ORIGINAL ARTICLE

Quadrivalent HPV Vaccination and the Risk of Adverse Pregnancy Outcomes

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ABSTRACT

BACKGROUND

The quadrivalent human papillomavirus (HPV) vaccine is recommended for all girls and women 9 to 26 years of age. Some women will have inadvertent exposure to vaccination during early pregnancy, but few data exist regarding the safety of the quadrivalent HPV vaccine in this context.

METHODS

We assessed a cohort that included all the women in Denmark who had a pregnancy ending between October 1, 2006, and November 30, 2013. Using nationwide registers, we linked information on vaccination, adverse pregnancy outcomes, and potential confounders among women in the cohort. Women who had vaccine exposure during the prespecified time windows were matched for propensity score in a 1:4 ratio with women who did not have vaccine exposure during the same time windows. Outcomes included spontaneous abortion, stillbirth, major birth defect, small size for gestational age, low birth weight, and preterm birth.

RESULTS

In matched analyses, exposure to the quadrivalent HPV vaccine was not associated with significantly higher risks than no exposure for major birth defect (65 cases among 1665 exposed pregnancies and 220 cases among 6660 unexposed pregnancies; prevalence odds ratio, 1.19; 95% confidence interval [CI], 0.90 to 1.58), spontaneous abortion (20 cases among 463 exposed pregnancies and 131 cases among 1852 unexposed pregnancies; hazard ratio, 0.71; 95% CI, 0.45 to 1.14), preterm birth (116 cases among 1774 exposed pregnancies and 407 cases among 7096 unexposed pregnancies; prevalence odds ratio, 1.15; 95% CI, 0.93 to 1.42), low birth weight (76 cases among 1768 exposed pregnancies and 277 cases among 7072 unexposed pregnancies; prevalence odds ratio, 1.10; 95% CI, 0.85 to 1.43), small size for gestational age (171 cases among 1768 exposed pregnancies and 783 cases among 7072 unexposed pregnancies; prevalence odds ratio, 0.86; 95% CI, 0.72 to 1.02), or stillbirth (2 cases among 501 exposed pregnancies and 4 cases among 2004 unexposed pregnancies; hazard ratio, 2.43; 95% CI, 0.45 to 13.21).

CONCLUSIONS

Quadrivalent HPV vaccination during pregnancy was not associated with a significantly higher risk of adverse pregnancy outcomes than no such exposure. (Funded by the Novo Nordisk Foundation and the Danish Medical Research Council.)

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Human papillomavirus (HPV) vaccines are recommended for all girls and women 9 to 26 years of age,[1,2] and more than 72 million girls and women have been vaccinated worldwide.[3] Although HPV vaccination is not recommended in pregnancy, a number of women will be inadvertently vaccinated early in the first trimester of unplanned or unrecognized pregnancies.[4,5] However, data on the safety of vaccination during pregnancy are limited.[6]

The clinical trials of HPV vaccines did not include women who were known to be pregnant. Consequently, analyses of safety during pregnancy that were based on data from clinical trials focused mainly on the risk of adverse pregnancy outcomes associated with HPV vaccines that were administered before pregnancy onset.[7-9] These studies did not identify a significant difference in the risk of adverse pregnancy outcomes, although one pooled analysis of two trials of the bivalent HPV vaccine showed a nonsignificantly higher risk of spontaneous abortion among women whose pregnancies were conceived less than 90 days from bivalent HPV vaccination than among those in the control group (13.7% vs. 9.2%, one-sided $P=0.03$).[7] However, this finding could not be substantiated in an updated, larger study that included a meta-analysis of the risk of spontaneous abortion among women whose pregnancies were conceived less than 90 days from bivalent HPV vaccination, as compared with women in the control group (relative risk, 1.11, 95% confidence interval [CI], 0.82 to 1.51).[9]

Observational studies have been few and limited by size and design and are therefore inadequate to evaluate the risks associated with HPV vaccination during pregnancy. A cohort study of the bivalent HPV vaccine, which included only 207 vaccinated women, showed that the risk of adverse pregnancy outcomes was not higher among women whose first day of gestation occurred between 30 days before vaccination and 45 days after vaccination than among women whose first day of gestation was between 120 days and 18 months after the last dose of bivalent HPV vaccine.[10] Two studies that were based on data from the same manufacturer-managed pregnancy register of the quadrivalent HPV vaccine also showed that the risk among vaccinated women was not greater than the expected risk in the general population, but these studies were based on voluntary reporting and lacked control groups.[11,12]

To investigate the safety of HPV vaccine further, we conducted a nationwide register-based cohort study involving all pregnant women in Denmark. We investigated the association between quadrivalent HPV vaccination during pregnancy and the risks of a major birth defect, spontaneous abortion, stillbirth, preterm birth, low birth weight, and small size for gestational age.

METHODS

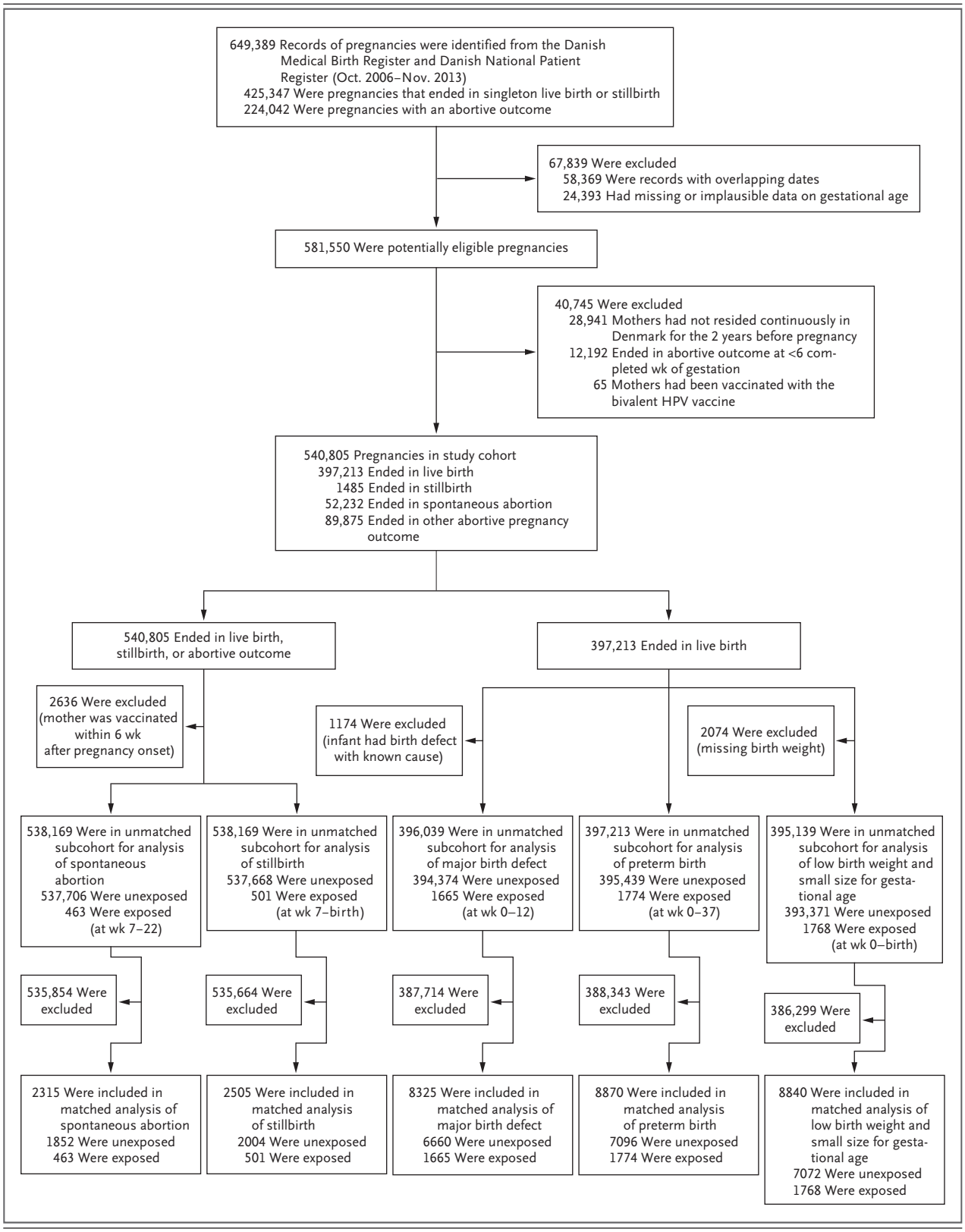
COHORT

Using the Medical Birth Register[13] and the National Patient Register,[14] we identified all pregnancies in Denmark that ended in a singleton birth or an abortive outcome between October 1, 2006, and November 30, 2013 (Fig. 1). We then estimated the date of pregnancy onset by subtracting the gestational age from the date of birth or abortion. Records of gestational age for live-born infants and stillbirths are based mainly on ultrasonography, whereas records of gestational age in abortive outcomes are based on ultrasonography or the first day of the last menstrual period.[15] The records of gestational age for live births were validated to be accurate within 1 week for 87% of the records.[16]

We subsequently excluded pregnancies with missing or implausible data on gestational age, pregnancies with multiple overlapping records, and pregnancies in women who had not lived continuously in Denmark for the 2-year period before pregnancy onset. In addition, for analyses of spontaneous abortion, we excluded all cases of

Figure 1 (facing page). Construction of the Two Study Cohorts and Propensity-Score Matching of Pregnant Women with Vaccine Exposure and Those without Vaccine Exposure, Yielding Five Outcome-Specific Cohorts.

Values regarding exclusions do not sum to the total number because some pregnancies were excluded for more than one reason. Between the unmatched and matched analyses, all the excluded records were from unmatched pregnancies that did not have vaccine exposure. All the outcome-specific analyses were matched for maternal age, calendar year of pregnancy onset, and propensity score, and the analyses of spontaneous abortion and stillbirth were also matched for gestational age.



spontaneous abortion that occurred within the first 6 weeks of gestation, because many cases of spontaneous abortion during the first weeks of gestation are likely to be clinically unrecognized. Consequently, for analyses of spontaneous abortion we also excluded women who had been exposed to vaccination within the first 6 weeks of gestation to ensure that immortal time (a follow-up period during which the study outcome, by design, could not occur) was not introduced.

We then defined two cohorts: one cohort included all the pregnancies that ended in live birth and was used for the analyses of major birth defect, preterm birth, low birth weight, and small size for gestational age; and the second cohort included all the pregnancies and was used for the analyses of spontaneous abortion and stillbirth (Fig. 1). The unique personal identification numbers that are given to every resident in Denmark enabled the individual-level linkage of our cohort with nationwide health and demographic registers containing information on vaccination, adverse outcomes, and potential confounders (see the Supplementary Appendix, available with the full text of this article at NEJM.org).[17]

The study was approved by the Danish Data Protection Agency. Informed consent is not required for registry-based research in Denmark. The funders and vaccine manufacturers had no role in the design and conduct of the study; the collection, management, analysis, and interpretation of the data; the preparation, review, or approval of the manuscript; or the decision to submit the manuscript for publication. The first and last authors take responsibility for the accuracy and completeness of the reported data and analyses.

VACCINATION

During the study period, the quadrivalent HPV vaccine (Gardasil, Sanofi Pasteur MSD [manufactured in the United States by Merck]) was the sole HPV vaccine used in the Danish national vaccination program (see the Supplementary Appendix). The few women who were vaccinated with the bivalent HPV vaccine (Cervarix, GlaxoSmithKline Biologicals) before or during pregnancy were excluded from our study (Fig. 1).

Information on quadrivalent HPV vaccinations that were given through the national vaccination program was obtained from the Danish Childhood Vaccination Database.[18] The quadrivalent HPV vaccine was also available by prescription, so additional data on vaccination status were obtained from the Danish National Prescription Registry.[19] We defined the date of exposure to the quadrivalent HPV vaccine as the date of the first vaccination or filled prescription. Exposure windows were categorized according to study outcome: the first trimester (pregnancy onset through week 12 of gestation) for the analysis of major birth defect, the start of week 7 through week 22 of gestation for the analysis of spontaneous abortion, the start of week 7 of gestation until birth for the analysis of stillbirth, before 37 completed weeks of gestation for the analysis of preterm birth, and at any time during pregnancy for the analyses of low birth weight and small size for gestational age. In each analysis, unexposed pregnancies were defined as pregnancies in women who were not vaccinated during the specified exposure window.

OUTCOMES

The outcome of major birth defects overall was defined as the first registered diagnosis of any major birth defect within the first year of life (see the Supplementary Appendix), as identified in the National Patient Register.[14] A validation study of the National Patient Register estimated a predictive value of 88% for diagnoses of birth defects overall.[20] Cases of spontaneous abortion (defined as fetal death occurring through 22 weeks of gestation) were identified in the National Patient Register. Validation has shown that the records were correct for 99% of the diagnoses of spontaneous abortion (details on abortive outcomes are provided in the Supplementary Appendix).[21] Information on stillbirth (defined as fetal loss after 22 completed weeks of gestation), preterm birth (delivery before 37 completed weeks of gestation), low birth weight (<2500 g), and small size for gestational age (birth weight in the lowest 10th percentile of gestational age–specific birth weight in the cohort) was obtained from the Medical Birth Register. For the analyses of low birth weight and small size for gestational age, we excluded records of all the pregnancies resulting in live birth for which information on birth weight was missing.

STATISTICAL ANALYSIS

Because different exposure windows and exclusion criteria were applied to the analysis set of each distinct outcome, we created five unmatched

outcome-specific cohorts (Fig. 1). Selected baseline characteristics were then identified for each woman at pregnancy onset, and missing values were imputed with the use of mode imputation (Table S1 in the Supplementary Appendix). To account for potential confounders, we calculated propensity scores using logistic regression, which estimated the probability of quadrivalent HPV vaccination in outcome-specific exposure time windows, given all baseline characteristics (and all two-way interactions between demographic variables).

For each of the five outcome-specific cohorts, we then matched vaccinated women and unvaccinated women in a 1:4 ratio according to age (5-year categories), calendar year of pregnancy onset, and propensity score, thus creating a distinct matched analysis set for each of the five outcome-specific cohorts (Fig. 1). We added gestational age as a matching criterion for the cohorts regarding spontaneous abortion and stillbirth, the risks of which are highly dependent on gestational age.

Matching was performed with the use of the nearest-neighbor matching algorithm (caliper width, 0.2 of the standard deviation of the logit score). We assessed the balance of covariates that was achieved from matching by evaluating standardized differences between vaccinated groups and unvaccinated groups. We considered covariates with a standardized difference of less than 10% to be well balanced.

We performed the analyses of spontaneous abortion and stillbirth using gestational age as the underlying time scale. Hazard ratios with 95% confidence intervals were estimated with the use of Cox proportional-hazards regression. The Wald test was used to assess the fulfillment of the proportional-hazards assumption, which was fulfilled for the analyses of stillbirth ($P=0.83$) and spontaneous abortion ($P=0.46$). In the analyses of major birth defect, preterm birth, low birth weight, and small size for gestational age, we used logistic regression to estimate prevalence odds ratios. The generalized estimating equation method was used for all analyses to account for the possible correlation between pregnancies within the same mother.

We performed a number of prespecified sensitivity analyses. In the analysis of major birth defect, we restricted the exposure window to weeks 4 to 10 of gestation, which corresponds to the period of maximal susceptibility to teratogenic agents.[22] Basing the analysis of major birth defect only on live births could potentially introduce misclassification by the exclusion of stillbirths and induced abortions that were caused by major birth defects, thus potentially biasing results toward no association. We therefore conducted an analysis of major birth defect that included stillbirths and induced abortions (see the Supplementary Appendix). Furthermore, we performed a complete-case analysis for each subcohort, which excluded all the participants who had missing data. To investigate the potential for residual confounding we performed two additional analyses: one analysis incorporated 1:1 matching, because 1:1 matching increases the comparability between exposure groups,[23] and a second analysis incorporated an increase in the granularity of the age variable that was used for matching, because vaccination and pregnancy outcomes are both heavily dependent on age.

Post hoc, we added two sensitivity analyses. First, we excluded pregnancies among women who were not exposed to vaccination during pregnancy but who had been vaccinated within 30 days before pregnancy onset. Second, since the date of filling a prescription may differ from the actual date of vaccination, we excluded women who were defined as having vaccine exposure when they filled a vaccine prescription during pregnancy as well as excluding their corresponding matches. SAS software, version 9.4 (SAS Institute), was used for all the analyses.

RESULTS

COHORTS

During the study period, we identified 649,389 records of pregnancies, of which 581,550 were eligible for inclusion in the study cohort (Fig. 1). Taking into account the use of the outcome-specific exposure windows and exclusion criteria, we created five unmatched subcohorts: for the analysis of spontaneous abortion, the subcohort included 538,169 pregnancies (463 women vaccinated during weeks 7 to 22 of gestation); for the analysis of major birth defect, the subcohort included 396,039 pregnancies (1665 women vaccinated in week 0 to week 12 of gestation); for the analysis of stillbirth, the subcohort included 538,169 pregnancies (501 women vaccinated in week 7 to birth); for the analysis of preterm

birth, the subcohort included 397,213 pregnancies (1774 women vaccinated in week 0 to week 37 of gestation); and for the analyses of low birth weight and small size for gestational age, the subcohorts included 395,139 pregnancies (1768 women vaccinated at any time during pregnancy).

Before matching was performed, we found that vaccinated women were younger, were more often nulliparous, had lower levels of education, were more likely to be in the two lowest quintiles of household income, were more likely to be unmarried, and were more likely to be smokers than were unvaccinated women (Table S3A and S3B in the Supplementary Appendix). The C-statistic for the propensity-score models ranged from 0.79 to 0.82, which indicated that substantial differences existed between the vaccinated group and the unvaccinated group. This finding highlighted the need to adjust for confounders.

After matching in a 1:4 ratio, we found that the included covariates were well balanced between the vaccinated group and the unvaccinated group in almost all outcome-specific subcohorts (Tables 1 and 2, and Table S5 in the Supplementary Appendix). Parity and number of hospital admissions during the previous year were not well balanced in the matched analysis of spontaneous abortion and stillbirth. Consequently, we conducted a sensitivity analysis with additional adjustment for these variables.

OUTCOMES

In unadjusted analyses before propensity-score matching, quadrivalent HPV vaccination during pregnancy was associated with significantly higher risks than no such exposure in the analyses of low birth weight (prevalence odds ratio, 1.26; 95% CI, 1.00 to 1.59), preterm birth (prevalence odds ratio, 1.38; 95% CI, 1.14 to 1.67), and major birth defect (prevalence odds ratio, 1.36; 95% CI, 1.06 to 1.75). However, quadrivalent HPV vaccination was not associated with a significantly higher risk of spontaneous abortion (hazard ratio vs. no vaccine exposure, 0.93; 95% CI, 0.60 to 1.44), small size for gestational age (prevalence odds ratio, 0.98; 95% CI, 0.83 to 1.14), or stillbirth (hazard ratio, 1.90; 95% CI, 0.48 to 7.61).

Table 3 shows analyses in the matched subcohorts. Quadrivalent HPV vaccination during pregnancy was not associated with a significantly higher risk than no such exposure in the analyses of major birth defect (prevalence odds ratio, 1.19; 95% CI, 0.90 to 1.58), spontaneous abortion (hazard ratio, 0.71; 95% CI, 0.45 to 1.14), preterm birth (prevalence odds ratio, 1.15; 95% CI, 0.93 to 1.42), small size for gestational age (prevalence odds ratio, 0.86; 95% CI, 0.72 to 1.02), or low birth weight (prevalence odds ratio, 1.10; 95% CI, 0.85 to 1.43). The matched analysis of stillbirth also showed that the risk with vaccine exposure was not significantly higher than the risk without vaccine exposure (hazard ratio, 2.43; 95% CI, 0.45 to 13.21). However, the analysis included only two cases among pregnancies with vaccine exposure and four among pregnancies without vaccine exposure.

SENSITIVITY ANALYSES

The results of the sensitivity analyses are presented in Table 4. In an analysis in which the exposure window was limited to weeks 4 to 10 of gestation, quadrivalent HPV vaccination was not associated with a significantly higher risk than no vaccine exposure in the analysis of major birth defect. Moreover, the inclusion of information on birth defects in pregnancies that ended in induced abortion or stillbirth did not materially change the results of analyses regarding major birth defect. Results for all the outcomes did not differ materially from the main analyses when the analyses were limited to women who had no missing values for any covariates, when 1:1 matching was used, or when we increased the granularity of the age categorization. Post hoc sensitivity analyses that excluded unexposed pregnancies in women who were vaccinated within 30 days before pregnancy onset, that excluded pregnancies that were defined as exposed owing to filling of a vaccine prescription, or that included variables that were not well balanced between groups also yielded results similar to those in the main analysis (Table S6 in the Supplementary Appendix).

DISCUSSION

In a nationwide cohort study conducted in Denmark, we found that quadrivalent HPV vaccination during pregnancy was not associated with significantly greater risks of adverse pregnancy outcomes. Given the upper limits of the confidence intervals in our study, relatively higher risks of more than 58% for major birth defect,

Table 1. Characteristics of Women Included in the Matched Analyses of Spontaneous Abortion and Stillbirth, According to Time Window of Quadrivalent Human Papillomavirus (HPV) Vaccination and Vaccination-Exposure Status during Pregnancy.*

Characteristic	7 to 22 Wk of Gestation for Analysis of Spontaneous Abortion		7 Wk of Gestation to Birth for Analysis of Stillbirth	
	No Vaccine Exposure (N=1852)	Vaccine Exposure (N=463)	No Vaccine Exposure (N=2004)	Vaccine Exposure (N=501)
Median no. of days that vaccination occurred after pregnancy onset (IQR)	—	54 (47–72)	—	55 (47–80)
Age at pregnancy onset — yr	24.8±4.0	24.6±3.5	24.9±4.1	24.6±3.6
Born in Denmark — %	92.5	91.8	92.6	92.0
Married or living with partner — %	59.9	59.6	59.0	60.7
Bachelor's degree or higher educational level — %	13.5	13.2	14.0	12.8
Household income in 3rd quintile — %†	17.4	16.8	16.7	17.6
Calendar year of delivery or pregnancy loss 2012 or 2013 — %	88.0	88.1	88.3	88.4
Parity — %				
0	71.0	65.7	69.7	65.3
≥1	29.1	34.4	30.3	34.7
Same outcome in previous pregnancy — %‡	9.3	11.9	0.4	0.4
Diabetes mellitus — %	2.6	2.8	3.4	3.2
Used in vitro fertilization drug — %	1.3	1.5	1.8	1.8
No. of hospital admissions in previous year — %				
1 or 2	6.0	6.7	5.6	6.4
≥3	15.2	17.3	13.3	17.0
No. of outpatient hospital visits in previous year — %				
1 or 2	18.2	17.3	16.1	17.4
≥3	23.9	25.3	24.1	25.5
No. of emergency hospital visits in previous year — %				
1 or 2	13.8	15.1	12.6	15.0
≥3	4.5	4.5	3.3	4.6
No. of prescriptions filled in previous 6 mo — %				
1 or 2	46.3	47.5	45.5	47.5
≥3	24.8	27.2	27.4	27.0

* Plus–minus values are means ±SD. Matching was done in a 1:4 ratio on the basis of maternal age, calendar year of pregnancy onset, propensity score, and gestational age. A complete table of baseline characteristics is provided in Table S4A in the Supplementary Appendix. IQR denotes interquartile range.

† The household income in the 3rd quintile was approximately 280,000 to 380,000 Danish kroner (U.S. $39,500 to $53,700).

‡ In each analysis of the outcome-specific cohorts, only the history of the same outcome in a previous pregnancy was included in the propensity score. Thus, for the analysis of spontaneous abortion, only history of spontaneous abortion was included, and for the analysis of stillbirth, only history of stillbirth was included (not history of other outcomes). Percentages were calculated as compared with all the pregnancies in each exposure group.

14% for spontaneous abortion, 42% for preterm birth, 43% for low birth weight, and 2% for small size for gestational age are unlikely to be associated with quadrivalent HPV vaccination. Our analysis of stillbirth included only two cases among pregnancies with vaccine exposure, which makes it impossible to draw clinically meaningful conclusions regarding this outcome on the basis of our data.

Our results are consistent with other evidence that does not indicate that the vaccination of pregnant women with inactivated virus, bacterial,

Table 2. Characteristics of Women Included in the Matched Analyses of Major Birth Defect, Preterm Birth, Low Birth Weight, and Small Size for Gestational Age, According to Time Window of Quadrivalent HPV Vaccination and Vaccination-Exposure Status during Pregnancy.*

Characteristic	0 to 12 Wk of Gestation for Analysis of Major Birth Defect		0 to 37 Wk of Gestation for Analysis of Preterm Birth		0 Wk of Gestation to Birth for Analyses of Small Size for Gestational Age and Low Birth Weight	
	No Vaccine Exposure (N=6660)	Vaccine Exposure (N=1665)	No Vaccine Exposure (N=7096)	Vaccine Exposure (N=1774)	No Vaccine Exposure (N=7072)	Vaccine Exposure (N=1768)
Median no. of days that vaccination occurred after pregnancy onset (IQR)	—	18 (8–29)	—	19 (9–32)	—	19 (9–32)
Age at pregnancy onset — yr	25.9±3.8	25.5±3.4	25.9±3.8	25.5±3.4	25.9±3.8	25.5±3.4
Born in Denmark — %	92.5	92.6	92.5	92.7	92.5	92.6
Married or living with partner — %	68.5	69.5	68.8	69.4	69.2	69.2
Bachelor's degree or higher educational level — %	20.1	18.5	19.1	18.0	19.7	18.0
Household income in 3rd quintile — %	23.8	24.3	23.8	24.0	24.0	24.0
Calendar year of delivery 2012 or 2013 — %	90.4	90.6	90.2	90.5	90.2	90.4
Parity — %						
0	67.9	66.0	68.7	65.8	68.0	65.8
≥1	32.1	34.0	31.3	34.1	32.0	34.1
Same outcome in previous pregnancy — %†	3.4	3.4	2.4	2.5	1.6	1.8
Smoking during pregnancy — %‡	14.8	16.3	15.2	17.0	15.3	17.1
Body-mass index of 18.5 to <25.0 — %‡	61.8	60.4	61.6	60.0	61.6	60.1
Diabetes mellitus — %	3.1	3.5	3.2	3.6	3.5	3.6
Used in vitro fertilization drug — %	2.2	2.6	2.4	2.6	2.5	2.7
No. of hospital admissions in previous year — %						
1 or 2	5.5	6.6	5.7	6.6	5.7	6.6
≥3	11.9	13.0	11.9	13.4	11.8	13.3
No. of outpatient hospital visits in previous year — %						
1 or 2	14.6	15.6	15.1	15.8	14.5	15.9
≥3	24.5	24.6	25.2	24.7	25.1	24.8

No. of emergency hospital visits in past year — %						
1 or 2	12.7	13.6	12.3	13.7	12.8	13.6
≥3	2.1	2.7	2.4	3.0	2.3	2.9
No. of prescriptions filled in previous 6 mo — %						
1 or 2	49.2	49.5	50.1	49.3	49.2	49.2
≥3	27.3	27.4	26.4	27.6	27.3	27.6

* Plus–minus values are means ±SD. Matching was done in a 1:4 ratio on the basis of maternal age, calendar year of pregnancy onset, and propensity score. A complete table of baseline characteristics is provided in Table S4B in the Supplementary Appendix.

† In each analysis of the outcome-specific cohorts, only the history of the same outcome in a previous pregnancy was included in the propensity score. Thus, for the analysis of major birth defect, only history of any birth defect was included, and for the analysis of preterm birth, only history of preterm birth was included (not history of other outcomes). However, for the analyses of low birth weight and small size for gestational age, history of low birth weight and history of small size for gestational age were both included. Percentages were calculated as compared with all the pregnancies in each exposure group.

‡ Data on smoking and body-mass index (the weight in kilograms divided by the square of the height in meters) were available only for women whose offspring were included in the analyses of major birth defect, preterm birth, small size for gestational age, and low birth weight (i.e., women whose pregnancies ended in live birth but not those whose pregnancies ended in stillbirth or abortive outcome).

or toxoid vaccines generally confers a higher risk of adverse pregnancy outcomes than no such vaccination.[24] Our results also confirm and considerably expand on results from previous studies of the quadrivalent HPV vaccine. A pooled analysis of five phase 3 clinical trials of quadrivalent HPV vaccine, including 1796 women who had been randomly assigned to receive quadrivalent HPV vaccine and 1824 women who had been randomly assigned to receive placebo, none of whom were known to be pregnant at the time of vaccination, did not show significant between-group differences in the rates of spontaneous abortion, stillbirth, or birth defects. Because the majority of women who subsequently became pregnant had been vaccinated at least 6 months before the date of conception,[8] the study was unable to evaluate the risks of quadrivalent HPV vaccination during pregnancy directly.[8] After the licensure of the quadrivalent HPV vaccine, a manufacturer-managed pregnancy register was created, but it relied on voluntary reporting. The final study of this pregnancy register included 1752 reports, with follow-up rates of spontaneous abortion and birth defects that were not greater than the expected rates in the general population.[12] However, analysis of data that were based on voluntary reporting can identify only potential risk signals and can neither estimate the risks relative to those in an unexposed population nor rule out risks with certainty.

Our study specifically investigated risks that are associated with vaccination during pregnancy in a large population-based cohort. The use of data from nationwide registers allowed comparison with a control group of women who did not receive vaccination in pregnancy, and the data provided detailed individual-level information on the characteristics of the participants. Furthermore, information on exposure and outcomes were collected in a prospective and independent manner that limited susceptibility to recall and selection bias.

The limitations of our study include the need to rely on the physician-assigned diagnoses recorded in the registry. Misclassification of these outcomes could bias results toward no association with quadrivalent HPV vaccination. However, previous validation studies have indicated a high degree of accuracy of reported diagnoses of spontaneous abortion and birth defects.[20,21] The accuracy of diagnoses of low birth weight, small

Table 3. Association between Exposure to Quadrivalent HPV Vaccination during Pregnancy and Adverse Pregnancy Outcomes.*

Outcome	No Vaccine Exposure		Vaccine Exposure		Measure of Association (95% CI)
	No. of Participants	No. of Events (%)	No. of Participants	No. of Events (%)	
Major birth defect	6660	220 (3.3)	1665	65 (3.9)	1.19 (0.90–1.58)
Spontaneous abortion	1852	131 (7.1)	463	20 (4.3)	0.71 (0.45–1.14)
Preterm birth	7096	407 (5.7)	1774	116 (6.5)	1.15 (0.93–1.42)
Low birth weight	7072	277 (3.9)	1768	76 (4.3)	1.10 (0.85–1.43)
Small size for gestational age	7072	783 (11.1)	1768	171 (9.7)	0.86 (0.72–1.02)
Stillbirth	2004	4 (0.2)	501	2 (0.4)	2.43 (0.45–13.21)

* For the analyses of spontaneous abortion and stillbirth, the reported measures of association are hazard ratios. For the analyses of major birth defect, preterm birth, low birth weight, and small size for gestational age, the reported measures of association are prevalence odds ratios. Differences in baseline characteristics between women who were vaccinated during pregnancy and those who were not were taken into account by means of matching in a 1:4 ratio, according to propensity-matched scores of selected variables, maternal age, and calendar year of pregnancy onset. Gestational age was included as a matching criterion in the analyses of spontaneous abortion and stillbirth.

Table 4. Sensitivity Analyses of the Association between Quadrivalent HPV Vaccination during Pregnancy and Adverse Pregnancy Outcomes.*

Outcome	No Vaccine Exposure		Vaccine Exposure		Measure of Association (95% CI)
	No. of Participants	No. of Events (%)	No. of Participants	No. of Events (%)	
Major birth defect					
With vaccination during gestational wk 4–10	6660	220 (3.3)	431	19 (4.4)	1.35 (0.84–2.18)
Including cases from induced abortions and stillbirths	6744	245 (3.6)	1686	70 (4.2)	1.15 (0.88–1.51)
All outcomes					
According to complete-case analysis					
Spontaneous abortion	1784	110 (6.2)	446	19 (4.3)	0.82 (0.50–1.33)
Major birth defect	6376	210 (3.3)	1594	63 (4.0)	1.21 (0.91–1.61)
Preterm birth	6788	386 (5.7)	1697	108 (6.4)	1.13 (0.90–1.41)
Low birth weight	6764	258 (3.8)	1691	70 (4.1)	1.09 (0.83–1.43)
Small size for gestational age	6764	739 (10.9)	1691	165 (9.8)	0.88 (0.74–1.05)
Stillbirth	1924	6 (0.3)	481	2 (0.4)	1.63 (0.33–8.05)
According to 1:1 matching					
Spontaneous abortion	463	33 (7.1)	463	20 (4.3)	0.70 (0.40–1.21)
Major birth defect	1665	53 (3.2)	1665	65 (3.9)	1.24 (0.85–1.79)
Preterm birth	1774	103 (5.8)	1774	116 (6.5)	1.14 (0.86–1.49)
Low birth weight	1768	78 (4.4)	1768	76 (4.3)	0.97 (0.70–1.34)
Small size for gestational age	1768	184 (10.4)	1768	171 (9.7)	0.92 (0.74–1.15)
Stillbirth	501	1 (0.2)	501	2 (0.4)	2.33 (0.22–25.18)
According to increased number of age categories†					
Spontaneous abortion	1849	111 (6.0)	463	20 (4.3)	0.84 (0.52–1.34)
Major birth defect	6655	219 (3.3)	1665	65 (3.9)	1.19 (0.90–1.58)
Preterm birth	7083	430 (6.1)	1773	116 (6.5)	1.08 (0.88–1.34)
Low birth weight	7059	301 (4.3)	1767	76 (4.3)	1.01 (0.78–1.31)
Small size for gestational age	7059	769 (10.9)	1767	171 (9.7)	0.88 (0.74–1.04)
Stillbirth	2001	3 (0.1)	501	2 (0.4)	3.16 (0.53–18.77)

* For the analyses of spontaneous abortion and stillbirth, the reported measures of association are hazard ratios. For the analyses of major birth defect, preterm birth, low birth weight, and small size for gestational age, the reported measures of association are prevalence odds ratios.
† To evaluate the effect of residual confounding, we increased the number of age categories to include the following: 10 to 15 years, 16 or 17 years, 18 or 19 years, 20 or 21 years, 22 or 23 years, 24 or 25 years, 26 or 27 years, 28 or 29 years, 30 or 31 years, 32 or 33 years, 34 or 35 years, 36 or 37 years, 38 or 39 years, and older than 39 years.

size for gestational age, stillbirth, and preterm birth has not been validated, although the previous validation of gestational-age reports in this cohort suggests that preterm birth is also likely to have been reported accurately.[16,25] Our analyses of birth defects included only live births. However, sensitivity analyses that included data on birth defects in aborted fetuses and stillborn infants showed similar results. We adjusted the analyses for a large number of relevant confounders, but we cannot rule out the possibility of residual confounding. To examine the potential for residual confounding, we performed post hoc sensitivity analyses to evaluate the effect of increased precision in matching and age adjustment; the results were materially unchanged. Finally, because many pregnancy outcomes are rare, our study did not have the statistical power to assess the risks of stillbirth and specific major birth defects associated with quadrivalent HPV vaccination. Larger studies would be needed to address these outcomes.

In conclusion, in this large nationwide study we found that the risks of spontaneous abortion, major birth defect, stillbirth, preterm birth, small size for gestational age, and low birth weight were not significantly higher with quadrivalent HPV vaccination during pregnancy than without vaccination.

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