ORIGINAL ARTICLE

RSV Prefusion F Protein–Based Maternal Vaccine — Preterm Birth and Other Outcomes

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ABSTRACT

BACKGROUND

Vaccination against respiratory syncytial virus (RSV) during pregnancy may protect infants from RSV disease. Efficacy and safety data on a candidate RSV prefusion F protein–based maternal vaccine (RSVPreF3-Mat) are needed.

METHODS

We conducted a phase 3 trial involving pregnant women 18 to 49 years of age to assess the efficacy and safety of RSVPreF3-Mat. The women were randomly assigned in a 2:1 ratio to receive RSVPreF3-Mat or placebo between 24 weeks 0 days and 34 weeks 0 days of gestation. The primary outcomes were any or severe medically assessed RSV-associated lower respiratory tract disease in infants from birth to 6 months of age and safety in infants from birth to 12 months of age. After the observation of a higher risk of preterm birth in the vaccine group than in the placebo group, enrollment and vaccination were stopped early, and exploratory analyses of the safety signal of preterm birth were performed.

RESULTS

The analyses included 5328 pregnant women and 5233 infants; the target enrollment of approximately 10,000 pregnant women and their infants was not reached because enrollment was stopped early. A total of 3426 infants in the vaccine group and 1711 infants in the placebo group were followed from birth to 6 months of age; 16 and 24 infants, respectively, had any medically assessed RSV-associated lower respiratory tract disease (vaccine efficacy, 65.5%; 95% credible interval, 37.5 to 82.0), and 8 and 14, respectively, had severe medically assessed RSV-associated lower respiratory tract disease (vaccine efficacy, 69.0%; 95% credible interval, 33.0 to 87.6). Preterm birth occurred in 6.8% of the infants (237 of 3494) in the vaccine group and in 4.9% of those (86 of 1739) in the placebo group (relative risk, 1.37; 95% confidence interval [CI], 1.08 to 1.74; P=0.01); neonatal death occurred in 0.4% (13 of 3494) and 0.2% (3 of 1739), respectively (relative risk, 2.16; 95% CI, 0.62 to 7.56; P=0.23), an imbalance probably attributable to the greater percentage of preterm births in the vaccine group. No other safety signal was observed.

CONCLUSIONS

The results of this trial, in which enrollment was stopped early because of safety concerns, suggest that the risks of any and severe medically assessed RSV-associated lower respiratory tract disease among infants were lower with the candidate maternal RSV vaccine than with placebo but that the risk of preterm birth was higher with the candidate vaccine. (Funded by GlaxoSmithKline Biologicals; ClinicalTrials.gov number, NCT04605159.)

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Respiratory syncytial virus (RSV) is a major cause of lower respiratory tract disease in young children,[1,2] with a disproportionate effect on infants younger than 6 month of age, especially in low- and middle-income countries.[3] Effective interventions to prevent RSV-associated lower respiratory tract disease in infants are needed.

Maternal immunization has safely prevented millions of cases of tetanus, influenza, and pertussis among infants worldwide[4-7] and could protect young infants against RSV disease.[8,9] GSK has been developing an RSV subunit vaccine for maternal use (RSVPreF3-Mat) that is based on the RSV fusion (F) protein stabilized in its prefusion conformation.[10,11] In 2020, GSK initiated a phase 3 trial (RSV MAT-009) to assess the safety and efficacy of RSVPreF3-Mat against RSV-associated lower respiratory tract disease in infants born to women who had received the vaccine during pregnancy. In mid-February 2022, the independent data monitoring committee overseeing the trial notified GSK of an imbalance in preterm births in the vaccine group as compared with the placebo group. After further investigation, GSK stopped enrollment and vaccination on February 25, 2022, in this trial and all other ongoing RSVPreF3-Mat trials involving pregnant women. GSK promptly informed the relevant regulatory authorities, the ethics committees at the trial sites, and the investigators; unblinded the trial-group assignments to the investigators; and stopped enrollment and vaccination in all RSVPreF3-Mat trials in nonpregnant women (see the Supplementary Appendix, available with the full text of this article at NEJM.org).

In the current article, we report the results of the primary efficacy analyses and safety assessments in the RSV MAT-009 trial. We also report findings of exploratory analyses in which we further assessed the safety signal of preterm birth.

Methods

Trial Design

RSV MAT-009 was a phase 3, double-blind, randomized, placebo-controlled trial conducted in 24 countries across six continents. The target enrollment was approximately 10,000 pregnant women and their infants, which was not reached because enrollment was stopped early. Eligible women were 18 to 49 years of age and in good general health and had a singleton fetus with a gestational age of 24 weeks 0 days to 34 weeks 0 days (as confirmed by the date of the last menstrual period and by ultrasonography; see the Supplementary Appendix) without known fetal genetic abnormalities or major congenital malformations (as defined in the protocol, available with the statistical analysis plan at NEJM.org). Women with clinically important complications (as judged by the investigator) during the current pregnancy or two or more previous stillbirths, neonatal deaths, or preterm births were excluded (see the Supplementary Appendix). Women were randomly assigned in a 2:1 ratio to receive one intramuscular injection of 120 μg of RSVPreF3-Mat or placebo.

Trial Oversight

The protocol was approved by the ethics committees at the trial sites. Written informed consent was obtained from all the maternal participants and from all the guardians of the infant participants. Safety was monitored by an independent data monitoring committee and by the safety review team at GSK. On identification of the preterm birth signal, GSK instituted urgent safety measures (see the Supplementary Appendix) and shared monthly updated analyses with the independent data monitoring committee, trial investigators, and relevant regulatory authorities. Authors employed by GSK designed the trial and participated in the collection, analysis, and interpretation of the data; the writing and critical revision of the manuscript; and the decision to submit the manuscript for publication. All the authors vouch for the accuracy and completeness of the data and for the fidelity of the trial to the protocol. Assistance with the writing and preparation of an earlier version of the manuscript was supported through GSK by Akkodis Belgium.

Efficacy and Safety Assessments

The primary outcomes were any medically assessed RSV-associated lower respiratory tract disease and severe medically assessed RSV-associated lower respiratory tract disease in infants from birth to 6 months of age and safety

in infants from birth to 12 months of age (Table S1). The interim analysis of the data at day 43 after birth (the database was locked on October 4, 2022) confirmed the safety signal of preterm birth, and these data were the primary source for subsequent investigations. Other reported outcomes are based on the analysis of data collected 6 months after delivery or birth (the database was locked on October 5, 2023) and include the primary efficacy outcomes, adverse events of special interest through week 6 after delivery or birth, and serious adverse events through month 6 after delivery or birth. Gestational age and adverse events of special interest (including preterm birth) were defined according to the guidelines of the Global Alignment of Immunization Safety Assessment in Pregnancy network (see the Supplementary Appendix).[12-14] After the safety signal was identified, an adjudication committee composed of three independent external experts reviewed all preterm births. Details about the surveillance methods for respiratory tract illness, case definitions for any and severe medically assessed RSV-associated lower respiratory tract disease, adverse event reporting, laboratory assessments, and other assessments are provided in the Supplementary Appendix.

STATISTICAL ANALYSIS

Vaccine efficacy (with a 95% credible interval) was calculated as 1 minus the relative risk with the use of a Bayesian model. The efficacy analyses included all the infants who were born at least 4 weeks after their mothers received RSVPreF3-Mat or placebo. After an interim safety assessment showed an imbalance in the incidence of preterm birth between the vaccine and placebo groups, GSK stopped enrollment and vaccination in the trial. The protocol was then amended to replace hypothesis testing with descriptive analyses in the assessment of efficacy.

The safety analyses included all the mothers who received RSVPreF3-Mat or placebo and their live-born infants. The percentages of participants with an adverse event of special interest or a serious adverse event were calculated with exact 95% confidence intervals. The investigation of the safety signal included post hoc analyses that were not controlled for multiplicity. The relative risks of preterm birth in the vaccine group as compared with the placebo group, along with Wald 95% confidence intervals, were calculated with stratification according to risk factors; multivariable analyses were performed. Laboratory analyses to investigate possible mechanisms to explain the preterm birth signal were performed in an investigation cohort that was created according to a matched case–cohort design (see the Supplementary Appendix).

RESULTS

PARTICIPANTS

Trial enrollment began on November 20, 2020. By February 25, 2022, when enrollment and vaccination were stopped because of the safety signal of preterm birth, 5328 pregnant women had been vaccinated: 3557 had received RSVPreF3-Mat, and 1771 had received placebo. The analysis of data available at day 43 after birth included data from 5233 infants: 3494 in the vaccine group and 1739 in the placebo group (Fig. S1 in the Supplementary Appendix).

Approximately 50% of the participants in each trial group were enrolled in low- or middle-income countries (Table 1). Characteristics of the mothers at baseline and those of the infants at birth were balanced between the two trial groups overall and according to country income level (low or middle income and high income) (Table 1 and Table S2). The representativeness of the trial population is shown in Table S3.

EFFICACY

A total of 3426 infants in the vaccine group and 1711 in the placebo group were followed from birth to 6 months of age and were included in the efficacy analysis. Between birth and 6 months of age, medically assessed RSV-associated lower respiratory tract disease occurred in 16 infants in the vaccine group and in 24 infants in the placebo group (vaccine efficacy, 65.5%; 95% credible interval, 37.5 to 82.0), and severe medically assessed RSV-associated lower respiratory tract disease occurred in 8 and 14 infants, respectively (vaccine efficacy, 69.0%; 95% credible interval, 33.0 to 87.6) (Table 2).

SAFETY

Adverse events of special interest occurred in a similar percentage of participants in the two trial

Table 1. Characteristics of the Pregnant Women at Baseline and the Infants at Birth.*

Characteristic	RSVPreF3-Mat Group	Placebo Group
Pregnant women		
No. of participants	3557	1771
Age at vaccination		
Overall — yr	29.0±6.0	29.0±6.0
Distribution — no. (%)		
18–34 yr	2866 (80.6)	1408 (79.5)
35–39 yr	581 (16.3)	300 (16.9)
≥40 yr	110 (3.1)	63 (3.6)
Gestation at vaccination — no. (%)		
<24 wk 0 days	1 (<0.1)	2 (0.1)
24 wk 0 days–28 wk 6 days	1469 (41.3)	727 (41.1)
29 wk 0 days–34 wk 0 days	2079 (58.4)	1036 (58.5)
>34 wk 0 days	8 (0.2)	6 (0.3)
Race or ethnic group — no. (%)†		
Asian	663 (18.6)	326 (18.4)
Black	517 (14.5)	251 (14.2)
White	1669 (46.9)	838 (47.3)
Mixed race or other ethnic group	707 (19.9)	356 (20.1)
Hispanic or Latino	1196 (33.6)	582 (32.9)
Not Hispanic or Latino	2360 (66.3)	1189 (67.1)
Country income level‡		
Low or middle income	1808 (50.8)	898 (50.7)
High income	1749 (49.2)	873 (49.3)
Infants		
No. of participants	3494	1739§
Gestational age		
Overall — wk	39.1±1.6	39.2±1.4
Distribution — no. (%)		
<37 wk	237 (6.8)	86 (4.9)
≥37 wk	3257 (93.2)	1653 (95.1)
Female sex — no. (%)	1741 (49.8)	866 (49.8)
Length — cm	49.8±2.6	49.8±2.5
Weight — g	3223.2±491.5	3222.4±494.7
Head circumference — cm	34.3±1.6	34.3±1.6
Apgar score of 7–10 at 5 min — no. (%)	3381 (96.8)	1685 (96.9)

* Plus–minus values are mean ±SD. This analysis was based on data available at day 43 after delivery or birth from pregnant women who received a single dose of the investigational respiratory syncytial virus prefusion F protein–based maternal vaccine (RSVPreF3-Mat) or placebo and their live-born infants (the database was locked on October 4, 2022). Data on race and ethnic group were missing for 1 pregnant woman in the vaccine group. Data on sex were missing for 2 infants in the vaccine group and 1 infant in the placebo group; data on length were missing for 52 and 15, respectively; data on weight were missing for 16 and 8, respectively; data on head circumference were missing for 107 and 49, respectively; and data on Apgar score were missing for 77 and 40, respectively.

† Race and ethnic group were reported by the investigator on the basis of information shared by the maternal participant. A total of 14 women in the vaccine group and 7 women in the placebo group were American Indian or Alaska Native; 8 and 4, respectively, were Native Hawaiian or other Pacific Islander; and 685 and 345 were of mixed race (in most women) or belonged to another ethnic group.

‡ Low- and middle-income countries included Argentina, Bangladesh, Brazil, Colombia, the Dominican Republic, Honduras, India, Mexico, Panama, the Philippines, South Africa, and Thailand. High-income countries included Australia, Belgium, Canada, Finland, France, Italy, New Zealand, South Korea, Spain, Taiwan, the United Kingdom, and the United States.

§ As of the database-lock date for the analysis at day 43, of the 1741 infants in the placebo group, 2 had missing information and were therefore not included in the analysis.

Table 2. Vaccine Efficacy against Medically Assessed RSV-Associated Lower Respiratory Tract Disease in Infants up to 6 Months of Age.*

Outcome	RSVPreF3-Mat Group (N=3426)		Placebo Group (N=1711)		Vaccine Efficacy (95% Credible Interval)
	Events	Incidence	Events	Incidence	
	no.	*no. of events/ 1000 person-yr*	*no.*	*no. of events/ 1000 person-yr*	%
Any medically assessed RSV-associated lower respiratory tract disease	16	9.7	24	29.2	65.5 (37.5–82.0)
Severe medically assessed RSV-associated lower respiratory tract disease	8	4.8	14	17.0	69.0 (33.0–87.6)

* This analysis was based on data available at month 6 after birth from all infant participants born at least 4 weeks after their mothers received a single dose of RSVPreF3-Mat or placebo (the database was locked on October 5, 2023). The difference in the incidence of any medically assessed RSV-associated lower respiratory tract disease and of severe medically assessed RSV-associated lower respiratory tract disease between the two trial groups was evaluated with the use of a Bayesian model that was conditional on the total number of cases. The analyses were not adjusted for multiplicity.

groups, except for pathways to preterm birth in mothers (premature preterm rupture of membranes, preterm labor, and provider-initiated preterm birth) and preterm birth and neonatal death in infants, which occurred in a higher percentage of participants in the vaccine group than in the placebo group (Table 3 and Table S4). Serious adverse events other than preterm birth or neonatal death occurred in similar percentages of mothers and infants in the RSVPreF3-Mat and placebo groups (Tables S5 and S6).

SAFETY SIGNAL OF PRETERM BIRTH

In February 2022, the independent data monitoring committee reported a higher incidence of preterm birth in the vaccine group (7.6%; 183 of 2419 infants) than in the placebo group (5.0%; 60 of 1199 infants), with a relative risk of 1.51 (95% confidence interval [CI], 1.14 to 2.01; P=0.004). The imbalance in the risk of preterm birth remained after all the mothers had delivered (last delivery, June 2022): analysis of the data set at day 43 after birth showed that preterm birth occurred in 6.8% of the infants in the vaccine group and in 4.9% of those in the placebo group (relative risk, 1.37; 95% CI, 1.08 to 1.74; P=0.01) (Table 4). Thus, for every 54 infants (95% CI, 32 to 214) born to women who received RSVPreF3-Mat rather than placebo during pregnancy, one additional preterm birth occurred. The evaluation of preterm births by the adjudication committee was consistent with our assessment (see the Supplementary Results section in the Supplementary Appendix).

The imbalance in the incidence of preterm birth between the vaccine group and the placebo group was predominantly observed in low- and middle-income countries (Table 4 and Table S7). The incidence of preterm birth in the two trial groups was below the background incidence in most of the participating countries (Table S8).

Among the infants who were born prematurely, very preterm birth (at 28 to <32 weeks of gestation) or extremely preterm birth (at <28 weeks of gestation) occurred in 5.5% (13 of 237) in the vaccine group and in 2.3% (2 of 86) in the placebo group (Table 4). The intervals between vaccination and delivery were similar in the vaccine and placebo groups among all the infants and among the preterm infants (Table S9), and no characteristic interval between the receipt of RSVPreF3-Mat and delivery was observed (Fig. S2). The imbalance in the incidence of preterm birth between the two trial groups was similar regardless of the gestational age of the fetus at the time of maternal vaccination (Table 4).

NEONATAL DEATH

Neonatal death occurred in 0.4% of the infants (13 of 3494) in the vaccine group and in 0.2% of those (3 of 1739) in the placebo group (relative

Table 3. Adverse Events of Special Interest Reported in Infants and Pregnant Women.*

Event	RSVPreF3-Mat Group		Placebo Group		Relative Risk (95% CI)
	Participants	Incidence (95% CI)	Participants	Incidence (95% CI)	
	no./total no.	%	*no./total no.*	%	
Infants					
Congenital anomalies					
Overall	51/3494	1.5 (1.1–1.9)	31/1741	1.8 (1.2–2.5)	0.82 (0.53–1.28)
With functional defects	5/3494	0.1 (0.0–0.3)	2/1741	0.1 (0.0–0.4)	1.25 (0.24–6.41)
With internal structural defects	23/3494	0.7 (0.4–1.0)	16/1741	0.9 (0.5–1.5)	0.72 (0.38–1.35)
With major external structural defects	24/3494	0.7 (0.4–1.0)	14/1741	0.8 (0.4–1.3)	0.85 (0.44–1.65)
Low, very low, or extremely low birth weight†					
<2500 g	204/3494	5.8 (5.1–6.7)	102/1741	5.9 (4.8–7.1)	1.00 (0.79–1.26)
1500 to <2500 g	196/3494	5.6 (4.9–6.4)	102/1741	5.9 (4.8–7.1)	0.96 (0.76–1.21)
1000 to <1500 g	5/3494	0.1 (0.0–0.3)	0/1741	0.0 (0.0–0.2)	NE
<1000 g	3/3494	0.1 (0.0–0.3)	0/1741	0.0 (0.0–0.2)	NE
Neonatal death‡					
Overall	13/3494	0.4 (0.2–0.6)	3/1741	0.2 (0.0–0.5)	2.16 (0.62–7.57)
After full-term birth	6/3494	0.2 (0.1–0.4)	3/1741	0.2 (0.0–0.5)	1.00 (0.25–3.98)
After moderate-to-late or very preterm birth	5/3494	0.1 (0.0–0.3)	0/1741	0.0 (0.0–0.2)	NE
After extremely preterm birth	2/3494	0.1 (0.0–0.2)	0/1741	0.0 (0.0–0.2)	NE
Preterm birth§	237/3494	6.8 (6.0–7.7)	86/1741	4.9 (4.0–6.1)	1.37 (1.08–1.75)
Small for gestational age	253/3494	7.2 (6.4–8.2)	161/1741	9.2 (7.9–10.7)	0.78 (0.65–0.95)
Maternal participants					
Chorioamnionitis	30/3557	0.8 (0.6–1.2)	14/1771	0.8 (0.4–1.3)	1.07 (0.57–2.01)
Fetal growth restriction¶	30/3557	0.8 (0.6–1.2)	26/1771	1.5 (1.0–2.1)	0.57 (0.34–0.97)
Gestational diabetes mellitus	42/3557	1.2 (0.9–1.6)	19/1771	1.1 (0.6–1.7)	1.10 (0.64–1.89)
Hypertensive disorders of pregnancy					
Overall	209/3557	5.9 (5.1–6.7)	99/1771	5.6 (4.6–6.8)	1.05 (0.83–1.33)
Gestational hypertension	85/3557	2.4 (1.9–2.9)	41/1771	2.3 (1.7–3.1)	1.03 (0.71–1.49)
Preeclampsia	74/3557	2.1 (1.6–2.6)	36/1771	2.0 (1.4–2.8)	1.02 (0.69–1.52)
Preeclampsia with severe features, including eclampsia	58/3557	1.6 (1.2–2.1)	28/1771	1.6 (1.1–2.3)	1.03 (0.66–1.61)
Maternal death	2/3557	0.1 (0.0–0.2)	1/1771	0.1 (0.0–0.3)	1.00 (0.09–10.97)
Pathway to preterm birth‖	248/3557	7.0 (6.2–7.9)	93/1771	5.3 (4.3–6.4)	1.33 (1.05–1.67)
Premature preterm rupture of membranes	53/3557	1.5 (1.1–1.9)	20/1771	1.1 (0.7–1.7)	1.32 (0.79–2.20)
Preterm labor	141/3557	4.0 (3.3–4.7)	57/1771	3.2 (2.4–4.2)	1.23 (0.91–1.67)
Provider-initiated preterm birth	54/3557	1.5 (1.1–2.0)	16/1771	0.9 (0.5–1.5)	1.68 (0.96–2.93)

Table 3. (Continued.)

* This analysis was based on data from women who received a single dose of RSVPreF3-Mat or placebo and their live-born infants (the database was locked on October 5, 2023). Adverse events of special interest were recorded through week 6 after delivery or birth; definitions are provided in the Supplementary Appendix. The incidence of some of these events was lower than that in the general population because women who were known to have clinically important pregnancy complications (including fetal growth restriction, gestational diabetes, gestational hypertension, preeclampsia, and eclampsia) before randomization and those whose fetus had major congenital malformations (as defined in the protocol) were excluded from the trial. The analyses were not adjusted for multiplicity. NE denotes could not be estimated.

† A weight of 1500 to less than 2500 g was defined as low birth weight, a weight of 1000 to less than 1500 g as very low, and a weight of less than 1000 g as extremely low.

‡ Full-term birth was defined as a gestational age of 37 weeks or more at birth, moderate-to-late preterm birth as a gestational age of 32 to less than 37 weeks, very preterm birth as a gestational age of 28 to less than 32 weeks, and extremely preterm birth as a gestational age of less than 28 weeks.

§ Preterm birth was defined as birth at a gestational age of less than 37 weeks.

¶ Fetal growth restriction was defined as an estimated fetal weight below the 10th percentile for a given gestational age on ultrasonography (see the Supplementary Methods in the Supplementary Appendix). Although mothers had to undergo ultrasonography before enrollment in the trial, after enrollment ultrasound examinations were performed as part of routine clinical management, with participants undergoing an ultrasound examination if warranted as part of a symptom-directed obstetrical examination.

‖ The number of maternal participants with a reported pathway to preterm birth was higher than the number of reported preterm births because some cases of premature preterm rupture of membranes resulted in birth at full term even though the rupture occurred before 37 weeks of gestation, some cases of preterm labor or provider-initiated preterm birth resulted in stillbirth or fetal death, and some maternal participants were lost to follow-up.

risk, 2.16; 95% CI, 0.62 to 7.56; P=0.23). Among the preterm infants, neonatal death occurred in 7 in the vaccine group and in none in the placebo group; neonatal death occurred in 6 and 3 full-term infants, respectively, a finding that reflected the 2:1 randomization ratio. No consistent interval between maternal vaccination and neonatal death and between birth and neonatal death was observed among the infants who died, and factors that led to neonatal death were recognized as complications of prematurity (Tables S10, S11, and S12), which suggests that any between-group difference in the incidence of neonatal death was attributable to the higher incidence of preterm birth in the vaccine group than in the placebo group.

TEMPORAL DISTRIBUTION OF PRETERM BIRTHS

The imbalance in the incidence of preterm birth between the two trial groups was consistently observed between April and December 2021 but not thereafter (Fig. 1 and Table S13). This temporal pattern occurred in low- and middle-income countries and in high-income countries, but the pattern was particularly apparent in low- and middle-income countries, where a peak occurred from August to December 2021. The imbalance was not differentially associated with any vaccine lot (Table S14).

The current trial was performed during the coronavirus disease 2019 (Covid-19) pandemic. Although the months when the imbalance in the incidence of preterm birth was observed did not match the months when most of the Covid-19 cases in the maternal participants were reported, the peak between-group difference in the incidence of preterm birth coincided with the Covid-19 wave that occurred when the B.1.617.2 (delta) variant was dominant (Fig. S3). However, only a small percentage of the preterm infants in the vaccine group were born to mothers who reported having had Covid-19 during pregnancy (6.3% of infants [15 of 237]) or who had evidence of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection, as indicated by seroconversion for antibodies to the SARS-CoV-2 nucleocapsid protein (7.7% of infants [17 of 222]), during pregnancy. The difference in the incidence of preterm birth between the vaccine and placebo groups could not be explained by reported cases of Covid-19, evidence of SARS-CoV-2 infection, or vaccination against Covid-19 during pregnancy in women who delivered at any time during the trial or when the delta variant was dominant (Tables S15, S16, and S17).

RECEIPT OF ADDITIONAL VACCINES DURING PREGNANCY

Maternal participants could receive additional vaccines at least 2 weeks before or 2 weeks after the receipt of RSVPreF3-Mat or placebo according to the standard of care at each trial site and the participant's choice. Nearly 70% of the women in

Table 4. Post Hoc Analysis of the Risk of Preterm Birth among Infants According to Risk Factor.*

Variable	RSVPreF3-Mat Group		Placebo Group		Relative Risk (95% CI)
	Infants	Incidence (95% CI)	Infants	Incidence (95% CI)	
	no. of preterm births/total no.	%	*no. of preterm births/total no.*	%	
Preterm birth†					
Overall	237/3494	6.8 (6.0–7.7)	86/1739	4.9 (4.0–6.1)	1.37 (1.08–1.74)
Moderate-to-late preterm	224/3494	6.4 (5.6–7.3)	84/1739	4.8 (3.9–5.9)	1.33 (1.04–1.69)
Very preterm	11/3494	0.3 (0.2–0.6)	2/1739	0.1 (0.0–0.4)	2.74 (0.61–12.34)
Extremely preterm	2/3494	0.1 (0.0–0.2)	0/1739	0.0 (0.0–0.2)	NE
Country income level					
Low or middle income	172/1753	9.8 (8.5–11.3)	55/875	6.3 (4.8–8.1)	1.56 (1.17–2.09)
High income	65/1741	3.7 (2.9–4.7)	31/864	3.6 (2.5–5.1)	1.04 (0.68–1.58)
Gestational age at vaccination					
≤28 wk 6 days	127/1428	8.9 (7.5–10.5)	49/714	6.9 (5.1–9.0)	1.30 (0.94–1.78)
≥29 wk 0 days	110/2066	5.3 (4.4–6.4)	37/1025	3.6 (2.6–4.9)	1.47 (1.02–2.12)
Mother reported Covid-19 during pregnancy‡					
Yes	15/178	8.4 (4.8–13.5)	5/83	6.0 (2.0–13.5)	1.40 (0.53–3.72)
No	222/3316	6.7 (5.9–7.6)	81/1656	4.9 (3.9–6.0)	1.37 (1.07–1.75)
Mother received ≥1 additional vaccine during pregnancy§					
In all countries					
No	101/1082	9.3 (7.7–11.2)	41/523	7.8 (5.7–10.5)	1.19 (0.84–1.69)
Yes	136/2412	5.6 (4.8–6.6)	45/1216	3.7 (2.7–4.9)	1.52 (1.10–2.12)
In low- and middle-income countries					
No	80/625	12.8 (10.3–15.7)	30/298	10.1 (6.9–14.1)	1.27 (0.86–1.89)
Yes	92/1128	8.2 (6.6–9.9)	25/577	4.3 (2.8–6.3)	1.88 (1.22–2.90)
In high-income countries					
No	21/457	4.6 (2.9–6.9)	11/225	4.9 (2.5–8.6)	0.94 (0.46–1.92)
Yes	44/1284	3.4 (2.5–4.6)	20/639	3.1 (1.9–4.8)	1.09 (0.65–1.84)

* This analysis was based on data in infants born to women who received a single dose of RSVPreF3-Mat or placebo (the database was locked on October 4, 2022). The relative risks were calculated by dividing the incidence of preterm birth in the vaccine group by the incidence of preterm birth in the placebo group, and the 95% confidence intervals were calculated with the use of the Wald method. The analyses were not adjusted for multiplicity.

† Preterm birth was defined as a gestational age of less than 37 weeks, moderate-to-late preterm birth as a gestational age of 32 to less than 37 weeks, very preterm birth as a gestational age of 28 to less than 32 weeks, and extremely preterm birth as a gestational age of less than 28 weeks.

‡ The occurrence of coronavirus disease 2019 (Covid-19) during pregnancy was reported by the maternal participants and included suspected, probable, or confirmed Covid-19.

§ Additional vaccines were those received during the second or third trimester of pregnancy. Pertussis-containing, Covid-19 messenger RNA, influenza, tetanus (in low- or middle-income countries), and tetanus–diphtheria (in low- or middle-income countries) vaccines were the most common additional vaccines received by the maternal participants.

each trial group received at least one additional vaccine during the second or third trimester. Pertussis-containing, Covid-19 messenger RNA, influenza, tetanus, and tetanus–diphtheria vaccines were the most common additional vaccines received (Table S18).

In the two trial groups, the incidence of preterm birth was lower among infants born to women who received at least one additional vaccine than among infants born to women who did not receive an additional vaccine, both overall and according to country income level (Table 4 and Table S19). The percent differences were less pronounced in the vaccine group than in the placebo group (Table S20). Thus, the relative risk for preterm birth associated with the receipt of RSVPreF3-Mat as compared with placebo appeared to be higher among infants born to mothers who received at least one additional vaccine, with the highest relative risk observed in low- and middle-income countries (Table 4). The imbalance in the incidence of preterm birth between the two trial groups did not increase with the number of additional vaccines received (Table S21), and the time between the receipt of the last additional vaccine and delivery was similar in the two trial groups (Fig. S4 and Table S22). However, the months when the imbalance peaked coincided with the months when at least one additional vaccine was received by the most women (Fig. S5).

We performed additional post hoc analyses to identify other factors that might explain the higher risk of preterm birth in the RSVPreF3-Mat group than in the placebo group (Tables S23, S24, and S25 and Figs. S6 and S7). No other factors were identified in these analyses.

DISCUSSION

The results of this multicenter, placebo-controlled trial suggest that the risks of any and severe medically assessed RSV-associated lower respiratory tract disease among infants were lower in the vaccine group than in the placebo group but that the risk of preterm birth was higher in the vaccine group. The findings regarding preterm birth led to the cessation of recruitment and vaccination in the current trial and to the discontinuation of RSVPreF3-Mat development.

Preterm birth has many potential sequelae, including neonatal death.[15] The difference in the incidence of neonatal death between the RSVPreF3-Mat and placebo groups in this trial was not significant and was likely attributable to the between-group imbalance in the incidence of preterm birth and the degree of prematurity. No other safety signal has been observed among infant or maternal participants in any trial of RSVPreF3-Mat. An adjuvanted RSV vaccine for older adults that contains the same antigen as RSVPreF3-Mat showed a strongly positive benefit-to-risk profile in persons 60 years of age or older.[16]

The mechanism by which RSVPreF3-Mat may have led to an increased risk of preterm birth as compared with placebo is unknown. The intervals from vaccination to preterm delivery generally ranged from weeks to months, which suggests the absence of a direct effect of vaccination on mechanisms that initiated preterm birth. Although inflammatory processes have been associated with premature birth,[17] post hoc analyses in the current trial showed no association between cytokine levels in maternal participants (as assessed in serum samples obtained before and 1 month after vaccination) and preterm birth. However, blood samples were not obtained soon enough after vaccination to reliably detect vaccine-related increases in cytokine levels.

A difference in the incidence of preterm birth between the RSVPreF3-Mat and placebo groups was predominantly observed in low- and middle-income countries, where approximately 50% of the trial population was enrolled and where the medical need for maternal RSV vaccines is the greatest. If a smaller percentage of participants from low- and middle-income countries had been enrolled in our trial, the relative risk of preterm birth in the vaccine group as compared with the placebo group might have been reduced in the overall trial population. Measures of socioeconomic status other than trial site in a low- or middle-income country were not associated with the preterm birth imbalance.

The imbalance in the incidence of preterm birth had a distinctive temporal distribution, particularly in low- and middle-income countries, where it peaked between August and December 2021 and was not evident afterwards.

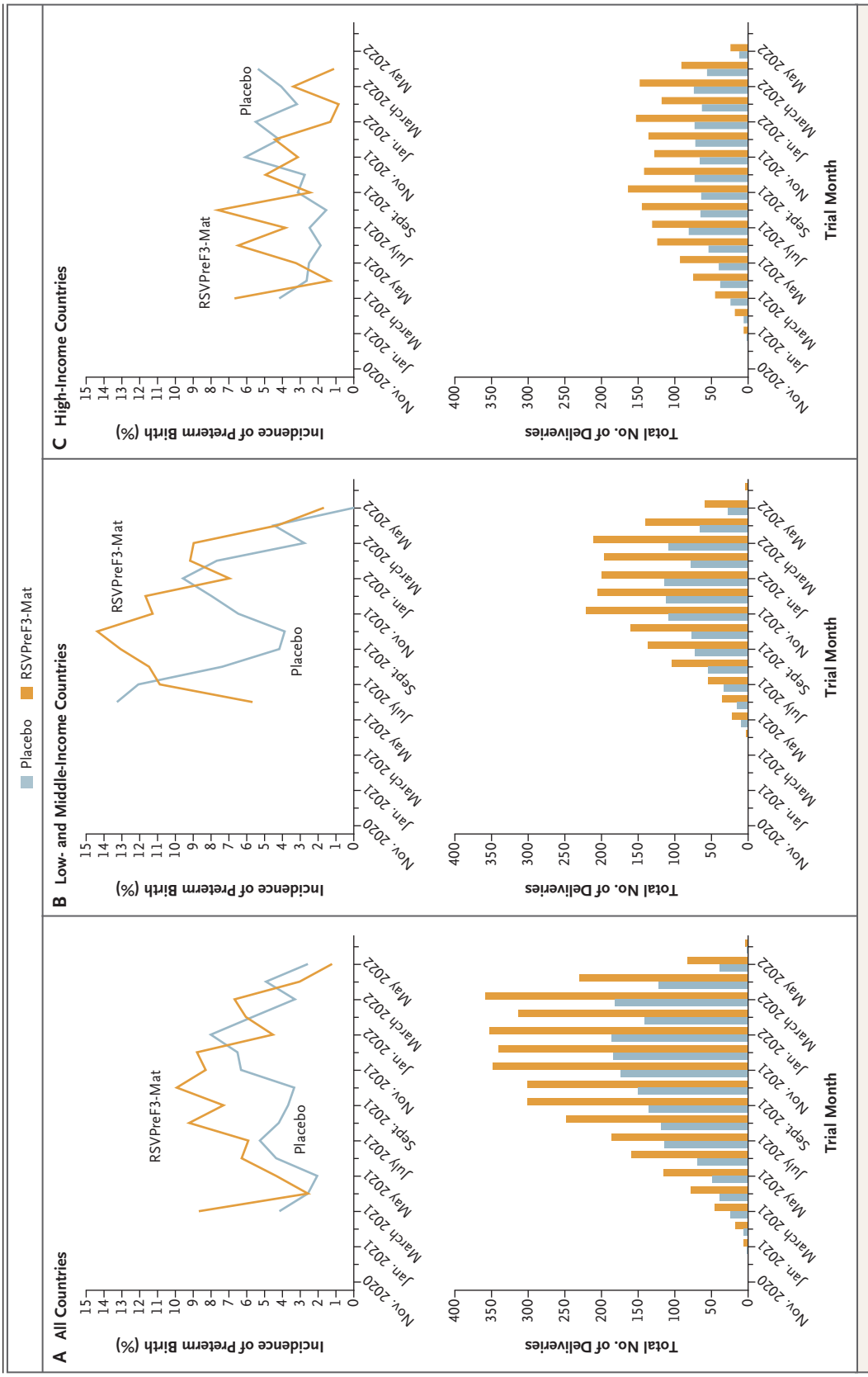

Figure 1. Monthly Incidence of Preterm Births and Number of Deliveries.

Shown is the noncumulative incidence of preterm births during months in which at least 50 deliveries occurred (upper panels) and the total number of deliveries per month (lower panels), overall and according to country income level, among pregnant women who were randomly assigned in a 2:1 ratio to receive an investigational respiratory syncytial virus prefusion F protein–based maternal vaccine (RSVPreF3-Mat) or placebo. Data for this post hoc analysis were locked as of October 4, 2022.

This temporal pattern suggests the possibility of a time-limited cofactor. The trial was conducted during the Covid-19 pandemic, and the peak imbalance occurred when the delta variant was dominant. SARS-CoV-2 infection during pregnancy — particularly during waves of infection with the delta variant — has been associated with an increased risk of preterm birth,[18,19] as we also observed in the current trial. However, participant-reported cases of Covid-19, serologic evidence of SARS-CoV-2 infection, or vaccination against Covid-19 could not account for the higher incidence of preterm birth in the vaccine group than in the placebo group among women who delivered at any time during the trial or when the delta variant was dominant. The spread of influenza virus remained suppressed during the trial. RSV infection during pregnancy was not associated with preterm birth.

In our trial, participants could receive standard-of-care vaccinations during pregnancy, except within the 2 weeks before or the 2 weeks after receipt of the trial vaccine or placebo. Additional vaccines received by the maternal participants included pertussis-containing, Covid-19, influenza, tetanus, and tetanus–diphtheria vaccines. The administration of these vaccines during pregnancy is common in many countries, has maternal and infant health benefits,[5,6,20,21] and has not been associated with increased risks of preterm birth.[7,20-23] In our analyses, the receipt of additional vaccines during pregnancy was associated with a lower incidence of preterm birth in the RSVPreF3-Mat and placebo groups; this finding could be explained by a lower incidence of infections or by differences in health-related behaviors that are associated with a lower risk of preterm birth among women who are vaccinated during pregnancy. However, the percent reduction in the risk of preterm birth associated with the receipt of additional vaccines was greater in the placebo group, such that the receipt of additional vaccines was paradoxically associated with an increased risk of preterm birth in the vaccine group as compared with the placebo group, especially in low- and middle-income countries. Participants were not randomly assigned to receive these additional vaccines, and the receipt of additional vaccines was not concealed from the participants or the investigators.

A higher risk of preterm birth in the vaccine group than in the placebo group was not seen in two smaller trials of RSVPreF3-Mat involving pregnant women. In the phase 2 RSV MAT-004 trial, conducted from November 2019 to May 2021, preterm birth occurred in 4 of 140 infants (2.9%) in the vaccine group and in 2 of 66 infants (3.0%) in the placebo group (relative risk, 0.94; 95% CI, 0.18 to 5.02).[24] In the phase 3 RSV MAT-012 trial (ClinicalTrials.gov number, NCT04980391) in women with a high-risk pregnancy, preterm birth occurred in 24 of 132 infants (18.2%) in the vaccine group and in 13 of 66 infants (19.7%) in the placebo group (relative risk, 0.92; 95% CI, 0.50 to 1.69) (unpublished interim data from August 2021 to September 2023). No imbalance in the incidence of premature delivery was observed in a phase 3 trial (conducted from December 2015 to July 2019 in the United States and South Africa) of a maternal vaccine containing the RSV F protein that was not stabilized in the prefusion state: the incidence was 5.7% in both the vaccine group (among 3045 pregnant women) and the placebo group (among 1581 pregnant women).[25]

However, results of interim analyses of a phase 2b trial and a phase 3 trial of a maternal RSV vaccine that contained RSV F protein in a prefusion-stabilized conformation suggested that the risk of preterm birth was higher in the vaccine groups than in the placebo groups. In the phase 2b trial, an analysis of data as of January 31, 2020, showed that 14 of 325 infants (4.3%) in the vaccine groups and 1 of 78 infants (1.3%) in the placebo group were born prematurely.[26] In the phase 3 trial, an analysis of data as of September 2, 2022, showed that 201 of 3568 infants (5.6%) in the vaccine group and 169 of 3558 infants (4.7%) in the placebo group were born prematurely.[27] Our phase 3 trial differed from the phase 3 trial by Kampmann et al.[27] in several ways, including the eligibility criteria (e.g., vaccination was allowed up to week 34 of gestation in our trial as compared with vaccination through week 36 of gestation in their trial) and the percentage of participants who were enrolled in low- or middle-income countries (50% in our trial as compared with 31% in their trial).

Limitations of our trial include the necessary use of post hoc analyses to assess the safety signal of preterm birth; also, in keeping with the standard approach in safety investigations, these

analyses were not controlled for multiplicity in order to minimize the chance of missing a true safety signal. Blood and tissue samples obtained at the most relevant time points (e.g., 1–2 weeks after vaccination for the detection of vaccine-related increases in cytokine levels) were not available. In addition, because sites in low- and middle-income countries were primarily in the Southern Hemisphere and sites in high-income countries were primarily in the Northern Hemisphere, it was not possible to distinguish the possible contributions of socioeconomic or geographic factors to the imbalanced incidence of preterm births. Finally, prespecified efficacy and safety outcomes could have been affected by performance bias or detection bias after the trial was unblinded.

The results of this trial, in which enrollment was stopped early because of safety concerns, suggest that the risks of any medically assessed RSV-associated lower respiratory tract disease and severe medically assessed RSV-associated lower respiratory tract disease among infants were lower with RSVPreF3-Mat than with placebo but that the risk of preterm birth was higher with RSVPreF3-Mat. The increased risk of preterm birth in the vaccine group was largely seen in low- and middle-income countries, was limited to a defined period in the trial, and remains unexplained.

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A data sharing statement provided by the authors is available with the full text of this article at NEJM.org.

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