



Immunogenicity and tolerability of an HPV-16/18 AS04-adjuvanted prophylactic cervical cancer vaccine in women aged 15–55 years

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ABSTRACT

The immunogenicity and safety of an HPV-16/18 AS04-adjuvanted vaccine were assessed in women aged 26–55 years and compared with women aged 15–25 years in a Phase III, non-randomised, open-label, age-stratified study. Overall the vaccine was well tolerated and 100% seropositivity was achieved 1 month after the third dose in all age groups. There was a high correlation between HPV-16 and HPV-18 antibody levels (IgG) in cervicovaginal secretions and sera, regardless of age. The HPV-16/18 AS04-adjuvanted vaccine induces a robust and persistent immune response in women >26 years of age and generates antibodies that transudate through the cervix epithelium.

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1. Introduction

Human papillomavirus (HPV) has been identified as the causal infectious agent in cervical cancer and in the development of cervical cytological abnormalities and pre-cancerous lesions [1,2]. Of the 15 oncogenic HPV types identified as being carcinogenic to the cervix, types 16 and 18 are most commonly found in cancerous cervical tissue [3–6].

Infection with oncogenic HPV can occur throughout a sexually active woman's lifetime, where infection may be the result of reactivation of infections acquired earlier in life, and infection from new sexual contacts later in life [7]. Persistent infection with oncogenic HPV types has been identified as the necessary cause for the development of cervical cancer. It has been suggested that up to 50% of oncogenic HPV infections persist in adult women [8]. Prevalent

HPV infection has shown to first peak in adolescent girls, with an observed second peak in women 45–50 years of age [8–10]. Pre-cancerous lesions are also significantly less likely to regress in older women (50–80%) compared with young adult women (>90%) [11], and a consistent number of cervical adenocarcinomas and squamous cell carcinomas occur in women aged 34–49 years [12].

Clinical trials assessing the use of prophylactic HPV L1 virus-like particle (VLP) vaccines have demonstrated that these vaccines are highly efficacious in the prevention and reduction of oncogenic HPV infections, persistent infections and related cervical lesions in women 15–29 years of age [13–15].

Local and systemic antibody responses are likely to represent an important aspect of the assessment of HPV vaccine-induced immune responses. In clinical trials, systemic responses induced after HPV immunization have been clearly demonstrated, although protective serological antibody levels have not been identified. As cervical cancers typically develop at the transformation zone, vaccine-induced antibodies at the site of infection, or in the mucosa of the genital tract, could play an important role in the prevention

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of oncogenic HPV-related (types 16 or 18) cervical disease [16,17]. The presence of antibodies at the site of infection has been previously observed after experimental HPV vaccination, suggesting transudation of systemic antibodies into the cervix [18].

In the present study, the immunogenicity and safety of the HPV-16/18 L1 VLP AS04-adjuvanted vaccine was assessed in women aged 26–55 years, compared with women aged 15–25 years.

2. Methods

2.1. Study design

This multicentre, open-labelled trial was conducted in two phases: an initial phase that included follow-up for 1 year after first vaccination (Months 0–12), and an extension phase that includes follow-up to Month 48. Interim data up to Month 24 are reported here.

In the initial phase, the primary and secondary objectives were to evaluate vaccine-induced immune response to HPV-16 and HPV-18, in terms of non-inferiority based on seroconversion rates, in women 26–45 and 46–55 years of age compared with women 15–25 years of age. In the extension phase, the primary objective was to evaluate HPV-16 and 18 seropositivity rates and antibody levels in the serum at all time-points, and the secondary objective was to compare HPV-16 and 18 antibody levels of women in the current study (by age group) to antibody levels in a previously published efficacy study in women 15–25 years of age. Cervicovaginal secretions were also collected in a subset of women at selected study sites, for evaluation of anti-HPV-16 and anti-HPV-18 IgG antibody levels in secretions at the cervix. The safety of the HPV-16/18 AS04 vaccine was assessed for all age groups during the entire study period.

Women between 15 and 55 years of age were included and, if of childbearing potential, were required to be abstinent from sexual activity or using adequate contraception for 30 days prior to vaccination and up to 2 months after completion of the vaccination series, and have a negative pregnancy test on the day of vaccination. Women were excluded from the study if they had used an investigational drug or vaccine within 30 days, chronic immune-modifying drugs within 6 months, immunoglobulins or blood products within 3 months, or planned to use any of these during the study period; were breastfeeding; or had previously received HPV or AS04-based vaccines. Women included in the extension phase were those who completed the full vaccination course during the initial study phase and did not meet exclusion study requirements.

For both phases of the study each participant provided written informed consent, or if below the legal age of consent, assented with written consent from a parent or legal representative prior to study enrolment. Cervical secretion samples were collected from consenting women who volunteered for this additional procedure. For each of the six study centres (three in Germany and three in Poland), each centre's Independent Ethics Committee or Review Board approved the protocol and informed consents. The study was conducted in accordance with Good Clinical Practice, all applicable local regulatory requirements, and the 1996 version of the Declaration of Helsinki. This study is registered with the European Clinical Trials Database.

2.2. Study procedures

Study visits were scheduled at Months 0, 1, 2, 6, 7, 12 and subsequent extension follow-up visits at Months 18 and 24. Study personnel were allocated individual treatment numbers using a central internet-based system at the investigator site. The treat-

ment numbers using this system were assigned according to subject number and age. Enrolment was stratified into three equally sized age strata: 15–25, 26–45 and 46–55 years of age. The second age group (26–45 years of age) was also stratified into equally sized age strata: 26–35 and 36–45 years of age.

Three doses of HPV-16/18 AS04 vaccine (*Cervarix*TM, Glaxo-SmithKline Biologicals, Rixensart, Belgium) were administered to women in the initial study phase at Months 0, 1 and 6. Each dose contained 20 µg each of HPV-16 and HPV-18 L1 protein VLPs adjuvanted with AS04 (50 µg 3-O-desacyl-4'-monophosphoryl lipid A [MPL] and 500 µg aluminium hydroxide). The vaccine was supplied in individual 0.5 mL pre-filled syringes and administered into the deltoid muscle.

2.3. Evaluation of Immunogenicity

Serum samples were collected at Months 0, 2, 7, 12, 18 and 24. HPV-16 and HPV-18 antibodies were measured using a type-specific enzyme-linked immunosorbent assay (ELISA) [14,19]. ELISA titres were calculated from a reference curve, generated from a reference pool of serum samples from human vaccinees, using a four-parameter logistic equation from *SoftMax*TMPro (Molecular Devices, Sunnyvale, CA, USA) and were expressed as EU/mL. Titres were obtained by averaging the values from all dilutions that fell within the working range of the reference curve. Seropositivity was defined as a titre of greater than or equal to 8 ELISA units/mL (EU/mL) for HPV-16 and 7 EU/mL for HPV-18. Women participating in the studies were not screened at entry based on their serological status. The geometric mean titres (GMTs) resulting from natural infection were determined by testing pre-vaccination blood samples obtained from women HPV-16 or 18 seropositive and DNA negative in another ongoing HPV vaccine efficacy study that demonstrated prevention of high-grade cervical intraepithelial lesions [19].

Cervicovaginal secretion (CVS) samples were collected at Month 24 using Merocel[®] ophthalmic sponges (Merocel[®] Eye Spear, Medtronic, Jacksonville, FL, USA) to measure anti-HPV-16 and 18 antibodies. To minimise blood contamination, collections were performed at least 2–3 days after completion of menstrual flow. The sponges were placed in contact with the cervix (cervicovaginal) for approximately 30 s to absorb any mucus. Two sponges were collected per woman and stored at –20 °C until antibody extraction.

The extraction protocol has been previously described by Castle et al. [20]. The only variation to the protocol was that CVS samples weighing <10 mg were excluded. To reduce any bias in mucosal IgG assessment that may be introduced by blood contamination due to sample collection, the presence of blood in CVS-extracted samples was determined using Hemastix[®] (Siemens Medical Solutions Diagnostics Europe Ltd., Dublin, Ireland), which measures the concentration of erythrocytes based on the peroxidase activity of haemoglobin. An aliquot of extracted sample (5 µL) was placed onto the Hemastix[®] and, based on internal validation, we selected samples showing no more than 25 erythrocytes per µL for antibody assessment.

Anti-HPV-16 and anti-HPV-18 IgG antibodies in the CVS-extracted samples were detected and quantified according to ELISA serum standardised protocols as previously published [19]. CVS extracted samples were serially diluted, starting from a 1/10. The final antibody titre for each CVS-extracted sample was calculated by multiplying the ELISA titre by the dilution factor obtained during the antibody extraction step.

To normalise hormonal fluctuations related to ovulation or menstruation [18], a methodology was developed to standardise HPV specific antibodies. The measured total IgG in serum and CVS samples was used to calculate a ratio based on the HPV-16 or HPV-18 IgG

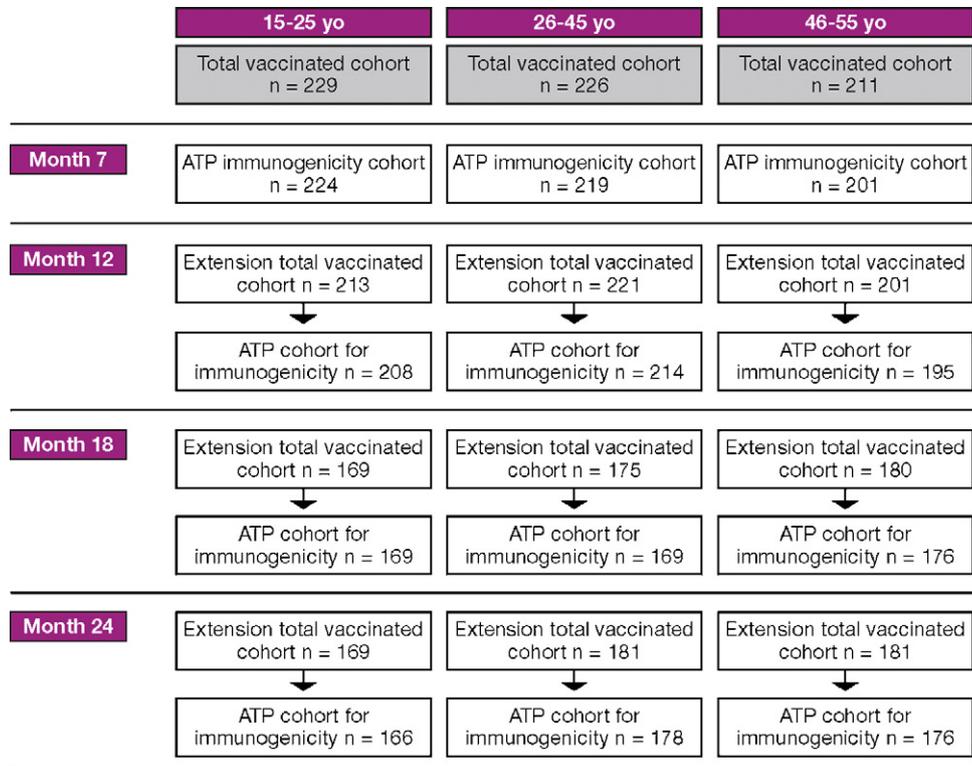


Fig. 1. Study design. Per study protocol, subjects who missed the first visit of the extension protocol (visit 7, Month 18) were still eligible to join the next scheduled visit (visit 8, Month 24). yo = years old.

titre divided by total IgG concentration. The results were stratified according to age group.

2.4. Evaluation of tolerability

After each vaccine dose, all study participants recorded solicited local and general symptoms on diary cards within a 7-day follow-up period (Days 0 through 6), and unsolicited symptoms within 30 days after vaccination. Solicited local adverse events (AEs) included pain, redness, and swelling at the injection site. Grade 3 solicited AEs were defined as pain that prevented normal activity, redness or swelling larger than 50 mm. Solicited general AEs included fever, headache, fatigue, gastrointestinal symptoms (e.g. nausea, vomiting, diarrhoea, abdominal pain), arthralgia, myalgia, rash, and urticaria. Grade 3 solicited general symptoms included fever higher than 39.0 °C (axillary temperature), urticaria distributed on at least four body areas, or events that prevented normal everyday activities. Information was collected on serious AEs (SAEs), medically significant AEs, new onset of chronic disease (NOCD), and pregnancies during the entire study period. A SAE was defined as any untoward medical occurrence that resulted in death, was life-threatening, required hospitalisation, resulted in disability or incapacity, was an important medical event or was a congenital anomaly/birth defect in the offspring of a study subject. Medically significant conditions were defined as AEs prompting emergency room or physician visits that were not related to common diseases or routine visits for physical examination or vaccination or SAEs not related to common disease. NOCDs included auto-immune conditions, allergies and asthma.

2.5. Statistical analysis

The study was powered to demonstrate non-inferiority, in terms of seroconversion, of the 26–45 years age group versus the 15–25

years age group for both HPV-16/18 antigens at the Month 7 visit. It was estimated that a total of 172 women per age category (15–25, 26–45, and 46–55 years) would be needed for the evaluation of primary and secondary endpoints at final analysis for each antigen. Two hundred and twenty women were enrolled per age group based on the assumption that 20% of the participants would be non-evaluable (10% drop-out rate and 10% initially seropositive for both HPV-16 and HPV-18).

For statistical analysis of primary and secondary non-inferiority endpoints in the initial study phase, with respect to seroconversion rates, two-sided 95% confidence intervals (CI) were computed [Proc StatXact Version 5.0, Cytel Inc., Cambridge, MA, USA] of the difference between the percentages of participants who seroconverted after administration of the HPV-16/18 AS04 vaccine in the 26–45 and 46–55-year-old age groups versus the 15–25-year-old age group. If the upper limit of the CI was below the predefined clinical limit of 10%, we concluded that non-inferiority was met.

GMTs (95% CI), antibody titre range and distribution were calculated for each antigen in all age groups at Months 0, 7, 12, 18 and 24.

Seroconversion rates were calculated for serum for each antigen in all age groups at each available timepoint.

All analyses included women from the defined total vaccinated cohort (TVC) or from the according-to-protocol (ATP) cohort (Fig. 1). The TVC included all participants who received at least one dose of vaccine.

The TVC cohort was used for analysis of the CVS samples in women with available samples. The \log_{10} values of the ratios calculated for both CVS and serum samples were used to establish the correlation coefficient using the Pearson coefficient between pairs of CVS and serum samples of each woman.

The ATP analysis for immunogenicity included all participants for whom data were available for at least one study vaccine antigen component after vaccination, and those who met all eligibility crite-

Table 1
Baseline serological status prior to vaccination (total vaccinated cohort).

| Baseline seropositivity status (n, %) | 15–25 Years old, N = 224 | 26–35 Years old, N = 226 | 46–55 Years old, N = 211 | Total, N = 666 |
|--|--------------------------|--------------------------|--------------------------|----------------|
| Neither HPV-16/18 | 189 (82.9) | 153 (68.0) | 136 (65.6) | 479 (72.4) |
| HPV-16 only | 23 (10.1) | 40 (17.8) | 45 (21.5) | 108 (16.3) |
| HPV-18 only | 10 (4.4) | 17 (7.6) | 8 (3.8) | 35 (5.3) |
| Both HPV-16/18 | 6 (2.6) | 15 (6.7) | 19 (9.1) | 40 (6.0) |
| Invalid result for HPV-16 or HPV-18 ^a | 1 (0.4) | 1 (0.9) | 2 (1.0) | 4 (0.8) |

HPV-16/18 refers to single or combined HPV-16 or 18 antigens.

^a Women with unavailable serologic results at baseline for either antigen were excluded from immune response analyses for that antigen.

ria and complied with the study protocol. Immunogenicity analyses were stratified according to three age categories (15–25, 26–45, and 46–55 years of age).

Safety outcomes were analysed in women included in the TVC for both initial and extension phases. Women were eliminated from this analysis if a scheduled vaccination was not administered and the woman continued to participate in the study. All analyses of safety were stratified according to three age categories (15–25, 26–45, and 46–55 years of age).

3. Results

3.1. Study population and baseline characteristics

The initial study phase (Month 0 through Month 12) took place between October 2004 and January 2006. A total of 667 participants were enrolled, of whom 666 received at least one dose of HPV-16/18 AS04 vaccine (Fig. 1). At Month 24, a total of 531 women were included in the extension phase follow-up. The mean age of the participants was 35 years (standard deviation [S.D.] = 12.40) and the majority (99%) of the study participants were White/Caucasian. Over 70% of women evaluated in the ATP cohort were seronegative for HPV-16 and 18 at baseline (Table 1).

3.2. Serological immune response

In all age groups, all initially seronegative women had seroconverted for both HPV-16 and HPV-18 one month after the third dose of vaccine (Month 7). In the ATP analysis, primary and secondary endpoints were achieved as a result of non-inferiority of seroconversion rates; 100% seroconversion in women aged 26–45 years and 46–55 years, compared to 100% seroconversion in women aged 15–25 years. Through Month 24, all women remained seropositive for both anti-HPV-16 and anti-HPV-18 antibodies regardless of their age.

Peak antibody titres in all age groups were observed at Month 7 (Fig. 2). The immunological kinetic profile was similar to those observed in other HPV-16/18 AS04 vaccine clinical efficacy trials with peak GMTs at Month 7 gradually decreasing to Month 18, followed by a plateau reached at Month 24 (Fig. 2) [14,19]. An age-dependent decrease in GMTs was observed with increasing age; however, absolute values were high in all age groups.

Peak GMTs at Month 7 in the 46–55 year old age group were 84-fold and 57-fold higher for HPV-16 and HPV-18, respectively, than those elicited after natural infection (29.8 EU/mL and 22.7 EU/mL for HPV-16 and HPV-18, respectively) [19]. GMTs remained 16-fold (HPV-16) and 8-fold (HPV-18) higher than those elicited after natural infection up to Month 24. GMTs at Months 7, 12 and 18 in the 46–55-year-old group were higher or in the same order of magnitude than GMTs for HPV-16 and HPV-18 achieved during the plateau phase (Month 45–50 timepoint) of an efficacy study in women 15–25 years of age [14].

3.3. Mucosal antibody levels

At Month 24 a total of 250 women had CVS tested and 149 women had CVS samples with no more than 25 erythrocytes per μL (Hemastix[®]).

The correlations between the serum and CVS anti-HPV-16 and anti-HPV-18 antibody titres (standardised for total IgG) by age at Month 24 in the TVC are presented in Fig. 3. Correlation coefficients were high regardless of the age group considered, and ranged from 0.73 to 0.90 for HPV-16 and from 0.82 to 0.93 for HPV-18.

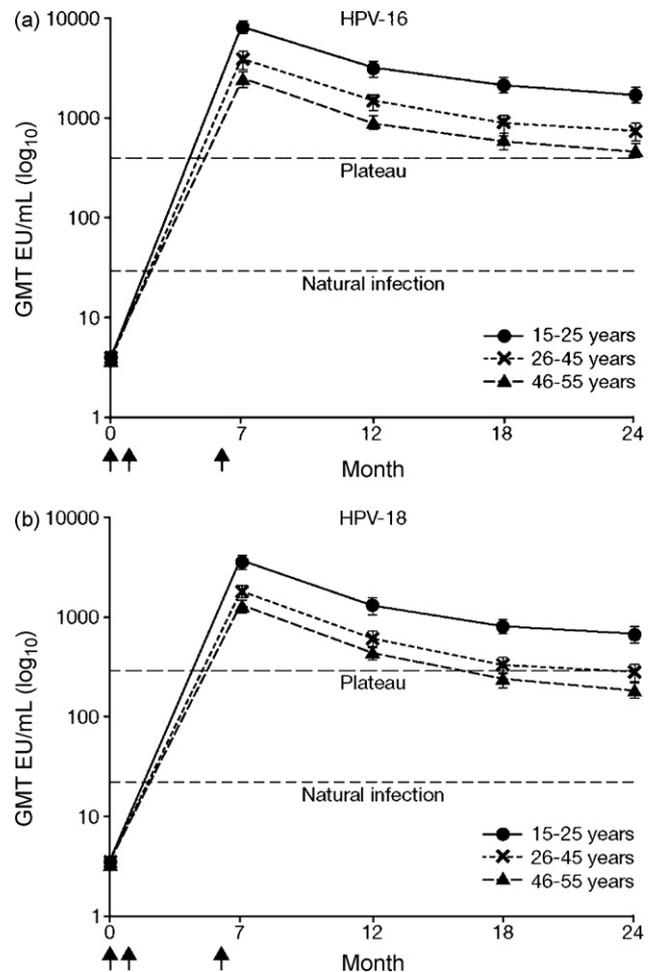


Fig. 2. Antibody levels in women between 15 and 55 years of age (ATP immunogenicity cohort) initially seronegative for HPV-16 or HPV-18 antigens. (a) HPV-16; (b) HPV-18; GMT = geometric mean titre; EU/mL = ELISA units per milliliter. Arrows indicate the vaccination timepoints (Months 0, 1 and 6). Geometric mean titres are shown with 95% confidence interval. Seropositivity defined as ≥ 8 EU/mL for anti-HPV-16 and ≥ 7 EU/mL for anti-HPV-18. Plateau = GMTs at Months 45–50 timepoint of the long-term efficacy study extension phase [14]. Natural infection = GMTs in women seropositive, DNA negative for HPV-16 and HPV-18 from the phase III trial [19].

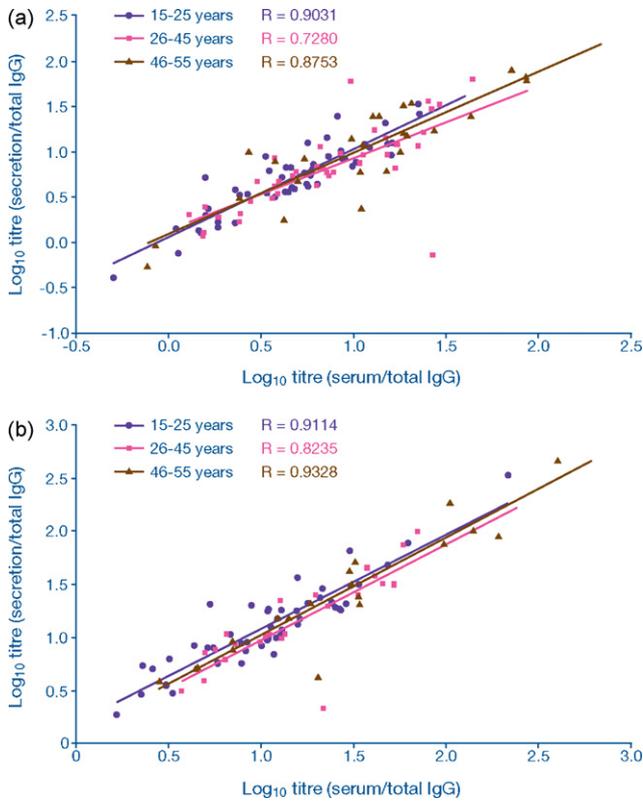


Fig. 3. Correlation of ratio between cervicovaginal secretion and serum samples by age for HPV-16 and HPV-18. (a) HPV-16; (b) HPV-18. Titres were measured using an ELISA assay for detection of anti-HPV-16 and anti-HPV-18 antibodies for both cervicovaginal secretion and serum samples. The scatter plots show the ratio [specific IgG/total IgG] transformed to linear log₁₀ values and plotted.

3.4. Tolerability evaluation

The total number of women evaluated for safety outcomes in the TVC are shown in Fig. 1. Pain of any grade was the most frequently reported injection-site symptom followed by redness and swelling

of any grade (Table 2). Grade 3 symptoms were reported at low frequencies. An age-dependent decrease in the frequency of reporting of local injection-site symptoms was observed for any grade and grade 3 symptoms (not shown). Overall, injection-site symptoms were mild and transient with symptoms lasting for 2–3 days in all women. Fatigue, headache, and myalgia were the most frequently reported solicited general symptoms (Table 2). Grade 3 symptoms were also reported at low frequencies. As observed for solicited local symptoms, an age-dependent decrease was observed in the frequency of solicited general symptoms (not shown). Compliance in terms of administration of doses according to the protocol was excellent in the three age groups, ranging from 98.7 to 99.3%.

Up to Month 24, the incidence of medically significant AEs was similar for all age strata (Table 2). The most frequently reported medically significant AEs (not shown) across all age groups were bronchitis (n = 8), depression (n = 6) and hypertension (n = 5).

Overall, 14 participants reported a total of 15 SAEs from Month 0 to Month 24 (Table 2). One SAE was considered by the investigator to be potentially related to vaccination: a 27-year-old woman was diagnosed with optic neuritis of moderate intensity. The condition developed in the left eye 9 days after the first vaccination dose, the vaccination course was discontinued but the subject remained in the study. The woman was treated with a high dosage of steroids and was free of symptoms four-and-a-half months later.

3.5. Pregnancy

Through the Month 24 visit, 15 pregnancies were reported in study participants. None of the pregnancies started during the vaccination course: two pregnancies were reported from Month 7 to Month 12, seven from Month 12 to Month 18, and six from Month 18 to Month 24. To date, 11 pregnancies have resulted in normal births, one pregnancy is ongoing and two pregnancies were interrupted (one elective and one spontaneous abortion).

4. Discussion

The results presented from this ongoing study are the first to describe the immunogenicity of the prophylactic HPV-16/18 AS04 vaccine in women over 26 years of age.

Table 2

Incidence of solicited local and solicited general symptoms during the 7-day follow-up period as well as medically significant and serious adverse events reported following administration of HPV-16/18 L1 VLP AS04 vaccine, overall per subject (total vaccinated cohort).

| Event | Age range (years) | | |
|---|-------------------|----------------|----------------|
| | 15–25, N = 229 | 26–45, N = 226 | 46–55, N = 211 |
| HPV-014 main study (Month 0 to Month 7 results) | | | |
| Solicited local AEs | | | |
| Pain | | | |
| Any grade n, % | 220 (96.9) | 210 (92.9) | 171 (82.6) |
| Grade 3 n, % | 23 (10.1) | 15 (6.6) | 9 (4.3) |
| Redness | | | |
| Any n, % | 133 (58.6) | 126 (55.8) | 101 (48.8) |
| Grade 3 n, % | 0 (0) | 6 (2.7) | 2 (1.0) |
| Swelling | | | |
| Any n, % | 96 (42.3) | 100 (44.2) | 83 (40.1) |
| Grade 3 n, % | 3 (1.3) | 3 (1.3) | 1 (0.5) |
| Solicited general symptoms n, % | | | |
| Fatigue n, % | 24 (10.5) | 29 (12.8) | 26 (12.3) |
| Fatigue n, % | 128 (56.4) | 116 (51.3) | 81 (39.1) |
| Headache n, % | 123 (54.2) | 100 (44.2) | 82 (39.6) |
| Myalgia n, % | 126 (55.5) | 97 (42.9) | 83 (40.1) |
| HPV-014 extension study (Month 0 to Month 24 cumulative results) | | | |
| Medically significant AEs n, % | 24 (10.5) | 29 (12.8) | 26 (12.3) |
| Serious AEs n, % | 4 (1.7) | 3 (1.3) | 8 (3.3) |

AEs, adverse events.

At study entry and across all age groups, the vast majority of women in this study were HPV-16/18 seronegative at entry (i.e. absence of detectable levels of antibodies prior to vaccination). This is in contrast to the notion that the vast majority of sexually active women have been infected with HPV and therefore must be seropositive. In all age groups, a robust HPV-16 and 18 antibody response was measured at all timepoints up to Month 24 with 100% seropositivity for both antigens in women initially seronegative for anti-HPV-16/18 antibodies. Initially seropositive women tended to have higher immune responses compared to initially seronegative subjects in the 26–45 and 46–55 age groups (data not shown).

Since even with increasing age the majority of women did not have serological evidence of past exposure to HPV types 16 or 18 and since they demonstrated robust immune responses to the AS04-adjuvanted cervical cancer vaccine, it is expected that women above 26 years of age are likely to benefit from HPV vaccination if exposed to HPV 16 or 18 in the future.

In the absence of an identified serological correlate of protection, GMTs in this study were evaluated using other benchmarks that have been previously demonstrated as relevant with respect to protection against virological infections and HPV-related disease. These benchmarks are GMTs achieved in vaccinated women in whom vaccine efficacy has been demonstrated and GMTs measured in women who were able to clear a natural infection (seropositive/DNA negative) [14,19]. As natural infection with HPV does not always confer protection against subsequent infections, it can be assumed that if a correlate of protection is identified, it will be between these two benchmarks. Since re-infection with the same HPV type and co-infection with multiple HPV types have been demonstrated [21–23], the benefit of prophylactic HPV vaccination may be considered relevant for all women, including those over 26 years of age.

Similar to data generated with other types of vaccines, lower GMTs were observed with increasing age in this study [24]. The HPV-16/18 AS04 vaccine induced a high level of antibodies in women 46–55-years of age up to the last reported timepoint (Month 24), which were several fold higher than those elicited after natural infection with HPV-16 and HPV-18. At Month 24, GMTs in women 26–55 years of age in this study were in the same range as GMTs achieved at the plateau level in the long-term (6.4 years) follow-up efficacy study [25,26]. In the oldest age category, the anti-HPV-18 GMT value was slightly lower than this plateau level but well above the natural infection level. Overall, we can conclude that the HPV-16/18 AS04-adjuvanted vaccine is likely to induce high antibody levels in an age group that was shown to have somewhat lower antibody levels (while vaccine responses are 100%) with increasing age. These high levels of vaccine-induced antibodies are sustained over time and well above those induced by natural infection alone.

HPV infection occurs at the cervix and local immunity plays a key role in whether the viral infection will be cleared or will persist, which may result in the development of cervical neoplasia [16,18]. One mechanism in which L1 VLP-based HPV vaccines are thought to provide protection against cervical infections involves the induction of serum neutralising antibodies that transudate across the cervical epithelium in sufficiently high concentrations to bind to HPV virus particles. Vaccine-induced antibodies found in mucosal secretions may prevent new infections, re-infection at another site in the cervical, vaginal or vulvar region, or reduce the viral load of viral particles shed from an active infection. In the long-term efficacy trial, an observed correlation between total IgG (as measured in this study by ELISA) and neutralising antibodies (as measured by pseudovirion neutralising assay) has been confirmed up to 5.5 years, suggesting that antibodies generated by the HPV-16/18 AS04

vaccine are neutralising in the serum and in the mucosal secretions (personal communication).

The current study shows that the HPV-16/18 AS04-adjuvanted vaccine induces a high level of total HPV-16/18 IgG antibodies in the serum and at the cervix up to 24 months after the first dose of vaccine. The levels of HPV-16 and HPV-18 antibodies in the CVS were highly correlated with antibody levels detected in serum samples in all age groups studied. Half of the women enrolled in the oldest age group were post-menopausal (for at least 1 year) and were therefore expected to produce significantly less CVS; however, a high correlation between CVS and serum antibodies in the 46–55-year-old age group was still observed. These results indicate that serum IgG antibodies likely transudate to the cervical epithelium, regardless of age, conferring site-specific immunity at the cervix. The combination of sustained HPV-16 and 18 antibody levels in the serum and cervical mucosa suggests that transudation is likely to play a role in overall vaccine efficacy.

Clinical studies have demonstrated that the HPV-16/18 AS04-adjuvanted vaccine-induced antibody levels that are at least two times higher than those induced with the same HPV-16/18 VLPs but formulated with conventional aluminium hydroxide [27]. The AS04 vaccine formulation also elicited a more robust memory B cell response, which may contribute to the sustained high level of antibody titres in all age groups studied. As adjuvants are used to enhance the immune response, the AS04 adjuvant in combination with high quality HPV-16 and HPV-18 VLPs could be expected to induce a robust immune response in adolescent girls and young women, in whom there is a high risk of acquiring an infection either shortly after sexual debut, or throughout the lifetime of all sexually active women, that would be maintained over a long period of time regardless of age.

In the present study, the HPV-16/18 AS04 adjuvanted vaccine was well tolerated across all age groups up to 2 years after the administration of the first vaccine dose, and the frequency of AE reports was similar between the three age categories. Other large studies including a control arm have demonstrated that this vaccine and other AS04 containing vaccines are generally safe and well tolerated [13,14,19,28–30].

Considering the increasing risk of HPV infections to become persistent and the general decline in immune function with advancing age, HPV vaccination for the prevention of cervical cancer may provide substantial benefit to women up to 55 years of age. Further studies are underway to extend our findings and to evaluate vaccine efficacy for the prevention of HPV-related disease and related cervical outcomes.

The HPV-16/18 AS04 vaccine was immunogenic and generally well tolerated in 15–55-year-old women. The vaccine was able to induce a high-level and sustained immune response in women over the age of 26 years for both HPV-16 and 18 for 2 years after the first vaccine dose. One hundred percent seropositivity was achieved at each timepoint through Month 24. Twenty-four months after the first vaccine dose HPV-16 and HPV-18 antibody levels in women older than 26 years of age reached a plateau that was comparable to the plateau of GMTs observed in women 15–25 years of age in an efficacy study over 6.4 years [14,26]; antibody levels were several fold higher than those associated with natural infection titres [19]. Furthermore, high levels of antibodies were also detected at the cervix for up to 2 years after the first vaccine dose, providing a basis for site-specific vaccine-induced immunity.

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