczema#	0·94	0·74-1·18	0·89	0·69	1·15
Secondary					
RR Cumulative number of days hospitalized	0·91	0·66-1·25	0·83	0·59	1·16
RR Cumulative number of days with ARI	1·06	1·04-1·09	1·06	1·03	1·09
Sensitivity					
HR for infectious diseases hospitalization*	0·93	0·56-1·60	0·96	0·58	1·62
HR for acute hospitalization until 6 months*	1·08	0·76-1·53	1·20	0·85	1·71
HR for acute hospitalization until 11 months*	0·87	0·65-1·16	0·95	0·71	1·27
IRR for ARI until 6 months#	1·05	0·89-1·24	0·94	0·78	1·14
IRR for ARI until 11 months#	1·10	0·99-1·22	1·03	0·92	1·15
IRR for eczema until 6 months#	0·99	0·71-1·41	0·91	0·62	1·33
IRR for eczema until 11 months#	0·96	0·73-1·24	0·89	0·66	1·19

* Adjusted for: gestational age, presence of congenital disorder, type of hospital care and seasonality. # Adjusted for: gestational age, daycare attendance, parental educational level, presence of sibling in the household, vaccinated according to NIP program and seasonality. Abbreviations: HR = hazard ratio IRR = incidence rate ratio RR = relative risk and, CI = confidence interval.

that inactivated childhood vaccinations abrogate the beneficial non-specific effects of earlier administered live vaccines. To investigate this, we conducted a sensitivity analysis in which we restricted the analysis to the time-period up to the next routine childhood vaccination at 11 months, but the effect estimate was unchanged. Jointly, our results do not support the existence of non-specific effects for rotavirus vaccines lowering overall hospitalization rates, infection incidence or duration.

For eczema, we did not find a significant effect of rotavirus vaccination either. Evidence on preventing atopy by vaccination so far, is based on one underpowered randomized clinical trial and several heterogeneous low-quality observational studies.[29] Unfortunately, our study did not collect information on risk factors for atopy and was underpowered to draw any firm conclusion on the effect of rotavirus vaccination on eczema incidence. Our point estimate of 0.89 suggests some benefit may exist, but this warrants confirmation in other controlled studies.

In this study the majority of infants was born prematurely. Non-specific effects have previously been investigated and detected among low birthweight infants in multiple randomized trials. [6,30,31] Premature infants have a higher risk of respiratory morbidity and hospitalization.[32] Therefore, it is likely that any non-specific effect on ARI from rotavirus vaccination would be detectable in this specific study population. On the other hand, we also observed very low and non-significant direct vaccine effectiveness in our study population.[25] Possibly, the immaturity of the infants (gut) immune system results in lower vaccine responses that affect both direct and non-specific effects in this population. Additional research into non-specific effects of rotavirus vaccination in a healthy term infant population, who are known to mount adequate vaccine responses to rotavirus vaccination could provide some further insights on this.

A few limitations of this study need to be addressed. First, we used complete cases only, missing information is documented and comparing participants with complete versus missing data did not reveal significant differences (data not shown). Loss to follow up occurred in approximately 30% of participants,[33] however mean follow-up was 14.5 months. By using time-to-event analysis and including an offset for observation time this was taken into account in our analyses and we therefore expect this has minimal effect on our results.

We were unable to assess severity of ARI, a discrimination in episodes by severity might further explain why the difference in acute hospitalizations was not reflected in occurrence of ARI. Instead, we assessed number of days with ARI complaints and observed no relevant difference between vaccinated and unvaccinated infants. Duration of disease could function as a proxy for severity. In addition, the quasi-experimental design (as opposed to randomization) can lead to residual confounding by inequality between groups. However, we corrected for most important and known confounders and overall differences between the groups were small. We therefore expect limited effect on the results of our study.

In conclusion, rotavirus vaccination in medical risk infants was not associated with beneficial non-specific effects on (acute hospitalization due to) non-target diseases. The study results suggest that beneficial non-specific effects, as observed for some other live attenuated vaccines, may not apply to oral rotavirus vaccines, but studies in healthy term infants are needed to further establish this.

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Trademark: Rotarix is a trademark owned by or licensed to the GSK group of companies.

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GlaxoSmithKline Biologicals SA was provided the opportunity to review a preliminary version of this manuscript for factual accuracy, but the authors are solely responsible for final content and interpretation. Sponsors had no role in study design, data collection, analysis, writing or submitting of the study.

Authors' Contribution Statement

Josephine van Dongen designed the study, collected data, carried out the initial analysis and drafted the initial manuscript. Elsbeth Rouers assisted in data collection and study set-up and reviewed the manuscript. Prof Bonten assisted in conceptualizing the study, reviewed and revised the manuscript. Dr. Patricia. Bruijning-Verhagen conceptualized and designed the study, coordinated and supervised data collection, assisted and supervised

data analyses and, critically reviewed and revised the manuscript. All other authors assisted in data collection, study set-up and reviewed the manuscript. All approved the final manuscript as submitted and agree to be accountable for all aspects of the work.

Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.vaccine.2021.08.067.

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