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Impact of baseline covariates on the immunogenicity of the 9-valent HPV vaccine – A combined analysis of five phase III clinical trials

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ABSTRACT

Background: The immunogenicity profile of the 9-valent HPV (9vHPV) vaccine was evaluated across five phase III clinical studies conducted in girls and boys 9–15 years of age and young women 16–26 years of age. The effect of baseline characteristics of subjects on vaccine-induced HPV antibody responses was assessed.

Methods: Immunogenicity data from 11,304 subjects who received ≥1 dose of 9vHPV vaccine in five Phase III studies were analyzed. Vaccine was administered as a 3-dose regimen. HPV antibody titers were assessed 1 month after dose 3 using a competitive Luminex immunoassay and summarized as geometric mean titers (GMTs). Covariates examined were age, gender, race, region of residence, and HPV serostatus and PCR status at day 1.

Results: GMTs to all 9 vaccine HPV types decreased with age at vaccination initiation, and were generally similar among the demographic subgroups defined by gender, race and region of residence. For all subgroups defined by race or region of residence, GMTs were higher in girls and boys than in young women. Vaccination of subjects who were seropositive at day 1 to a vaccine HPV type resulted in higher GMTs to that type, compared with those in subjects who were seronegative for that type at day 1.

Conclusions: 9vHPV vaccine immunogenicity was robust among subjects with different baseline characteristics. It was generally comparable across subjects of different races and from different regions. Greater immunogenicity in girls and boys versus young women (the population used to establish 9vHPV vaccine efficacy in clinical studies) indicates that the anti-HPV responses generated by the vaccine in adolescents from all races or...
1. Introduction

Human papillomavirus (HPV) is the cause of nearly all cervical cancers and a substantial proportion of anal, vulvar, vaginal, penile and oropharyngeal cancers; thus, it is responsible of approximately 5% of the global cancer burden [1]. The identification of HPV as a primary cause of anogenital cancers created an opportunity for cancer prevention through vaccination. First generation HPV vaccines, including the quadrivalent HPV (types 6/11/16/18) (qHPV) vaccine and the bivalent HPV (types 16/18) vaccine were initially developed [2]. A 9-valent HPV (types 6/11/16/18/31/33/45/52/58) (9vHPV) vaccine (Gardasil 9, Merck & Co., Inc., Kenilworth, NJ) was subsequently developed to provide protection against the HPV types already covered by the qHPV vaccine and the next five most common oncogenic types associated with cervical cancer worldwide (types 31/33/45/52/58) [3]. The 9vHPV vaccine could potentially prevent approximately 90% of cervical cancers, HPV-related vulvar, vaginal and anal cancers and genital warts worldwide [4–9]. The 9vHPV vaccine was licensed in 2014 in the US, in 2015 in Canada, the EU and Australia, and in 2015 and 2016 in other countries.

In a clinical trial conducted in women 16–26 years of age, the 9vHPV vaccine prevented infection and disease caused by HPV 31/33/45/52/58. It also induced anti-HPV 6/11/16/18 antibody responses that were non-inferior to responses induced by the qHPV vaccine; efficacy of the 9vHPV vaccine against infection and disease caused by HPV 6/11/16/18 was inferred based on these results [10–12]. In another clinical trial, the 9vHPV vaccine induced non-inferior antibody responses to HPV 6/11/16/18/31/33/45/52/58 in girls and boys 9–15 years of age vs. women 16–26 years of age; efficacy of the 9vHPV vaccine against infection and disease caused by the 9v HPV vaccine types in girls and boys 9–15 years of age was inferred based on these results [13].

HPV infection is a global health concern; prophylactic HPV vaccination is included in the national immunization programs of at least 80 countries [14], and used in diverse settings worldwide. It is anticipated that the 9vHPV vaccine will be widely licensed and recommended. Thus, it is useful to evaluate the impact of demographic parameters on the immunogenicity of the 9vHPV vaccine. Of relevant note, a similar study examining the impact of demographic parameters on the immunogenicity of the qHPV vaccine was published shortly after the initial licensure of the qHPV vaccine [15]. This report summarizes a combined analysis of five Phase III clinical trials conducted in girls and boys 9–15 years of age and women 16–26 years of age to examine antibody responses in subgroups for which individual studies may have had limited sample size. Thus, these analyses are novel and may be of interest to many as the 9vHPV vaccine becomes more widely available.

Immunogenicity of the 9vHPV vaccine in young men 16–26 years of age was not included in these analyses; it will be the topic of another report so that the additional complexities specific to that population (i.e., lower HPV antibody responses in men having sex with men than in heterosexual men [16,17]) can be fully explored.

2. Materials and methods

2.1. Enrollment and vaccination

An analysis of the combined immunogenicity database of Phase III studies submitted to regulatory agencies in support of the licensure of the 9vHPV vaccine was conducted. This analysis included 11,304 subjects who received 9vHPV vaccine in five Phase III studies (Table 1). These studies contained three separate populations: virgin girls 9–15 years of age, virgin boys 9–15 years of age, and young women 16–26 years of age, most of whom were sexually active. Eligible subjects received a 3-dose vaccination regimen given as intramuscular injections at day 1, month 2 and month 6. Each study was conducted in accordance with principles of Good Clinical Practice and was approved by the institutional review board at each participating institution and by regulatory agencies. Written informed consent was provided by all adult subjects and by a parent or legal guardian of subjects who were minors, assent was also obtained from minors in conformity with

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**Table 1**

| Study   | Key objectives                               | Experimental arm                                                                 | Control arm                                                                      | Included in analyses* |
|---------|----------------------------------------------|----------------------------------------------------------------------------------|----------------------------------------------------------------------------------|-----------------------|
| 001     | Immunogenicity, efficacy vs. qHPV vaccine     | Women age 16–26 years randomized to 9vHPV vaccine (N=6799)                       | Women age 16–26 years randomized to qHPV vaccine (N=6799)                        | N=6792               |
| 002     | Adult-to-adolescent immunobridging           | Girls and boys age 9–15 years (N=2604) enrolled to receive 9vHPV vaccine        | Girls and boys age 11–15 years randomized to concomitant arm (N=621)            | N=3066               |
| 005     | Co-administration with Menactra/Adacel       | Girls and boys age 11–15 years randomized to concomitant arm (N=526)            | Girls and boys age 11–15 years randomized to non-concomitant arm (N=620)       | N=618                |
| 007     | Co-administration with Repevax               | Girls and boys age 9–15 years randomized to 9vHPV vaccine (N=300)               | Girls and boys age 9–15 years randomized to non-concomitant arm (N=528)       | N=528                |
| 009     | qHPV-to-9vHPV immunobridging                 | Girls age 9–15 years randomized to 9vHPV vaccine (N=300)                        | Girls age 9–15 years randomized to qHPV vaccine (N=300)                        | N=300                |

*Subjects who received at least one vaccination with 9vHPV vaccine. A total of 11,304 subjects who received at least one 9vHPV vaccination are included in these analyses. Most subjects (97.7% [11,046 of 11,304]) received the three vaccinations.

**Study 001:** NCT00543543 [10].
**Study 002:** NCT00945722 [13].
**Study 005:** NCT00988884 [22].
**Study 007:** NCT01073293 [23].
**Study 009/GDS01C:** NCT01034498 [12].

Subjects who received the low-dose, mid-dose or high-dose formulation of 9vHPV vaccine during the dose selection portion of the study [10,43] are not included; immunogenicity results in these subjects are reported in [44].

Subjects randomized to the 9vHPV vaccine who received ≥1 dose of vaccine.

Subjects randomized to the non-concomitant arm who received ≥1 dose of 9vHPV vaccine. Subjects randomized to the concomitant arm of studies 005 and 007 are not considered in this report; immunogenicity results in these subjects are reported in [22,23].
applicable national and local requirements. Baseline characteristics for the overall population of subjects who were randomized to receive the 9vHPV vaccine are presented in Table 2.

2.2. Immunogenicity evaluation

Serum samples were obtained at day 1 and month 7 for anti-HPV antibody testing. The serum samples were assessed for antibodies to HPV VLP types 6/11/16/18/31/33/45/52/58 by a multiplexed competitive LumineX Immunoassay (cLIA; HPV-9 cLIA Version 2.0; performed by PPD Vaccines and Biologics Lab, Wayne, PA, USA), as described previously [18]. Antibody titers for each individual HPV type were determined through competition with type-specific monoclonal antibodies, so it is not possible to directly compare assay results across HPV types. In addition, cervical and external genital swabs collected at day 1 and month 7 in young women 16–26 years of age for testing by polymerase chain reaction (PCR) for type-specific detection of HPV DNA; PCR testing included the 9 vaccine types and 5 additional oncogenic HPV types (HPV 35/39/51/56/59)[19,20]. HPV seropositivity at day 1 or PCR positivity at day 1 and month 7 was not a reason for exclusion from the study; however, the results were part of the criteria to define analysis populations.

2.3. Data analysis

The serum samples from day 1 and PCR samples from day 1 and month 7 were analyzed for each vaccine HPV type prior to enrollment to identify participants who were positive to one or more HPV types, and these participants were subsequently excluded from the per-protocol immunogenicity analysis for the corresponding HPV type(s). To be included in the HPV type specific per-protocol immunogenicity analysis populations, subjects had to meet the following requirements: (1) be seronegative at day 1 and (for 16- to 26-year-old women) PCR-negative from day 1 through month 7 only for the HPV type being analyzed (for HPV 6 and HPV 11 immunogenicity analyses, because of extensive cross-reactivity due to the high amino acid sequence identity [92%] between HPV 6 and HPV 11 L1 proteins [21], subjects had to be seronegative and, for women 16–26 years of age, PCR-negative for both HPV 6 and HPV 11); (2) receive all 3 doses of the correct clinical material; and (3) have a post-dose 3 serology result a day 1 serology result and a day 1 PCR result; (2) receive all 3 doses of the correct clinical material; and (3) have a post-dose 3 serology result and a day 1 PCR result; and (3) have a post-dose 3 serology result and a day 1 PCR result. This analysis, subjects had to meet the following requirements: (1) have baseline HPV positive and HPV negative subjects. To be included in the HPV type specific per-protocol immunogenicity analyses, because of extensive cross-reactivity due to the high amino acid sequence identity [92%] between HPV 6 and HPV 11 L1 proteins [21], subjects had to be seronegative and, for women 16–26 years of age, PCR-negative for both HPV 6 and HPV 11); (2) receive all 3 doses of the correct clinical material; and (3) have a post-dose 3 serology result and a day 1 PCR result; (2) receive all 3 doses of the correct clinical material; and (3) have a post-dose 3 serology result and a day 1 PCR result.

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Geometric mean titers (GMTs) with associated 95% confidence intervals were computed and compared across categories of baseline subject characteristics. Cohorts analyzed included subjects given the 9vHPV vaccine stratified into the following three age/sex groups: boys 9–15 years of age, girls 9–15 years of age, and young women 16–26 years of age. Baseline covariates analyzed included age, sex, race, region of residence, and baseline HPV seropositivity and PCR-positivity. All of these evaluations were exploratory in nature; therefore, no statistical tests of hypotheses were performed. Non-overlapping 95% confidence intervals were used as indicators of differences of immune response.

It has been previously observed that HPV antibody response to HPV vaccine declines with increasing age [2,13,15]. Analyses were conducted to explore whether this relationship varies with race and geographic region. Linear regression model was fitted on the logarithm (base 10) of HPV antibody titer at Month 7 as a function of age at vaccination dose 1 to model the relationship of HPV antibody response at Month 7 with age. Graphical methods were used to display the regression line representing the estimated mean log_{10}HPV antibody response as a function of age together with the 95% confidence band around the regression line. The regression line and 95% confidence band were displayed by race and geographic region to graphically compare trends of the relationship of HPV antibody response by age across race and geographic region. For each HPV type, two analyses were conducted: an analysis of mean HPV antibody response by race irrespective of geographic region (this analysis is relevant given that race is a reasonable surrogate for geographic region); a second analysis of mean HPV antibody response by race and geographic region. No formal statistical testing was done on the trends of the relationship of mean HPV antibody response by age across race or race and geographic region.

In studies 001 and 009, subjects were randomized to receive 9vHPV vaccine or qHPV vaccine [10,12]. This report considers only subjects who received the 9vHPV vaccine. Immunogenicity of qHPV vaccine by baseline covariates has already been reported [15].

| Race       | Females 9–15 years of age | Males 9–15 years of age | Females 16–26 years of age |
|------------|---------------------------|------------------------|---------------------------|
| Asian      | 470 (16.7)                | 224 (18.0)             | 1113 (15.3)               |
| Black      | 182 (6.5)                 | 60 (4.8)               | 281 (3.9)                 |
| White      | 1749 (62.3)               | 665 (53.5)             | 4004 (55.1)               |
| Other      | 408 (14.5)                | 294 (23.7)             | 1971 (25.7)               |
| Region     |                           |                        |                           |
| Africa     | 95 (3.4)                  | 30 (2.4)               | 40 (0.6)                  |
| Asia-Pacific| 458 (16.3)               | 219 (17.6)             | 998 (13.7)                |
| Europe     | 1102 (39.2)               | 373 (30.0)             | 2531 (34.8)               |
| Latin America| 545 (19.4)              | 286 (23.0)             | 2319 (31.9)               |
| North America| 609 (21.7)              | 335 (27.0)             | 1381 (19.0)               |

Table 2

Study participants were from 26 countries (Austria, Belgium, Brazil, Canada, Chile, Colombia, Costa Rica, Denmark, Finland, Germany, Hong Kong, India, Italy, Japan, Korea, Mexico, New Zealand, Norway, Peru, Poland, South Africa, Spain, Sweden, Taiwan, Thailand, and the United States [including Puerto Rico]).

The category 'Other' for the race variable includes Multi-Racial, American Indian or Alaska Native, Native Hawaiian or Pacific Islander, Unknown and missing race information. Most subjects in that category are Multi-Racial.
Per-protocol summary of month 7 anti-HPV geometric mean titers in subjects who received 3 doses of 9vHPV vaccine.

| Population | N   | n  | % Seropositive (95% CI) | GMT (95% CI) |
|------------|-----|----|-------------------------|--------------|
| Anti-HPV 6  |     |    |                         |              |
| 9–15-year-old girls | 2805 | 2349 | 99.7 (99.4, 99.9) | 1744.6 (1684.7, 1806.7) |
| 9–15-year-old boys | 1239 | 1055 | 99.9 (99.5, 100) | 2085.3 (1984.2, 2191.6) |
| 16–26-year-old women | 7260 | 4321 | 99.8 (99.6, 99.9) | 893.7 (873.5, 914.3) |
| Anti-HPV 11 |     |    |                         |              |
| 9–15-year-old girls | 2805 | 2350 | 99.9 (99.7, 100) | 1289.7 (1244.3, 1336.8) |
| 9–15-year-old boys | 1239 | 1055 | 100 (99.7, 100) | 1469.2 (1397.7, 1544.4) |
| 16–26-year-old women | 7260 | 4327 | 100 (99.9, 100) | 669.3 (653.6, 685.4) |
| Anti-HPV 16 |     |    |                         |              |
| 9–15-year-old girls | 2805 | 2405 | 99.9 (99.7, 100) | 7159.9 (6919.7, 7408.5) |
| 9–15-year-old boys | 1239 | 1076 | 100 (99.7, 100) | 8444.9 (8054.2, 8854.5) |
| 16–26-year-old women | 7260 | 4361 | 100 (99.9, 100) | 3159.0 (3088.6, 3231.1) |
| Anti-HPV 18 |     |    |                         |              |
| 9–15-year-old girls | 2805 | 2420 | 99.9 (99.6, 100) | 2085.5 (2002.2, 2172.3) |
| 9–15-year-old boys | 1239 | 1074 | 100 (99.7, 100) | 2620.4 (2474.3, 2775.2) |
| 16–26-year-old women | 7260 | 4884 | 99.8 (99.7, 99.9) | 809.9 (789.2, 831.1) |
| Anti-HPV 31 |     |    |                         |              |
| 9–15-year-old girls | 2805 | 2397 | 100 (99.8, 100) | 1883.3 (1811.3, 1958.1) |
| 9–15-year-old boys | 1239 | 1069 | 100 (99.7, 100) | 2173.5 (2057.0, 2296.6) |
| 16–26-year-old women | 7260 | 4806 | 99.8 (99.6, 99.9) | 664.8 (647.4, 682.6) |
| Anti-HPV 33 |     |    |                         |              |
| 9–15-year-old girls | 2805 | 2418 | 99.9 (99.7, 100) | 960.6 (927.5, 994.9) |
| 9–15-year-old boys | 1239 | 1076 | 100 (99.7, 100) | 1178.6 (1120.9, 1239.4) |
| 16–26-year-old women | 7260 | 5056 | 99.7 (99.5, 99.8) | 419.2 (409.6, 429.1) |
| Anti-HPV 45 |     |    |                         |              |
| 9–15-year-old girls | 2805 | 2430 | 99.8 (99.6, 100) | 728.7 (697.6, 761.2) |
| 9–15-year-old boys | 1239 | 1079 | 100 (99.7, 100) | 841.7 (790.6, 896.7) |
| 16–26-year-old women | 7260 | 5160 | 99.6 (99.4, 99.7) | 254.1 (247.0, 261.5) |
| Anti-HPV 52 |     |    |                         |              |
| 9–15-year-old girls | 2805 | 2426 | 99.9 (99.7, 100) | 978.2 (942.8, 1015.0) |
| 9–15-year-old boys | 1239 | 1077 | 100 (99.7, 100) | 1062.2 (1007.2, 1120.2) |
| 16–26-year-old women | 7260 | 4792 | 99.8 (99.6, 99.9) | 382.4 (373.6, 392.0) |
| Anti-HPV 58 |     |    |                         |              |
| 9–15-year-old girls | 2805 | 2397 | 99.9 (99.7, 100) | 1306.0 (1259.8, 1354.0) |
| 9–15-year-old boys | 1239 | 1072 | 100 (99.7, 100) | 1545.8 (1470.6, 1624.8) |
| 16–26-year-old women | 7260 | 4818 | 99.8 (99.6, 99.9) | 489.2 (477.5, 501.2) |
Fig. 1. Plots of month 7 anti-HPV geometric mean titers (GMTs) responses in females to component human papillomavirus (HPV) vaccine types, by age at enrollment. GMTs with associated 95% confidence intervals are presented for the per-protocol immunogenicity population. cLIA, competitive Luminex-based immunoassay; mMU, milli-Merck units.
defined by day 1 HPV serostatus and PCR status. Inclusion of subjects regardless of baseline HPV status permitted a comparison of vaccine-induced immune responses with those generated in response to an HPV infection. Robust antibody responses were observed in all groups for all HPV types. GMTs appeared to be the highest in the group which was seropositive and PCR negative on day 1 (i.e., subjects who were seropositive at enrollment likely due to a prior exposure to HPV). GMTs were analyzed over time in the per-protocol population and in subjects seropositive and PCR negative at day 1 in study 001. Table 4 enumerates anti-HPV GMTs at all time-points from month 3 (1 month post-dose 2) and month 7 (1 month post-dose 3) to month 42 in subjects that were seropositive and PCR negative at day 1. Notably, in

| Race  | Test | n | GMT (95% CI) | n | GMT (95% CI) | n | GMT (95% CI) | n | GMT (95% CI) |
|-------|------|---|-------------|---|-------------|---|-------------|---|-------------|
|       | Asian | 785 | 986.1 (763.5, 1443.9) | 758 | 827.4 (775.1, 883.2) | 1584 | 578.7 (552.7, 605.9) | 1616 | 369.1 (381.1, 431.4) |
|       | Black | 520.3 | 507.5 (431.9, 562.9) | 493.1 (431.9, 562.9) | 485.6 (468.2, 499.4) | 480.8 (457.2, 505.5) |
|       | White | 814 | (450.5, 551.9) | 161 | (431.9, 562.9) | 2720 | (468.2, 499.4) |
|       | Other | 15 | 442.9 (300.3, 653.3) | 782 | (381.8, 431.9) | 1635 | (381.1, 431.4) | 1505 | (421.0, 475.0) |
|       |       | 24 | 518.1 (345.1, 782.3) | 810 | (249.8, 284.7) | 1714 | (197.9, 218.2) | 1604 | (276.7, 306.1) |
|       |       | 20 | 552.8 (376.5, 811.7) | 717 | (319.4, 363.2) | 1620 | (349.8, 381.0) | 1465 | (373.7, 408.8) |
|       |       | 5 | 706.9 (458.1, 1090.8) | 743 | (467.5, 528.9) | 1647 | (449.2, 487.9) | 1425 | (461.7, 504.7) |

GMT, geometric mean titer (given in milli-Merck units per milliliter). CI, confidence interval.

Table 5

Per-protocol summary of anti-HPV geometric mean titers at month 7 by race in women 16–26 years of age who received 3 doses of 9vHPV vaccine.

| Race  | Test | n | GMT (95% CI) | n | GMT (95% CI) | n | GMT (95% CI) |
|-------|------|---|-------------|---|-------------|---|-------------|
|       | Asian | 691 | 817.9 (772.7, 865.8) | 1454 | 850.9 (818.2, 884.9) | 1292 | 946.6 (908.1, 988.8) | 868 | 953.2 (906.1, 1002.8) |
|       | Black | 722 | 588.9 (555.0, 624.8) | 1412 | 2929.3 (2816.1, 3047.1) | 1336 | 3355.0 (3221.8, 3493.8) | 871 | 3412.8 (3245.7, 3585.8) |
|       | White | 758 | 827.4 (775.1, 883.2) | 1604 | 714.2 (682.9, 747.0) | 1537 | 887.6 (847.9, 922.9) | 964 | 883.4 (793.1, 888.3) |
|       | Other | 785 | 673.6 (631.0, 719.0) | 1584 | 578.7 (552.7, 605.9) | 1616 | 369.1 (381.1, 431.4) | 1005 | 450.9 (428.1, 475.0) |
|       |       | 810 | 267.7 (249.8, 284.7) | 1714 | 207.8 (197.9, 218.2) | 1604 | 291.0 (276.7, 306.1) | 1008 | 272.0 (255.2, 289.8) |
|       |       | 717 | 340.6 (319.4, 363.2) | 1620 | 385.1 (349.8, 381.0) | 1465 | 390.8 (373.7, 408.8) | 970 | 432.1 (409.0, 456.7) |
|       |       | 743 | 497.3 (467.5, 528.9) | 1647 | 468.2 (449.2, 487.9) | 1425 | 482.8 (461.7, 504.7) | 988 | 527.2 (499.8, 556.1) |
this population, the anti-HPV GMTs generated by the 9vHPV vaccine were dramatically increased after 2 doses (month 3) or 3 doses (month 7), and were substantially higher (as evidenced by non-overlapping 95% CI) than GMTs observed in the per-protocol population for all time points from month 3 to month 42.

### 4. Discussion

A combined analysis of the immunogenicity in five Phase III clinical studies showed that a 3-dose regimen of the 9vHPV vaccine was highly immunogenic in girls and boys 9–15 years of age and young women 16–26 years of age, with seroconversion rates at 1 month post-dose 3 > 99% in these three populations. GMTs at 1 month post-dose 3 in the combined database were higher in girls and boys than in young women, due to these types. It must be noted though, regardless of the HPV type day 1 seropositivity status prior to vaccination, all subjects exhibited a robust boost in GMTs post-vaccination.

In a Phase III clinical study (study 001), the 9vHPV vaccine prevented infection and disease related to HPV 31/33/45/52/58 in young women 16–26 years of age from multiple races and regions [10]. In the same study, the efficacy findings established with qHPV vaccine for HPV 6/11/16/18 in earlier clinical studies [27–29] were extended

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Table 6

| HPV type | Day 1 serostatus | Day 1 PCR status | n       | GMT (mMU/mL) (95% CI) |
|----------|------------------|-----------------|---------|-----------------------|
| HPV 6    | Negative         | Negative        | 4720    | 901.4 (881.8, 921.4) |
|          | Negative         | Positive        | 113     | 1025.2 (881.8, 1192.0) |
|          | Positive         | Negative        | 807     | 1876.0 (1742.2, 2020.1) |
|          | Positive         | Positive        | 109     | 1599.5 (1287.3, 1770.1) |
| HPV 11   | Negative         | Negative        | 4723    | 675.7 (660.3, 691.4) |
|          | Negative         | Positive        | 13      | 618.4 (331.1, 1155.3) |
|          | Positive         | Negative        | 188     | 1063.9 (935.0, 1215.0) |
|          | Positive         | Positive        | 14      | 1110.9 (652.9, 1890.2) |
| HPV 16   | Negative         | Negative        | 4799    | 3177.3 (3109.4, 3246.7) |
|          | Negative         | Positive        | 323     | 2941.4 (2676.1, 3233.1) |
|          | Positive         | Negative        | 492     | 5248.6 (4848.6, 5681.5) |
|          | Positive         | Positive        | 260     | 4374.0 (3968.9, 4820.6) |
| HPV 18   | Negative         | Negative        | 5334    | 815.9 (796.0, 836.3) |
|          | Negative         | Positive        | 178     | 941.2 (829.7, 1067.7) |
|          | Positive         | Negative        | 266     | 1917.2 (1714.3, 2144.0) |
|          | Positive         | Positive        | 86      | 1472.5 (1236.3, 1753.8) |
| HPV 31   | Negative         | Negative        | 5254    | 668.2 (651.5, 685.3) |
|          | Negative         | Positive        | 184     | 625.3 (553.5, 708.4) |
|          | Positive         | Negative        | 327     | 964.4 (877.3, 1060.1) |
|          | Positive         | Positive        | 112     | 798.5 (690.3, 923.7) |
| HPV 33   | Negative         | Negative        | 5503    | 424.1 (414.8, 433.7) |
|          | Negative         | Positive        | 101     | 443.3 (375.7, 523.1) |
|          | Positive         | Negative        | 215     | 665.7 (582.5, 760.7) |
|          | Positive         | Positive        | 59      | 616.0 (482.4, 786.7) |
| HPV 45   | Negative         | Negative        | 5620    | 255.0 (248.1, 262.0) |
|          | Negative         | Positive        | 130     | 256.9 (216.8, 304.4) |
|          | Positive         | Negative        | 82      | 354.0 (279.1, 449.1) |
|          | Positive         | Positive        | 29      | 285.8 (195.7, 417.4) |
| HPV 52   | Negative         | Negative        | 5259    | 385.1 (376.1, 394.3) |
|          | Negative         | Positive        | 275     | 303.5 (272.8, 337.5) |
|          | Positive         | Negative        | 220     | 547.1 (488.5, 612.7) |
|          | Positive         | Positive        | 121     | 319.8 (271.4, 376.7) |
| HPV 58   | Negative         | Negative        | 5267    | 493.8 (482.5, 505.3) |
|          | Negative         | Positive        | 158     | 423.3 (376.1, 476.3) |
|          | Positive         | Negative        | 373     | 618.9 (559.6, 684.6) |
|          | Positive         | Positive        | 82      | 669.8 (548.4, 818.0) |

N=number of subjects who received at least 1 injection of 9vHPV vaccine; n=number of subjects contributing to the analysis.

GMT, geometric mean titer (given in milli-Merck units per milliliter). CI, confidence interval.

* This analysis population includes subjects who received all 3 doses of correct clinical material, had serology & PCR results at day 1 for the relevant HPV type and had a post-dose 3 or month 7 serology result within acceptable day ranges.
### HPV 6

**Day 1**
- n: 3993
- GMT (95% CI): < 16 (< 16, < 16)

**Month 3**
- n: 788
- GMT (95% CI): 734.0 (692.8, 777.7)

**Month 7**
- n: 3993
- GMT (95% CI): 893.1 (871.7, 915.1)

**Month 12**
- n: 800
- GMT (95% CI): 330.6 (312.2, 350.1)

**Month 24**
- n: 715
- GMT (95% CI): 208.6 (195.5, 222.7)

**Month 36**
- n: 685
- GMT (95% CI): 163.9 (153.0, 175.6)

**Month 42**
- n: 692
- GMT (95% CI): 147.2 (137.3, 157.8)

**Day 1**
- n: 3995
- GMT (95% CI): < 6 (< 6, < 6)

**Month 3**
- n: 790
- GMT (95% CI): 529.1 (499.7, 560.1)

**Month 7**
- n: 3995
- GMT (95% CI): 666.3 (649.6, 684.3)

**Month 12**
- n: 810
- GMT (95% CI): 212.4 (200.1, 225.6)

**Month 24**
- n: 778
- GMT (95% CI): 529.7 (487.4, 573.4)

**Month 36**
- n: 695
- GMT (95% CI): 386.5 (356.3, 419.4)

**Month 42**
- n: 696
- GMT (95% CI): 84.9 (79.0, 91.3)

### HPV 11

**Day 1**
- n: 4032
- GMT (95% CI): < 12 (< 12, < 12)

**Month 3**
- n: 794
- GMT (95% CI): 2435.8 (2303.5, 2575.6)

**Month 7**
- n: 4032
- GMT (95% CI): 311.1 (3057.1, 3206.9)

**Month 12**
- n: 814
- GMT (95% CI): 1041.7 (979.9, 1107.4)

**Month 24**
- n: 778
- GMT (95% CI): 529.7 (487.4, 573.4)

**Month 36**
- n: 695
- GMT (95% CI): 386.5 (356.3, 419.4)

**Month 42**
- n: 696
- GMT (95% CI): 84.9 (79.0, 91.3)

### HPV 16

**Day 1**
- n: 4032
- GMT (95% CI): < 12 (< 12, < 12)

**Month 3**
- n: 794
- GMT (95% CI): 2435.8 (2303.5, 2575.6)

**Month 7**
- n: 4032
- GMT (95% CI): 311.1 (3057.1, 3206.9)

**Month 12**
- n: 814
- GMT (95% CI): 1041.7 (979.9, 1107.4)

**Month 24**
- n: 778
- GMT (95% CI): 529.7 (487.4, 573.4)

**Month 36**
- n: 695
- GMT (95% CI): 386.5 (356.3, 419.4)

**Month 42**
- n: 696
- GMT (95% CI): 84.9 (79.0, 91.3)

### HPV 31

**Day 1**
- n: 4466
- GMT (95% CI): < 4 (< 4, < 4)

**Month 3**
- n: 881
- GMT (95% CI): 437.6 (406.7, 470.8)

**Month 7**
- n: 4466
- GMT (95% CI): 658.4 (636.7, 680.9)

**Month 12**
- n: 909
- GMT (95% CI): 196.5 (183.5, 210.4)

**Month 24**
- n: 863
- GMT (95% CI): 101.9 (94.9, 109.5)

**Month 36**
- n: 778
- GMT (95% CI): 78.5 (71.9, 85.6)

**Month 42**
- n: 806
- GMT (95% CI): 70.8 (64.8, 77.3)

### HPV 33

**Day 1**
- n: 4702
- GMT (95% CI): < 4 (< 4, < 4)

**Month 3**
- n: 937
- GMT (95% CI): 437.6 (406.7, 470.8)

**Month 7**
- n: 4466
- GMT (95% CI): 658.4 (636.7, 680.9)

**Month 12**
- n: 909
- GMT (95% CI): 196.5 (183.5, 210.4)

**Month 24**
- n: 863
- GMT (95% CI): 101.9 (94.9, 109.5)

**Month 36**
- n: 778
- GMT (95% CI): 78.5 (71.9, 85.6)

**Month 42**
- n: 806
- GMT (95% CI): 70.8 (64.8, 77.3)

### HPV 45

**Day 1**
- n: 4792
- GMT (95% CI): < 3 (< 3, < 3)

**Month 3**
- n: 937
- GMT (95% CI): 160.4 (151.7, 169.7)

**Month 7**
- n: 4792
- GMT (95% CI): 252.8 (246.2, 259.6)

**Month 12**
- n: 976
- GMT (95% CI): 69.2 (65.4, 73.3)

**Month 24**
- n: 928
- GMT (95% CI): 33.0 (31.0, 35.0)

**Month 36**
- n: 835
- GMT (95% CI): 22.9 (21.4, 24.4)

**Month 42**
- n: 846
- GMT (95% CI): 21.1 (19.8, 22.5)

### HPV 52

**Day 1**
- n: 4455
- GMT (95% CI): < 3 (< 3, < 3)

**Month 3**
- n: 895
- GMT (95% CI): 241.3 (229.7, 253.4)

**Month 7**
- n: 4455
- GMT (95% CI): 379.7 (371.6, 388.0)

**Month 12**
- n: 916
- GMT (95% CI): 118.9 (113.0, 124.0)

**Month 24**
- n: 867
- GMT (95% CI): 57.9 (54.7, 61.2)

**Month 36**
- n: 777
- GMT (95% CI): 47.9 (45.0, 50.9)

**Month 42**
- n: 791
- GMT (95% CI): 43.2 (40.6, 46.0)
to the 9vHPV vaccine based on the demonstration of non-inferior HPV 6/11/16/18 antibody responses. In additional analyses, both qHPV and 9vHPV vaccine were found to be highly efficacious against infection and disease in subgroups of young women 16–26 years of age differing by age, race, and region of residence [29–35]. Therefore, the small differences in 9vHPV vaccine immunogenicity by age, race or region of residence shown in this report in young women 16–26 years of age are unlikely to have a clinical significance.

As seen in this report, anti-HPV GMTs at month 7 are substantially higher in all subgroups of girls and boys 9–15 years of age defined by age, race, and region of residence compared with HPV antibody responses in young women 16–26 years of age in the combined database (Table 3) or previously reported in study 001 [10]. As previously reported, prophylactic administration of the 9vHPV vaccine to 16–26-year-olds was highly effective in preventing infection and disease due to vaccine HPV types [10]. Thus, the anti-HPV responses generated by the vaccine in adolescents were sufficient to induce high-level protective efficacy. Overall, 9vHPV vaccine efficacy can be inferred in all subgroups and the small differences in 9vHPV vaccine immunogenicity by age, race or region of residence shown in this report are unlikely to have a clinical significance in girls and boys 9–15 years of age.

There are several limitations to this combined analysis of immunogenicity. Even though the studies included in these analyses enrolled subjects from six continents, they enrolled only a limited number of subjects from Africa and South Asia. Therefore, it will be important to further evaluate the immunogenicity of the 9vHPV vaccine in these regions, especially given the prevalence of HIV infection or other co-infections and malnutrition in these regions which may impact immune response to the vaccine. Of note, studies of the qHPV vaccine in sub-Saharan Africa, India, and Vietnam, and a study in HIV-infected children demonstrated robust immunogenicity in these populations [36–39]. Given that the 9vHPV vaccine and qHPV vaccine have comparable immunogenicity profiles [10–12], similar results are expected with the 9vHPV vaccine.

This combined analysis assessed the immunogenicity of a 3-dose regimen of 9vHPV vaccine. The use of an alternative 2-dose regimen for HPV vaccines has been recommended in 2014 by the World Health Organization in 9- to 14-year-olds [40]. The 2-dose schedule for the previously developed bivalent and quadrivalent HPV vaccines has been implemented in several countries [41]. A Phase III study to assess a 2-dose regimen of the 9vHPV vaccine in girls and boys 9–14 years of age has recently provided relevant immunogenicity data [42].

In summary, the 9vHPV vaccine induced robust HPV antibody responses to all 9 vaccine HPV types in subjects from all ages, races, and geographic regions represented in the aforementioned five Phase III studies of the vaccine. This comprehensive immunogenicity profile provides compelling evidence for administration of the 9vHPV vaccine at an early age (i.e., before exposure to HPV) and supports a widespread 9vHPV vaccination program regardless of race or region of residence.

5. Conclusion

In clinical trials, the 9vHPV vaccine was highly immunogenic in subjects aged 9–26 years. The 9vHPV vaccine immunogenicity was robust among subjects with differing baseline characteristics (age, gender, race, region of residence, and HPV serostatus and PCR status at day 1). Its immunogenicity profile was similar to that of the qHPV vaccine. The demonstrated efficacy, safety and immunogenicity profile of the 9vHPV vaccine supports widespread vaccination programs.

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Author contribution

Study concept and design: RM, EM, AL.

Acquisition of data: LKP, EDM, OEI, PP, PVD, EAJ, S-EO, DF, SB, ARG.

Analysis and interpretation of data: All authors.

Manuscript Preparation: All Authors.

Statistical analysis: OMB, DH, RM.

Conflicts of interest

LKP: was advisory board member for Sanofi Pasteur and investigator at the vaccine trials funded by MSD.

JAR: Nothing to disclose.

EDM: has received research grants from and is a member of a speaker’s bureau for Merck & Co., Inc, Kenilworth, NJ, USA.

O-EI: has received compensation from Merck to conduct vaccine clinical trials and scientific advisory board fees.

PP: No potential conflicts of interest to disclose.

PVD: Acts as chief and principal investigator for vaccine trials conducted on behalf of the University of Antwerp, for which the University obtains research grants from vaccine manufacturers; speak-
ers fees for presentations on vaccines are paid directly to an educational fund held by the University of Antwerp. PVD receives no personal remuneration for this work.

EAJ: Reports having received grant support paid to his institution from Merck and GlaxoSmithKline and advisory board fees from Merck and Sanofi Pasteur MSD.

S-EO: has received grants from Merck according to contracts to perform studies with HPV-vaccines.

DF: has received grant support from Merck through his institution and personal fees for consultancy and advisory boards for Merck.

SB: has received research grants from and is a member of a speaker’s bureau for Merck & Co., Inc., Kenilworth, NJ, USA and has served as a paid expert witness and consultant for Merck.

ARG: has received fees for serving on an advisory board and grant support through her institution from Merck.

XB: has received institutional research and educational grants from Sanofi Pasteur MSD and GlaxoSmithKline and personal travel grant and speakers honorarium from Sanofi Pasteur MSD and GlaxoSmithKline.

SP: has received travel expenses from Sanofi Pasteur MSD.

JC: has received fees for serving on advisory boards from Merck, Abbott, Gen-Probe Hologic, and Becton Dickinson, lecture fees from GlaxoSmithKline, and grant support from Roche, Abbott, Gen-Probe Hologic, Becton Dickinson, and Qiagen.

SMG: has received Grants to her institution from Commonwealth Dept. of Health for HPV genoprevalence surveillance post-vaccination, Merck and GSK to perform phase 3 clinical vaccine trials: Merck to evaluate HPV in RRP post-vaccination program & CSL for HPV in cervical cancer study, & VCA for a study on effectiveness of public health HPV vaccine study plus a study on associations of early onset cancers. Received speaking fees from MSD and SEPMED for work performed in her personal time. Merck paid for travel & accommodation to present at HPV Advisory board meetings.

WH: has received fees for serving on advisory board from Merck, Abbott, Gen-Probe Hologic, and Becton Dickinson. SSK:has received speaker’s and advisory board fees from Sanofi Pasteur MSD and Merck, advisory board fees from BD, and unrestricted research grants through her institution from Merck.

OMB, DH, EM, HQ, AL: are employees of Merck & Co., Inc., Kenilworth, NJ, USA.

RM, CR: were employees of Merck & Co., Inc., Kenilworth, NJ, USA at the time of the study.

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Appendix A. Supplementary material

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