Efficacy of a quadrivalent HPV (types 6, 11, 16, 18) L1 VLP vaccine against cervical intraepithelial neoplasia grades 1–3 and adenocarcinoma in situ
A combined analysis

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Abstract
There are solid research data to support the construction of a quadrivalent (HPV6, 11, 16 and 18) L1 VLP vaccine. Accordingly, the two HR–HPV types (16 and 18) cover around 70% of all CCs, 50–70% of high-grade CIN lesions, 70% of other HPV-related genital cancers, and some 25% of low-grade CIN lesions. Similarly, in males, 70% of all anal cancers are caused by HPV16 and HPV18, as are 70% of all AIN2/3 lesions. There are several potential surrogate endpoint markers that can be used in evaluating the efficacy of prophylactic HPV vaccines in prevention of CC. The most important of these surrogate markers is CIN2/3 (CIS), because (a) it is clearly a CC precursor, (b) its prompts rapid treatment, and (c) eradication of CIN2/3 leads to alleviation of CC burden. The combined analysis of the clinical GARDASIL 

1 trials show that the burden of HPV-related disease is significant. Accordingly, 27% of enrolled subjects were either seropositive or PCR-positive to HPV6, 11, 16 or 18. Altogether, 8.7% of placebo recipients developed CIN during the follow-up period, of which a significant proportion was due to HPV6, 11, 16 or 18. Importantly, GARDASIL is 100% effective in preventing CIN3 and AIS as well as CIN2 in the per-protocol population. In the modified-intention-to-treat population, vaccine efficacy against CIN3 and AIS was also 100% (95% CI 91–100). In all these studies, GARDASIL has been well-tolerated. It can be anticipated that this vaccine will lower a woman’s life-time risk for developing CC and substantially reduces the morbidity, mortality and clinical costs of HPV-related disease.

Keywords: Human papillomavirus; CIN; Adenocarcinoma in situ (AIS); HPV vaccine; Quadrivalent; VLP; GARDASIL; Phase III clinical trials; Cervical cancer; Life-time risk; Combined analysis

1. Background on HPV and Merck’s quadrivalent HPV vaccine program
Cervical cancer (CC) is a major public health problem, second only to breast cancer in terms of global disease burden. Taken worldwide, HPV is estimated to be related to 0.5 million annual new cases of CC [1], estimated 10 million cases of high-grade CIN [2], 30 million low-grade CIN [2], as well as 30 million genital warts (GW) [3]. In addition, it has been estimated that there are 300 million new cases of HPV infections without clinically detectable (cytological) abnormalities [2]. The life-time risk of mucosal HPV infections has been estimated to be at least 70% [4,5]. Most importantly, oncogenic HPV types seem to be involved in the development of 99.7% of all CCs [6].

2. Rational for a quadrivalent HPV (types 6, 11, 16 and 18) L1 VLP vaccine
There are solid research data to support the construction of a quadrivalent (HPV 6, 11, 16 and 18) L1 VLP vaccine [1–6]. Accordingly, the two HR–HPV types (16 and 18) cover around 70% of all CCs, 50–70% of high-grade CIN lesions, 70% of other HPV-related genital cancers, and some 25% of...
low-grade CIN lesions. Similarly, in males, 70% of all anal cancers are caused by HPV 16 and HPV 18, as are 70% of all AIN2/3 lesions. Furthermore, there are other HPV-associated cancers in males, and the potential for the virus transmission to female sexual partners must be considered as well.

Concerning the HPV types 6 and 11, these are involved in 5–25% of low-grade CIN lesions, and 90% of all GWs. Transmission of the virus from an infected mother to her child can cause recurrent respiratory papillomatosis (RRP). In the males, we should again consider the transmission of these viral types to women, with the above consequences; 90% of GWs and the potential for RRP. Thus, these four HPV types are responsible for a major disease burden worldwide.

3. Surrogate endpoints for evaluation of prophylactic HPV vaccines

According to Pagliusi and Aguado, there are several potential surrogate endpoint markers that can be used in evaluating the efficacy of prophylactic HPV vaccines in prevention of CC [7]. The most important of these surrogate markers is CIN2/3 (CIS), because (a) it is clearly a CC precursor, (b) it prompts rapid treatment, and (c) eradication of CIN2/3 leads to alleviation of CC burden [7]. The process of developing a new vaccine proceeds from the proof-of-concept, to evaluation of its effects on disease burden, and finally ends up with regulatory and public health approval.

4. Clinical trial programmes with GARDASIL®

Based on the above rational, Merck & Co. developed their quadrivalent (HPV 6, 11, 16 and 18) L1 VLP vaccine GARDASIL®, which has been extensively tested in different settings since 2003 (Fig. 1). This figure also depicts the trial programme planned with this vaccine extending until January 2010. The vaccine efficacy in the per-protocol (PP) populations has been analysed in a series of recent papers, and also discussed elsewhere in this volume [8–10]. Instead, what the author will do here is to report the combined analysis of these multinational trials spanning Europe, North America, Latin America and Asia pacific. The aims are: (a) to provide an assessment of the prevalence of HPV 6, 11, 16 and 18 infection, (b) to show the burden of observed HPV disease, and (c) to improve the precision of the estimates on vaccine efficacy against vaccine-related CIN and/or AIS.

5. Baseline characteristics of study participants

In its basic construction, the Merck’s quadrivalent HPV vaccine is a recombinant vaccine. GARDASIL® is a quadrivalent HPV (types 6, 11, 16 and 18) L1 virus-like particle (VLP) vaccine, for which the VLPs are manufactured in Saccharomyces cerevisiae. Aluminum adjuvant is used, 225 μg per dose, while the injection
Composite HPV Status by PCR and Serology at Enrollment

<table>
<thead>
<tr>
<th></th>
<th>GARDASIL® (N = 9,087)</th>
<th>Placebo (N = 9,087)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive to HPV 6, 11, 16 or 18 (%)</td>
<td>(%)</td>
<td>(%)</td>
</tr>
<tr>
<td>By serology</td>
<td>19.8</td>
<td>19.7</td>
</tr>
<tr>
<td>By PCR</td>
<td>14.9</td>
<td>14.8</td>
</tr>
<tr>
<td>By serology or PCR</td>
<td>27.2</td>
<td>26.9</td>
</tr>
</tbody>
</table>

N = Number of subjects randomized

Fig. 2. HPV status among the women in the vaccine and placebo groups.

Analysis of Prophylactic Efficacy Against HPV 6, 11, 16, or 18-Related CIN

Per-Protocol Population

<table>
<thead>
<tr>
<th></th>
<th>GARDASIL® (n = 7,858)</th>
<th>Placebo (n = 7,861)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Cases</td>
<td>Rate*</td>
</tr>
<tr>
<td>CIN or worse</td>
<td>4</td>
<td>&lt;0.1</td>
</tr>
<tr>
<td>CIN 1</td>
<td>4</td>
<td>&lt;0.1</td>
</tr>
<tr>
<td>CIN 2</td>
<td>0</td>
<td>0.0</td>
</tr>
<tr>
<td>CIN 3/AIS</td>
<td>0</td>
<td>0.0</td>
</tr>
</tbody>
</table>

The 4 vaccine cases were HPV 16-related CIN 1

Subjects are counted once in each applicable endpoint category. A subject may appear in more than one category.

*Cases per 100 person years at risk
n = received 3 vaccinations within 1 year; no major protocol violations; HPV 6, 11, 16 or 18 DNA(-) at Day 1 and HPV 6, 11, 16 or 18 DNA(-) Day 1 through one month post-dose 3; Cases counted starting one month post-dose 3

Fig. 3. Prophylactic efficacy analysis in the per-protocol population.
volume is 0.5 ml. Thus, it is produced in recombinant yeast, like the hepatitis B vaccine, and adsorbed on a proprietary aluminum adjuvant, like the tetanus vaccine.

In their baseline characteristics, the study populations in all these trials are very similar in the vaccine group and the placebo group. This applies to the age of the study subjects, their median number of lifetime sexual partners, past pregnancy, using hormonal contraception. The same also applies to the baseline PAP smear status of these women, which is almost identical in the vaccine- and the placebo groups. Importantly, also the HPV status determined by PCR and serology is very similar in these two groups, as shown in Fig. 2. When considered together, the entire series of 18.174 women usually had somewhat higher prevalence of HPV antibodies to each individual HPV types than DNA determined by PCR. This difference was most marked for HPV 6, showing 8.1% seroprevalence and 4.0% DNA detection rate by PCR.

6. Burden of HPV-related disease: incidence of CIN over the follow-up period in the placebo cohort

Altogether, 784 women in the placebo group developed CIN (any grade) during the follow-up period. This consists of 637 CIN1 lesions, 328 CIN2 or worse, and 190 CIN3 or worse. The incidence rate was highest (≥5100 person years at risk or pyr) among the 16–18 years age group, and slightly declined thereafter. Of the four geographic regions, the incidence of CIN was highest among the women in Latin America (5.5/100 pyr), and lowest (4/100 pyr) in Europe.

When related to the baseline HPV status of these women, the rate of CIN was highest among those who were PCR+/Sero+, caused by the HPV type with which the person was infected (rate = 9.2). On the other hand, the incidence was very low (0.2 and 0.8) among those who were baseline PCR−/Sero+ or HPV 6, 11, 16 and 18 naïve at day 1, respectively.

When the incidence of CIN related to HPV types 6, 11, 16, and 18 was considered, these rates were substantially lower, however, ranging from 1.8% for all CIN to 0.7 for CIN3 or worse (126/17.551 pyr). However, of all CIN3 or worse lesions, the proportion of HPV 16 and HPV 18 was 64%.

Accordingly, 40% of subjects in the placebo group who developed CIN 1–3 during the follow-up period had a lesion that was due to HPV 6, 11, 16 or 18. Altogether, 64% of subjects in the placebo group who developed CIN 3 or worse during the follow-up period had a lesion that was due to HPV 16 or 18. In general, CIN in the placebo group was observed across all ages, races and regions.

7. Vaccine efficacy using the combined phase II/III datasets

Prophylactic efficacy analysis was performed separately for the per-protocol (PP) arm and the modified intention-to-treat population.

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Please cite this article as: Nubia Munoz, Efficacy of a quadrivalent HPV (types 6, 11, 16, 18) L1 VLP vaccine against cervical intraepithelial neoplasia grades 1–3 and adenocarcinoma in situ, European Journal of Obstetrics & Gynecology and Reproductive Biology (2006), doi:10.1016/j.ejogrb.2006.06.027
to-treat (MITT) arm. In the PP arm, the vaccine proved to be 95% effective against all CIN, being lower (93%) for CIN 1 than for CIN 2 and CIN 3 (both 100%) (Fig. 3). Importantly, however, the vaccine was 100% effective against all HPV 16 and HPV 18 related CIN lesions.

As to the MITT population, the efficacy of GARDASIL® is summarised in Fig. 4. The vaccine was 100% effective against CIN 3/AIS, and somewhat less (92–98%) against the lower grade CIN lesions. Altogether, there were only eight cases of HPV 6, 11, 16 or 18 associated CIN cases in the vaccine group, as compared with 126 in the placebo group. The majority were found between 12 and 24 months post-dose 1.

8. Conclusions

This combined analysis of the clinical GARDASIL® trials show that the burden of HPV-related disease is significant. Accordingly, 27% of enrolled subjects were either seropositive or PCR-positive to HPV 6, 11, 16, or 18. Altogether, 8.7% of placebo recipients developed CIN during the follow-up period, of which a significant proportion was due to HPV 6, 11, 16 or 18. Importantly, GARDASIL® is 100% effective in preventing CIN 3 and AIS as well as CIN 2 in the per-protocol population. In the modified-intention-to-treat population, vaccine efficacy against CIN 3 and AIS was also 100% (95% CI: 91–100). In all studies, GARDASIL® has been well tolerated. It can be anticipated that this vaccine will lower a woman’s life-time risk for developing CC and substantially reduces the morbidity, mortality and clinical costs of HPV-related disease.

References