Strongly Decreased Risk of Genital Warts After Vaccination Against Human Papillomavirus: Nationwide Follow-up of Vaccinated and Unvaccinated Girls in Denmark

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Background. A reduction in the incidence of genital warts (GWs) is one of the first markers of the effectiveness of vaccination against human papillomavirus (HPV) at the population level. The aim of this cohort study was to use individual information on HPV vaccination status to assess the effect on risk of GWs.

Methods. Population-based registries were used to identify all girls in the birth cohorts 1989–1999 in Denmark, and information about HPV vaccination was obtained for the period 2006–2012. The cohort was linked to incident cases of GWs, and vaccinated and unvaccinated girls were compared using Cox proportional hazards models.

Results. A total of 248 403 girls were vaccinated. The relative risk of GWs among girls who had received at least 1 dose of vaccine compared with unvaccinated girls was 0.12, 0.22, 0.25, and 0.62 for those born in 1995–1996, 1993–1994, 1991–1992, and 1989–1990, respectively (P for trend <.0001). No GWs occurred among vaccinated girls in the youngest birth cohort (1997–1999).

Conclusions. The strong, highly significant reduction in the occurrence of GWs among vaccinated girls indicates an early and marked population effect of the national HPV vaccination program and may forecast a similar effect on cervical precancerous lesions.

Keywords. genital warts; condyloma acuminata; human papillomavirus; vaccination; epidemiology.

Human papillomavirus (HPV) is one of the most common sexually transmitted infections worldwide. The lifetime risk of acquiring an HPV infection is approximately 75% [1], but most infections are transient and without symptoms. More than 120 HPV types have been identified, and about 40 have a special affinity for the anogenital epithelium.

Two vaccines against HPV have been licensed. Both are active against HPV16 and 18, the types responsible for 70% of all cervical cancers and a large proportion of other anogenital cancers and head-and-neck cancers [2]. The quadrivalent HPV vaccine (licensed in Denmark in October 2006) also provides protection against HPV6 and 11, which cause 90% of genital warts (GWs) [3]. With the primary aim of preventing cervical cancer, several countries are now offering HPV vaccination to young girls in national vaccination programs. Denmark has done so through its national childhood vaccination program, which provides vaccination against 10 serious infectious diseases [4]. The quadrivalent HPV vaccine was included in this program in January 2009 for girls aged 12 years (birth cohorts 1996–1999), with catch-up vaccination for girls aged 13–15 years (birth cohorts 1993–1995) since October 2008. General practitioners administer these vaccinations, and HPV vaccine acceptance has been high (with coverage rates between 87% and 90% for at least 1 dose) [5]. Girls and women born
before 1993 have had to pay for HPV vaccination, and the coverage has been much lower (<30% for at least 1 dose).

A reduction in the incidence of GWs is one of the first markers of the effectiveness of HPV vaccination at a population level, as they develop over a few months, whereas precancerous lesions and cancer usually develop over several years. In randomized clinical trials (FUTURE I and II), the quadrivalent HPV vaccine reduced the incidence of HPV6-, 11-, 16-, and 18-related GWs by 99% in a per protocol susceptible population (women given 3 vaccinations who were seronegative on day 1 and DNA negative on day 1 through month 7 to the respective HPV type) and by 79% in an intent-to-treat population (women with past or current HPV exposures and those naive to HPV who received >1 vaccination) [3]. Subsequent ecological studies reported a decline in the occurrence of GWs in specific age groups following licensure of the quadrivalent HPV vaccine, indicating a population effect of HPV vaccination [6–9]. However, individual vaccination status was not known in these studies and therefore a direct demonstration of the effect of HPV vaccination on GW occurrence has until now not been possible, and so far no studies linking nationwide, individual information on HPV vaccination to subsequent risk of GWs have been published.

With one of the highest HPV vaccine coverage rates in the world (88% of birth cohorts 1993–1999 have received at least 1 and 80% all 3 vaccinations by 24 February 2013) [5] and the ability to identify and link registry information on all residents, Denmark is an ideal country to assess population effects of HPV vaccination. The aim of this study was therefore to assess the effect of HPV vaccination on subsequent risk of GWs in the general Danish population from individual information on HPV vaccination status.

METHODS

Data Sources and Linkage of Data Between Nationwide Registers

In Denmark, each resident is allocated a unique personal identification number comprising information on sex and date of birth. The numbers are registered in the computerized Civil Registration System and are used as identifiers in all national registries, allowing accurate linkages between registries. From this database, we identified all female Danish residents born between 1961 and 1999 and retrieved their date of birth and, if applicable, date of emigration or death. Boys and men were not included, as extremely few have been vaccinated (<1% of birth cohorts 1989–1999).

To categorize the girls and women according to HPV vaccination status, the cohort was linked to the National Health Insurance Service Register and the Prescription Registry. The Health Insurance Service Register holds information on the activities of health professionals paid by the public healthcare system, such as general practitioners and practicing medical specialists. Using the service codes for quadrivalent HPV vaccination (8328, 8329, and 8330) we retrieved information on all girls vaccinated against HPV in the children’s vaccination and catch-up program since the initiation of national HPV vaccination. In order to identify nonprogram vaccinations, we identified in the Prescription Registry girls and women who had purchased HPV vaccine with a prescription since licensing of the quadrivalent HPV vaccine. This was done by extracting the Anatomical Therapeutic Chemical (ATC) category J07BM01 (papillomavirus vaccine, human types 6, 11, 16, 18).

We obtained information on dates of vaccination for all vaccinated girls and women. We included only birth cohorts 1989–1999, which had a vaccine coverage rate (at least 1 dose) >10%. Among the excluded birth cohorts (1961–1988), only the cohorts 1983–1988 had coverage rates >5%; the vaccine coverage rate was <1% in the birth cohorts 1961–1970. Vaccinated and unvaccinated girls were followed for the occurrence of incident GWs by linkage to the National Patient Register, which contains information on all hospital admissions and outpatient visits since 1977 and 1995, respectively. Data on GWs could be retrieved until 22 May 2012. Diagnoses were classified according to the International Classification of Diseases, Tenth Revision, and we used code A63.0 to identify incident cases of GWs. The girls were followed until first episode of GWs, date of emigration, or death or 22 May 2012, whichever came first. Girls with a GW diagnosis before licensure of the HPV vaccine (October 2006) were excluded (n = 197). The analyses were also performed with the inclusion of girls with GWs before vaccine licensure and of all episodes of GWs, but as these inclusions changed the estimates minimally, only the results from the most stringent analysis are presented.

To identify potential differences in the frequency of cervical cancer screening between vaccinated and unvaccinated girls—and thereby the likelihood of coincidental detection of GWs—we linked the cohort to the Pathology Data Bank, to which all departments of pathology report pathology data. From this register, we retrieved information on all cervical cytology and histological examinations. The study was approved by the Danish Data Protection Agency.

Statistical Analysis

We compared the occurrence of incident GWs among unvaccinated and vaccinated girls in a Cox proportional hazards model with age as the underlying time scale. In this analysis, we stratified on birth cohort and allowed the effect of vaccination to be different in each stratum. Girls were considered vaccinated 28 days after the first registered date of vaccination (start of case-counting) and were thus unvaccinated until this date. Each girl contributed person-time to each state (vaccinated/unvaccinated), corresponding to the time spent in each state. To assess how
sensitive our results were for the selected time interval between first vaccination and time to counting of GW cases for vaccinated girls (ie, when unvaccinated girls changed to having a status as vaccinated), we also performed analyses with the start of case counting ranging from 7 days after the first vaccination and up until the second dose was given (60 days). Cervical cancer screening rates were calculated for vaccinated and unvaccinated girls, respectively. The statistical analyses were performed with the statistical software R, version 2.15.1 [10].

RESULTS

Identification of the Cohort and Vaccination Status
We identified a total of 399,967 girls born between 1989 and 1999. Of these, 248,800 (62.2%) girls were vaccinated with the quadrivalent HPV vaccine between 1 October 2006 and 22 May 2012—214,904 (86.4%) in the children’s vaccination or catch-up program, and 33,896 (13.6%) via self-payment. We excluded 63 vaccinated girls and 134 unvaccinated girls who had GWs before vaccine licensure. An additional 334 vaccinated girls were registered as unvaccinated because they developed GWs before vaccine licensure. An additional 33,896 (13.6%) via self-payment. We also analyzed risk by age at vaccination and found a similar pattern (data not shown). When changing the time interval for the cohorts 1993–1994, 3.0 and 5.5 for cohorts 1995–1996, and 0.0 and 2.8 for cohorts 1997–1999 for vaccinated and unvaccinated girls, respectively.

In the main analyses where age was taken into account, we considered the relative risk of GWs among vaccinated girls (ie, having received at least 1 dose) compared to unvaccinated girls. The risk was significantly decreased in vaccinated girls, and as shown in Table 2 the risk varied between 0.12 (95% confidence interval [CI], 0.04–0.36, \( P < .001 \)) in girls born during 1995–1996 and 0.25 (95% CI, 0.19–0.32, \( P < .001 \)) in girls born during 1991–1992, 29.4 and 34.6 for cohorts 1993–1994, 3.0 and 5.5 for cohorts 1995–1996, and 0.0 and 2.8 for cohorts 1997–1999 for vaccinated and unvaccinated girls, respectively.

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Risk of Genital Warts
The median follow-up was 3.1 years (range, 0–5.5 years) for vaccinated and 3.5 years (range, 0–5.6 years) for unvaccinated girls. During this time, GWs were diagnosed in 229 of 248,403 girls who received at least 1 dose and in 2241 of 151,367 girls who were not vaccinated by the end of follow-up. The crude incidence rates of GWs in birth cohorts 1989–1990 were 256.0 per 100,000 person-years for vaccinated and 385.9 per 100,000 person-years for unvaccinated girls. The corresponding figures were 87.5 and 264.7 per 100,000 person-years for cohorts 1991–1992, 29.4 and 34.6 for cohorts 1993–1994, 3.0 and 5.5 for cohorts 1995–1996, and 0.0 and 2.8 for cohorts 1997–1999 for vaccinated and unvaccinated girls, respectively.

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Table 1. Human Papillomavirus (HPV) Vaccination Status of Girls From Denmark Included in the Present Study (Quadrivalent HPV Vaccine, 1 October 2006–22 May 2012)

<table>
<thead>
<tr>
<th>Birth Cohort</th>
<th>Years (95% Tolerance Interval)</th>
<th>Total (No.)</th>
<th>Unvaccinated (No.)</th>
<th>Vaccinated (at Least 1 Dose, No. (%))</th>
</tr>
</thead>
<tbody>
<tr>
<td>1989–1990</td>
<td>18.7 (16.8–21.9)</td>
<td>78,691</td>
<td>67,495</td>
<td>11,196 (14)</td>
</tr>
<tr>
<td>1991–1992</td>
<td>16.6 (15.0–19.2)</td>
<td>74,547</td>
<td>54,711</td>
<td>19,836 (27)</td>
</tr>
<tr>
<td>1993–1994</td>
<td>14.9 (13.9–16.2)</td>
<td>72,581</td>
<td>87,12</td>
<td>63,869 (88)</td>
</tr>
<tr>
<td>1997–1999</td>
<td>12.2 (11.7–13.5)</td>
<td>10,272</td>
<td>1312</td>
<td>89,606 (87)</td>
</tr>
</tbody>
</table>

Table 2. Risk of Genital Warts Among Girls Vaccinated Against Human Papillomavirus Types 6, 11, 16, and 18 (at Least 1 Dose) Versus Unvaccinated Girls Stratified by Birth Cohort, 1 October 2006–22 May 2012

<table>
<thead>
<tr>
<th>Birth Cohort</th>
<th>Hazard Ratio</th>
<th>95% CI</th>
<th>( P ) Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1989–1990</td>
<td>0.62</td>
<td>.50–.76</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>1991–1992</td>
<td>0.25</td>
<td>.19–.32</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>1993–1994</td>
<td>0.22</td>
<td>.15–.33</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>1995–1996</td>
<td>0.12</td>
<td>.04–.36</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>1997–1999</td>
<td>n.e.</td>
<td></td>
<td>&lt; .0001</td>
</tr>
</tbody>
</table>

A Cox proportional hazards model was used with age as the underlying time scale. Stratification on birth cohort. Unvaccinated girls are reference. Abbreviations: CI, confidence interval; n.e., not estimable (no events yet among vaccinated).

\( a \) \( P \) value for trend.
We found a significantly decreased risk of GWs in young girls after vaccination with quadrivalent HPV vaccine. The strong impact of HPV vaccination was observed at population level shortly after initiation of a national HPV vaccination program. Earlier indications of a rapid onset of effect of quadrivalent HPV vaccination have come primarily from ecological studies. In Australia, where vaccination coverage has reached a similar level as in Denmark, Read and colleagues [11] showed a marked decrease in the proportion of young girls with GWs attending a sexual health center, beginning immediately after implementation of a national school and community HPV vaccination program. Similar findings were reported from Germany and the United States [9, 12], and also from Denmark and Sweden, where the national incidences of GWs were assessed from population-based registries [6, 7]. In general, ecological studies are limited by having no information on changes in other risk factors for the disease in question, and studies of the proportion of sexually transmitted infections diagnosed in a given sexual health center are limited by changes in the frequency of other sexually transmitted diseases. Therefore it is essential that the results of these ecological studies are now finally confirmed by this study where there is a direct linkage between vaccination status and subsequent occurrence of GWs on the individual level.

Our findings provide hope for a similar early effect on pre-cancerous cervical lesions, but the effect on GWs in itself should not be neglected. Although GWs are not malignant, they represent a substantial burden to both the individual and to society. In the United States, the cost of treating GWs has been estimated to be between US$520 and US$735 per episode of care, and in Europe between US$241 and US$491 (year 2009 values) [13], so the observed strong and early effect of HPV

**Figure 1.** Risk of genital warts among girls vaccinated against human papillomavirus types 6, 11, 16, and 18 (at least 1 dose) versus unvaccinated girls according to start of case counting (time interval between first vaccination and initiation of time to counting of genital wart cases for vaccinated girls). Hazard ratio (Cox proportional hazards model with age as the underlying time scale; y-axis) versus start of case counting (x-axis). Dashed lines indicate 95% confidence limits. Stratified on birth cohorts.
vaccination on GWs is clearly of great importance. As a result of the high vaccine coverage achieved among girls in Denmark (birth cohorts 1993–1998), it could be hypothesized that boys in the same and some older age groups could also benefit from the substantial immunization of girls, resulting in a subsequently reduced burden of GWs.

The risk of GWs was lower for vaccinated compared to unvaccinated girls in all birth cohorts, but the risk reduction was most pronounced among the younger birth cohorts. We cannot rule out that this is because of biological differences occurring with age that make it better to be vaccinated at a younger age. However, as women in the older birth cohorts are not covered by the vaccination program and therefore would have to pay for the vaccination themselves, we observe much lower coverage rates in these cohorts, and consequently the vaccinated women in these older birth cohorts are most likely not representative for the respective birth cohort. This may imply self-selection bias in the oldest birth cohorts, with women at high risk being more likely to be vaccinated. We chose to start the case counting of GWs among the vaccinated women 28 days after first vaccination. In this way, some of the prevalent infections would be excluded from the analyses, but because the incubation time of GWs is often longer [14, 15], some cases may still be caused by prevalent HPV infection, underestimating the effect of the vaccine. The same phenomenon was seen in an intent-to-treat population of quadrivalent vaccine– and placebo-vaccinated girls, where no significant difference in the incidence of HPV6/11/16/18-related cervical intraepithelial neoplasia grade 3/adenocarcinoma in situ was found in the initial months of the trial, presumably because of prevalent infections, which cleared as time passed [16].

To test whether our risk estimates were affected by factors other than HPV vaccination, we also analyzed differences in cervical cancer screening. Because GWs can be difficult to detect due to both the natural anatomy of women and the sometimes symptomless nature of the warts, screening against cervical cancer is one way in which GWs can be coincidentally detected. Differences in screening rates between vaccinated and unvaccinated girls could therefore influence the relative risks of GWs. However, in this study, screening rates were higher for vaccinated than for unvaccinated girls, so the frequency of screening does not contribute to the decreased risk of GWs found in vaccinated girls.

The strengths of the study were that we were able to identify virtually all girls and women vaccinated against HPV types 6, 11, 16, and 18 in Denmark. Moreover, due to the unique personal identification numbers and the nationwide registries, we had virtually no loss to follow-up. The study was limited by the fact that we could identify only GWs diagnosed at hospitals and outpatient clinics, so the results may not be generalizable to diagnoses by general practitioners and practicing specialists. It seems unlikely, however, that these diagnoses would be different.

Given the population-based design and the individual source of the data, this study adds accurate and valid evidence to the past ecological approach on the effectiveness of HPV vaccination in the occurrence of GWs. The study shows that the effect of the quadrivalent HPV vaccine and the Danish national HPV vaccination program on the risk of GWs is already apparent and substantial. This rapid and marked effect on risk of GWs may suggest that a similar prompt reduction in HPV16/18-associated precancerous lesions is likely to occur.

Notes

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All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

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