GYNECOLOGY

Risk of progression of cervical intraepithelial neoplasia grade 2 in human papillomavirus–vaccinated and unvaccinated women: a population-based cohort study

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BACKGROUND: Many countries have implemented active surveillance (ie, leaving the lesion untreated) as an option among younger women with cervical intraepithelial neoplasia grade 2 because regression rates are high and excisional treatment increases the risk for preterm birth in subsequent pregnancies. However, early identification of women at increased risk for progression to cervical intraepithelial neoplasia grade 3 or worse is important to ensure timely treatment. Because women who have received a human papillomavirus vaccine have a lower risk for cervical cancer, they may have a lower risk for progression of untreated cervical intraepithelial neoplasia grade 2 to cervical intraepithelial neoplasia grade 3 or worse.

OBJECTIVE: This study aimed to investigate if women who received a human papillomavirus vaccine and who are undergoing active surveillance for cervical intraepithelial neoplasia grade 2 are less likely to progress to cervical intraepithelial neoplasia grade 3 or worse when compared with women who did not receive the vaccine.

STUDY DESIGN: We conducted a population-based cohort study in Denmark using data from national health registers. We identified all women aged 18 to 40 years who were undergoing active surveillance for cervical intraepithelial neoplasia grade 2 from January 1, 2007, to December 31, 2020. Women with a previous record of cervical intraepithelial neoplasia grade 2 or worse, hysterectomy, or a loop electrosurgical excision procedure were excluded. Exposure was defined as having received ≥1 dose of a human papillomavirus vaccine at least 1 year before the cervical intraepithelial neoplasia grade 2 diagnosis. We used cumulative incidence functions to estimate the risk for progression to cervical intraepithelial neoplasia grade 3 or worse within 28 months using hysterectomy, emigration, and death as competing events. We used modified Poisson regression to calculate crude and adjusted relative risks of progression during the 28-month surveillance period. Results were stratified by age at vaccination and adjusted for index cytology, disposable income, and educational level.

RESULTS: The study population consisted of 7904 women of whom 3867 (48.9%) were vaccinated at least 1 year before a diagnosis of cervical intraepithelial neoplasia grade 2. At the time of cervical intraepithelial neoplasia grade 2 diagnosis, women who were vaccinated were younger (median age, 25 years; interquartile range, 23–27 years) than those who were not (median age, 29 years; interquartile range, 25–33 years). The 28-month cumulative risk for cervical intraepithelial neoplasia grade 3 or worse was significantly lower among women who were vaccinated before the age of 15 years (22.9%; 95% confidence interval, 19.8–26.1) and between the ages of 15 and 20 years (31.5%; 95% confidence interval, 28.8–34.3) when compared with women who were not vaccinated (37.6%; 95% confidence interval, 36.1–39.1). Thus, when compared with women who were not vaccinated, those who were vaccinated before the age of 15 years had a 35% lower risk for progression to cervical intraepithelial neoplasia grade 3 or worse (adjusted relative risk, 0.65; 95% confidence interval, 0.57–0.75), whereas women who were vaccinated between the ages of 15 and 20 years had a 14% lower risk (adjusted relative risk, 0.86; 95% confidence interval, 0.79–0.95). For women who were vaccinated after the age of 20 years, the risk was comparable with that among women who were not vaccinated (adjusted relative risk, 1.02; 95% confidence interval, 0.96–1.09).

CONCLUSION: Women who were vaccinated and who were undergoing active surveillance for cervical intraepithelial neoplasia grade 2 had a lower risk for progression to cervical intraepithelial neoplasia grade 3 or worse during 28 months of follow-up when compared with women who were not vaccinated but only if the vaccine was administered by the age of 20 years. These findings may suggest that the human papillomavirus vaccination status can be used for risk stratification in clinical management of cervical intraepithelial neoplasia grade 2.

Key words: active surveillance, cervical cancer, cervical cancer screening, epidemiology, HPV vaccination, public health, risk stratification

Introduction

Historically, most countries have recommended surgical excision of cervical intraepithelial neoplasia grade 2 (CIN2) to avoid progression to cancer.[1] However, excisional treatment is associated with an increased risk for preterm birth,[2] and the regression rates of CIN2 are high, especially among younger women (ie, 50%–60%).[3,4] Therefore, active surveillance for CIN2 is now an option among younger women in several developed countries, including the United States, England, and the Nordic countries.[1] Although active surveillance likely reduces the risk for overtreatment, some women progress to CIN3 or cervical cancer (CIN3+). Furthermore, studies have demonstrated that active surveillance can cause anxiety, insecurity, and self-doubt.[5,6] Therefore, identification of women at increased risk for progression is of clinical relevance because it may improve clinical

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AJOG at a Glance

Why was this study conducted?

Many countries have implemented active surveillance for cervical intraepithelial neoplasia grade 2 (CIN2) because of high regression rates, but early identification of women at increased risk for progression to CIN3+ is important. We investigated the use of human papillomavirus (HPV) vaccination status for risk stratification of cases with CIN2.

Key findings

HPV-vaccinated women with untreated CIN2 had a significantly lower risk for progression to CIN3+ than unvaccinated women if the vaccine was administered before the age of 20 years.

What does this add to what is known?

HPV-vaccinated women have a lower risk for CIN2+, CIN3+, and cancer, and the greatest impact is observed among women who received the vaccine before the first sexual encounter. We demonstrated that HPV vaccination by the age of 20 years is also associated with a reduced risk for progression among women with untreated CIN2.

counseling and ensure timely treatment. Given that several studies have demonstrated that women who have received a human papillomavirus (HPV) vaccine have a lower risk for CIN2+, CIN3+, and cervical cancer,[7−12] it is important to explore whether HPV vaccination status may be associated with the risk for progression of CIN2.[4]

Therefore, we aimed to investigate if women who have received an HPV vaccine and who are undergoing active surveillance for CIN2 have a lower risk for progression to CIN3+ than women who are not vaccinated.

Material and Methods

Setting

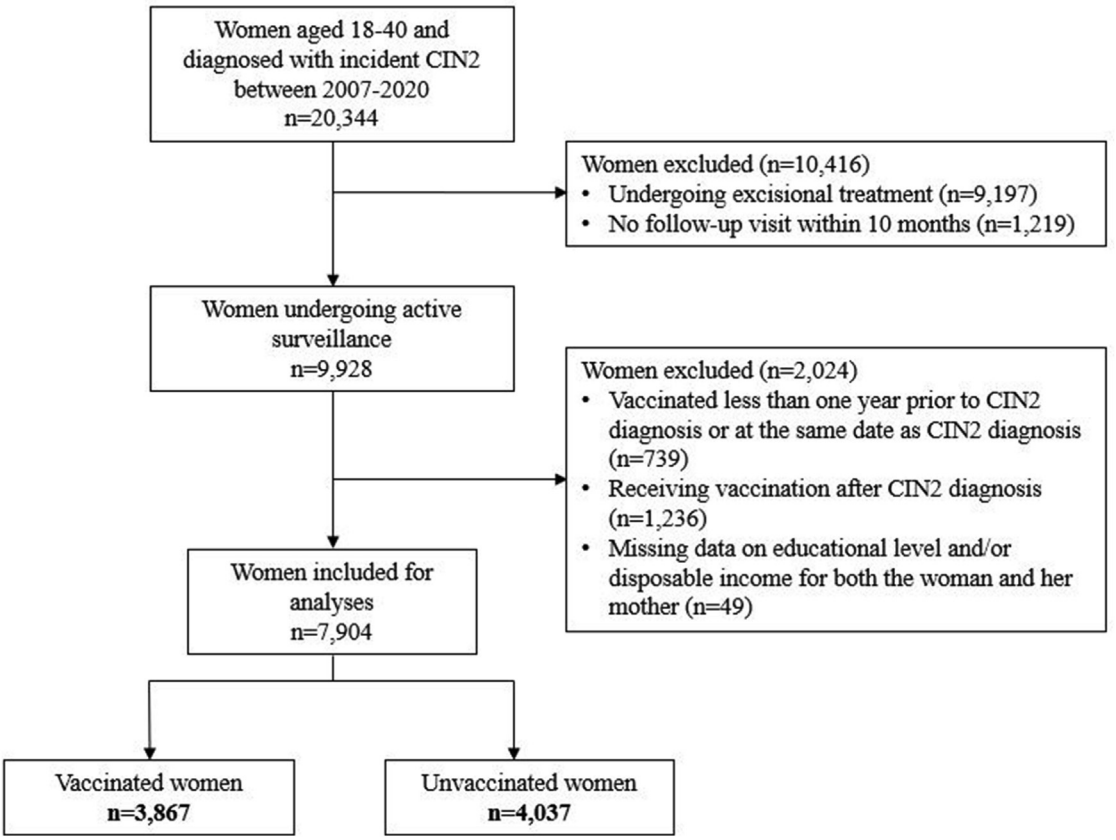
We conducted a nationwide, population-based cohort study in Denmark where all citizens have equal access to medical care at no cost owing to a tax-funded healthcare system. Cervical cancer screening was introduced in Denmark in the mid-1960s,[13] and since 2007, all women aged 23 to 64 years have been invited for screening every 3 to 5 years.[14] Women with an abnormal screening test result may undergo repeated testing or may be referred for a colposcopy depending on the test result. According to national Danish guidelines from 2013, all women referred for a colposcopy should have a minimum of 4 biopsies collected, including random biopsies if no cervical lesion is detected.[15] Women diagnosed with CIN2 are recommended to undergo active surveillance if they are of reproductive age and have a future childbearing desire without any consideration of other risk factors, whereas women of reproductive age without a future pregnancy wish may choose between active surveillance or excisional treatment.[15] Active surveillance consists of a semiannual colposcopy, cytology, and/or cervical biopsies for up to 2 years. Excisional treatment, namely a loop electrosurgical excision procedure (LEEP), is recommended in case of progression or persistent disease after 2 years of follow-up. In Denmark, cold knife conization is not routinely used for excisional treatment of CIN. This study was registered at the Central Denmark Region (748860, 09-29-2021). According to Danish legislation, ethical approval is not required for register-based research.

Study population

At birth or upon immigration, all Danish residents are assigned a unique personal identification number, which allows linkage across all Danish registers at the individual level.[16] The study cohort was identified through the Danish Pathology Register, which is considered complete since 1998.[17] This register contains information on all samples collected for cytology and histopathology in Denmark, including cervical cytology samples, cervical biopsies, LEEP samples, and hysterectomy samples. The study population consisted of all women diagnosed with incident CIN2 from January 1, 2007, to December 31, 2020 (Figure 1). Women were eligible for inclusion if they were aged between 18 and 40 years at the date of the CIN2 diagnosis and if they underwent active surveillance. We included women from the age of 18 years because a colposcopy is recommended for women with postcoital bleeding, irrespective of age, and because opportunistic screening may have been performed in relation to a gynecologic examination for other reasons. We defined active surveillance as having a subsequent record of cervical cytology being performed and/or cervical biopsies being collected at the first visit within 10 months after a CIN2 diagnosis. Women who had a record of undergoing a LEEP at the first visit within 10 months after a CIN2 diagnosis were excluded, as well as women with a previous record of CIN2+, hysterectomy, or LEEP. We also excluded women who were vaccinated less than 1 year before a CIN2 diagnosis and women who were vaccinated after a CIN2 diagnosis. Furthermore, we excluded women for whom information on both personal and maternal educational level and/or disposable income was missing. Women who were excluded because of missing information (n=49; 0.5%) were comparable with those who were included with respect to age and index cytology. However, the majority was not vaccinated (n=44; 89.8%).

Information on a previous CIN2+ diagnosis was obtained through the Danish Pathology Register and the Danish Cancer Register. The Danish Cancer Register was established in 1943 and captures all incident cases of cancer.[18] Information on LEEP and hysterectomies was obtained through the

FIGURE 1
Flowchart of the study population



Flowchart of the study population for the primary outcome (CIN3 or worse).

CIN2, cervical intraepithelial neoplasia grade 2.

Krog. Risk of progression of cervical intraepithelial neoplasia grade 2 by human papillomavirus vaccination status. Am J Obstet Gynecol 2024.

Danish National Patient Register, which was established in 1977 and holds information on all in- and out-patient contacts and procedures since 1995.[19]

Exposure and outcomes

Exposure was defined as having received ≥1 dose of the HPV vaccine at least 1 year before the CIN2 diagnosis. Following the introduction of the HPV vaccine, 3 different vaccine types have been used (quadrivalent, bivalent, and nonavalent). Because of the age of our study population, >95% of the vaccinated women received the quadrivalent vaccine, which was the only 1 available in Denmark from 2006 to 2016.[12] The bivalent and nonavalent vaccines were used in the Danish Childhood Vaccination Program from 2016 to 2017 and from 2017 onward, respectively.[12]

Because of different vaccine regimens in Denmark, information on HPV vaccination status was obtained through 2 registers, namely the Danish National Health Service Register and the Danish National Prescription Register. Free-of-charge HPV vaccination was implemented in the Danish Childhood Vaccination Program in 2009, targeting 12-year-old girls (birth cohorts 1996−1997), and was accompanied by 2 catch-up programs that targeted women born in 1985 to 1995.[7,11,20] For girls and women vaccinated in the childhood vaccination program or the catch-up programs, vaccination was performed by a general practitioner at no cost. Of note, the vaccine was not available free of charge if administered before the age of 12 years. Through the Danish National Health Service Register, which contains data on all services supported by the Danish Health Insurance System since 1990,[21] we identified women who were vaccinated at their general practitioner using service codes 8328, 8329, 8330, 8334, 8335, and 8336. For women not included in any of the programs, self-payment was required to receive the vaccine.[7,22] Through the Danish National Prescription Register, which contains data on all prescription drugs redeemed in pharmacies since 1994,[23] we identified women who had redeemed a prescription of the HPV vaccine (ie, self-payment) using the Anatomical Therapeutical Chemical Code (ATC) J07BM*.

Our primary outcome was the development of CIN3+ (progressive disease) and our secondary outcome was CIN2+ (nonregressive and progressive disease).

Information on the outcomes was obtained through the Danish Pathology Register and the Danish Cancer Register. The size of the study population varied based on the outcome because of our exclusion criteria (Figure 1 and Supplemental Figure). We excluded women who were vaccinated during the follow-up period if the vaccine was administered before an eventual outcome. However, because CIN2+ was a more common outcome than CIN3+, more women in the CIN2+ population developed an outcome before getting vaccinated, leading to a larger study population. We observed no difference in the baseline characteristics between the 2 groups.

Covariates

We obtained information on personal and maternal educational level and disposable income from Statistics Denmark. Educational level was classified as basic if the woman or her mother had ≤10 years of education, as vocational if at least one of them had 11 to 15 years of education, and as higher if at least one of them had >15 years of education. Income was divided into the lowest, middle, and highest tertile.[24] For the analyses, we used the highest educational level and income of the women and their mothers because the women in our study population may not have reached their final education or income level yet. Of note, information on vital status, age, emigration, and identification of the mothers of the included women was obtained through the Civil Registration System. Information on the index cytology was obtained through the Danish Pathology Register. Index cytology was defined as the most recent cytology sample collected within 6 months before a CIN2 diagnosis, and it was classified according to the Bethesda classification system.[25]

Statistical analyses

For our main analysis, we followed women for 28 months after their CIN2 diagnosis to allow the use of the LEEP specimen result among women who underwent excisional treatment after the 2 year follow-up visit. We used the cumulative incidence function (CIF) to estimate the absolute risks of CIN3+ and CIN2+ in vaccinated and unvaccinated women in which hysterectomy, emigration, and death were considered competing events. We used a modified Poisson regression model with robust variances to estimate the crude and adjusted relative risks (RR and aRR, respectively) of CIN3+ and CIN2+ within 28 months using unvaccinated women as reference. The results were stratified by age at HPV vaccination (<15, 15−20, and >20 years), age at CIN2 diagnosis (18−22, 23−29, and 30−40 years), and index cytology (normal, low grade, high grade, and other or missing). Age categories for HPV vaccination were based on the classification used in previous Danish studies because the vaccine effectiveness has been shown to vary with age with the greatest impact observed if administered before the age of 15 years.[10] With respect to age at the CIN2 diagnosis, we found that it was important to separate out women younger than the screening age (<23 years). Women who met screening age were split into 2 groups (23−29 and 30−40 years) because a meta-analysis reported that there was a difference in the regression rates of CIN2 across age groups.[3] We adjusted for index cytology (normal, low grade, high grade, and other or missing), highest personal or maternal disposable income (lowest, middle, or highest), and highest personal or maternal educational level (basic, vocational, or higher). The statistical programs Stata, version 17.0, (StataCorp LLC, 2017, College Station, TX) and SAS, version 9.4, (SAS Institute, Cary, NC) were used for data processing, data management, and statistical analyses.

Results

We identified 20,344 women with incident CIN2, 9928 of whom had undergone active surveillance. We included 7904 (38.9%) women in our analysis for the primary outcome (CIN3+) (Figure 1). Among these, almost half were vaccinated against HPV. When compared with women who were unvaccinated, those who were vaccinated were younger, however, we found no meaningful differences in the index cytology, disposable income, and educational level between women who were vaccinated and those who were not (Table 1). Nearly all vaccinated women (94.5%) received more than 1 dose of the HPV vaccine. One-fifth of the vaccinated cohort received the first vaccine dose before the age of 15 years (17.5%), whereas more than half were vaccinated after the age of 20 years (54.2%).

The cumulative risk of CIN3+ among women who were vaccinated before the age of 15 year (CIF, 22.9%; 95% confidence interval [CI], 19.8−26.1) and among women vaccinated between the ages of 15 and 20 years (CIF, 31.5%; 95% CI, 28.8−34.3) was lower than the risk among women who were not vaccinated (CIF, 37.6%; 95% CI, 36.1−39.1) (Table 2, Figure 2). Thus, when compared with women who were not vaccinated, women who were vaccinated before the age of 15 years had a 35% lower risk for progression to CIN3+ (aRR, 0.65; 95% CI, 0.57−0.75), whereas women who were vaccinated between the ages of 15 and 20 years had a 14% lower risk (aRR, 0.86; 95% CI, 0.79−0.95). For women vaccinated after the of age 20 years, the risk was comparable with that among women who were not vaccinated (aRR, 1.02; 95% CI, 0.96−1.09). There was no difference in risk for CIN3+ between women who were not vaccinated and those who were vaccinated at the age of 21 to 26 years (aRR, 1.02; 95% CI, 0.95−1.10) and at >26 years (aRR, 1.01; 95% CI, 0.86−1.18) (data not shown).

When we stratified by age at CIN2 diagnosis, the results were similar; that is, HPV vaccination before the age of 15 was associated with the lowest risk for progression (Table 2). We were unable to assess risk differences among women diagnosed at an age of 30 years or older because no women had received the vaccine before 20 years of age. With respect to index cytology, we found no difference between women who were vaccinated before the age of 15 years and those who were not vaccinated with a normal cytology (aRR, 0.86; 95% CI, 0.61−1.22). However, for women with either a low- or high-grade cytology,

TABLE 1
Baseline characteristics of Danish women aged 18 to 40 years who underwent active surveillance for CIN2 during 2007 to 2020

Characteristics	Total, n (%)	Unvaccinated, n (%)	Vaccinated, n (%)
Total, n	7904 (100.0)	4037 (100.0)	3867 (100.0)
Age at diagnosis (y), median (IQR)	26 (24–30)	29 (25–33)	25 (23–27)
Age at diagnosis (y)			
18–22	783 (9.9)	279 (6.9)	504 (13.0)
23–29	4738 (59.9)	1870 (46.3)	2868 (74.2)
30–40	2383 (30.2)	1888 (46.8)	495 (12.8)
Age at vaccination (y)			
<15	—	—	677 (17.5)
15–20	—	—	1095 (28.3)
>20	—	—	2095 (54.2)
Vaccine doses			
1	—	—	214 (5.5)
>1	—	—	3653 (94.5)
Time from HPV vaccination to CIN2 diagnosis (y)			
1–4	—	—	1932 (50.0)
5–9	—	—	1596 (41.3)
10–12	—	—	339 (8.8)
Index cytology[a]			
Normal	981 (12.4)	437 (10.8)	544 (14.1)
Low-grade[b]	2826 (35.8)	1418 (35.1)	1408 (36.4)
High-grade[c]	3606 (45.6)	1913 (47.4)	1693 (43.8)
Other[d] or missing	491 (6.2)	269 (6.7)	222 (5.7)
Educational level[e]			
Basic	636 (8.1)	422 (10.5)	214 (5.5)
Vocational	3448 (43.6)	1744 (43.2)	1704 (44.1)
Higher	3820 (48.3)	1871 (46.4)	1949 (50.4)
Disposable income[e]			
Lowest, first	1165 (14.7)	697 (17.3)	468 (12.1)
Middle, second	2643 (33.4)	1378 (34.1)	1265 (32.7)
Highest, third	4096 (51.8)	1962 (48.6)	2134 (55.2)

CIN2, cervical intraepithelial neoplasia grade 2; *HPV*, human papillomavirus; *IQR*, interquartile range.

[a] Up to 6 months before and 7 days after the CIN2 diagnosis; [b] Includes atypical squamous cells of undetermined significance (ASCUS) and low-grade squamous intraepithelial lesion (LSIL); [c] Includes atypical squamous cells that cannot rule out high-grade squamous lesion (ASC-H), atypical glandular cells (AGC), high-grade squamous intraepithelial lesion (HSIL), carcinoma in situ (CIS), adenocarcinoma in situ (AIS), and carcinoma; [d] Includes nonrepresentative samples; [e] Highest among the participant or her mother.

Krog. Risk of progression of cervical intraepithelial neoplasia grade 2 by human papillomavirus vaccination status. Am J Obstet Gynecol 2024.

women who were vaccinated before the age of 15 years had a lower risk for progression to CIN3+ than women who were not vaccinated (aRR, 0.61; 95% CI, 0.47–0.78; and aRR, 0.59; 95% CI, 0.47–0.74, respectively).

Of note, we identified 20 cases (0.25%) of cervical cancer during the follow-up period of 28 months. In the unvaccinated cohort, 15 women (0.37%) developed cancer in comparison with 5 women (0.13%) in the vaccinated cohort. In the vaccinated cohort, all cancer cases were observed among women who were vaccinated after the age of 20 years.

In an ancillary analysis, we estimated the risk for CIN2+ within 28 months

TABLE 2
Risk for CIN3 or worse among women who were HPV vaccinated or unvaccinated and who underwent active surveillance for CIN2, overall and stratified by age at CIN2 diagnosis and index cytology

Characteristics	HPV vaccination status	Total, n	CIN3+ within 28 mo, n (%)	CIF (%) (95% CI)	Crude RR (95% CI)	Adjusted RR (95% CI)[a]
Cohort	Total	7904	2822 (35.7)	35.6 (34.6–36.7)		
	Unvaccinated	4037	1519 (37.6)	37.6 (36.1–39.1)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	677	156 (23.0)	22.9 (19.8–26.1)	0.61 (0.53–0.71)	0.65 (0.57–0.75)
	Vaccinated at age 15–20 y	1095	347 (31.7)	31.5 (28.8–34.3)	0.84 (0.77–0.93)	0.86 (0.79–0.95)
	Vaccinated after age 20 y	2095	800 (38.2)	38.2 (36.1–40.3)	1.01 (0.95–1.09)	1.02 (0.96–1.09)
Age at CIN2 diagnosis						
18–22	Unvaccinated	279	87 (31.2)	31.2 (25.8–36.7)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	302	60 (19.9)	19.5 (15.3–24.2)	0.64 (0.48–0.85)	0.70 (0.53–0.93)
	Vaccinated at age 15–20 y	179	49 (27.4)	26.8 (20.6–33.5)	0.88 (0.65–1.18)	0.96 (0.71–1.30)
	Vaccinated after age 20 y	23	5 (21.7)	21.7 (7.9–39.9)	0.70 (0.31–1.54)	0.72 (0.33–1.57)
23–29	Unvaccinated	1870	691 (37.0)	37.0 (34.8–39.1)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	375	96 (25.6)	25.6 (21.3–30.1)	0.69 (0.58–0.83)	0.73 (0.61–0.88)
	Vaccinated at age 15–20 y	910	296 (32.5)	32.4 (29.4–35.5)	0.88 (0.79–0.98)	0.91 (0.82–1.01)
	Vaccinated after age 20 y	1583	621 (39.2)	39.2 (36.8–41.6)	1.06 (0.97–1.16)	1.08 (1.00–1.18)
30-40	Unvaccinated	1888	741 (39.3)	39.1 (36.9–41.3)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	—	—	—	—	—
	Vaccinated at age 15–20 y	—	—	—	—	—
	Vaccinated after age 20 y	489	174 (35.6)	35.6 (31.4–39.8)	0.91 (0.79–1.03)	0.91 (0.79–1.03)
Index cytology						
Normal	Unvaccinated	437	116 (26.5)	26.5 (22.5–30.8)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	129	30 (23.3)	23.3 (16.4–30.8)	0.88 (0.62–1.24)	0.86 (0.61–1.22)
	Vaccinated at age 15–20 y	151	24 (15.9)	15.9 (10.6–22.2)	0.60 (0.40–0.89)	0.61 (0.41–0.90)
	Vaccinated after age 20 y	264	84 (31.8)	31.8 (26.3–37.5)	1.20 (0.95–1.52)	1.21 (0.96–1.54)
Low grade	Unvaccinated	1418	451 (31.8)	31.7 (29.3–34.2)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	287	56 (19.5)	19.5 (15.2–24.3)	0.61 (0.48–0.79)	0.61 (0.47–0.78)
	Vaccinated at age 15–20 y	404	105 (26.0)	25.7 (21.6–30.1)	0.82 (0.68–0.98)	0.81 (0.68–0.98)
	Vaccinated after age 20 y	717	230 (32.1)	32.1 (28.7–35.5)	1.01 (0.88–1.15)	1.01 (0.89–1.15)
High grade	Unvaccinated	1913	867 (45.3)	45.3 (43.1–47.5)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	213	57 (26.8)	26.3 (20.6–32.3)	0.59 (0.47–0.74)	0.59 (0.47–0.74)
	Vaccinated at age 15–20 y	479	201 (42.0)	41.8 (37.3–46.1)	0.93 (0.82–1.04)	0.94 (0.83–1.05)
	Vaccinated after age 20 y	1001	451 (45.1)	45.1 (42.0–48.1)	0.99 (0.91–1.08)	1.00 (0.92–1.09)
Other or missing	Unvaccinated	269	85 (31.6)	31.2 (25.8–36.8)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	48	13 (27.1)	27.1 (15.5–40.0)	0.86 (0.52–1.41)	0.84 (0.51–1.39)
	Vaccinated at age 15–20 y	61	17 (27.9)	27.9 (17.3–39.4)	0.88 (0.57–1.37)	0.90 (0.58–1.39)
	Vaccinated after age 20 y	113	35 (31.0)	31.0 (22.7–39.6)	0.98 (0.71–1.36)	1.03 (0.73–1.44)

CI, confidence interval; *CIF*, cumulative incidence function; *CIN2*, cervical intraepithelial neoplasia grade 2; *CIN3+*, cervical intraepithelial neoplasia grade 3 or worse; *HPV*, human papillomavirus; *RR*, relative risk.

[a] Adjusted for index cytology (normal, low grade, high grade, other or missing), highest disposable income tertile of the individual or her mother (lowest, middle, highest), and highest educational level of the individual or her mother (basic, vocational, higher).

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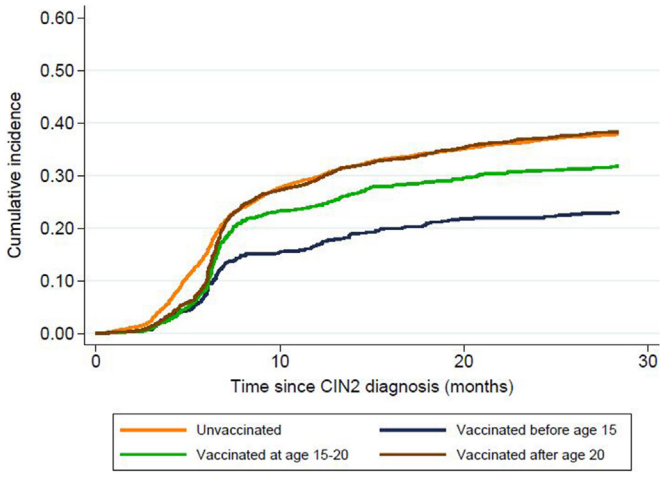
FIGURE 2
Risk for CIN3 or worse by HPV vaccination status



Risk for CIN3 or worse among 7904 women who underwent active surveillance for CIN2 by HPV vaccination status.

CIN3+, cervical intraepithelial neoplasia grade 3 or worse; *HPV*, human papillomavirus.

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after a CIN2 diagnosis. The cumulative risk for CIN2+ among women vaccinated before the age of 15 years (CIF, 36.6%; 95% CI, 33.0−40.3) and women vaccinated between the ages of 15 and 20 years (CIF, 45.7%; 95% CI, 42.7−48.6) was lower than that among women who were not vaccinated (CIF, 54.9%; 95% CI, 53.4−56.4) (Table 3, Figure 3). Women who were vaccinated before the age of 15 years had a 31% lower risk for progression to CIN2+ than women who were not vaccinated (aRR, 0.69; 95% CI, 0.63−0.77), whereas women who were vaccinated between the ages of 15 and 20 years had a 16% lower risk (aRR, 0.84; 95% CI, 0.79−0.90) (Table 3).

Discussion

Principal findings

In this population-based, nationwide cohort study of women who underwent active surveillance for CIN2, we found that HPV vaccination by the age of 20 years was associated with a lower risk for CIN3+ and CIN2+ during 28 months of follow-up. For women who were vaccinated before the age of 15 years, the risk for CIN3+ and CIN2+ was 35% and 31% lower, respectively, when compared with women who were unvaccinated. These findings suggest that HPV vaccination status may be used to identify women at higher risk for progression, thereby enabling risk stratification at the time of CIN2 diagnosis.

Results in the context of what is known

Several studies have demonstrated high regression rates of CIN2, especially among younger women. After 2 years, the regression and progression rates of CIN2 are approximately 50% to 55% and 18%, respectively.[3,26,27] Furthermore, women who received an HPV vaccine have a significantly lower risk for infection with HPV types included in the HPV vaccines and a lower risk for CIN2+, CIN3+, and cervical cancer than women who are unvaccinated.[7−12] However, the vaccine is most effective when administered before exposure, that is, before becoming sexually active.[9,10,28−30] Our results support these studies in that women with CIN2 who were vaccinated before the age of 15 years had a lower risk for progression to CIN3+ than women who were unvaccinated. However, we also observed a lower risk among women who were vaccinated between the ages of 15 and 20 years. For those vaccinated after the age of 20 years, we found no effect of HPV vaccination on the risk for progression. Our results are important because an increasing number of women will have received the HPV vaccine in the childhood vaccination program, that is, before becoming sexually active. Unfortunately, the underlying reasons for the observed findings are unclear because we had no information on the causal HPV types in the CIN2 lesions. One explanation may be that, among women who received an HPV vaccine, CIN2 lesions may be attributed to HPV types not included in the vaccines, types that are less oncogenic and therefore may have a lower tendency to progress. Supporting this, a study observed that CIN lesions among young women who had previously received the quadrivalent or bivalent HPV vaccine were mainly attributed to non-HPV16/18 types.[31] Furthermore, studies have observed that untreated CIN2 lesions associated with HPV16 infection were more likely to progress than lesions attributed to non-HPV16 types.[4,32]

In our study, >95% of the women who were vaccinated had received the quadrivalent HPV vaccine. Consequently, we were unable to stratify by HPV vaccine type. The quadrivalent HPV vaccine has been replaced by the nonavalent HPV vaccine in vaccination programs in many developed countries, thereby potentially increasing the protection against cervical cancer from 70% to 90%.[33] Consequently, risk of progression will likely be lower among women with CIN2 who have received the nonavalent HPV vaccine before the age of 15 years. In addition, the incidence of CIN2 will likely decline in the years to come as a growing proportion of women become vaccinated.[9,10]

Clinical and research implications

Several countries have now adopted active surveillance as an option among younger women with CIN2. However, active surveillance implies that the lesion is left untreated with the potential risk of not detecting prevalent cancers or progression during the surveillance period. To ensure timely treatment and to avoid

TABLE 3
Risk for CIN2 or worse among women who were HPV vaccinated or unvaccinated and who underwent active surveillance for CIN2, overall and stratified by age at CIN2 diagnosis and index cytology

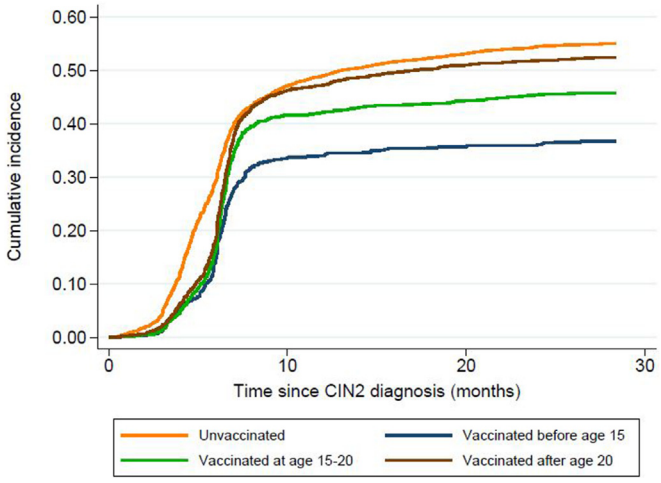
Characteristics	HPV vaccination status	Total, n	CIN2+ within 28 mo, n (%)	CIF (%) (95% CI)	Crude RR (95% CI)	Adjusted RR (95% CI)[a]
Cohort	Total	8100	4172 (51.5)	51.5 (50.4–52.6)		
	Unvaccinated	4233	2326 (55.0)	54.9 (53.4–56.4)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	677	248 (36.6)	36.6 (33.0–40.3)	0.67 (0.60–0.74)	0.69 (0.63–0.77)
	Vaccinated at age 15–20 y	1095	500 (45.7)	45.7 (42.7–48.6)	0.83 (0.77–0.89)	0.84 (0.79–0.90)
	Vaccinated after age 20 y	2095	1098 (52.4)	52.4 (50.3–54.5)	0.95 (0.91–1.00)	0.96 (0.91–1.01)
Age at CIN2 diagnosis						
18–22	Unvaccinated	303	162 (53.5)	53.5 (47.7–58.9)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	302	102 (33.8)	33.8 (28.5–39.1)	0.63 (0.52–0.76)	0.67 (0.55–0.81)
	Vaccinated at age 15–20 y	179	72 (40.2)	40.2 (33.0–47.3)	0.75 (0.61–0.93)	0.79 (0.64–0.97)
	Vaccinated after age 20 y	23	6 (26.1)	26.1 (10.6–44.7)	0.49 (0.24–0.98)	0.49 (0.25–0.98)
23–29	Unvaccinated	1995	1096 (54.9)	54.9 (52.7–57.1)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	375	146 (38.9)	38.9 (34.0–43.8)	0.71 (0.62–0.81)	0.73 (0.64–0.83)
	Vaccinated at age 15–20 y	910	426 (46.8)	46.8 (43.5–50.0)	0.85 (0.79–0.92)	0.87 (0.80–0.94)
	Vaccinated after age 20 y	1583	839 (53.0)	53.0 (50.5–55.4)	0.96 (0.91–1.03)	0.98 (0.92–1.04)
30–40	Unvaccinated	1935	1068 (55.2)	55.1 (52.9–57.3)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	—	—	—	—	—
	Vaccinated at age 15–20 y	—	—	—	—	—
	Vaccinated after age 20 y	489	253 (51.7)	51.7 (47.2–56.1)	0.94 (0.85–1.03)	0.94 (0.86–1.04)
Index cytology						
Normal	Unvaccinated	456	197 (43.2)	43.2 (38.6–47.7)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	129	43 (33.3)	33.3 (25.4–41.5)	0.77 (0.59–1.01)	0.77 (0.59–1.00)
	Vaccinated at age 15–20 y	151	50 (33.1)	33.1 (25.8–40.6)	0.77 (0.60–0.98)	0.77 (0.60–0.99)
	Vaccinated after age 20 y	264	123 (46.6)	46.6 (40.5–52.5)	1.08 (0.91–1.27)	1.08 (0.91–1.28)
Low grade	Unvaccinated	1488	754 (50.7)	50.7 (48.1–53.2)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	287	93 (32.4)	32.4 (27.1–37.9)	0.64 (0.54–0.76)	0.63 (0.53–0.76)
	Vaccinated at age 15–20 y	404	161 (39.9)	39.9 (35.1–44.6)	0.79 (0.69–0.90)	0.78 (0.69–0.89)
	Vaccinated after age 20 y	717	327 (45.6)	45.6 (41.9–49.2)	0.90 (0.82–0.99)	0.90 (0.82–0.99)
High grade	Unvaccinated	2011	1242 (61.8)	61.8 (59.6–63.8)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	213	95 (44.6)	44.6 (37.8–51.1)	0.72 (0.62–0.84)	0.72 (0.62–0.84)
	Vaccinated at age 15–20 y	479	265 (55.3)	55.3 (50.8–59.7)	0.90 (0.82–0.98)	0.90 (0.83–0.99)
	Vaccinated after age 20 y	1001	595 (59.4)	59.4 (56.3–62.4)	0.96 (0.90–1.02)	0.97 (0.91–1.03)
Other or missing	Unvaccinated	278	133 (47.8)	47.5 (41.5–53.2)	1.0 (ref)	1.0 (ref)
	Vaccinated before age 15 y	48	17 (35.4)	35.4 (22.3–48.8)	0.74 (0.50–1.11)	0.72 (0.48–1.08)
	Vaccinated at age 15–20 y	61	24 (39.3)	39.3 (27.2–51.3)	0.82 (0.59–1.15)	0.81 (0.58–1.14)
	Vaccinated after age 20 y	113	53 (46.9)	46.9 (37.5–55.8)	0.98 (0.78–1.24)	0.98 (0.77–1.25)

CI, confidence interval; *CIF*, cumulative incidence function; *CIN2*, cervical intraepithelial neoplasia grade 2; *CIN2+*, cervical intraepithelial neoplasia grade 2 or worse; *HPV*, human papillomavirus; *RR*, relative risk.

[a] Adjusted for index cytology (normal, low grade, high grade, other or missing), highest disposable income tertile of individual or her mother (lowest, middle, highest), and highest educational level of individual or her mother (basic, vocational, higher).

Krog. Risk of progression of cervical intraepithelial neoplasia grade 2 by human papillomavirus vaccination status. Am J Obstet Gynecol 2024.

FIGURE 3
Risk for CIN2 or worse by HPV vaccination status



Risk for CIN2 or worse among 8100 women who underwent active surveillance for CIN2 by HPV vaccination status.

CIN2+, cervical intraepithelial neoplasia grade 2 or worse; *HPV*, human papillomavirus.

Krog. Risk of progression of cervical intraepithelial neoplasia grade 2 by human papillomavirus vaccination status. Am J Obstet Gynecol 2024.

progression to cancer, it is important to identify risk markers that may accurately identify women at increased risk. As demonstrated in the current study, HPV vaccination status may be used for risk stratification seeing that women who were vaccinated before the age of 15 years had a 35% lower risk for progression. Furthermore, 0.13% of the vaccinated cohort developed cancer during 28 months of follow-up, and all cancer cases were observed among women vaccinated after the age of 20 years. This information is important for clinical management because women and their healthcare providers would want to know the risks associated with abstaining from excisional treatment.

Recent studies have explored the use of biomarkers for risk stratification, such as HPV genotyping and host-cell DNA methylation.[4,34,35] A Swedish study reported that HPV16 was associated with a higher risk for progression than non-HPV16 types,[4] whereas a study from the Netherlands reported a lower likelihood of regression among women with hypermethylation of certain host-cell genes when compared with those without.[34] Thus, with better knowledge on individual-level risk factors, clinicians will be able to provide better counseling to women diagnosed with CIN2 in the future. In addition, information on risk for progression is often used to identify eligibility criteria for active surveillance. For example, some countries do not allow active surveillance among women who are positive for HPV16, whereas other countries preclude women with high-grade cytology.[1] Because the risk for progression was lower among women who were vaccinated before the age of 15 years, irrespective of a low-grade or high-grade index cytology or age at CIN2 diagnosis, HPV vaccination status may be used as an eligibility criterion in settings in which information on vaccination status is reliable.

Strengths and limitations

The strength of this study is the availability of individual-level data covering the entire Danish population, thereby enabling identification of all Danish women diagnosed with incident CIN2 during the study period. Furthermore, we were able to obtain information on screening history, HPV vaccination status, and personal and maternal socioeconomic status. According to national Danish guidelines, all women should have a minimum of 4 biopsies collected during the colposcopy,[15] and because of this, we expect that detection bias would be of minor importance. In addition, risk for detection bias is supposed to be similar among women who were vaccinated and those who were not, and therefore misclassification would likely be nondifferential.

With respect to limitations, we had limited information on potential confounding variables, such as smoking, immunosuppressive conditions, and age at sexual debut, and we therefore cannot rule out residual confounding. Second, we cannot rule out selection bias because we only included women who were managed with active surveillance. Therefore, we cannot be sure that our results also apply to those who underwent immediate excisional treatment, because these groups are not completely comparable. Third, we had no information on the HPV types that caused the cervical lesions. Fourth, we had no information on the stage of disease among those who developed cancer.

Conclusion

In this study, HPV vaccination by the age of 20 years was associated with a 14% to 35% lower risk for CIN3+ among women who underwent active surveillance for CIN2, and the greatest protection was achieved when the vaccine was administered before the age of 15 years. Therefore, HPV vaccination status may be used for risk stratification of CIN2. ■

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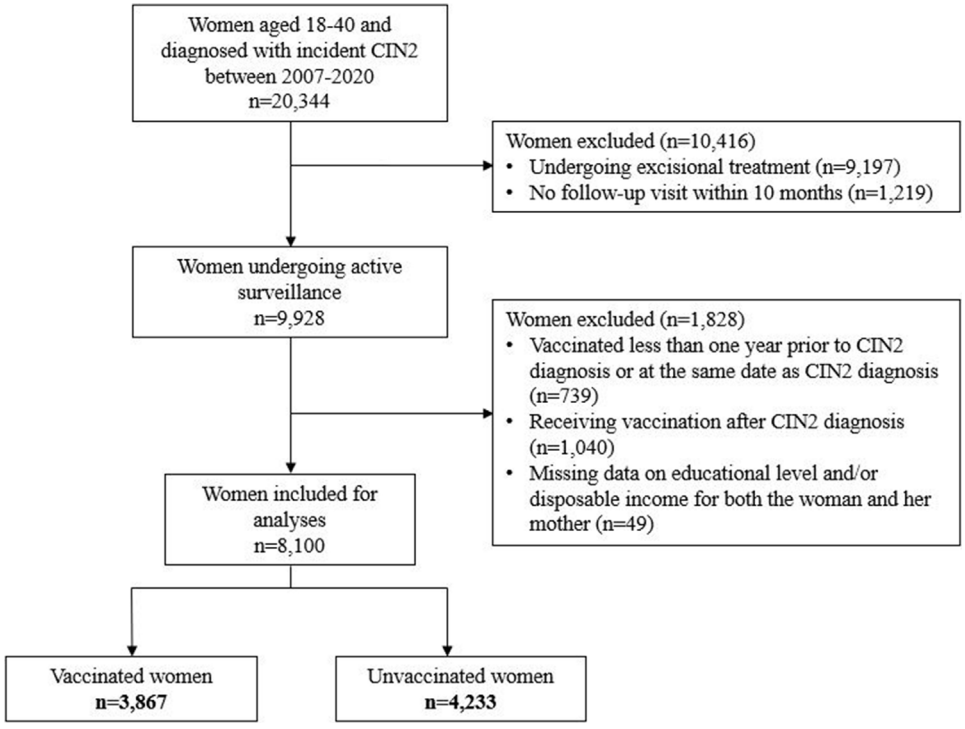
SUPPLEMENTAL FIGURE
Flowchart of the study population for the secondary outcome



Flowchart of the study population for the secondary outcome (CIN2 or worse).

CIN2, cervical intraepithelial neoplasia grade 2.

Krog. Risk of progression of cervical intraepithelial neoplasia grade 2 by human papillomavirus vaccination status. Am J Obstet Gynecol 2024.