SCIENTIFIC DISCUSSION

1. Introduction

Human papillomavirus (HPV) infection is currently the most common sexually transmitted disease worldwide. By 5 years after sexual debut, ~50% of young women will have been infected with at least one of the 40 HPV types that preferentially infect the genitals. Thirteen of these HPV types are highly carcinogenic. Although a consistent picture of the epidemiology and pathogenesis of genital infections in women has developed during the past two decades, less is known about these infections in men. However, studies suggest a similar infection pattern in men, who are the most important vectors for transmission of HPV disease to women. The peak incidence of HPV infection occurs in young adults between the ages 16 and 23 years.

Human papillomaviruses are double-stranded DNA viruses that infect epithelial cells and are significantly associated with low-grade cervical intraepithelial neoplasia (CIN), genital condyloma and cervical cancer. HPVs are judged to be the primary cause of cervical cancer, which is the second most common type of cancer causing deaths in women worldwide. Other HPV-related cancers in young women include vulvar and vaginal cancers, which are preceded by dysplastic lesions (vulvar intraepithelial neoplasia (VIN) and vaginal intraepithelial neoplasia (VaIN). In men anal cancer is the most common HPV-related cancer. The virus is also related to penile and certain oropharyngeal cancers. Other benign HPV-associated conditions include condyloma acuminata (genital warts) located in the genital or perianal region and juvenile recurrent respiratory papillomatosis (JRRP) primarily located in the larynx. JRRP is thought to occur by transmission of the virus from an infected mother to her child.

HPV types are classified into different categories based on their association with cancer in humans. Epidemiological studies on the prevalence of HPV types in cervical cancer show some geographical differences. However, all over the world the majority of cervical cancers are related to two types, HPV 16 (~55%) and HPV 18 (~15%). The contributions of HPV 16 and HPV 18 to high-grade CIN and to HPV-related vulvar, vaginal and anal cancers are similar to those found in cervical cancer. HPV 6, 11, 16 and 18 together cause ~35% of CIN 1 cases. HPV 6 and HPV 11 cause approximately 90% of genital warts and RRP and 10% and 20% of CIN 1 lesions, respectively, but are not associated with cervical or anal carcinoma. These data provide the basis for the selection of the four HPV types 6, 11, 16 and 18 in the current HPV vaccine.

Mechanism of action

There are no animal models for human papillomavirus infection. In animal models vaccination with LI Virus-like Particles (VLPs) derived from species-specific papillomaviruses protected against acquisition of infection and disease. Successful passive transfer experiments have also been performed suggesting that type-specific antibody responses are virus-neutralizing. The quadrivalent HPV vaccine was developed based on these animal data that suggest that a systemic neutralizing anti-HPV response by vaccination with type-specific HPV L1 VLPs result in protective immunity against type-specific HPV infection and disease.

The vaccine also elicits cell-mediated responses as detected by in vitro stimulation of PBMCs, Th1 and Th2 cytokines and immunoglobulin subclasses. The LI responses are not expressed in the cellular department and therefore it is not expected CTLs will assist in vaccine-induced protective immunity. The induced T-cell responses may play an important role in establishing long-lived B-cell immune memory.

The exact role of various immune mechanisms in the protective efficacy of the HPV L1 VLP vaccine remains to be determined. It is believed that the vaccine provides protection by inducing type-specific antibodies that interfere with transmission by binding to and neutralizing contaminating HPV prior to entry into basal cells.

Sanofi Pasteur MSD SNC submitted a Marketing Authorization Application for Gardasil in accordance with Article 8.3(i) of Directive 2001/83/EC as a complete application.

The approved indications are as follows: “Gardasil is a vaccine for the prevention of high-grade cervical dysplasia (CIN 2/3), cervical carcinoma, high-grade vulvar dysplastic lesions (VIN 2/3), and external genital warts (condyloma acuminata) causally related to Human Papillomavirus (HPV) types 6, 11, 16 and 18. The indication is based on the demonstration of efficacy of Gardasil in adult females 16 to 26 years of age and on the demonstration of immunogenicity of Gardasil in 9- to 15-year old children and adolescent. Protective efficacy has not been evaluated in males”.

2. Quality aspects

Introduction

Gardasil contains virus-like particles (VLPs) of the recombinant major capsid (L1) protein of HPV types 6, 11, 16, and 18 as active substance. The recombinant proteins forming the VLPs are produced by separate fermentation in recombinant *Saccharomyces cerevisiae.* VLPs of each type are adsorbed on amorphous aluminium hydroxyphosphate sulfate adjuvant, and the formulation also includes sodium chloride, L-histidine, polysorbate 80, sodium borate, and water for injection. The final product is presented as a sterile suspension either in a single-dose vial or in a prefilled syringe for intramuscular injection.

Active Substance

The drug substance consists of the four Monovalent Bulk Adsorbed Products (MBAPs), one for each of the four human papillomavirus (HPV) types included in the final product. The active components in each MBAP are virus-like particles (VLPs) made up of the recombinant major capsid (L1) protein for that HPV type, produced in recombinant *S. cerevisiae*. L1 is the major structural protein of the human papillomavirus viral capsid.
Native papillomavirus virions have an icosahedral symmetry consisting of 72 pentamers of L1 protein, and are nearly spherical with an approximate diameter of 60 nm. The pentamers are stabilized by intra- and inter-L1 disulfide bonds, and there is also evidence of interpentamer disulfide bonds stabilizing the capsid. The HPV VLPs mimic the structure of the virion capsid.

- **Manufacture**

Genetic Development

The pGAL110 yeast expression vector was used for expression of all four HPVL1 proteins. Differences in the cell substrates among the HPV types were introduced when cloning the particular HPVL1 open reading frame (ORF), when inserting the ORF into the pGAL110 vector, and when transforming the yeast host strain with the resulting vector. Plasmid or phage libraries were constructed from DNA obtained from human clinical specimens or cell lines positive for HPV types 6, 11, 16, or 18. The L1 genes were derived by a direct cloning protocol. However, the coding sequence for HPV11L1 was synthetically rebuilt based on HPV6L1 nucleotide sequences that supported good VLP expression in yeast and that were appropriately changed to encode the HPV11L1 polypeptide. Following sub-cloning of either the whole HPV genomes or parts thereof into standard cloning vectors, polymerase chain reaction (PCR) was used to subclone the L1 genes into the yeast expression vector pGAL110, which contains the divergent yeast GAL1-GAL10 promoter and the yeast ADH1 terminator (ADH1t) for transcription termination and polyadenylation.
The pGAL110-related yeast expression vectors for each of the four HPV types were used to transform spheroplasts of the recombinant *S. cerevisiae*. Transformations were conducted independently for each of the HPV types by a published method.
Based on analyses of HPV VLP expression productivity, one high-producing isolate of each type was selected as the source seed for preparation of a premaster seed.

Cell Banking

<u>Master Cell bank (MCB)</u>

Four cell substrates were derived from *Saccharomyces cerevisiae*, which were transformed by pGAL110 based yeast expression vectors, each containing the gene of interest coding for HPVL1 types 6, 11, 16, or 18 respectively. A clone for each type was selected to establish the four MCBs.
All cGMP master seed stocks were prepared using a two-stage expansion. Optical density, pH, and residual dextrose were monitored to assure consistency of growth among all cGMP seed stocks.
A series of characterization tests have been performed on the MCB to verify culture purity, species identity, host strain identity, viable count, plasmid identity and integrity by restriction mapping, DNA sequencing of the L1-encoding genes and junctions, and specific productivity.

Working Cell bank (WCB)
Master seed vials are used to inoculate the HPV Shakeflask Medium. Future working seeds will be manufactured using the same process steps and similar equipment used for the original working seed. Following freezing, seed supplies are stored at ≤-60 °C.

End of Production Cells (EOP)
The genetic stability has been analysed for one full-scale fermentation for each type of end of production cells (culture purity, species identity, host strain identity, restriction endonuclease mapping, plasmid retention and DNA sequencing).

Fermentation

The manufacturing process of the drug substance consists of two main steps: 1) fermentation and harvest of the recombinant yeast cell slurry, and 2) purification of the VLPs and adsorption of the purified VLPs onto aluminium-containing adjuvant to form the MBAP.

The fermentation process consists of a seed fermentation and a production fermentation.
During the seed fermentation process all HPV types use the same Seed Fermentation Medium. Culture growth and dextrose utilization are monitored. Upon completion of this stage, the seed culture is transferred to the production fermentor, which contains the HPV Production Fermentation Medium.
Following fermentation, the cells are then harvested by microfiltration to produce the cell slurry, which is dispensed and frozen for storage.

Purification

The downstream processing is initiated by thawing the frozen cell slurry and releasing the VLPs from yeast cells by homogenization/microfiltration run. The cell lysates are then incubated. The VLPs are purified by cross-flow membrane filtration, chromatography and ultrafiltration. The final steps in the purification process for all four types are buffer exchange and sterile filtration to produce the final aqueous product (FAP). The FAP for each type is then adsorbed onto amorphous aluminium hydroxyphosphate sulfate to produce the MBAP. The MBAP for each type is then filled into bulk storage containers and stored at 2-8°C.
The amorphous aluminium hydroxyphosphate sulfate adjuvant used in the manufacture of the MBAP is manufactured Merck & Co. at the same site as the drug substance. The adjuvant is manufactured by adding sodium hydroxide to a solution of aluminium potassium sulfate and collecting the precipitate.

Process control and validation

The validation studies were executed according to prospectively approved protocols.
Selection of Critical process parameter (CPP) and Critical quality attribute (CQA) and their respective ranges were established based on data at lab, pilot, and full scale, including data from laboratory experiments specifically designed to test process parameter ranges. Some of the CPPs and CQAs were later defined as “critical controls”.

Separate validation studies for filters and column sanitization and reuse are presented. Validation of the aseptic processing is described. Bulk media challenges have been performed by simulating all steps in the manufacturing of the MBAP, including holding times, after the sterile filtration. After the initial qualification consisting of three consecutive media challenges, the aseptic process is re-qualified by one yearly media challenge.

Manufacturing process development

Initial clinical trials focused on monovalent Type 11 or 16 VLP vaccines, because Type 11 had the only known assay for neutralising antibody and type 16 is the HPV type predominantly associated with cervical cancer. Type 6 and 18 VLPs were added later.
Process development proceeded in parallel with clinical development, culminating in the Final Manufacturing Processes (FMPs), which were validated. Most clinical trials of the quadrivalent vaccine (trials No. 011, 012, 015, 016, and 018) used the final manufacturing process for both fermentation and purification.

Elucidation of structure and other characteristics

Testing was carried out on the Final Aqueous Product (FAP), Monovalent Bulk Adsorbed Product (MBAP), and in some cases, intermediate. The characterization testing was performed on a minimum of three full-scale lots per HPV type for each assay.

Monomers were analyzed by peptide map (LysC + AspN) followed by MALDI-MS, Reduced SDS-PAGE (intact monomer), Isoquant (deamidation), free thiol groups, circular dichroism, and FT-IR spectroscopy. Size exclusion chromatography was used to analyse oligomers of L1 proteins. No evidence has been found of N-linked glycosylation.

Virus-like particles were analyzed by CD and FT-IR spectroscopies, Transmission Electron Microscopy (TEM), Cryo-electron microscopy (cryoEM), Dynamic Light Scattering (DLS), epitope mapping, inhibitory concentration at 50% response (IC50) by competitive ELISA and affinity of monoclonal Antibody (mAb) to the different epitopes.
Epitope mapping was performed on the different HPV types prior assembly and after reassembly (FAP) using a surface plasmon resonance (SPR) technique with a panel of monoclonal antibodies (mAbs) that recognizes conformational or linear epitopes.
Adsorbed VLPs (MBAP) were analyzed by differential scanning calorimetry (DSC), mouse potency, and In-Vitro Relative Potency (IVRP). It has been concluded that adsorption to the aluminium adjuvant does not result in significant changes in VLPs structure.
The mouse potency assay was performed on BALB/c mice in which anti-HPV antibodies were measured using an ELISA based assay four weeks post-immunisation. This in vivo potency assay was used to establish the immunogenicity of MBAP in mice and the assay was used to release early clinical lots and to characterise the stability of MBAP.
The IVRP assay is the proposed potency assay for release. It is a sandwich-type ELISA used to measure the antigenicity of drug substance and was shown to be specific for the four types.

Impurities

Non-L1 protein impurities originated from yeast host cells were analyzed by SDS-PAGE, Western blot, and protease activity. These assays are typically performed on FAP. Purity results show successive clearance of protein impurities through the process.

- Specification

The tests applied for release of the cell slurry are as follows: culture purity and host strain ID.
The release specifications for drug substance (MBAP) include tests for protein concentration, percent purity (SDS-PAGE), percent intact monomer (SDS-PAGE), in vitro relative potency (ELISA), identity, sterility, endotoxin, aluminium and pH. Satisfactory validation of the analytical procedures has been performed in accordance with current ICH guidelines.
The drug substance batch analysis was provided for at least 4 batches of each type produced with the final manufacturing process. The tests parameters applied are the ones retained in the drug substance specification, completed with the analysis of polysorbate 80 for FAP, and completeness of adsorption, freezing point, mouse potency and calculated protein concentration for MBAP.

The proposed IVRP limits are based on the variability of the manufacturing process and the variability of the analytical method, and they represent the lower bounds on IVRP values expected to be observed at release under routine manufacturing and testing conditions.

The reference standard used for the routine IVRP testing is a batch of drug product (quadrivalent final container product: QFCP) stored at 2-8 °C. In addition to this working standard, four FAPs, one per type, have been implemented as primary standards and stored at -60°C.

- Stability

Stability studies were performed on cell slurry (one batch per type) and three full-scale lots of MBAP per type under long-term storage conditions (2-8 °C) and on MBAP under accelerated conditions at 23-27 °C. An additional study was initiated using one lot of MBAP per type manufactured by the full manufacturing process. Results of stability studies performed on cell slurry support the proposed holding time for cell slurry when stored frozen.
For the MBAP, results are provided on three lots per HPV type stored at 2−8 °C. No change in physical appearance, pH, or completeness of adsorption is observed. A certain degree of variability is observed by IVRP, mouse potency and percent intact monomer, but no clear and consistent degradation trend was observed for all types.
All tests in the release specification for drug substance were included in the stability studies, except for protein concentration, percent purity, identity, and aluminium. In addition, the following tests were included: physical appearance, completeness of adsorption, and mouse potency.

Finished Product

The vaccine is a sterile liquid suspension prepared from the Type 6, Type 11, Type 16, and Type 18 Monovalent Bulk Adsorbed Products (MBAPs) combined with a histidine buffer and a suspension of the aluminium-containing adjuvant. It is filled into single-dose vials or syringes with a minimum recoverable volume of 0.5 mL.

- Pharmaceutical Development

Formulation development

The early clinical studies were made with monovalent vaccines, however, the majority of the studies on quadrivalent vaccine (011, 012, 015, 016, 018) were all made with the final formulation described in this application.
The choice of adjuvant was based on preclinical studies showing that vaccine formulations containing this adjuvant induce substantially higher anti-HPV responses than vaccine formulations without adjuvant. The other excipients and their respective concentrations were selected based on stability considerations.

Manufacturing process development

The most significant changes to the manufacturing process were changes to the filling process from a manual to an automated process (Type 6 study, Protocol 004) and introducing the quadrivalent formulation (Quadrivalent study, Protocol 007). Only minor modifications were made upon transfer from the research pilot facilities to full manufacturing scale.

There are no overages. An overfill is provided to ensure that a minimum recoverable volume of 0.5 mL/dose is achieved.

- Manufacture of the Product

The manufacturing process consists of two main steps: formulation and aseptic filling.
The Quadrivalent Bulk Adsorbed Product (QBAP) formulation is prepared from six sterile ingredients: histidine buffer, adjuvant diluent, and the four MBAPs (Types 6, 11, 16, and 18). The

QBAP is then mixed to ensure homogeneity, aseptically sampled to test sterility and aluminium concentration. The portable formulation tank is maintained under positive pressure before it is transferred to the appropriate fill line for subsequent filling.
Aseptic filling and stoppering of QFCP in vials or syringes occurs in a barrier isolator system. Before filling, homogeneity is ensured by mixing the QBAP by agitation and recirculation. The QBAP is aseptically filled into vials or syringes such that each vial or syringe contains a minimum recoverable volume of 0.5 ml. After each vial or syringe is filled, it exits the isolator and is inspected for defects. Quality control samples for in vitro relative potency (IVRP), identity, sterility, pH, endotoxin, aluminium, package identity, and recoverable volume are taken during the fill. After inspection, the vials are placed into trays and stored at 2-8°C until they are labeled and packaged.

Process Controls and validation

For the purpose of process validation, six formulation lots were manufactured to prepare three lots of QFCP in vials and three lots of QFCP in syringes. All validation lots satisfied the validation criteria and met all release tests for both QBAP and QFCP.

Control of Excipients

The vaccine contains the following excipients: amorphous aluminium hydroxyphosphate sulfate adjuvant, sodium chloride, L-histidine, polysorbate 80, sodium borate and water for injection.
The adjuvant amorphous aluminium hydroxyphosphate sulfate is a non-compendial insoluble aluminium based precipitate to which the VLPs are adsorbed. This adjuvant has been used in HPV VLP vaccine clinical lots and in four other licensed vaccines manufactured by Merck & Co., Inc.
All other excipients comply with the Ph.Eur. None of the excipients are of animal origin, and no animal-derived materials are used in the manufacture of any excipient.

- Product Specification

The specifications for drug product include tests for in vitro relative potency (IVRP, same assay as for drug substance), identity, sterility, endotoxin, aluminium, pH, package identity, recoverable volume and syringeability (syringes only).
The acceptance criteria for IVRP at release and end-expiry for the drug product take into account process capability, as well as assay variability and stability data. The results from a clinical substudy of Protocol 016 (designed to place in context these limits with respect to immunogenicity) support the proposed limits, all of which are the lowest dose evaluated in clinical trials.
An upper IVRP specification for individual type is not proposed; a limit has been established for the Total IVRP and is a safety limit derived from the highest dose used in clinical studies (Clinical Protocol 007).

Batch analysis data for both the Quadrivalent Bulk Adsorbed Product (QBAP) and the Quadrivalent Final Container Product (QFCP) in vials and in syringes are provided and analyzed according to the drug product specification.

The reference standard used for routine testing is a quadrivalent final container product (QFCP) lot, which is referred to as the working standard. The same standard is used for both IVRP and completeness of adsorption. The same reference standard is used to test the drug substance lots.

- Stability of the Product

The proposed shelf-life for both container type is considered acceptable. No difference in the product stability profile has been observed among the different final contained images, including both vials and syringes. No change in physical appearance, sterility, endotoxin, pH, or completeness of adsorption is anticipated during the proposed shelf-life.

A photostability study was performed to assess the stability of the drug product to cool, white light and UV radiation.

- Adventitious Agents

Nonviral Adventitious Agents (TSE aspects)

D-galactose, which is used in the fermentation culture medium, is the only raw material identified as being of ruminant origin using in the manufacturing process. D-galactose is obtained from bovine milk sourced from healthy animals in the same manner as milk for human consumption, and no other ruminant material are used in its production. It is thus compliant with the EMEA *Note for Guidance on Minimising the Risk of Transmitting Animal Spongiform Encephalopathy Agents via Human and Veterinary Medicinal Products* (EMEA/410/01 rev 2, July 2004). Other materials used in the purification process, and four amino acids, used in the HPV fermentation culture media, are products of microbial fermentation where the fermentation media contained material of animal origin.

Viral Adventitious Agents

There are no live viruses and no cell lines of human or animal origin used in the manufacture of Quadrivalent HPV VLP Vaccine.

2. Non-clinical aspects

Introduction

There is no animal model for human papillomavirus infection, however there are other species-specific papillomaviruses that have been studied in animal models. Cottontail rabbit papillomavirus (CRPV) induce tumours, which are cutaneous rather than mucosal. Canine oral papillomavirus (COPV) infects and induces lesions at a mucosal site (oral mucosa). These studies with species-specific papillomaviruses have demonstrated the possibility to vaccinate against infection and development of tumour lesions using virus-like particles formed by recombinant viral capsid proteins (Breitburd et al 1995, Jansen et al 1995, Suzich et al 1995) and have been used as models for vaccine development.

African green monkeys were immunised with HPV-11 VLPs. Sera from immunised monkeys neutralised HPV-11 in an ex vivo model for HPV infection (human foreskin tissue was infected with HPV and then implanted into athymic mice). Significant levels of HPV-11-neutralising antibodies were observed in cervicovaginal secretions (Lowe et al 1997).

GLP

Pharmacology studies were not performed under GLP conditions. All 5 toxicology studies were carried out under GLP conditions.

Pharmacology

- Primary pharmacodynamics

A series of studies which were performed to determine the immunogenicity of the monovalent HPV 6, 11, 16 and 18 L1 VLP vaccines and/or the quadrivalent HPV 6, 11, 16 and 18 L1 VLP vaccine (Gardasil) in non-human primates were submitted. The monovalent L1 VLP and quadrivalent Gardasil vaccine formulations tested in these studies were made by the same manufacturing process as the GMP lots and are, therefore, similar to lots that were used in the clinic. Monovalent HPV VLP vaccines or Gardasil were administered by intramuscular injection on 3 or 4 occasions during the course of 52 weeks followed by an immunogenicity assessment of antibody titres raised against the relevant HPV types. Studies were conducted in rhesus macaques, chimpanzees, and African Green monkeys.

In each of the 3 non-human primate species tested, the intramuscular administration of Gardasil, or it's monovalent components, was found to elicit immune responses resulting in production of antibodies against the HPV VLP types present in the vaccine. These studies showed that Merck's aluminium adjuvant is necessary to induce an increased immune response against the vaccine antigens. Serum

antibodies to all 4 HPV types were shown to neutralize pseudovirus infection of a cervical carcinoma cell line (C33A) indicating the potential of HPV VLP vaccines to protect against HPV infection. High titres of total IgG are induced in addition to noteworthy IgA levels and measurable IgM, IgG1, and IgG4. The isotype of the antibodies were indicative of a TH2 response.

- Safety pharmacology programme

Safety pharmacology studies were not performed. However in the toxicological studies safety daily monitoring for physical signs did not reveal any notable respiratory problems. This approach is in accordance with Note for guidance on preclinical pharmacological and toxicological testing of vaccines (CPMP/SWP/465/95).

- Pharmacodynamic drug interactions

Such studies are not required according Note for guidance on preclinical pharmacological and toxicological testing of vaccines (CPMP/SWP/465/95).

Pharmacokinetics

Experimental studies to demonstrate absorption, distribution, metabolism, and excretion of the active ingredients in Gardasil have not been performed for any of the component viruses. This is in line with Note for guidance on preclinical pharmacological and toxicological testing of vaccines (CPMP/SWP/465/95).

The Merck Aluminium Adjuvant (aluminium hydroxyphosphate sulphate adjuvant) is used in other vaccines, which are approved in Europe, and it is agreed that no further studies on the adjuvant are required according Guideline on adjuvants in vaccines for human use (CHMP/VEG/134716/2004).

Toxicology

- Single dose toxicity

Single-dose toxicity of Gardasil was assessed in 2 GLP studies in mice and rats. The dose administered intramuscular represents approximately 1200-fold excess in mice and 300-fold excess in rats of the human dose. The vaccine was well tolerated and there were no treatment-related effects on mortality, physical signs, or body weight over a 14-day observation period.

- Repeat dose toxicity (with toxicokinetics)

A 10-week repeat-dose toxicity study was performed in BALB/c mice. Vaccine-treated mice received 1 or 3 doses of Gardasil at a concentration of 160/160/80/160 µg/ml of the HPV types 6/11/16/18 L1 VLPs. Control mice received 1 or 3 doses of Merck's aluminium adjuvant placebo control. For each dose, approximately 50 µl of the vaccine or placebo control was administered. On a body weight basis, the dose of the vaccine administered to mice was approximately 1450-fold the projected human dose. All animals were dosed with either vaccine or placebo on Day 1. Necropsy was performed at day 8, 29, 57 and 60.

There were no treatment-related changes in physical signs, body weight gain, food consumption, haematology, or serum biochemistry. There was a treatment-related enlargement of the iliac lymph nodes. Treatment-related microscopic changes were noted in lymph nodes (hyperplasia) and in the muscle at the injection sites (inflammation). Despite the slightly increased severity of the cellular infiltration in the muscle in some vaccine-injected animals, the overall damage at the injection sites was not more severe in these animals as compared to controls.

- Genotoxicity

No genotoxicity studies were conducted, and this is in line with CPMP/SWP/465/95.

- Carcinogenicity

No carcinogenicity studies were conducted, and this is in line with CPMP/SWP/465/95.

- Reproduction Toxicity

The reproductive and developmental toxicity was assessed in a study in female Sprague-Dawley rats addressing all phases of reproduction and foetal development.

An immune response to the vaccine was observed, and antibodies were shown to be transferred to the offspring during gestation and also lactation. There were no adverse findings in the study.

- Local tolerance

Local tolerance was assessed by intramuscular administration of the vaccine in rabbits. Intramuscular administration of Gardasil caused minimal irritation at injection site and the changes observed were similar in severity to those produced by the injection of the Merck aluminium adjuvant as placebo control.

- Other toxicity studies

Two non-GLP exploratory immunogenicity studies were performed in rats. The purpose of these studies was to confirm that Gardasil induced an immune response in the animal model used for reproductive and developmental toxicology testing.

There were no deaths or treatment-related physical signs during the study. All animals mounted specific antibody responses to all 4 HPV VLPs. The immune response was consistent with a typical prime-boost effect. The highest HPV type-specific antibody titres were observed with a regimen which included 3 initial administrations of the vaccine, 3 weeks apart, followed by a fourth administration 10 weeks after the third dose.

A second exploratory immunogenicity study was carried out in order to confirm the immune response in rats. In both exploratory studies, it was demonstrated that the vaccine was immunogenic in rats and that an anti-HPV antibody response was seen for each HPV type present in the vaccine. These results show that the rat is an appropriate animal model for investigating the potential toxic effects related to administration of Gardasil.

Ecotoxicity/environmental risk assessment

It is anticipated that the environmental impact of the Gardasil vaccine would be minimal to none for the following reasons:

- The vaccine is comprised of Virus-like Particles (VLPs) and therefore does not contain any live or attenuated virus. The VLPs are simply protein sub-units found in the virus capsid.
- The VLP vaccine is not capable of replicating. Therefore, there is no risk of transmission from an immunised host to a susceptible host.
- The VLP vaccine is specific for humans and will not impact the organisms of a sewage treatment plant or the effluent receiving stream.

The environmental risk assessment has shown that there is no environmental risk associated with the use of Gardasil.

4. Clinical aspects

Introduction

The clinical development programme to support licensure of the quadrivalent HPV vaccine consisted of 12 clinical studies. Approximately 21,514 subjects (11,813 HPV vaccine/9,701 placebo) were included and vaccinated.

Initial phase I/IIa studies evaluated the immunogenicity and safety of monovalent vaccine precursors in 3,160 study subjects. Monovalent HPV 11 LI VPL vaccine was evaluated in one study (Protocol

001), HPV 16 L1 VPL vaccine in four studies (Protocols 002, 004, 005 and 012) and HPV 18 L1 VPL vaccine in one study (Protocol 006). Anti-HPV responses were followed up for 1.5 to 3.5 years.

The quadrivalent HPV vaccine programme was divided into 2 sets of studies based on the age range of enrolled subjects: 1) efficacy studies were performed in female subjects 16 to 26 years of age and 2) studies to bridge efficacy, immunogenicity and safety in 16- to 23-year old female subjects to younger age cohorts. Seven phase II/III studies evaluated the quadrivalent HPV L1 VLP vaccine (Protocols 007, 011, 012, 013, 015, 016 and 018). Protocols 011 and 012 were sub-studies of Protocol 013.

- Immunogenicity of the quadrivalent HPV vaccine was evaluated in Protocols 007 (dose selection study), 011 (concomitant hepatitis B vaccine), 012 (monovalent HPV 16 bridging), 015V1 (consistency lots), 016V1 (adolescent bridging), 016V2 (end-expiry dose) and 018 (preadolescent /adolescent) and involved a total of 12,345 subjects.
- Efficacy was assessed in 4 randomised placebo-controlled clinical studies including a total number of 20,541 subjects. There were two phase II studies, 005 (n=2,391) that evaluated the HPV 16 component and 007 (n=551) that evaluated the quadrivalent HPV (types 6, 11, 16, 18) LI VPL vaccine. The two pivotal phase III studies, termed FUTURE (Female United To Unilaterally Reduce Endo/Ectocervical Disease) I (Protocol 013; n=5,442) and FUTURE II (Protocol 015; n=12,157) evaluated the quadrivalent HPV vaccine in the prevention of HPV 6/11/16/18-related CIN/external genital lesions (EGLs) and HPV 16- or HPV 18-related CIN 2/3 or AIS (Cervical Adenocarcinoma In situ), respectively. The efficacy trials recruited 16- to 26-year-old adolescent and young adult women, who were mostly HPV naïve, but were at risk for HPV infection. The Phase II and Phase III studies did not have screening phases; women were enrolled regardless of their baseline HPV status and Pap test (Papanicolau's test) result.
- Safety of the different HPV vaccine formulations has been evaluated in a total of 16,041 subjects. Of these 11,813 received quadrivalent HPV vaccine and the remaining monovalent vaccine formulations. All studies were placebo controlled and the total population that received placebo included 9,701 subjects (the placebo was aluminium adjuvant in all studies except study 018 (pre-/adolescent safety study) which used a non-aluminium-containing placebo).

GCP

The Clinical trials were performed in accordance with GCP as claimed by the applicant.

The applicant has provided a statement to the effect that clinical trials conducted outside the community were carried out in accordance with the ethical standards of Directive 2001/20/EC.

Pharmacokinetics

Pharmacokinetic studies were not performed in accordance with the Note for guidance on clinical evaluation of new vaccines (CPMP/EWP/463/97).

Pharmacodynamics

The pharmacodynamics of the vaccine relate to its interaction with the immune system. Therefore, this section will discuss the data on the systemic immune response to vaccination.

Initial phase I/IIa studies evaluated the immunogenicity of **monovalent** HPV vaccine precursors.

Overview phase I and phase II immunogenicity studies (monovalent HPV vaccines)

Study Protocol	No. of study centres/ locations/date	Study vaccine No/study arm	No subjects and age group	Primary Endpoint	Duration follow-up
P001 Phase I Dose escalating study	US (n=2 sites) 1997-2002	HPV **11** VPL vaccine (10/20/50/100 mcg)/ Placebo N=112 / 28 3 doses 0, 2, 6 months + subset	N=140 18- to 25- year old women (HPV 11/6-naïve)	- Percentage of subjects achieving anti-HPV 11 serum RIA* levels ≥200mMU/ml** and neutralizing antibody (xenograft anti-HPV 11 NT-assay) at month 7	3 years

		dose 4 at month 12		- Safety and tolerability	
P002 Phase I Dose escalating study	US (n=1 site) 1998-2001	HPV **16** VLP vaccine (10/40/80mcg) /Placebo N=82 / 27 3 doses 0, 2, 6 months	N=109 18- to 25- year old women (HPV-naïve)	- Percentage of subjects achieving anti-HPV 16 serum RIA levels ≥20mMU/ml at month 7 -Persistence of anti-HPV 11 - Safety and tolerability	3 years
P006 Phase I	US (n=3 sites) 2000-2001	HPV **18** VPL vaccine (80mcg) / Placebo N=27 /13 3 doses 0, 2, 6 months	N=40 16- to 23- year old women	- Percentage of subjects achieving anti-HPV 18 serum RIA levels ≥200mMU/ml at month 7 - Safety and tolerability	7 months
P004 Phase IIa Dose escalating study	US (n=15 sites) 1998-2001	HPV **16** VLP vaccine (10/20/40/80mcg)/ Placebo N=428 /52 3 doses 0,2,6 months	N=480 18- to 25- year old women	- Percentage of subjects achieving anti-HPV 16 serum cRIA levels ≥20mMU/ml and neutralizing antibody at month 7 - Safety and tolerability	2 years
P005 Phase IIb	US (n=16 sites) 1998-2004	HPV **16** VLP vaccine (40mcg) /Placebo N=1204 / 1205 3 doses 0,2,6 months	N=2409 16- to 23- year old women	**Primary:** Efficacy: Prevention of persistent HPV16 infection cf placebo- Safety and tolerability -Immunogenicity: Anti-HPV 16 levels (RIA) and correlate of protection. -Long-term persistence of anti- HPV 16 antibodies	3.5 years

* RIA: Radioimmunoassay

** mMU/ml: milli Merck Units per milliliter

Immunogenicity of the **quadrivalent** HPV (Types 6, 11, 16, 18) L1 VLP vaccine was evaluated in a Phase II dose selection (study Protocol 007, see below), and six phase III studies 011, 012, 013, 015, 016 and 018, a total of 12,344 subjects 9 to 26 years of age were enrolled. The studies were conducted in 33 countries and 5 continents with 3,453 subjects recruited from Europe. All studies were randomised, double-blind, and placebo-controlled (except P016).

Overview of phase II/III immunogenicity studies (quadrivalent HPV vaccine)

Study Protocol	No. of study centres / locations/date	Study vaccine No/study arm	No subjects and age group	Primary Endpoint	Duration
P007 Phase IIb *Part A*: Dose-escalation *Part B*: Dose-ranging	US, Brazil, Scandinavia (n=23 sites) 2000-2004	Quadrivalent HPV (Types 6,11, 16,18) L1 VLP vaccine (20/40/40/20mcg 40/40/40/40mcg 80/80/40/80mcg) /Placebo *Part A* n=52 *Part B* n=1106	N=1158 16- to 23- year old females	*Part A:* General tolerability *Part B:* Identify a formulation with acceptable type specific anti-HPV responses (cRIA) for phase 3 Investigate anti cLIA levels: - Ab kinetics/ seroconversion - General tolerability **Secondary:** Efficacy endpoints. HPV 6/11/16/18–related persistent infection, EGL, CIN, AIS and or cervical cancer	Mean: 2.4 yrs Median 3.0 yrs
P011 Phase III ***Substudy to P013*** Concomitant Hep B vaccine	US, Europe, Peru, Brazil (n=21) 2001-2004	Quadrivalent HPV (Types 6,11, 16, 18) L1VLP vaccine (20/40/ 40/20mcg) / Hepatitis B vaccine/ Placebo 3 doses 0, 2, 6 months	N=1877 16- to 23- year old females	-Evaluation of concomitant administration of HPV vaccine and hepatitis B vaccine regarding immune interferences as measured by GMTs *(HPV) and serocon-version rates at Week 4 postdose 3 -Safety and tolerability of co-administration	7 months (FU within P013)

P012 Phase III ***Substudy to P013*** Bridging study to P005	US, Canada, Europe, Latin Am, Australia, New Zealand, Asia, Russia, (n=48 sites) 2002-2004	Quadrivalent HPV (Types 6,11, 16, 18) L1 VLP vaccine (20/40/ 40/20mcg) / HPV16 L1 VLP vaccine/ Placebo N=1784/ 304/ 1794	N=3882 16- to 23- year old females	-Non-inferiority of the monovalent HPV 16 L1 vaccine to quadrivalent HPV vaccine with respect to anti-HPV 16 response (GMTs) at 4 weeks post-dose 3	7 months (FU within P013)
P013 Phase III efficacy study		Quadrivalent HPV (Types 6,11, 16, 18) L1 VLP (20/40/40/20 mcg) / placebo N=2732 / 2717	N=5455 16- to 23-year old females	Persistence of immune response	1.5 years (FU 48 months)
P015 Phase III ***Substudy***: Lot-to-lot consistency	North America, Europe, Latin Am, Singapore (n=80 sites) -Feb to May 2003	Quadrivalent HPV (Types 6,11, 16,18) L1 VLP vaccine (20/40/ 40/20mcg) 3 vaccine lots N=500 / 510 / 504	N=1514 16- to 23- year old females	*Substudy:* - Demonstration that 3 vaccine lots induce a consistent anti-HPV 6,11,16,18 response 4 weeks post-dose 3 - Long-term persistence of antibody response	1.5 years (FU 48 months)
P016 Phase III ***Substudies:*** **A)** Adolescent Bridging study **B)** End Expiry	US, Canada, Europe, Latin America, Asia (n=61 sites) 2002-2004	**A)** Quadrivalent HPV (Types 6,11, 16,18) L1 VLP vaccine (20/40/ 40/20mcg) **B)** 20%/ 40%/ 60%/ 100% formulations N=503/ 513/ 508/ 1017	Total: 3,055 **A)** N=1525 10-15 -year old **males** (n=508) 10-15 -year old females (n=506) 16-23-year old females (n=511) **B)** N=2545 10-15 -year old females 16-23-year old females	*A) Adolescent substudy*:- Non-inferiority of the younger age group cf 16-23 year- old females with respect to GMTs at Week 4 post-dose 3 - Safety and tolerability *B) Expiry dose substudy*: -Identify the minimum partial dose formulation that is similar to full dose (GMTs at Week 4 postdose 3)	7 months
P018 Phase III	Europe, North America, Latin America, Asia (n=47 sites) 2003-2005	Quadrivalent HPV (Types 6,11, 16,18) L1 VLP vaccine (20/40/ 40/20mcg)/ Placebo (non-Alum) 1184 / 597	N=1781 9-15 year-old females (n=939) 9-15 year-old **males** (n=842)	Primary: Safety and tolerability -Non-inferiority of boys to girls with respect to GMTs at Week 4 post-dose 3. -Persistence of anti-HPV response	7 month (18 months)

*GMTs: Geometric Mean Titres

The immunogenicity of the HPV vaccines was measured using three methods:

- A competitive radio-immunoassay (cRIA)
- A competitive Luminex-based immunoassay (cLIA) and
- A xenograft based HPV 11 neutralization (NT) assay.

During the development programme, the early cRIA assay was transitioned to cLIA assay, which was subsequently used during the phase III trials. The immune response to each vaccine HPV type was measured separately.

Primary study population was the per-protocol immunogenicity (PPI) population defined as:

- Subjects who were seronegative and PCR negative to the relevant HPV type(s) at Day 1, remained HPV PCR negative through 1 month post dose 3 (month 7), received all 3 vaccinations within pre-specified time intervals, and no deviation from the study protocol.

The following immunogenicity objectives were focused on:

- To evaluate vaccine-induced serum anti-HPV responses during the vaccination regimen, 4 weeks following the completion of a the 3-dose regimen as well as persistence of antibody response for up to 3.5 years
- To define impact of base-line covariates (e.g. age, gender, ethnicity) and deviations from vaccination regimen at 4 weeks post-dose 3

- To bridge the efficacy data obtained in female subjects aged 16 to 26 years of age at enrolment to subjects 10 to 15 years of age at enrolment by demonstrating that the post dose 3 anti-HPV responses in the younger age group are non-inferior to those observed in the older group
- For HPV 11 to demonstrate that immune responses are virus neutralizing
- To establish that vaccine-induced responses are comparable or superior to immune responses to natural infection
- To establish that the HPV vaccine can generate memory responses in subjects seropositive for one or more HPV types at baseline
- To investigate potential immune correlates of vaccine efficacy
- The phase III trials also compared immune responses between treatment groups to address questions of concomitant administration of recombinant hepatitis B vaccine (P011), consistency of manufacturing process (P015) and evaluation of partial-dose formulations (P016)

Primary immunological endpoints were:

- Geometric mean titres (GMTs) of anti-HPV 6, anti-HPV 11, anti-HPV 16 and anti-HPV 18, 4 weeks after the third dose (Month 7)
- Proportion of subjects who seroconverted to each of the four antigens 4 weeks after the third dose.

Since the minimum anti-HPV levels associated with protection from acquisition of HPV is not known, the cut-off value of validated assays was used as a surrogate for seropositive level. The cLIA cutoffs for seropositivity were as follows: $\geq$20 mMU/ml for HPV 6 and 16, $\geq$16 mMU/ml for HPV 11 and $\geq$24 mMU/ml for HPV 18.

- **Monovalent HPV vaccines (n=3,160 subjects)**

Initial phase I/IIa studies evaluated the immunogenicity of monovalent vaccine precursors in 3,160 16- to 26-year-old female subjects (1,842 vaccine/1,318 placebo). All studies were randomised, double blind and placebo-controlled and all vaccine candidates were given in a 3-dose schedule (0, 2, 6 months). Monovalent HPV 11 LI VPL vaccine was evaluated in one study (P001).

The HPV 11 L1 VLP vaccine was chosen as the first vaccine for evaluation in humans, since it was possible to use the nude mouse xenograft model for HPV 11 infection to determine if the vaccine could induce neutralizing antibodies. A correlation was demonstrated between functional (neutralizing) antibodies and cRIA/cLIA antibodies. The vaccine proved immunogenic with 90-100% of subjects reaching the pre-specified cRIA level of $\geq$200 mMU/ml 4 weeks postdose 3. Anti-HPV levels were shown to decline after Month 7, but were still detectable at Month 36 at levels above those induced by natural infection. A fourth dose at Month 12 was shown to induce higher anti-HPV levels than postdose 3, but the differences dissipated after one year and therefore it was decided to use the 3-dose regimen in subsequent trials. Anti-HPV levels in cervicovaginal mucosa was measured in lavage, but proved difficult to standardise due to intrinsic problems encountered in accurate sampling of the mucosal surface. Therefore, it was determined not to include this assay in subsequent studies.

The other phase I/IIa studies (P002, P004 and P006) confirmed the immunogenicity results obtained in P001. It is to be noted that the pre-specified cRIA cut-off levels varied by study. The kinetics of vaccine-induced anti-HPV responses were evaluated, demonstrating a priming effect of 2 doses with peak titres achieved at Month 7, after which titres declined rapidly by around 1 log to reach a plateau at Month 24, which remained stable to Month 36. Based on dose-ranging data from P002 and P004, the 40mcg-dose for the HPV 16 vaccine was selected for the first efficacy trial (P005).

Mode of action

A substudy in an early monovalent HPV 11 L1 VLP vaccine clinical trial was conducted to measure HPV 11 L1-VLP vaccine priming of humoral and cellular immune responses in seronegative, HPV DNA-negative, college-age women. The results of this study were consistent with those observed in the non-human primate study; Th1 or Th2 cytokines (IFN-γ or IL-5) were detected in response to HPV 11 L1 VLP in vitro re-stimulation for all vaccine recipients and none of the placebo recipients tested. The predominant T-cell population with detectable IFN-γ and IL-5 production was the T-cell subpopulation depleted of $CD8^+$ T-cells. The overall HPV-specific T-cell activity was observed as a discrete proliferative response consistent with homeostasis in memory T-cell responses. In addition the

immunoglobulin isotype and subclass profiles elicited by vaccination demonstrated the generation of both Th1 and Th2 responses (IgG1 and IgG2).

In a separate sub study several cohorts, including a cohort of 10- to 15-year-old virgin females who participated in one of the quadrivalent HPV vaccine trials were evaluated for Th1 T-cell immunity. The results demonstrated no response prior to vaccination, however post-vaccination strong HPV 16 L1 peptide-specific T-cell responses were observed in all subjects. Vaccination of young females with the quadrivalent HPV L1 VLP vaccine resulted in induction of a strong L1 peptide-specific IFNγ-associated T-cell response and a concomitant B-cell response producing L1 VLP-specific antibodies.

- **Quadrivalent HPV vaccines (n=12,344 subjects)**

Study 012

This substudy to study P013, aimed at bridging anti-HPV 16 responses between the monovalent HPV 16 vaccine used in the first efficacy trial 005 (Pilot Manufacturing Material, PMM) and the quadrivalent HPV vaccine (Final Manufacturing Process, FMP) used in the pivotal efficacy trials. Non-inferiority of the anti-HPV 16 GMTs and seroconversion responses at Month 7 in the FMP quadrivalent HPV vaccine group relative to the PMM HPV 16 vaccine was demonstrated. This allowed including the P005 trial in the pooling of efficacy data from all efficacy trials.

Study 015V1

Study P015V1, a substudy to study 015, was a lot consistency study. The primary objective was to demonstrate that 3 separate lots of the quadrivalent vaccine induced similar GMTs to HPV types 6, 11, 16 and 18 at Week 4 postdose 3. Based on the non-inferiority criteria defined, consistency of the anti-HPV responses in the 3 vaccine lot groups was demonstrated.

Study 016

This study consisted of two immunogenicity studies, the adolescent substudy and the end-expiry substudy that partly used the same study participants.

The adolescent ***study 016V1*** was designed to bridge the efficacy findings in the older age groups to virginal aged 15 years and younger by demonstrating that 10- to 15-year-old subjects had immune responses to a 3-dose regimen that were non-inferior to those observed in the 16- to 23-year olds. In the absence of an immune correlate of efficacy, immune responses in the demographic groups were compared using GMTs and the proportion of subjects who were naïve to HPV vaccine types and who became seropositive to the relevant HPV types at Month 7.

Summary of vaccine immune response by demographic group

Assay	Time point	Quadrivalent HPV vaccine								
		10- to 15 year-old females N=506			10- to 15-year-old males N=506			16- to 23-year-old females N=506		
		n	GMT	Sero.conv	n	GMT	Sero.conv	n	GMT	Sero.conv
Anti-HPV 6	Day 1	426	<8		431	<8		320	<8	
	Month 3	417	635.2	100	430	673.5	100	315	437.7	100
	Month 7	426	989.9	100	431	1118.6	100	320	603.0	100
Anti-HPV 11	Day 1	426	<8		431	<8		320	<8	
	Month 3	418	781.1	100	430	794.7	100	315	549.7	100
	Month 7	426	1270.6	100	431	1399.6	100	320	739.2	100
Anti-HPV 16	Day 1	427	<12		431	<12		306	<12	
	Month 3	419	2842.3	99.8	429	2993.1	100	302	1.705.6	100
	Month 7	427	4873.0	100	430	5962.1	100	306	2,753.0	100
Anti-HPV 18	Day 1	429	<8		432	<8		340	<8	
	Month 3	421	369.1	98.8	431	413.1	98.6	334	216.6	97.6
	Month 7	429	957.7	100	432	1241.6	99.8	340	470.5	99.1

For each of the 4 HPV types the vaccine induced numerically higher GMTs 4 weeks postdose 2 and 3 in adolescents than in adult females. Postdose 2 anti-HPV 6/11/16/18 levels among adolescents were comparable to those observed postdose 3 among 16- to 23-old females. Numerically higher GMTs were also observed in the male adolescent group compared with the female adolescent group consistently across the HPV vaccine types. The results of the statistical analyses demonstrated that the quadrivalent HPV vaccine induced non-inferior immune responses in the 10- to 15-year-old females and 10- to 15-year-old males compared with 16- to 23-year-old females.

The primary objective of the end-expiry ***study 016V2*** was to identify the minimum partial-dose formulation of quadrivalent HPV vaccine among the 20, 40, 60% partial-dose formulations, given in a 3-dose regimen, that induced immune responses non-inferior to administration of 100% full-dose formulation. The key assumption in this study was that testing partial doses of the vaccine could approximate the impact of manufacturing process loss in addition to vaccine potency loss over time on the immunogenicity of the vaccine. The primary immunogenicity hypothesis compared GMTs within 3 pairs of vaccination groups 20% formulation vs. 100% formulation, 40% vs. 100% and 60% vs. 100%. The criteria for declaring success required success on at least 1 of the 3 comparisons. A nested testing procedure was performed in 3 stages using an alpha level of 0.025 (1-sided) at each stage. In the first stage the 20% partial-dose formulation of the HPV vaccine was compared with the full-dose formulation. Since this comparison fulfilled the criteria for non-inferiority, testing stopped at this stage and all partial-dose formulations were declared non-inferior to the 100% formulation. All lower confidence bounds exceeded 0.5 and correspondingly, all p-values were <0.025.

Study 018

This study was conducted to further define the safety and immunogenicity of the quadrivalent HPV vaccine in 9- to 15-year-old subjects. Study P018 also provided a comparison to non-aluminium-containing placebo. Enrolment was stratified by gender and age (1:1 male/female; 2:1 9-12 year-old/13-15 year-old). The study population contained almost 700 subjects aged 9 to 12 years.

Primary objective was to assess safety, but the study also included secondary immunogenicity objectives to demonstrate that the 4-week postdose 3 anti-HPV 6/11/16/18 immune responses (GMTs and percentage of subjects who seroconvert) in pre-/adolescent boys were non-inferior to the responses observed in pre-/adolescent girls All subjects demonstrated a strong antibody response to the HPV vaccine. GMTs at Month 7 were higher among boys than girls. The Month 7 GMTs for all vaccine types were higher among 9- to 12-year-old subjects than among 13- to 15-year-old subjects. The statistical criteria for non-inferiority with respect to both GMTs and seroconversion rates were met.

Dose response study

Study 007

Study 007 was designed as a dose-ranging study evaluating immunogenicity (cRIA) of 3 vaccine formulations. The dose ranging concerned HPV 6 and HPV 18, whereas HPV 11 was tested only at a higher dose (80mcg) in one formulation. The dose of HPV 16 (40mcg) was fixed, based on results from previous phase I/II trials. The primary objective of P007 was to select a dose for the phase III trials, which was performed in a pre-planned interim analysis after 50% of the subjects had received Dose 3. Antibody kinetics was also evaluated and demonstrated a priming effect of the first 2 doses. All 3 doses were demonstrated to induce high anti-HPV levels without any clear dose response pattern, which favoured the lowest dose 20/40/40/20mcg. Moreover, a modest increase in injection-adverse events was observed with the higher dose formulations.

Preliminary data on an extension of study P007 up to 5 years were provided. Subjects in the quadrivalent HPV vaccine 20/40/40/20-mcg dose formulation group and placebo subjects who were enrolled in Europe and Brazil (n=241) were followed through Month 60. The plateau of anti-HPV GMTs reached at Month 24 was shown to remain stable through the follow-up period up to Month 60. Administration of a pre-planned fourth dose (booster dose) at Month 60 was evaluated in 78-87 subjects. A strong anamnestic response was observed for all 4 HPV types included in the vaccine, suggesting that the quadrivalent HPV vaccine induces an immune memory. The booster dose appeared well tolerated.

Co-administration

Study 011

This substudy of study 013 was designed to evaluate the immunogenicity and safety of the HPV vaccine when administered alone or concomitantly with the recombinant hepatitis B vaccine. The primary objective was to demonstrate that concomitant administration of the two vaccines did not interfere with the immune response to either vaccine. Non-inferiority of the anti-HPV responses (GMTs and seroconversion rates) after the third dose in the concomitant group relative to the HPV vaccine only group was demonstrated. Also with regard to the anti-HBV seroprotection rates in the concomitant group relative to hepatitis B vaccine alone, non-inferiority criteria were met. However, with respect to the anti-HBs GMTs, lower titres were observed in the concomitant group (535 vs 792 mIU/ml). Thus, the HPV vaccine appears to have a negative impact on the hepatitis B GMTs, although the clinical significance of these lower titres is not known. Overall, the anti-HBs responses seemed lower than would be expected in this group of young healthy females, which might be associated with the low immunogenicity of the hepatitis B component. Additional analyses of the data were provided, which did not add further concern.

A statement, regarding the reduced anti-HBs titres following the concomitant vaccination with Gardasil is included in the SPC section 4.5. The applicant was strongly recommended to perform a non-inferiority study with the new upgraded hepatitis B vaccine and also include an additional arm with the other recombinant hepatitis B vaccine available on the EU market.

Clinical efficacy

Efficacy was assessed in 4 randomised double-blind placebo-controlled phase II and phase III clinical studies:

Study Protocol	No. of study centres / locations/dates	Study vaccine No/study arm	No subjects and age group	Primary Endpoint	Duration Post-7 mo FU
P005 Phase IIb	USA (n=16 sites) Oct 1998 - Sep 2001	HPV 16 L1 VLP vaccine (40mcg)/ Placebo (1193 / 1198)	N=2,409 16- to 23-year-old women Mean 21.5 yrs	1. Safety and tolerability of vaccine, 3 doses 2. Efficacy in prevention of persistent HPV 16 infection cf placebo	Mean: 3.1 years Median: 4.0 years

P007 Phase IIb Dose-ranging study	USA, Europe Latin America (n=23 sites) May 2000 - May 2004	Quadrivalent HPV VLP vaccine (20/40/40/20mcg 40/40/40/40mcg 80/80/40/80mcg) or Placebo Part A n=52 Part B n=1106	N=1,158 16- to 23-year-old women Mean 20.0 yrs	1. Identify formulations with acceptable type specific anti-HPV responses 2. Efficacy in prevention of persistent HPV 6,11, 16, 18 infection and clinical disease cf placebo 3. General tolerability	Mean: 2.4 years Median 3.0 years
P013 Phase III **FUTURE I**	North Am, Latin America, Europe, Asia-Pacific (n=62 sites) Dec 2001 - July 2005	Quadrivalent HPV VLP vaccine 20/40/40/20mcg / Placebo (2717 / 2725)	N=5,455 16- to 23-year-old women Mean 20.3yrs	Co-primary endpoint: i) External genital lesion: efficacy in reducing HPV 6,11,16,18-related genital warts, VIN, VaIN, vulvar or vaginal cancer cf placebo ii) Cervical endpoint: efficacy in reducing the incidence of HPV 6,11, 16,18-related CIN (any grade), AIS or cervical cancer cf placebo - Safety and tolerability	Mean: 1.7 years Median: 2.4 years
P015 Phase III **FUTURE II**	North Am, Latin America, Europe, Asia-Pacific (n=90 sites) June 2002 - June 2005	Quadrivalent HPV VLP vaccine 20/40/40/20mcg / Placebo (6082 / 6075)	N=12,167 16- to 23 (26) - year-old women Mean 19.9 yrs	Primary Cervical endpoint: Efficacy in reducing the incidence of HPV 6,11,16,18-related CIN 2/3, AIS or invasive cervical cancer in HPV naïve subjects - Safety and tolerability	Mean : 1,4 years Median: 2.0 years (Sentinel planned 10 years follow-up)

A total of 20,887 subjects were enrolled in studies 005, 007, 013, and 015. This includes 304 subjects who received monovalent HPV 16 L1 VLP vaccine in study 012, a sub-study of study 013. The subjects were enrolled from 5 continents and 22 countries, with Europe well represented.

Study 005 was the proof-of-concept study, evaluating efficacy of a monovalent HPV 16 L1 VLP 40 mcg vaccine in preventing persistent HPV 16 infection.

Study 007 was the dose-ranging study of the quadrivalent HPV (Types 6, 11, 16, 18) L1 VLP vaccine, and involved 551 female subjects evaluated for efficacy of the phase III formulation (20/40/40/20 mcg) in preventing persistent HPV 6/11/16/18-related persistent infection or disease.

Studies 013 and 015 are the pivotal studies of the efficacy of the quadrivalent vaccine against vaccine HPV-related EGL and CIN and enrolled 5,746 and 12,157 female subjects, respectively. All studies were double-blind, randomised and placebo-controlled. The study subjects were healthy 16- to 23 (26)-year-old women.

METHODS

Study Participants

The study subjects were healthy 16- to 23 (26)-year-old women. The studies did not include a screening phase. Thus, both naïve individuals and individuals who had been exposed to HPV prior to enrolment were included. All subjects had at inclusion:

- Serum anti-HPV testing
- Pap test
- Cervicovaginal sampling for HPV typing
- Colposcopy if Pap test showed some abnormalities

Irrespectively of results of these examinations, subjects were randomised to either HPV vaccine or placebo. Five populations were considered for HPV-specific efficacy analysis:

Per-protocol efficacy (all studies):

- Received all 3 doses of study vaccine
- Were seronegative to relevant vaccine HPV type(s) at Day 1
- Were PCR negative to relevant vaccine HPV type(s) Day 1 to Month 7
- Did not have general protocol violations
- Cases counted starting 30 days postdose 3 (Month 7).

Since subjects were not pre-screened for HPV and were included regardless of HPV status and Pap test at baseline, only 64% to 84% (depending on HPV type) of all study subjects were included in the primary per-protocol efficacy (PPE) cohort.

The Per-Protocol Efficacy (PPE) population was used as the primary efficacy population.

MITT-populations (modified Intend to Treat Populations):

- MITT-1 population included protocol violators, but was otherwise similar to the PPE
- MITT-2 included HPV-vaccine-type naïve subjects who received at least 1 dose
- MITT-4 included HPV-vaccine-type naïve subjects who received at least 2 doses (only P013)

MITT-3 population (all studies)

- Received at least 1 injection
- Pap test: normal or abnormal at Day 1
- Regardless of HPV serology /PCR status at Day 1

The MITT-3 population represents the general (female) population in this age group.

For the analyses that were not HPV-vaccine-type specific (population benefit analyses) the following populations were defined:

Restricted MITT-2 population (P013, P015)

- Received at least 1 injection
- Were seronegative to all 4 vaccine HPV types at Day 1
- Were PCR negative to all 4 vaccine HPV types at Day 1
- Pap test: normal at Day 1 (used as a surrogate for non-vaccine HPV types)

This population represents virginal HPV-naïve adolescents.

Restricted MITT-3 population (P013, P015, combined analysis)

- Received at least 1 injection
- Pap test: normal at Day 1
- Regardless of HPV serology/PCR status at Day 1

For potential therapeutic benefit of the vaccine (P013, P015) the following subpopulations were evaluated:

- Subjects who were seronegative and PCR positive at Day 1 to the relevant vaccine HPV type
- Subjects who were seropositive and PCR negative at Day 1 to the relevant vaccine HPV type
- Subjects who were seropositive and PCR positive at Day 1 to the relevant vaccine HPV type

Treatments

All subjects received either quadrivalent HPV VLP vaccines (HPV LI VLP vaccine 40 mcg in study 005) or placebo at day 0, month 2 and month 6.

All subjects underwent Pap testing and cervicovaginal sampling for HPV testing every 6 months (studies 005, 007, and 013) or every 12 months (study 015). Referral to colposcopy was based on Pap

test diagnosis. All colposcopies were to be performed at study site by an experienced colposcopist. Referral to definitive therapy during the trial (surgical ablation and histological assessment) was based on Pap test (repeated CIN 1, high-grade squamous intraepithelial lesions (HSIL)) +/-abnormal colposcopy.

Objectives

The study objectives of the clinical program for the HPV vaccine were to demonstrate that administration of the HPV vaccine would reduce the incidence of:

- HPV 16/18-related high-grade cervical intraepithelial neoplasia (CIN 2/3) or cervical adeno-carcinoma in situ (AIS) as surrogate for cervical cancer
- HPV 6/11/16/18-related CIN of any grade
- HPV 6/11/16/18-related external genital lesions (EGLs) - condyloma acuminata (genital warts) vulvar intraepithelial neoplasia (VIN), vaginal intraepithelial neoplasia (VaIN)
- HPV 16/18-related VIN 2/3 or VaIN 2/3 as surrogates for vaginal cancer and vulvar cancer

And that administration of the HPV vaccine will reduce the overall incidence of HPV-related cervical and genital disease.

Outcomes/endpoints

The Applicant defined two types of endpoints:

Persistent HPV infection: used in study 005 (primary endpoint) and study 007, defined as:

- Persistent vaccine-type HPV infections without confirmed CIN: detection of vaccine-type HPV DNA by PCR on at least 2 consecutive visits spaced for at least 4 months
- Detection of vaccine-type HPV DNA by PCR on the last visit without confirmation of persistence
- Vaccine-type HPV infection with confirmed CIN: histologically confirmed CIN 1, 2, 3, AIS, or cervical carcinoma due to vaccine-type HPV

Disease endpoints: were included in all efficacy studies

- The incidence of HPV 16-related CIN 1, CIN 2, CIN 3 or worse (study 005)
- The combined incidence of CIN 1, CIN 2, CIN 3 or worse and combined incidence of external genital lesions related to HPV 6, 11, 16 and 18 (study 007)
- The combined incidence of CIN 1, CIN 2, CIN 3 or worse related to HPV 6, 11, 16 and 18 and combined incidence of condyloma acuminata VIN 1, VIN 2/3, VaIN 1, VaIN 2/3 or worse related to HPV 6, 11, 16 and 18 (two co-primary endpoints: external genital lesion endpoint and cervical lesion endpoint) (study 013)
- The incidence of AIS, CIN 2/3 or worse related to HPV 16 or HPV 18 (study 015).

Study 005 used a primary virological endpoint, whereas histological endpoints were used in study 007 and the two phase III trials (P013 and P015).

Disease endpoint definitions differed among the studies due to differences in the vaccines tested, i.e. 005 evaluating the HPV 16 vaccine, 007 evaluating different quadrivalent HPV vaccine doses and 013/015 evaluating the quadrivalent HPV vaccine. Endpoint definitions also differed among studies 005, 007, 013, and 015 due to the evolution of the method used to assess the causal HPV type within a lesion.

All HPV endpoints were histologically classified. The primary CIN and EGL endpoints of the studies were based on biopsies and required HPV DNA to be detected in the same tissue specimen by thin-section PCR. An independent blinded Pathology Panel was used for adjudication of all study endpoints. A comprehensive testing program was used for HPV-related disease. The results of the ThinPrep Pap test were used to identify subjects with cervical HPV disease and thorough genital

inspection to identify subjects with EGLs. The protocol-specified genital examinations and mandatory guidelines for triage of abnormal Pap test to colposcopy ensured consistency and standardised ascertainment of HPV-related lesions among study sites.

Sample size / Statistical methods

Neither study 013 nor study 015 was individually powered to provide substantial evidence to demonstrate that the HPV vaccines reduce the incidence of HPV 16/18-related CIN 2/3 or AIS, efficacy data were to be combined from all 4 studies in a pre-specified analysis.

Studies 013 and 015 were designed as fixed-case studies. The vaccine efficacy (VE) evaluations were to be performed when a pre-specified number of endpoint cases had been observed. Final efficacy analyses were to be performed at the end of studies. The requisite numbers of endpoint cases for both studies were obtained in August 2005, when the majority of subjects had been followed for at total of 2 years (1.5 years after completion of the 3-dose regimen). Data available at this time point were unblinded and analysed and form the basis of this application. Study 013 and study 015 are currently ongoing and vaccine efficacy is to be re-estimated at the end of the 4-year follow-up.

RESULTS

Overall 19,321 subjects (93% of subjects who received at least 1 one vaccine dose) continued in the study from the time of enrolment through the date at which the database was closed (Aug 2005). Except for the study 005, where 84.6% of patients received the 3 injections (in the vaccine group), in other studies 93 to 98% of patients received 3 injections.

There were regional differences between studies. Study 005 was recruiting subjects only in the US; study 013 had the largest recruitment in Latin America (41%) and study 015 in Europe (65%).

Overall 13% of the combined population had a test compatible with CIN at Day 1 and 27% of the combined population was either seropositive or PCR positive to a vaccine HPV type at baseline.

The median durations of follow-up were 4.0, 3.0, 2.0 and 2.0 years for P005, P007, P013 and P015, respectively.

Baseline data

The demographic characteristics were generally comparable between vaccine and placebo groups regarding the age, race, geographic region, smoking status, alcohol consumption, and history of sexually transmitted diseases at enrolment.

Sexual demographics differed by geographic region. Age at sexual debut occurred at a mean age of 16.7 years. Hormonal contraception was the most common form of contraception (59%) and highest in Europe (68%).

The demographic profile of subjects in the PPE population was general comparable to the overall population.

Outcomes and estimation

Primary efficacy results of individual studies

Study 005: The primary efficacy endpoint of study 005 was the incidence of new persistent HPV 16 infection. Two efficacy analyses were performed, a fixed-case analysis and a final analysis at the end of the 4-year study follow-up. In the first analysis in the PPE population, all 41 cases of persistent HPV 16 infection were in the placebo group. At end-of-study, 7 cases in the vaccine group had HPV 16 detected at the last study visit (without confirmed persistence). There were no cases of HPV-related CIN in the vaccine group in the fixed-case or end-of-study analyses versus 9 and 24, respectively in the placebo group. The MITT-2 results supported those of the PPE analyses.

Primary efficacy endpoint: persistent HPV 16 infection (P005)

Population	HPV 16 vaccine (n=1193)			Placebo (n=1198)			Observed efficacy % (95%CI)
	Number of cases	Person years at risk	Infection rate/100 PY at risks	Number of cases	Person years at risk	Infection rate/100 PY at risks	
Fixed-case analysis							
PPE	0 / 753	1083.2	0.0	41 / 750	1047.2	3.9	100.0 (90.0, 100)
MITT-2	7 / 824	1560.7	0.4	76 / 839	1516.3	5.0	91.0 (80.7, 96.5)
MITT-3	54 / 1004	1833.2	2.9	131 / 1044	1823.6	7.2	59.0 (43.3, 70.7)
End of study analysis							
PPE	7 / 755	2466.8	0.3	111 / 750	2245.9	4.9	94.3 (87.8, 100)
MITT-2	16 / 824	3016.0	0.5	150 / 839	2779.0	5.4	90.2 (83.5, 94.5)
MITT-3	67 / 1004	34932	1.9	217 / 1044	3325.7	6.5	70.6 (61.2, 78.0)

In the MITT-3 population, at the end of study, there was a significant reduction of HPV 16-related persistent infection and CIN in the vaccine group as compared to placebo; out of 67 cases of persistent HPV infection in the vaccine group, 9 patients had HPV 16-related CIN. In the placebo group, out of 217 cases of persistent HPV infection, 44 patients had HPV 16-related CIN. There was a significant reduction of HPV 16-related CIN 2/3 or worse in the vaccine group as compared to placebo (VE: 77.9% (95% CI: 40.6; 93.4)).

A re-analyses using the appropriate WHO definition of persistent infection ($\geq$6-12 and $\geq$12 months) were provided for study 005. It was demonstrated that one-third to one-fourth of the observed HPV infections were of shorter duration than 6 months. Including only cases $\geq$6 months in the MITT-2 analysis reduced the estimates of efficacy, but vaccine efficacy still remained high (5 vaccine vs. 88 placebo cases). For the $\geq$12 month definition there were 3 vs. 43 placebo cases (VE: 93.3% (95%CI: 79.1, 100).

Study 007: In the dose-ranging study 007, the 20/40/40/20mcg dose was selected for the phase III studies and only this dose was formally evaluated for vaccine efficacy. The primary endpoint was the incidence of new HPV6/11/16/18-related persistent infection, CIN and/or EGL. The efficacy estimate at the end of study (2.5 years follow-up) was 89.5% in the PPE population.

Efficacy against HPV 6/11/16/18-related persistent infection or disease (P007)

Primary endpoint	Quadrivalent HPV vaccine (20/40/40/20) (n=276)			Placebo (Aluminium adjuvant) (225 and 450mcg) (n=275)			Observed Efficacy% (95%CI)
	Number of cases	Person years at risk	Incidence rate/100 PY at risk	Number of cases	Person years at risk	Incidence rate/100 PY at risk	
Per-protocol	4 / 235	566.8	0.7	36 / 233	536.5	6.7	89.5 (70.7, 97.3)
HPV 6-related endpoints	0 / 214	517.5	0	13 / 209	501.2	2.6	100.0 (68.2, 100.)
HPV 11-related endpoints	0 / 214	517.5	0	3 / 209	503.7	0.6	100.0 (<0, 100.0)
HPV 16-related endpoints	3 / 199	484.4	0.6	21 / 198	465.4	4.5	86.3 (54.0, 97.4)
HPV 18-related endpoints	1 / 224	541.8	0.2	9 / 224	536.9	1.7	89.0 (20.5, 99.7)
MITT-2	6 / 266	723.6	0.8	48 / 263	667.1	7.2	88.5 (73.0, 96.0)
MITT-3	23 / 268	690.6	3.3	61 / 269	650.9	9.4	64.5 (41.7, 79.0)

Of the 4 endpoint cases in the vaccine group there were 3 infections without confirmed persistence and 1 case of confirmed persistent HPV 18 infection. There were no cases of CIN or external genital disease in the vaccine group versus 5 cases in the placebo group. In the two higher vaccine groups (40/40/40/40 mcg, 80/80/40/80 mcg), the incidence rates of persistent HPV 6, 11, 16 and 18 infection or related disease were comparable to those observed with the final formulation. Only results for PPE population were provided for the higher dose groups. There were 7 cases of persistent infection (2 confirmed) and 3 cases (1 confirmed) in the higher dose groups, of which two were HPV 18-related and one was HPV 16-related.

In the MITT-3 population there were 23 patients with persistent infection in the vaccine group, of whom 3 had HPV-related CIN (two were HPV 16-related CIN 3, one was HPV 18-related CIN 1). In the placebo group, there were 61 patients with persistent HPV infection, of whom 4 had external genital lesions (three were condyloma (one HPV 6-related, one HPV 6/16-related, one HPV 11-related), and one was a VIN 2/3 (HVP 16-related)) and 12 had HPV-related CIN (one was HPV 6-related CIN 3, one was HPV 18-related CIN 2, two were HPV 16-related CIN 1, three were HPV 16-

related CIN 2, three were HPV 16-related CIN 3, one was HPV 18-related CIN 1, one was HPV 18-related CIN 2).

Preliminary efficacy data through a study 007 extension up to 5 years were provided. After the end of study 007 (Month 36), subjects were re-evaluated at Month 54 and 60. Results at Month 60 are presented in the table below and suggest persistence of efficacy. Sensitivity analyses were performed to assess impact of the gap of 18 months (between month 36 and 54) during which no scheduled visits occurred. A supplementary analysis was also performed restricted to the subjects that participated in the 5-year extension (n=204).

Analysis of efficacy against HPV 6/11/16/18-related persistent infection or disease from Month 7 (PPE) and from 30 days (MITT-2, MITT-3) through Month 60

HPV 6/11/16/18-related infection or disease	Gardasil			Placebo			Observed efficacy % (95%CI)
	Number of cases	Person years at risk	Incidence rate/100 PY at risks	Number of cases	Person years at risk	Incidence rate/100 PY at risks	
By study population							
PPE	2 / 235	767.9	0.3	46 / 233	747.4	6.2	95.8 (83.8, 99.5)
MITT-2	4 / 266	945.0	0.4	59 / 263	879.5	6.7	93.7 (83.0, 98.3)
MITT-3	21 / 268	945.0	2.3	74 / 269	850.7	8.7	73.2 (56.1, 84.3)

A re-analyses using the appropriate WHO definition of persistent infection (≥6-12 and ≥12 months) were provided for. It was demonstrated that one-third to one-fourth of the observed HPV infections were of shorter duration than 6 months. Including only cases ≥6 months in the MITT-2 analysis reduced the estimates of efficacy, but vaccine efficacy still remained high (1 vs. 23). For the ≥12 month definition there were 0 vs.11 cases (VE: 100% (95%CI: 43.3, 100). However, vaccine efficacy was only established for persistent HPV 16 infection, whereas for persistent HPV 18 infection there were too few cases to obtain significant results. Since according to the WHO expert statement (Vaccine 23, (2004) 569-578) this virological endpoint is not considered as a valid primary efficacy endpoint for HPV vaccines, the claim for persistent infection in the indication cannot be accepted.

Study 013 (Future I): Study 013 covered the entire spectrum of clinical genital disease and had 2 co-primary efficacy endpoints, one EGL endpoint (incidence of HPV 6/11/16/18-related genital warts, VIN, VaIN, vulvar or vaginal cancer), and one cervical endpoint (incidence of HPV 6/11/16/18-related CIN (1,2,3), AIS or cervical cancer). Only the results of the fixed case analysis were provided in this application. At this time point, the majority of patients have completed 2 years of follow-up. The results of the primary efficacy analysis in the PPE population are shown below.

Summary of primary efficacy analysis (PPE population) (P013)

Primary endpoint	Quadrivalent HPV vaccine (n=2717)			Placebo (n=2725)			Observed efficacy % (95%CI)
	Number of cases	Person years at risk	Incidence rate/100 PY at risks	Number of cases	Person years at risk	Incidence rate/100 PY at risks	
HPV 6/11/16/18-related CIN	0 / 2240	3779.8	0.0	37 / 2258	3787.4	1.0	100 (87.4, 100.0)
HPV 6/11/16/18-related EGL	0 / 2261	3865.2	0.0	40 / 2279	3868.4	1.0	100 (88.4, 100.0)

Efficacy against cervical disease

Primary efficacy was also analysed by HPV type and CIN lesion and demonstrated that most endpoint cases were HPV 16-related (~59%). Approximately half of subjects with a CIN lesion had a pathological diagnosis of CIN 2 or worse. Vaccine efficacy (VE) against CIN 3/AIS was 100% with the lower bound of the 95% CI of 55%. No cases of invasive cervical cancer were detected among subjects in any population. In the MITT-2 population, VE was 96.5% with 2 cases of CIN detected in the vaccine group versus 57 in the placebo group.

Efficacy by HPV type and by CIN lesion (PPE population) (P013)

Endpoint	Quadrivalent HPV vaccine (n=2717)			Placebo (n=2725)			Observed efficacy % (95%CI)
	Number of cases	Person years at risk	Incidence rate/100 PY at risks	Number of cases	Person years at risk	Incidence rate/100 PY at risks	
HPV 6/11/16/18-related CIN							
PPE	0 / 2240	3779.8	0.0	37/ 2258	3787.4	1.0	100.0 (87.4, 100.0)
MITT-2	2 / 2557	5490.1	0.0	57 / 2573	5489.0	1.0	96.5 (86.7, 100.0)
By HPV type							
HPV 6-related CIN	0 / 1960	3316.0	0.0	7 / 1975	3332.6	0.2	100.0 (30.3, 100.0)
HPV 11-related CIN	0 / 1960	3316.0	0.0	3 /1975	3334.9	0.1	100.0 (<0, 100.0)
HPV 16-related CIN	**0 / 1887**	**3201.0**	**0.0**	**22 /1847**	**3130.6**	**0.7**	**100.0 (82.1, 100.0)**
HPV 18-related CIN	0 / 2101	3557.9	0.0	8 /2120	3569.1	0.2	100.0 (41.2, 100.0)
By lesion type							
CIN 1	0/2240	3779.8	0.0	25 / 2258	3789.7	0.7	100.0 (84.1, 100.0)
CIN 2 or worse	0/2240	3779.8	0.0	20 / 2258	3794.4	0.5	100.0 (79.7, 100.0)
CIN 2	0/2240	3779.8	0.0	14 / 2258	3794.8	0.4	100.0 (69.7, 100.0)
CIN 3/AIS	**0/2240**	**3779.8**	**0.0**	**10 / 2258**	**3796.2**	**0.3**	**100.0 (55.2, 100.0)**

In the MITT-3 population that included subjects seropositive/PCR positive to relevant HPV type(s) at baseline, vaccine efficacy was much lower, 43% against the combined endpoint and non-significant against CIN 2 or worse (23%) and CIN 3/AIS (0.2%).

Analysis of efficacy against HPV 6/11/16/18-related CIN in MITT-3 population (P013)

Endpoint	Quadrivalent HPV vaccine (n=2717)			Placebo (n=2725)			Observed efficacy % (95%CI)
	Number of cases	Person years at risk	Incidence rate/100 PY at risks	Number of cases	Person years at risk	Incidence rate/100 PY at risks	
HPV 6/11/16/18-related CIN	65 / 2607	5566.5	1.2	113 / 2611	5525.4	2.0	**42.9 (21.9, 58.6)**
By HPV type							
HPV 6-related CIN	4 / 2607	5593.5	0.1	18 / 2611	5570.6	0.3	77.9 (32.8, 94.6)
HPV 11-related CIN	0 / 2607	5597.2	0.0	9 / 2611	5574.5	0.2	100.0 (49.5, 100)
HPV 16-related CIN	54 / 2607	5577.4	1.0	79 / 2611	5551.6	1.4	32.0 (2.6, 52.8)
HPV 18-related CIN	8 / 2607	5590.0	0.1	22 / 2611	5570.5	0.4	63.8 (15.5, 86.1)
By lesion type							
CIN 1	41 / 2607	5576.2	0.7	83 / 2611	5534.5	1.5	51.0 (27.9, 67.1)
CIN 2 or worse	48 / 2607	5585.0	0.9	62 / 2611	5570.4	1.1	**22.8 (<0, 48.2)**
CIN 2	35 / 2607	5590.4	0.6	40 / 2611	5573.7	0.7	12.8 (<0, 46.2)
CIN 3/AIS	35 / 2607	5588.8	0.6	35 / 2611	5579.0	0.6	**0.2 (<0, 39.3)**

The cumulative incidence of HPV vaccine type-related CIN was 4.3% in the placebo and 2.5% in the vaccine group. The vaccine reduced the risk of diagnosis with HPV 6/11/16/18-related CIN from 1 in 23 to 1 in 40 over the 2-year mean duration of follow-up.

Efficacy against external genital lesions

In the primary analysis of the PPE population, all cases of EGL and of VIN/VaIN 2/3 occurred in the placebo group (see below). In the MITT-2 population VE against HPV6/11/16/18-related EGL was 94.9% (95% CI: 84.4, 99.0).

Efficacy by HPV type and by EGL lesion (PPE population) (P013)

Endpoint	Quadrivalent HPV vaccine (n=2717)			Placebo (n=2725)			Observed efficacy % (95%CI)
	Number of cases	Person years at risk	Incidence rate/100 PY at risks	Number of cases	Person years at risk	Incidence rate/100 PY at risks	
HPV 6/11/16/18-related EGL	0 / 2261	3865.2	0.0	40/ 2279	3868.4	1.0	100 (88.4, 100.0)
By HPV type							
HPV 6-related EGL	0 / 1978	3378.7	0.0	23 / 1991	3391.1	0.7	100 (82.5, 100)
HPV 11-related EGL	0 / 1978	3378.7	0.0	10 / 1991	3399.1	0.3	100 (55.1, 100)
HPV 16-related EGL	0 / 1890	3232.7	0.0	10 / 1855	3166.6	0.3	100 (56.3, 100)
HPV 18-related EGL	0 / 2120	3627.5	0.0	3 / 2136	3647.81	0.1	100 (<0.0, 100)
By lesion type							
Condyloma, VIN1 or VaIN 1	0 / 2261	3865.2	0.0	34 / 2279	3870.7	0.9	100 (88.5, 100)
VIN 2/3 or VaIN 2/3	**0 / 2261**	**3865.2**	**0.0**	**7 / 2279**	**3887.5**	**0.2**	**100 (30.2, 100)**

In the MITT-3 population, further EGL cases were observed, but vaccine efficacy (VE) against the combined incidence was still significant (68%, lower bound of 95% CI >20%). VE was non-significant against VIN/VaIN 2/3 (lower bound of 95% CI <0%). Cumulative incidence of HPV 6/11/16/18-related EGL was 3% in the placebo group and 1% in the vaccine group. Vaccination reduced the risk of EGL diagnosis from 1 in 33 to 1 in 103 over the 2 years of follow-up.

Efficacy by HPV type and by EGL lesion (MITT-3 population) (P013)

Endpoint	Quadrivalent HPV vaccine (n=2717)			Placebo (n=2725)			Observed efficacy % (95%CI)
	Number of cases	Person years at risk	Incidence rate/100 PY at risks	Number of cases	Person years at risk	Incidence rate/100 PY at risks	
HPV 6/11/16/18-related EGL	26 / 2671	5697.6	0.5	80 / 2668	5648.4	1.4	**67.8 (49.3, 80.1)**
By HPV type							
HPV 6-related EGL	19 / 2671	5707.2	0.3	51 / 2668	5673.4	0.9	63.0 (36.2,79.4)
HPV 11-related EGL	2 / 2671	5728.0	0.0	16 / 2668	5708.2	0.3	87.5 (47.0, 98.6)
HPV 16-related EGL	5 / 2671	5724.6	0.1	19 / 2668	5708.5	0.3	73.8 (27.3, 92.3)
HPV 18-related EGL	1 / 2671	5728.9	0.0	8 / 2668	5731.2	0.1	87.5 (7.0, 99.7)
By lesion type							
Condyloma, VIN1 or VaIN 1	22 / 2671	5701.8	0.4	72 / 2668	5653.1	1.3	69.7 (50.6, 82.1)
VIN 2/3 or VaIN 2/3	4 / 2671	5726.9	0.1	11 / 2668	5715.5	0.2	**63.7 (<0.0, 91.6)**

Study 015 (Future II): This study was focused on HPV 16/18-related high-grade cervical lesions and had one primary endpoint, the combined incidence of HPV 16- or 18-related CIN 2, CIN 3, AIS or cervical cancer. The interim analysis of vaccine efficacy demonstrated that the vaccine was 100% efficacious in the PPE population.

Interim analysis of efficacy against HPV16/18-related CIN 2/3 (PPE population)

Primary endpoint	Quadrivalent HPV vaccine (n=6082)			Placebo (n=6075)			Observed efficacy (95%CI)
	Number of cases	Person years at risk	Incidence rate/100 PY at risks	Number of cases	Person years at risk	Incidence rate/100 PY at risks	
HPV 16/18-related CIN 2/3 or worse	0 / 5301	7435.1	0.0	21 / 5258	7385.5	0.3	100 (75.8, 100.0)
By HPV type							
HPV 16-related CIN ≥ 2/3	0 / 4552	6407.9	0.0	16 / 4405	6215.7	0.3	100 (74.8, 100)
HPV 18-related CIN ≥ 2/3	0 / 5051	7083.2	0.0	8 / 4968	6980.2	0.1	100 (42.3, 100)
By lesion type							
CIN 2	0 / 5301	7435.1	0.0	15 / 5258	7386.3	0.2	100 (72.3, 100)
CIN 3/AIS	0 / 5301	7435.1	0.0	16 / 5258	7386.5	0.2	100 (74.2, 100)
Cervical cancer	0 / 5301	7435.1	0.0	0 / 5258	7387.4	0.0	NA

The efficacy in the MITT-2 population (VE: 97%) was supportive of the PPE results. The additional 13 placebo cases and one vaccine case (HPV 16) observed in the MITT-2 were subjects who became infected with vaccine HPV types during the vaccination period prior to Month 7.

The magnitude of efficacy was substantially lower in the MITT-3 population, but still significant against CIN 2 (VE: 51%) and CIN 3/AIS (VE: 44%). Compared with MITT-2, there were 141 additional cases (66 vaccine, 75 placebo). The great majority of these cases occurred among subjects who were HPV 16 and 18 PCR positive and/or seropositive at baseline. Over a 2-year follow-up the cumulative incidence of HPV16/18-related CIN 2/3 in the placebo and vaccine group were 1.9% and 1.1%, respectively with a total of 1 in 54 subjects and 1 in 89 subjects, respectively, developing such a lesion.

Summary of primary efficacy analysis in the MITT populations (P015)

Endpoint	Quadrivalent HPV vaccine (n=6082)			Placebo (n=6075)			Observed efficacy (95%CI)
	Number of cases	Person years at risk	Incidence rate/100 PY at risks	Number of cases	Person years at risk	Incidence rate/ 100 PY at risks	
HPV 16/18-related CIN 2/3 or worse:							
PPE	0 / 5301	7435.1	0.0	21 / 5258	7385.5	0.3	100 (75.8, 100.0)
MITT-2	1 / 5736	10797.2	0.0	36 / 5766	10885.5	0.3	97.2 (83.4 100.0)
CIN 2	1 / 5736	10797.2	0.0	28 / 5766	10883.1	0.2	96.3 (77.4, 100.0)
CIN 3 /AIS	0 / 5736	10797.2	0.0	27 / 5766	10885.2	0.2	100 (85.2, 100.0)
MITT-3	67 / 5947	11159.5	0.6	111 / 5973	11243.9	1.0	**39.2** (16.9, 55.8)
By HPV type							
HPV 16-related CIN	62 / 5947	11161.1	0.6	99 / 5973	11247.5	0.9	36.9 (12.4, 54.8)
HPV 18-related CIN	5 / 5947	11176.5	0.0	22 / 5973	11264.5	0.2	77.1 (38.0, 93.2)
By lesion type							
CIN 2	36 / 5947	11169.5	0.3	74 / 5973	11254.8	0.7	**51.0** (26.0, 68.0)
CIN 3/AIS	47 / 5947	11167.5	0.4	85 / 5973	11256.5	0.8	**44.3 (**19.5, 61.8)
Cervical cancer	0 / 5947	11178.0	0.0	0 / 5973	11267.9	0.0	NA

In the secondary efficacy analysis the vaccine was demonstrated to reduce the incidence of EGLs in all populations studied with VE of 99% (95% CI: 91.8, 100) in the PPE population and 71% (95% CI: 58.8, 79.9) in the MITT-3 population. With respect to VIN/VaIN 2/3, there were 0 cases in the vaccine group versus 6 cases in the placebo group in the PPE population (VE 100% (95% CI: 15.2, 100)) and 4 versus 18 cases, respectively, in the MITT-3 population (VE 78% (95% CI: 32.2, 94.5)).

Ancillary analyses

- *Analysis performed across trials (pooled analyses and meta-analysis)*

The data on the separate EGL diseases were given for separate studies only in the PPE population but for the integrated efficacy dataset (P007, P013, and P015) in the PPE-, MITT-2 and MITT-3 populations. In the PPE combined analyses statistically significant vaccine efficacy was demonstrated against both vulvar and vaginal condyloma acuminata, VIN 1, VIN 2/3 and VaIN 1. VE against VaIN 2/3 was not established due to very few cases. However for single studies only efficacy against genital warts (P013, P015) and VIN 1 (P015) was established. The majority of condyloma was HPV 6 and 11-related and located in the vulva.

Analysis of efficacy against HPV 6/11/16/18-related EGL by disease severity, PPE population (P007, 013, 015)

	Gardasil (n=9075)			Placebo (n=9075)			Observed efficacy % (95%CI)
	Number of cases	Person years at risk	Incidence rate/100 PY at risks	Number of cases	Person years at risk	Incidence rate/100 PY at risks	
HPV 6/11/16/18-related condyloma	**1 / 7897**	**11977,9**	**0.0**	**91 / 7899**	**11953.4**	**0.8**	**98.9 (93.7, 100)**
Vulvar condyloma	1 / 7897	11977.9	0.0	88 / 7899	11955.2	0.7	98.9 (93,5, 100)
Vaginal condyloma	0 / 7897	11979.2	0.0	8 / 7899	11986.8	0.1	100 (41.4, 100)
HPV 6/11/16/18-related VIN 1	0 / 7897	11979.2	0.0	10 / 7899	11986.3	0.1	100 (55.4, 100)
HPV 6/11/16/18-related VIN 2/3	0 / 7897	11979.2	0.0	8 / 7899	11988.3	0.1	100 (41.4, 100)
HPV 6/11/16/18-related VaIN 1	0 / 7897	11979.2	0.0	7 / 7899	11987.5	0.1	100 (30.6, 100)
HPV 6/11/16/18-related VaIN 2/3	0 / 7897	11979.2	0.0	5 / 7899	11989.9	0.0	100(<0.0, 100)

With respect to the most important endpoint, HPV 16/18-related VIN 2/3 and VaIN 2/3, vaccine efficacy was statistically significant only in the MITT-2 population for the combined analysis. Overall, there were very few cases of VaIN 2/3. However, data on VaIN 1 of which the majority were HPV 16/18-related and vaginal condyloma, the same distribution of cases were seen as for VaIN 2/3 with almost all in the placebo group (0 vs. 13 and 2 vs.15, respectively).

Analysis of efficacy of against HPV 16/18-related VIN 2/3 and VaIN 2/3 (P007, P013, P015)

	Gardasil (n=9075)			Placebo (n=9075)			Observed efficacy % (95%CI)
	Number of cases	Person years at risk	Incidence rate/100 PY at risks	Number of cases	Person years at risk	Incidence rate/100 PY at risks	
Per-Protocol‡							
HPV 16/18-Related VIN 2/3	0 / 7,769	11,786.6	0.0	5 / 7,741	11,754.3	0.0	100 (<0.0, 100.0)
HPV 16/18-Related VaIN 2/3	0 / 7,769	11,786.6	0.0	5 / 7,741	11,754.2	0.0	100 (<0.0, 100.0)
MITT-2							
HPV 16/18-Related VIN 2/3	0 / 8,641	17,079.0	0.0	17 / 8,667	17,163.3	0.1	100 (75.6, 100.0)
HPV 16/18-Related VaIN 2/3	0 / 8,641	17,079.0	0.0	7 / 8,667	17,169.4	0.0	100 (30.3, 100.0)
MITT-3‖							
HPV 16/18-Related VIN 2/3	7 / 8,954	17,673.1	0.0	18 / 8,962	17,730.0	0.1	61.0 (2.1, 86.2)
HPV 16/18-Related VaIN 2/3	2 / 8,954	17,678.4	0.0	9 / 8,962	17,734.5	0.1	77.7 (<0.0, 97.7)

Integrated summary of efficacy –HPV 6/11/16/18-related CIN and EGL of all grades

An integrated summary of vaccine efficacy with respect to CIN and EGL outcomes in studies 007, 013 and study 015 was pre-planned. Data from study 005 contributed only with respect to HPV 16-related CIN. A total of 20,887 women were included in this analysis. The summary report focused on the incidence of HPV 6/11/16/18-related CIN and EGL of all grades.

The integrated data from all efficacy studies, with regard to CIN endpoints, demonstrated that the vaccine in the PPE population:

- Reduced the incidence of HPV 6/11/16/18-related CIN/AIS (4 vs. 83 placebo cases; VE: 95.2% (95% CI: 87.2, 98.7)
- Reduced the incidence of CIN 1 caused by vaccine HPV types (4 vs. 58 placebo cases; VE: 93.1 % (95% CI: 81.4, 98.2))
- Prevented the development of International Federation of Gynaecology and Obstetrics (FIGO) Stage 0 (non-invasive) cervical cancer (CIN 3/AIS) caused by vaccine HPV types (0 vs. 26 placebo cases; VE: 100% (95% CI: 84.8, 100))

The vaccine efficacy against CIN endpoints in the MITT-3 population was 46.4% (95% CI: 35.2, 55.7) (170 vs. 317 placebo cases), 54.4% (95% CI: 41.8, 64.5) (97 vs. 213 placebo cases) and 33.1% (95% CI: 11.1, 49.8) (84 vs. 126 placebo cases), respectively.

The vaccine efficacy appeared comparable with respect to each HPV type included in the vaccine.

The integrated data from all efficacy studies in the PPE population with regard to EGL endpoints provided evidence that prophylactic administration of the HPV vaccine:

- Reduced the incidence of HPV 6/11/16/18-related EGLs (1 vs. 113 placebo cases; VE 99.1% (95% CI: 95.0, 100)) and most importantly
- Prevented the development of VIN 2/3 or VaIN 2/3 caused by vaccine HPV types (0 vs.13 placebo cases; VE: 100% (95% CI: 67.2, 100)).

The vaccine efficacy against EGL endpoints in the MITT-3 population was 70.4% (95% CI: 61, 77.7) (68 vs. 229 placebo cases) and 73.3% (95% CI: 40.3, 89.4) (8 vs. 30 placebo cases), respectively.

The majority of the VIN/VaIN 2/3 were caused by HPV 16/18 (PPE: 10 of 13 and MITT-3: all 8 vaccine and 26 of 30 placebo).

Genital warts predominated among EGLs. The vaccine was efficacious against HPV 6/11/16/18-related genital warts (+ VIN/VaIN 1) (VE 99% (95% CI: 94.4, 100), and those caused by HPV 16/18 (VE 100% (95% CI: 83.4, 100) in the PPE population. In the MITT- 3 population VE was 70.1% and 69.1%, respectively.

The vaccine efficacy appeared comparable with respect to each type included in the vaccine, although there were few HPV 18-related EGLs (95% CI lower bound at 50%).

- *Combined interim efficacy analysis - HPV 16/18-related CIN 2/3*

The composite endpoint of HPV 16/18-related CIN 2/3 and AIS was chosen as the primary surrogate clinical outcome against which the efficacy of Gardasil in preventing cervical cancer was evaluated. To increase the precision of vaccine efficacy estimate, a combined analysis of data from all 4 efficacy trials was pre-planned. This study population included in this analysis was 20,541 subjects. Data from study 005 contributed only to the analysis of HPV 16-related CIN. The primary efficacy analysis was based on 53 CIN 2/3 cases and all occurred in the placebo group; vaccine efficacy was 100% in the PPE population.

Efficacy against HPV 16/18-related CIN 2/3 or worse (P007, P005, P013, P015, combined) (PPE population)

Study population	Quadrivalent HPV vaccine (n=10268)			Placebo (n=10273			Observed Efficacy (95%CI)
	Number of cases	Person years at risk	Incidence rate/100 PY at risks	Number of cases	Person years at risk	Incidence rate/100 PY at risks	
Per-Protocol							
Combined protocols	**0/ 8,487**	**14,178.1**	**0.0**	**53 / 8,460**	**14,060.6**	**0.4**	**100 (92.9, 100.0)**
P005	0 / 755	2,471.9	0.0	12 / 750	2,393.9	0.5	100 (65.1, 100.0)
P007	0 / 231	554.4	0.0	1 / 230	545.2	0.2	100 (<0.0, 100.0)
P013	0 / 2,200	3,716.7	0.0	19 / 2,222	3,736.0	0.5	100 (78.5, 100.0)
P015	0/ 5,301	7,435.1	0.0	21 / 5,258	7,385.5	0.3	100 (80.9, 100.0)
By HPV type							
HPV 16	0 / 7,393	12,558.6	0.0	44 / 7,200	12,218.1	0.4	100 (91.5, 100.0)
HPV 18	0 / 7,376	11,179.1	0.0	14 / 7,312	11,080.8	0.1	100 (70.1, 100.0)
By lesion type							
CIN 2	0 / 8,487	14,178.1	0.0	36 / 8,460	14,064.0	0.3	100 (89.3, 100.0)
CIN 3 /AIS	0 / 8,487	14,178.1	0.0	**32** / 8,460	14,066.2	0.2	**100 (87.9, 100.0)**
MITT-3							
Combined protocols	**122 / 9,831**	**21,107.3**	**0.6**	**201/ 9,896**	**21,228.4**	**0.9**	**39.0 (23.3, 51.7)**
By lesion type							
CIN 2	76/ 9,831	21,123.1	0.4	131 / 9,896	21,246.6	0.6	41.8 (22.1, 56.7)
CIN 3 /AIS	85 /9,831	21,119.4	0.4	134 / 9,896	21,256.6	0.6	**36.3** (15.7, 52.0)

In the MITT-3 population VE was lower 39%. When analysed by disease severity, vaccine efficacy was 100% against CIN 3/AIS (stage 0 cancer) in the PPE population and 36% in the MITT-3 population. Most CIN 2/3 cases were caused by HPV 16, but comparable vaccine efficacy could be

demonstrated for HPV 18. The results in relevant MITT-1 and MITT-2 populations were supportive of the primary PPE conclusion. Various sensitivity analyses confirmed the robustness of data.

- *Therapeutic efficacy*

There was no evidence that the quadrivalent HPV vaccine has early therapeutic efficacy against CIN and EGL vaccine HPV-type endpoints in subjects who were baseline seronegative/PCR positive or serpositive/PCR positive.

- *Population benefit integrated summary of efficacy*

The population benefit of the HPV vaccine was measured in terms of the vaccine's impact on the overall rates of CIN and EGL disease due to any HPV type, incidence of Pap test abnormalities, and cervical procedures. Three MITT populations were defined to evaluate these parameters: RMITT-2 (HPV naïve and negative Pap test at Day 1 that received at least 1 vaccine dose), RMITT-3 (all subjects regardless of baseline HPV status with negative Pap test at Day 1 that received at least 1 vaccine dose) and MITT-3 (all subjects regardless of HPV status that received at least 1 vaccine dose).

Among the RMITT-2 population a significant reduction in the risk of CIN 2 and worse (37.9%) and CIN 3 or worse (45.5%) and EGL (66%) due to any type was demonstrated.

In the RMITT-2 population there was a reduction in patients vaccinated with Gardasil compared to placebo with respect to number of colposcopies (15%), cervical biopsies (17.2%) and cervical definitive therapies (28.1%). Results observed in the MITT-3 population were consistent, but the impact of the vaccine was lower.

In the RMITT-3 and MITT-3 population no vaccine efficacy against CIN 2 and worse could be established. Regarding Pap test abnormalities a modest reduction in the overall incidence was observed in the vaccine group compared to placebo.

- **Discussion on clinical efficacy**

Clinical data demonstrate the prophylactic efficacy of Gardasil in 16- to 23-year old female subjects in preventing the incidence of HPV16/18-related CIN 2/3 or AIS, as surrogate markers for cervical cancer and HPV 6/11/16/18-related external genital warts. Based on the similarities between the efficacy studies with regard to design, inclusion/exclusion criteria and detection system for HPV-related CIN, the combined efficacy analysis is acceptable.

The efficacy results were consistent among studies and showed that the vaccine was highly efficacious against HPV 6/11/16/18-related CIN and EGLs in the PPE population. Consistent results were obtained in three MITT populations. However, in the MITT-3 population that included subjects already infected at baseline, vaccine efficacy was much lower. Robustness of data was confirmed in various sensitivity analyses. Individual endpoints were also evaluated in all studies, such as efficacy by HPV type and by lesion type, which is commented on further below. Data on the MITT-2 and -4 populations indicated that the vaccine was efficacious already after 2 doses. However, only 0.3% of study subjects received less than 3 doses (n=39 vaccinees) and therefore no meaningful efficacy analyses after 2 vaccine doses could be made.

Immunogenicity was evaluated in pre-/adolescent females. It was shown that the anti-HPV responses increased as the age at which vaccination was initiated decreased. For each 4 HPV types, the vaccine induced numerically higher GMTs 4 weeks postdose 2 and 3 in 9- to 15-year-old females than in 16- to 26-year old females. Immunogenicity was related to age and Month 7 anti-HPV levels were significantly higher in younger individuals below 12 years of age than in those above that age. The results of the statistical analyses demonstrated that the vaccine induced non-inferior immune responses as measured by GMTs and seroconversion rates in the adolescent cohort compared with young adult women. Since efficacy of the HPV vaccine cannot be evaluated in virginal subjects, these data are considered acceptable to allow the bridging of efficacy data from the young adult women to the pre-/adolescent girls. However, the durability of response in this target group as well as long-term persistence of efficacy and immunogenicity requires close monitoring for 10 to 15 years. This will be critical for the decision of the optimal age to vaccinate sexually naïve subjects. Long-term immunogenicity will be monitored in the target adolescent population during the post-authorisation

period. The applicant also outlines a structured booster program. These issues are addressed in the risk management plan.

The maximum health benefits of the HPV vaccine would be obtained if both genders are vaccinated to break the cycle of transmission of HPV infection and induce herd immunity. The immunogenicity of Gardasil was assessed in male pre-/adolescents 9 to 15 years of age. Anti-HPV responses at Month 7 among 9- to 15-year-old boys were non-inferior to anti-HPV responses in 16- to 26-year-old young women for whom efficacy was established in the phase III studies.

Males were not included in the initial efficacy studies. An ongoing clinical study in 16- to 26-year-old male subjects is evaluating the impact of Gardasil on the incidence of persistent infection as well as precancerous penile and anal lesions and will be completed in the coming years.

With regard to the serotype replacement issue, it was shown that the incidence of disease due to non-vaccine types were 5.5% higher overall in the vaccine group compared to placebo. However time to event curves did not reveal any trend of an increasing event rate in the vaccine group compared with the placebo group. The issue of type replacement is addressed in the Risk Management Plan.

Potential therapeutic effects of the vaccine must be re-analysed at the completion of the 4-year follow-up. At this early stage no beneficial effect could be seen in seronegative/PCR positive subjects. It would also be important to evaluate whether the vaccine could be used to boost natural immunity to maintain or augment protection. This issue will be further addressed at the time of the final analysis of studies 013 and 015.

Results of the population benefit analyses appear promising, but have to be further evaluated on a longer-term basis. The primary analyses of population benefit are planned for later in the course of the phase III trials and will give more reliable estimates of the impact of the HPV vaccine on public health parameters. The analyses performed illustrated that the burden of HPV disease is substantial in these young adult women and that a large amount of disease is caused by non-vaccine HPV types.

Clinical safety

- Patient exposure

The safety of Gardasil and its monovalent precursors was assessed in 12 trials. Of these, 7 trials evaluated the quadrivalent HPV (types 6, 11, 16, 18) L1 VLP vaccine (P 007, P011, P012, P013, P015, P016 and P018). P011 and P012 are substudies of P013.

The data presented on the use of HPV vaccine comprises 16,014 subjects. The overall exposure database includes: 2146 subjects receiving monovalent HPV L1 VLP vaccines; 11,813 subjects receiving Gardasil; 1524 subjects receiving quadrivalent HPV (Types 6, 11, 16, 18) L1 VLP vaccine formulations containing VLP doses lower than those in Gardasil and 552 subjects receiving quadrivalent HPV (Types 6, 11, 16, 18) L1 VLP vaccine formulations containing VLP doses higher than those in Gardasil.

- Adverse events

Overall more subjects that received Gardasil reported an adverse event, which primarily was due to a higher incidence of injection site reactions.

Number (%) of subjects who reported Injection-site Adverse Experience by Maximum Intensity Rating (Days 1 to 5 following Any Vaccination Visit) (P 007, 011, 013, 015, 016 and 018)

	Gardasil (N =6,160) n (%)	Placebo (n=4064) n (%)
Subjects with follow up	6,069	3,992
Subjects without injection-site AEs	1,039	1,067
Subjects with injection-site AEs	5,030 **(82.9)**	2,927 (**73.3**)
Subjects with maximum intensity rating of injection-site AEs		
Mild	3,162 (52.1)	2,125 (53.2)
Moderate	1,586 (26.1)	724 (18,1)
Severe	271 (4.5)	75 (1.9)
Unknown	11 (0.2)	2 (0.1)

N = number of subjects who received 1, 2 or 3 doses of only the clinical material in the given column
n= number of subjects in the respective category

There was also a modest increase in systemic adverse events among those who received Gardasil as compared with placebo.

Numbers (%) of Subjects With Systemic Clinical Adverse Experiences (Incidence ≥ 1% in One or More Vaccination Groups) by System Organ Class (Days 1 to 15 following Any Vaccination Visit) (Protocols 007,011,012,013,015,016 and 018)

	Gardasil (n= 6160)				Placebo (n=4064)			
	All AEs		VR		All AEs		VR	
	N	%	N	%	N	%	N	%
Subjects in analysis population	6160				4064			
Subjects without follow up	91				70			
Subjects with follow up	6069				3994			
Number (%) of subjects with one or more systemic AEs	3591	59.2			2414	60.4		
Number (%) of subjects with no systemic AEs	2478	40.8			1580	39.6		
Ear and Labyrinth Disorders	**70**	**1.2**	**24**	**0.4**	**38**	**1.0**	**14**	**0.4**
Eye disorders	**54**	**0.9**	**9**	**0.1**	**49**	**1.2**	**9**	**0.2**
Gastrointestinal Disorders	**1051**	**17.3**	**418**	**6.9**	**730**	**18.3**	**313**	**7.8**
Abdominal pain	157	2.6	49	0.8	115	2.9	47	1.2
Abdominal pain upper	193	3.2	61	1.0	136	3.4	50	1.3
Diarrhoea	224	3.7	224	1.2	144	3.6	58	1.5
Nausea	370	6.1	370	3.7	251	6.3	157	3.9
Toothache	78	1.3	78	0.0	52	1.3	2	0.1
Vomiting	147	2.4	147	0.9	82	2.1	26	0.7
General Disorders and administration Site Conditions	**1116**	**18.4**	**817**	**13.5**	**726**	**18.2**	**515**	**12.9**
Asthenia	48	0,8	29	0.5	39	1.0	28	0.7
Fatigue	157	2.6	117	1.9	154	3.9	112	2.8
Malaise	75	1.2	49	0.8	46	1.2	33	0.8
Pyrexia	782	12.9	611	10.1	440	11.0	336	8.4
Infections and Infestations	**1046**	**17.2**	**184**	**3.0**	**735**	**18.4**	**143**	**3.6**
Influenza	192	3.2	46	0.8	154	3.9	51	1.3
Nasopharyngitis	353	5.8	87	1.4	245	6.1	61	1.5
Pharyngitis	50	0.8	6	0.1	40	1.0	6	8.4
Upper respiratory tract infection	93	1.5	4	0.1	59	1.5	3	0.1
Injury, Poisoning and Procedural Complications	**143**	**2.4**	**6**	**0.1**	**85**	**2.1**		
Musculoskeletal and Connective Tissue Disorders	**499**	**8.2**	**191**	**3.1**	**352**	**8.8**	**137**	**3.4**
Arthralgia	74	1.2	25	0.4	39	1.0	14	0.4
Back pain	116	1.9	21	0.3	99	1.0	26	0.7
Myalgia	119	2.0	66	1.1	81	2.5	49	1.2
Pain in extremity	118	1.9	49	0.8	95	2.4	39	1.0
Nervous System Disorders	**1782**	**29.4**	**1257**	**20.7**	**1231**	**30.8**	**877**	**22.0**
Dizziness	214	3.5	153	2.5	142	3.6	101	2.5
Headache	1602	26.4	1136	18.7	1101	27.6	796	19.9
Somnolence	49	0,8	34	0.6	43	1.1	27	0.7

Psychiatric Disorders	**106**	**1.7**	**31**	**0.5**	**77**	**1.9**	**23**	**0,6**
Insomnia	60	1.0	23	0.4	34	0.9	14	0.4
Reproductive System and breast Disorders	**352**	**5.8**	**41**	**0.7**	**266**	**6.7**	**44**	**1.1**
Dysmenorrhoea	178	2.9	18	0.3	152	3.8	26	0.7
Respiratory Thoracic and Mediastinal Disorders	**490**	**8.1**	**96**	**1.6**	**321**	**8.0**	**77**	**1.9**
Cough	117	1.9	18	0.3	63	1.6	15	0.4
Nasal congestion	67	1.1	13	0.2	39	1.0	6	0.2
Pharyngolaryngeal pain	266	4.4	49	0.8	190	4.8	45	1.1
Skin and Subcutanous Tissue Disorders	**210**	**3.5**	**76**	**1.3**	**143**	**3.6**	**53**	**1.3**
Percentages are calculated based on the number of subjects with follow-up VR= vaccine related								

The overall profile of systemic adverse events, based on the proportions and types of adverse events reported were generally similar between the 2 vaccination groups. Pyrexia, respiratory disorders, infections and infestations and nervous system disorders prevailed. However, most of these adverse events were not serious. The proportions of subjects reporting any systemic clinical adverse experience or a vaccine related such event were comparable between the vaccination groups.

Immunological adverse events

A summary of the adverse experiences conditions considered immunological events are displayed in the table below: anaphylactic reaction; bronchospasm/wheezing and urticaria.

Summary of Adverse Experiences Considered to be Immunological Events

Adverse Experience (AE)	Gardasil (n= 11,778)	Placebo (n=9,686)
Anaphylactic Reaction	0	1
Vaccine Related	0	0
Intensity		
Mild	0	0
Moderate	0	0
Severe	0	1*
Bronchospasm/Wheezing	4	1
Vaccine related°	1	1
Intensity		
Mild	0	1
Moderate	3	0
Severe	1	0
Urticaria	12	28
Vaccine Related°	7	17
Intensity		
Mild	7	16
Moderate	4	7
Severe	1	5

° determined to be possible, probably or definitely vaccine related * determined to be a serious adverse experience (SAE)

A higher number of the subjects in the Gardasil group reported bronchospasm or wheezing. However, only one subject in each vaccination group had a study-vaccine related adverse event. The number of subjects who reported urticaria was higher in the placebo group. The proportions of subjects with urticaria that was considered study vaccine related were comparable in both groups.

Systemic Autoimmune Disorder – Musculoskeletal and Connective Tissue Disorder

The table below displays the "Overall Incident Conditions of Systemic Autoimmune Connective Tissue Diseases" reported in the clinical trials program for Gardasil by vaccination group. In addition, non-specific diagnoses that could represent systemic inflammatory conditions are also included.

Summary of Subjects Who reported an Incident Condition Potentially Indicative of Systemic Autoimmnue Disorder After Enrolment in Clinical Trials of Gardasil

Potential Autoimmune Disorder	Gardasil (n= 11,813)	Placebo (n=9,701)
Specific terms	3 (0.025%)	1 (0.010%)
Juvenil arthritis	1	0

Rheumatoid arthritis	2	0
Systemic lupus erythematosus	0	1
Other terms	6 (0.051%)	2 (0.021%)
Arthritis	5	2
Reactive arthritis	1	0
Polyarthritis	0	0

The proportions of subjects who reported a history of musculoskeletal and connective tissue disorders at enrolment or after enrolment were comparable between the vaccination groups. Except for juvenile arthritis, the incidences of these conditions were consistent with the incidences described in the literature. One case of juvenile arthritis was reported in a subject who received Gardasil.

- Serious adverse event/deaths/other significant events

Few subjects reported serious adverse events and the proportions were comparable between the vaccination groups within the safety population. The types of serious adverse experiences reported between the vaccination groups were also comparable. The most common serious adverse events in both vaccination groups were infections and pregnancy. Overall 6 serious adverse experiences were determined to be possibly, probably or definitively related to the study vaccine/placebo.

Listings of Subjects with Serious Vaccine Related Clinical Adverse Experiences (Entire study period*)
Safety Population (P 007, 011, 012, 013, 015, 016 and 018)

Gender	Race	Age at first vac	Rel Day from start Trial	Dose Number Vaccine given	Rel Day of onset post dose	Adverse experience	Duration of AE	Intensity/ Size	Vaccine Relation ship	Outcome
Gardasil										
F	Hisp	20y	162	3 Gardasil	1	**Bronchospasm**	2 day	Severe	Poss	Recov
F	Asian	22y	47	2 Gardasil	5	**Gastroenteritis**	15 day	Severe	Poss	Recov
F	Hisp	22y	156	3 Gardasil	1	**Headache**	5 day	Severe	Def	Recov
			156	3 Gardasil	1	**Hypertension**	1day	Severe	Def	Recov
F	White	21y	43	2 Gardasil	1	**Injection site joint movements impairment**	5.09 mo	Mod	Prob	Recov
			43	2 Gardasil	1	**Injection site pain**	5.09 mo	Mod	Prob	Recov
Placebo										
F	White	20y	58	2 Placebo	1	**Hypersensitivity**	3day	Mod	Poss	Recov
F	Hisp	21y	54	2 Placebo	1	**Chills**	1day	Mod	Poss	Recov
			54	2 Placebo	1	**Headache**	1day	Mod	Poss	Recov
			54	2 Placebo	1	**Pyrexia**	1day	mod	Poss	Recov
Gardasil										
F	Asian	13y	26	1 Gardasil	26	**Vaginal haemorrhage**	1.71 mo	Mod	Prob	Recov
F	Asian	13y	223	3 Gardasil	42	**Vaginal haemorrhage**	2.30 mo	Severe	Prob	Recov

Eight subjects in the Safety Population group that received Gardasil (0.07%) and 6 subjects in the placebo group (0,06%) died during the course of trials. Three subjects died within 15 days and 11 died following any vaccination. None of the deaths was considered to be related to vaccine/ placebo or procedure. Five deaths were due to trauma, 3 were due to intentional overdose (non-study medications) or suicide, and 2 were due to pulmonary embolus; most likely related to use of hormonal contraceptives. There was 1 case each of cancer, infection, and complication of Caesarean section, asphyxia and arrhythmia.

- Discontinuation due to adverse events

Few subjects (0.1% in each vaccination group) discontinued due to an adverse experience.

- Safety in special populations

Pregnancy, lactating women, infants to lactating women, adolescent boys and girls

The numbers of subjects who reported an injection-site adverse event were higher in the groups that received Gardasil compared with the placebo groups; and the number of subjects who reported systemic clinical adverse events were comparable between vaccination groups. The adverse event profiles in 18- to 26-year-old women, 9- to 17-year-old girls and 9- to 15-year-old boys who received Gardasil showed that the proportions of subjects who experienced a serious adverse event or who discontinued due to an adverse event were comparable among the 3 groups. The proportions of subjects who reported an adverse event over all, an injection-site event, and a systemic event were highest among 18- to 26-year-old women and lowest in 9 to 15 year old boys. Among subjects who received Gardasil, the proportions of subjects who reported a fever were comparable.

Pregnancy

Although HPV is not a teratogenic and there were no theoretical concerns or experimental data to suggest that HPV L1 VLPs or aluminium adjuvant are teratogenic, the clinical studies within the clinical development program prohibited vaccination of pregnant subjects. Thus, there are no studies that directly randomized pregnant women to receive Gardasil or placebo.

In order to evaluate the interaction of vaccination and pregnancy outcomes, the outcomes of pregnancy were summarised according to whether the ECDn (Estimated Day of Conception) occurred within 30 days of a study vaccination or whether it occurred beyond this time period. The methodology used to determine the estimated date of conception was based on data described in the literature, and ranked source data in order of accuracy in the ability to estimate the gestational age and from this the EDCn.

During the course of immunisation with Gardasil, overall 2,266 women (vaccine: 1,115 vs. placebo: 1,151) reported at least one pregnancy. Among these pregnancies outcomes were known for 78.1% of all pregnancies. With few exceptions, the pregnancies with unknown outcomes represented either ongoing pregnancies or subjects that discontinued or were lost to follow-up.

- *Live born infants*

The proportion of live births that were accompanied by other medical conditions was slightly higher in the group that received Gardasil than in the placebo group. The most common medical condition observed during the neonatal period other than congenital anomaly were prematurity (9 and 8 infants, of mothers that received Gardasil and placebo, respectively) neonatal respiratory distress symptom (2 and 5 infants of mothers who received Gardasil or placebo, respectively), and neonatal jaundice (6 and 4 infants of mothers who received Gardasil or placebo, respectively).

- *Congenital anomalies*

The proportion of live births that resulted in congenital anomalies was slightly higher in the group that received Gardasil compared with the placebo group. However, the number of pregnancies that resulted in a congenital anomaly was small and well within the 3-4% incidence reported in studies of pregnancy in large-scale health care systems. If pregnancies resulting in foetal loss due to a congenital anomaly and pregnancies with live birth in which congenital anomaly was detected after the immediate neonatal period are included, a total of 13 pregnancies in the group that received Gardasil and 12 in the group that received placebo, resulted in a congenital anomaly.

Further sub-analyses were done to evaluate pregnancies with estimated onset within 30 days or more than 30 days from administration of a dose of Gardasil or placebo. For pregnancies with estimated onset within 30 days of vaccination, 5 cases of congenital anomaly were observed in the group that received Gardasil compared to 0 cases of congenital anomaly in the group that received placebo. Conversely, in pregnancies with onset more than 30 days following vaccination, 8 cases of congenital anomaly were observed in the group that received Gardasil compared with 12 cases of congenital anomaly in the group that received placebo. No trend of specific effect on any organ system in relation to the week of gestational development could be observed. An independent expert in Teratology and in the impact of environmental pregnancy outcomes concluded that the congenital anomaly events were not associated with exposure to Gardasil or to aluminium-placebo.

Congenital anomalies in vaccinated subjects, cases occurring within 30 days of vaccination

#1: Twin birth, Twin A: Hip dysplasia	Study vaccine Dose 1

#2: Pyloric stenosis, Ankyloglossia	Study vaccine Dose 1
#3: Congenital hydronephrosis	Study vaccine Dose 2
#4 Congenital megacolon	Study vaccine Dose 2
#5 Left talipes equinaovarus	Study vaccine Dose 1

Congenital anomalies not detected at birth were also found in 3 infants of mothers given placebo and in one infant born to a mother given Gardasil.

- *Spontaneous abortion*

Analyses were also performed to evaluate pregnancies with estimated onset within 30 days or beyond 30 days from administration of Gardasil or placebo. For pregnancies with estimated onset within 30 days of vaccination, the proportion of pregnancies that resulted in spontaneous pregnancy loss was lower in subjects who received Gardasil (23.1%) as compared to subjects who received placebo (28.3%). The opposite pattern was seen in pregnancies with onset beyond 30 days from any vaccination. The proportion of pregnancies with spontaneous pregnancy loss was higher in subjects who received Gardasil (34.2%) compared with subjects who received placebo (31.9%).

- *Adverse events reported by study subjects during pregnancy*

Overall 40 and 41 subjects in the group that received Gardasil or placebo, respectively (4.2% and 4.3% of all subjects who reported a pregnancy in respective vaccine group) experienced an adverse event. The most common events reported were conditions that can result in Caesarean sections, premature onset of labour and pregnancy related medical problems. The proportions of pregnant subjects who experienced such events were comparable between the vaccination groups.

- *Other adverse events in infants of study subjects*

Overall, 9 and 13 infants born to women who received Gardasil or placebo experienced such adverse event. None of these, however, had a causal relationship to the vaccine or placebo.

Administration of Gardasil to lactating women

Lactating women were also included in the studies. Medical events meeting criteria for serious adverse events were collected in both mother and her infant(s) from the time of the possible exposure of the infant to the vaccine via breast milk until the child was weaned.

A total of 995 subjects (500 and 495, who received Gardasil and placebo, respectively) in the safety population group were breastfeeding during the vaccination period. In the subjects with serious, vaccine related clinical adverse events, 4 subjects, 2 subjects and 1 subject had at least 1 serious adverse experience that was determined to be possibly, probably, or definitely related to the vaccine, respectively.

- *Infants to the lactating mothers*

A total of 17 (3.4%) and 9 (1.8%) infants of subjects, respectively, who were breast-feeding during the period when they received vaccine or placebo, experienced a serious adverse event. None of these were judged to be related to the vaccine

Male Subjects

All male subjects in the clinical development program for Gardasil were 9 to 15 years of age at study enrolment. In boys the proportion of subjects reporting any adverse experience and any injection-site adverse event were higher in the group that received Gardasil compared with the non-aluminium-containing placebo group.

Number of subjects who reported any clinical adverse experience by maximum intensity rating (Days 1 to 15 following any vaccination visit) – male subjects 9to 15 years of age at enrolment (P016 and 18)

	Gardasil (N= 1,071) n= (%)	**Placebo (Non- aluminium containing) (N=274) n (%)**
Subjects with follow-up	1,056	269

Subjects without adverse experiences	186	96
Subjects with adverse experiences	870 **(82.4)**	173 (64.3)
Subjects by maximum intensity rating of adverse experiences		
Mild	437 (41.1)	96 (35,7)
Moderate	313 (29.6)	60 (22.3)
Severe	108 (10.2)	15 (5.6)
Unknown	12 (1.1)	2 (0.7)
N = number of subjects who received 1, 2 or 3 doses of only the clinical material indicated in the given column n=number of subjects in respective category		

Serious adverse event and discontinuations due to an adverse event were rare, but all these events occurred in boys who received Gardasil. Boys who received Gardasil reported somewhat more systemic clinical adverse experiences compared with boys who received non-aluminium-containing placebo. The serious adverse event profile was similar to those seen in the other age cohorts and the incidence of injection site adverse event and fever predominated in this group.

More boys who received Gardasil reported specific injection-site adverse event. Somewhat more male subjects reported systemic clinical adverse event in the group who received Gardasil compared with the placebo group. The most common systemic reactions were headache, pyrexia, diarrhoea and pharyngolaryngeal pain. A higher proportion of male subjects, who received Gardasil, reported pyrexia compared with subjects in the non-aluminium containing placebo group.

Number of subjects with injection site adverse experiences (incidence ≥1% in one ore more vaccination groups) (Day 1 to 5 following Any Vaccination visit) Detailed Safety Population) – Male subjects 9 to 15 years of age at study enrolment (P016 and P 018)

	Gardasil N=1071				**Placebo (non-aluminium containing) N=274**			
	All adverse experiences		**VR**		**All adverse experiences**		**VR**	
	N	%	**N**	**%**	**N**	**%**	**N**	**%**
Number of subjects	1071				274			
Subjects without follow up	15				5			
Subjects with follow up	1056				269			
Number (%) of subjects with one or more injection-site adverse experiences	757	71.7			128	47.6		
Injection site Erythema	196	18.6	196	18.6	39	14.5	39	14.5
Injection site Haemorrhage	19	1.8	19	1.8	10	3.7	10	3.7
Injection site pain	731	69,2	731	69,2	112	41.6	112	41.6
Injection site pruritus	9	0.9	9	0.9	3	1.1	3	1.1

Nine to 15 years old male subjects who received Gardasil reported fewer adverse events compared with 18 to 26 year old female subjects. The proportions of male and female subjects who reported serious adverse experiences were comparable.

Fewer male subjects who received Gardasil reported injection-site reactions or systemic clinical adverse events compared with 9- to 17-year-old female subjects. The proportions of 9- to 15-year-old subjects who reported elevated temperature were comparable to those reported by 9- to 17-year-old female subjects.

- Safety related to drug-drug interactions and other interactions

No significant difference in reactogenicity and safety was seen between subjects receiving Gardasil co-administered with hepatitis B vaccine (recombinant) and subjects receiving placebo co-administered with hepatitis B vaccine (recombinant). The use of Gardasil concomitant with vaccines other than hepatitis B vaccine has not been studied. The safety profile for the concomitant

administration of Gardasil and hepatitis B vaccine was not different from that generally seen in the safety summary for other studies and no safety concerns can be raised.

In clinical studies, 57.5% of women (age 16 to 26 years) who received Gardasil used hormonal contraceptives. Use of hormonal contraceptives did not appear to affect the immune responses to Gardasil.

- Post marketing experience

Not applicable

- Discussion on clinical safety

There was no major increase in the reactogenicity following Gardasil vaccination as compared to the placebo (aluminium-containing) administration. Pyrexia, pain, erythema and swelling at the injection site were the most common symptoms observed in both groups. Of note, local pain was less frequent after non-aluminium-containing placebo administration. Moreover, the data from the studies do not indicate that the vaccine gives rise to anaphylactic reactions.

All fatalities were assessed as not related to the study vaccines by the investigators. Most of the fatalities can be explained by the underlying medical condition of the subject. Five fatalities were caused by accidental events.

Regarding the outcome of pregnancy occurring during the vaccination period it could be considered that administration of Gardasil had no impact on fertility. The rate of spontaneous abortions was higher in the group of 16 to 26 years of age compared with the placebo group. The elective abortions performed in recipients of Gardasil appeared not to be induced by any effects of the vaccine. There was no evidence that Gardasil reduced the proportion of pregnancies that resulted in a live birth of a normal infant. The rate of late foetal deaths was not different in the Gardasil group compared with placebo. Congenital anomalies were in expected ranges. Although there were differences in distribution of congenital anomalies between the vaccination groups with EDCn within and not within 30 days from any vaccination, respectively the overall proportions were comparable between the vaccination groups. Furthermore, the recorded anomalies were all different and no prediction for any specific organ system could be identified.

The data on Gardasil administered during pregnancy did not indicate any safety signal. However, these data are insufficient to recommend use of Gardasil during pregnancy. Vaccination should, therefore, be postponed until after completion of pregnancy.

5. Pharmacovigilance

Detailed description of the Pharmacovigilance system

The CHMP considered that the Pharmacovigilance system as described by the applicant fulfils the legislative requirements.

Risk Management Plan

The applicant submitted a risk management plan.

Summary of the risk management plan

Safety issue	Proposed pharmacovigilance activities	Risk Minimisation Activities
Vaccination during pregnancy	Pregnancy Registry, on a voluntary basis, in USA and France. Post-marketing Safety Surveillance Study, an observational short-term database study in USA (outcome of pregnancy exposure as a part of the study).	N/A

	Norwegian Vaccine Registry Study. Norway will establish a national HPV vaccine registry, which can be used in conjunction with medical birth registries.	
Potential for replacement of vaccine HPV types with non-vaccine HPV types	Nordic Cancer Registry Study. Monitoring of possibility of HPV-type replacement will be a part of the Nordic Cancer Registry Study.	N/A
Duration of protection and the need for a booster dose.	Nordic Cancer Registry Study. The long-term effectiveness and immunogenicity will be studied longitudinally. Adolescent Sentinel Cohort Study: Longterm immunogenicity and effectiveness (from 16th birthday)	N/A
Unanticipated Safety Signals	Short-term (60 days following vaccination) safety: Post-marketing Safety Surveillance Study, an observational short-term database study, in USA. Long-term Safety: Nordic Cancer Registry Study. Longterm safety: Adolescent Extension Study	N/A

The CHMP, having considered the data submitted in the application, is of the opinion that no additional risk minimisation activities are required beyond those included in the product information.

6. Overall conclusions, risk/benefit assessment and recommendation

Quality

During the evaluation of Gardasil, no major objections were identified. Minor concerns have been adequately addressed, however several commitments are made by the applicant, and several follow-up measures are defined to provide further information post-approval. In conclusion all quality issues are resolved.

Non-clinical pharmacology and toxicology

Studies in animal models with species-specific papillomaviruses have demonstrated the possibility to vaccinate against infection and development of tumour lesions using virus-like particles formed by recombinant viral capsid proteins. Protection could be transferred with serum, suggesting that the efficacy of Gardasil is mediated by development of humoral immune response.

A limited toxicological program has been performed with Gardasil. There are no toxicological concerns arising from the non-clinical studies with the vaccine. Theoretically, the vaccine could give rise to antibodies, which cross-react to self-structures, resulting in autoimmune events. Such possibility is not readily studied in experimental animal models, but can only be studied in the clinical situation.

A reproductive toxicity study was performed in female rats, addressing all phases of reproduction and development. An immune response to the vaccine was observed, and antibodies were shown to be transferred to the offspring during gestation and possibly also lactation. There were no adverse findings in the study.

Efficacy

The clinical development program for the Gardasil vaccine included adequately sized studies and involved altogether more than 20,000 women.

The goal of the clinical development program for Gardasil was to prevent cancer lesions related to HPV 16 and 18. High-grade cervical lesion CIN 2/3 was the primary efficacy endpoint, used as a surrogate marker for cervical cancer. Gardasil was administered without pre-screening for presence of HPV infection. The primary efficacy analysis was evaluated in the per-protocol efficacy population (PPE), consisting of subjects who received all 3 vaccinations, did not have protocol deviations and were naïve to the relevant HPV type(s) through 1 month postdose 3. In the two pivotal trials (FUTURE I and II) including over 17,500 young women aged 16 to 26 years, Gardasil was shown to prevent 100% of high-grade cervical precancers (CIN 2/3) and non-invasive cervical cancers (CIN 3/adenocarcinoma in situ (AIS)) associated with HPV types 16 and 18 in the PPE population. The vaccine also proved efficacious against low-grade cervical lesions (CIN 1). Several modified-intention-to-treat analyses including study participants who violated the protocol, received at least one dose or received at least two vaccine doses, supported the primary results. Efficacy was demonstrated for each of the 4 vaccine HPV types. In the ITT population (MITT-3), representing the general female population with or without HPV disease at enrolment, vaccine efficacy was substantially lower, 51% against CIN 2 and 44% against CIN 3/AIS.

The vaccine was also evaluated for the prevention of persistent HPV infection, HPV 6/11/16/18-related vulvar and vaginal intraepithelial neoplasia (VIN and VaIN) and genital warts. For genitals warts, vaccine efficacy was shown in all study populations. With regard to HPV 16/18-related VIN 2/3 and VaIN 2/3, there were too few cases in the per-protocol population to establish protective efficacy, but all cases occurred in the placebo group. In the MITT-2 population, representing the primary target population in clinical practice, consistent and statistically significant results were obtained in the combined study dataset for VIN 2/3 and VaIN 2/3 (VE: 100% with lower bound of the 95% CI at >20%). For VIN 2/3, vaccine efficacy was also established in several other relevant study populations, whereas there were too few cases of VaIN 2/3 to obtain significant results. In the MITT-3 population vaccine efficacy against vulvar and vaginal dysplastic lesions was substantially lower. The vaccine was also demonstrated to prevent low-grade lesions, VIN 1 and VaIN 1 (VE: 86%-100%) although these are not considered clinically relevant endpoints. Vaccine efficacy against persistent HPV infection was established for HPV 16 but not for HPV 18 due to too few cases. It can be concluded that with regard to external genital lesions, Gardasil was shown to be highly efficacious against genital warts and VIN 2/3. For VaIN 2/3 there were very limited number of cases, but taken all data into consideration there was sufficient evidence supporting vaccine efficacy also against these lesions.

Since the efficacy of HPV vaccines cannot be evaluated in sexually naïve subjects, immunogenicity was studied in 2800 virginal boys and girls aged 9 to 15 years to provide a basis for bridging the efficacy of Gardasil obtained in young adult women. Anti HPV responses at Month 7 among 9- to 15-year-old girls were non-inferior to anti-HPV responses in 16- to 26-year-old young women for whom efficacy was established in the phase III studies. At Month 18 GMTs remained 2- to 2.5-fold higher in adolescents compared with GMTs in adult women. The kinetics of immune responses up to Month 18 were similar in both groups. Modelling demonstrated a strong positive relationship between Month 7 GMTs and Month 18 GMTs, and for women Month 60 GMTs, suggesting that anti-HPV levels in adolescents will remain higher than those associated with protective efficacy in adults over the long-term. Five-year follow-up of adult women in Protocol 007 showed sustained efficacy with no breakthroughs due to waning immunity. A booster administered at Month 60 to young women resulted in a strong anamnestic response suggesting that the vaccine induces an immune memory. Furthermore, mathematic modelling predicted long-term persistence of detectable anti-HPV levels. The Applicant has agreed to perform a long-term post-marketing follow-up of immunogenicity and safety in an Adolescent Sentinel Cohort. Taking all these data into consideration efficacy bridging from adult women to girls is considered justified.

Bridging immunogenic data to males is less obvious since there is no efficacy bridge to adult males. There are no data on the possible correlation between immunogenicity and efficacy of Gardasil in males. Ongoing efficacy studies in males will address this. Although HPV-related infection and disease in men and women share many similarities there are some differences, such as lower prevalence of specific HPV types and lower antibody response to natural HPV infection in men. However, the data from the clinical trials demonstrate that the HPV vaccine is immunogenic and well tolerated in boys. Anti-HPV responses at Month 7 among 9- to 15-year old boys were higher than those in girls of the same age group and non-inferior to the anti-HPV responses in 16- to 26 -year old females for whom efficacy was established in the phase III studies. Genital warts affect males at the same age-related rates as females, which could justify bridging. Furthermore, men are perceived as the

most important vectors for transmission of HPV infection to women. To break the cycle of sexual transmission and attain herd immunity, males will have to be included in an immunisation program. At this stage it was deemed acceptable to mention in the SPC sections 4.1 and 5.1 that immunogenicity and safety of Gardasil has been shown in boys, but that there are no data on protective efficacy in males.

The efficacy of Gardasil has only been demonstrated against diseases that are caused by HPV types 6, 11, 16 and 18 and not against high-grade dysplastic genital lesions and cancers caused by non-vaccine types. Therefore routine cervical screening remains critically important and should follow local recommendations. It was considered important to include this information in the SPC section 4.4 with a statement that vaccination is not a substitute for routine cervical screening.

Safety

The general safety profile of Gardasil is considered favourable. The methodologies used for safety evaluation are established and appropriate. The safety database is large although for some separate age cohorts it might not sufficiently detect possibly precluded unexpected adverse reactions.

From the safety database all the adverse reactions reported in clinical trials have been included in the Summary of Product Characteristics.

Specific studies of the vaccine in pregnant women were not conducted. However, during the clinical development program, 2,266 women (vaccine: 1,115 vs. placebo: 1,151) reported at least one pregnancy. Overall, the numbers of pregnancies with an adverse outcome were comparable in subjects who received Gardasil and subjects who received placebo. For pregnancies with estimated onset within 30 days of vaccination, 5 cases of congenital anomaly were observed in the group that received Gardasil compared to 0 cases of congenital anomaly in the group that received placebo. Conversely, in pregnancies with onset more than 30 days following vaccination, 10 cases of congenital anomaly were observed in the group that received Gardasil compared with 16 cases of congenital anomaly in the group that received placebo. The types of anomalies observed were consistent with those generally observed in pregnancies in women aged 16 to 26 years.

The data on Gardasil administered during pregnancy did not indicate any safety signal. However, these data are insufficient to recommend use of Gardasil during pregnancy. Vaccination should, therefore, be postponed until after completion of pregnancy.

A total of 995 breastfeeding mothers were given Gardasil or placebo during the vaccination period of the clinical trials. The rates of adverse reactions in the mother and the breastfed infant were comparable between the vaccination and the placebo groups. In addition, vaccine immunogenicity was comparable among breastfeeding mothers and women who did not breastfeed during the vaccine administration. Gardasil can be given to breastfeeding women.

Gardasil has not been studied in children below 9 years of age. Thus the use of vaccine should be avoided in that age group. There is need for more safety data in boys aged 9 to 16 years and also in the older group (16 to 23 years) of men. The safety profile of the adolescent group of boys was, however, comparable with the two female groups.

Having considered the safety concerns in the risk management plan, the CHMP considered that the proposed activities described in section 3.5 adequately addressed these.

- User consultation

The applicant performed readability testing (“user consultation”) and a satisfactory report has been provided.

Risk-benefit assessment

A risk management plan was submitted. The CHMP, having considered the data submitted, was of the opinion that:

Pharmacovigilance activities in addition to the use of routine pharmacovigilance were needed to investigate further some of the safety concerns.
No additional risk minimisation activities were required beyond those included in the product information.

Recommendation

Based on the CHMP review of data on quality, safety and efficacy, the CHMP considered by consensus that the risk-benefit balance of Gardasil in "the prevention of high-grade cervical dysplasia (CIN 2/3), cervical carcinoma, high-grade vulvar dysplastic lesions (VIN 2/3), and external genital warts (condyloma acuminata) causally related to Human Papillomavirus (HPV) types 6, 11, 16 and 18. The indication is based on the demonstration of efficacy of Gardasil in adult females 16 to 26 years of age and on the demonstration of immunogenicity of Gardasil in 9- to 15-year old children and adolescents. Protective efficacy has not been evaluated in males" was favourable and therefore recommended the granting of the marketing authorisation.