European Medicines Agency
Evaluation of Medicines for Human Use

Doc Ref : EMEA/285631/2008

CHMP ASSESSMENT REPORT
FOR
Pandemrix

Common Name:
Pandemic influenza vaccine (H5N1) (split virion, inactivated, adjuvanted)
A/VietNam/1194/2004 NIBRG-14

Procedure No. EMEA/H/C/832

Assessment Report as adopted by the CHMP with
all information of a commercially confidential nature deleted.

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1. BACKGROUND INFORMATION ON THE PROCEDURE

1.1 Submission of the dossier

The applicant GlaxoSmithKline Biologicals S.A. submitted on 2 February 2007 an application for Marketing Authorisation to the European Medicines Agency (EMEA) for Pandemrix, through the centralised procedure falling within the Article 3(1) and point 1 of Annex of Regulation (EC) No 726/2004 .

The legal basis for this application refers to:

A - Centralised / Article 8(3) / New active substance.

Article 8.3 of Directive 2001/83/EC, as amended - complete and independent application

The application submitted is a complete dossier: composed of administrative information, complete quality data, non-clinical and clinical data based on applicants' own tests and studies and/or bibliographic literature substituting/supporting certain test(s) or study(ies)

The applicant applied for the following indication: Prophylaxis of influenza in an officially declared pandemic situation.

Licensing status:
The product was not licensed in any country at the time of submission of the application.

The Rapporteur and Co-Rapporteur appointed by the CHMP and the evaluation teams were:
Rapporteur: **Matthew Thatcher** Co-Rapporteur: **Barbara van Zwieten-Boot**

CHMP Peer reviewer(s): Germany (PEI)

1.2 Steps taken for the assessment of the product

- The application was received by the EMEA on 2 February 2008.
- Accelerated Assessment procedure was agreed-upon by CHMP on 14 December 2006.
- The procedure started on 21 February 2007.
- The Rapporteur's first Assessment Report was circulated to all CHMP members on 11 May 2007. The Co-Rapporteur's first Assessment Report was circulated to all CHMP members on 9 May 2007.
- During the meeting on 22 May 2007, the CHMP agreed on the consolidated List of Questions to be sent to the applicant. The final consolidated List of Questions was sent to the applicant on 22 May 2007.
- The applicant submitted the responses to the CHMP consolidated List of Questions on 31 October 2007.
- The Rapporteurs circulated the Joint Assessment Report on the applicant's responses to the List of Questions to all CHMP members on 28 December 2007.
- During a meeting of BWP Working Party on 14-15 January 2008, experts were convened to address the outstanding quality issues identified in the Joint response assessment report.
- During the CHMP meeting on 24 January 2008, the CHMP agreed on a list of outstanding issues to be addressed in writing by the applicant.
- The applicant submitted the responses to the CHMP list of outstanding issues on 31 January 2008.
- The Rapporteurs circulated the Joint Assessment Report on the applicant's responses to the List of Outstanding issues to all CHMP members on 8 February 2008.

- During a meeting of BWP Working Party on 11-13 February 2008, experts were convened to address the responses to the outstanding quality questions.
- During the meeting on 18-21 February 2008, the CHMP, in the light of the overall data submitted and the scientific discussion within the Committee, issued a positive opinion for granting a Marketing Authorisation under exceptional circumstances to Pandemrix on 21 February. The applicant provided the letter of undertaking on the specific obligations and follow-up measures to be fulfilled post-authorisation on 19 February 2008.

2 SCIENTIFIC DISCUSSION

2.1. Introduction

An influenza pandemic is a global outbreak of influenza disease that occurs when a type A influenza strain to which most or all humans are immunologically naïve emerges to cause clinically apparent illness, and then spreads easily from person to person worldwide. Pandemics are different from seasonal outbreaks of influenza, as the latter are caused by subtypes of influenza viruses that are already circulating in the world whereas pandemics are caused by new subtypes or by subtypes that have not circulated among people for a long time.

Specific guidance has been developed for the fast track assessment procedure for pandemic influenza vaccines[1], which can only be used once WHO/EU have officially declared the pandemic (WHO Phase 6 onwards). The procedure involves the submission and evaluation of a core pandemic dossier during the inter-pandemic period, followed by a fast track assessment of the data for replacing the mock-up vaccine strain with the recommended pandemic strain as a variation to the MAA.

GlaxoSmithKline Biologicals has submitted a Marketing Authorisation Application (core pandemic dossier) for Pandemrix in line with the above mentioned guidelines. Pandemrix is a split virion inactivated influenza vaccine, containing the mock-up strain H5N1 (NIBRG-14) derived by reverse genetics from the avian influenza virus A/Viet Nam/1194/2004. The final formulation contains 3.75 µg haemagglutinin (HA) per 0.5 ml dose adjuvanted by AS03. Pandemrix is indicated for prophylaxis of influenza in an officially declared pandemic situation. Pandemic influenza vaccine should be used in accordance with official guidance.

From an epidemiological point of view it is very unlikely that influenza strain A/Vietnam /1194/2004 would be the next pandemic strain, since the H5N1 virus continues to undergo antigenic drift. It is also possible that the next pandemic will not be caused by a H5N1 virus but will be due to another subtype of influenza virus (e.g. with haemagglutinin of type H2, H7 or H9). In line with the core dossier concept, a variation would therefore have to be submitted to introduce the WHO/EU recommended strain, prepared from the influenza virus causing the pandemic, prior to use of Pandemrix in a pandemic. Pandemrix is not indicated for prophylactic use during the prepandemic period.

2.2. Quality aspects

Introduction

Pandemrix is a split virion inactivated influenza vaccine. The final formulation contains 3.75 µg haemagglutinin (HA) of A/VietNam/1194/2004 NIBRG-14 (H5N1) per 0.5 ml dose adjuvanted by AS03.

The reference virus described in the current MAA is A/Vietnam/1194/2004 (H5N1) NIBRG-14 which was developed using reverse genetics. The reassortment strain combines the H5 and N1 segments to the PR8 strain backbone. In addition the H5 was engineered to eliminate the polybasic stretch of amino-acids at the HA cleavage site that is responsible for high virulence of the original strains.The virus is propagated in fertilised hens' eggs.

[1] *Guideline on Submission of Marketing Authorisation Applications for Pandemic Influenza Vaccines through the Centralised Procedure* (CPMP/VEG/4986/03).
Guideline on Dossier Structure and Content for Pandemic Influenza Vaccine Marketing Authorisations Application (CPMP/VEG/4717/03).

The vaccine consists of a suspension vial with the H5N1 antigen and an oil–in-water emulsion vial with the AS03 adjuvant, which are mixed extemporaneously. Thiomersal, 10 µg/ml (5 µg per dose), is added because of the multi-dose presentation.

Each 0.5 ml dose of vaccine has the following composition:

Active Ingredient:	
Purified antigen fractions of inactivated split virion A/Vietnam/1194/2004 NIBRG-14 (H5N1)	3.75 µg HA
Adjuvant:	
Squalene	10.68 mg
Alpha-tocopherol	11.86 mg
Polysorbate 80	4.86 mg
Other Ingredients:	
Octoxynol 10	
Sodium chloride	
Disodium phosphate	
Potassium dihydrogen phosphate	
Potassium chloride	
Magnesium chloride	
Thiomersal	
Water for injections	

Drug Substance

- *Manufacture*

A master and working seed were prepared from the NIBRG-14 reference virus, received from the WHO Reference centre (NIBSC, UK). The manufacturing process for the monovalent bulks is similar to the manufacturing process for the monovalent bulks of the licensed product Fluarix (seasonal influenza virus) and can be divided into four main parts:

- Propagation of the working seed in fertilised hen's eggs, harvesting and pooling of infected allantoic fluids
- Purification of the whole virus bulk
- Splitting of the monovalent with sodium deoxycholate
- Inactivation of the monovalent split virus using sodium deoxycholate and formaldehyde, followed by ultrafiltration and sterile filtration

The process parameters are generally derived from the Fluarix process. Where needed these have been specifically determined for this H5N1 strain.

- *Control of Materials*

The following starting materials used in the production of the monovalent bulk are of biological origin: influenza seed virus, eggs and sodium deoxycholate (derived from bovine bile). The H5N1 working seed is derived from the A/Viet Nam/1194/2004 (H5N1) NIBRG-14 vaccine virus strain.

The testing of the virus and the eggs is the same as for Fluarix. Sufficient detailed information has been provided on the control and source of these starting materials. The genetic stability of the A/VietNam/1194/2004 (H5N1) NIBRG-14 obtained through Reverse Genetics has been satisfactorily addressed.

- *Process validation*

Critical steps of the drug substance production process have been identified and are sufficiently controlled. Process consistency has been demonstrated on three commercial scale batches.

Inactivation studies performed on different H5N1 strains have demonstrated that the proposed inactivation method (Sodium deoxycholate and formaldehyde) results in complete inactivation. Based on experience with Fluarix, the ability of the manufacturing process to inactivate avian leucosis and mycoplasma has been demonstrated.

- *Characterisation and specifications*

The structure of the inactivated split monovalent bulks was studied by transmission electron microscopy and confirmed the predominance of disrupted particles after splitting.
Controls include HA content, neuraminidase identity, sterility tests, tests for residual infectious viruses, residual sodium deoxycholate and test for disrupted virus particles (not routine) and are in line with Ph.Eur. monograph <0158>.
All analytical methods have been appropriately validated.
The monovalent bulks are filled and stored in 10 l glass Type I containers with polypropylene closures.

- *Stability*

Eighteen months of stability data at 2-8°C has been generated for three relevant batches.

Drug Product

The drug product is described in three parts: The drug product containing H5N1 antigen, the AS03 adjuvant and the mixed AS03 adjuvanted H5N1 influenza vaccine which is the preparation to be administered within 24 hours.

Drug Product (H5N1 vial)

- *Pharmaceutical Development*

Developmental changes implemented since the first clinical studies have been stated and clinical studies have provided reassurance of product remaining comparable.

- *Manufacture of the Product*

Manufacture for the antigen component consists of formulation of the final bulk with the excipients at SSW (Germany), transport from SSW (Germany) to GSK Biols (Belgium: Rixensart or Wavre sites) for filling into final containers and then finally labelling and packaging at GSK Biols (Wavre).

Antigen bulk received is sterile. Bioburden is adequately controlled throughout the manufacturing process.
The sterile filtered H5N1 monovalent bulk (SSW, Dresden) and non-sterile excipients, are sterilised in-line, just prior to entering the mixing tank (SSW, Dresden). The resultant bulk is then transported in HDPE containers, initially to GSK Wavre for cold storage, and then for filling at GSK Rixensart or GSK Wavre. Formulation of final formulated bulk and subsequent filling into HDPE containers, and later, pooling of transported HDPE containers into the filling tank and subsequent filling is conducted under aseptic conditions. Stability data have been submitted to support the maximum storage period of the formulated bulk filling into bulk HDPE containers (30 days) and in the filling tanks (14days).

- *Product Specification*

Compliance with the product specifications has been shown on three batches representative of the final formulation and commercial scale manufacture
Specifications for excipients and analytical procedures are in line with the Ph.Eur. Controls of final bulks (sterility, HA, total protein, residual ovalbumin, thiomersal, residual formaldehyde and residual sucrose) and final containers (sterility, bacterial endotoxins, pH, volume, thiomersal and HA) of the antigen vial are acceptable (Ph.Eur. or in line with Fluarix). Methods are either in line with Ph.Eur. or are validated. Specificity in presence of thiomersal has been demonstrated.

HA content, sterility, thiomersal content, endotoxin content, pH and description, formaldehyde, ovalbumin and protein content are measured as part of the stability studies. Test methods and specifications are identical to those at release.
The Company proposes an 18 months expiry date - The proposed shelf life is accepted on basis of the commitment to provide updated stability results.

An overage of 10 % HA will be applied at formulation of the commercials lots and supporting data and satisfactory justification have been provided.

Drug Product (AS03 adjuvant vial)

- Pharmaceutical Development

Developmental changes implemented since the first clinical studies have been stated and non-clinical and clinical studies have provided reassurance of product remaining comparable.

- *Manufacture of the AS03 adjuvant vial*

Formulation of the AS03 adjuvant consists of the preparation of the bulk (formation of O/W emulsion using high shear and pressure homogenisation) followed by filling into glass vials. Process parameters are identified. No routine in-process tests are conducted. Bioburden is adequately controlled throughout the manufacturing process.

- *Specifications of the AS03 adjuvant*

With the exception of squalene, all excipients are described and controlled in line with the Ph.Eur. Adequate quality control of squalene is performed by the supplier and by GSK (according to an internal GSK monograph which is in line with the Ph.Eur. monograph for squalane).

Emulsion bulk and AS03 final containers are tested at release for Description, Identity and Content of adjuvant components (polysorbate 80, α-tocopherol and squalene), pH, Endotoxin content, Sterility, Particle size, Polydispersity index and Volume (final containers only).

Tests for sterility and bacterial endotoxins are performed in line with the Ph.Eur. and tests for polysorbate 80, α-tocopherol and squalene are validated. The method used for particle size analysis and associated system suitability measurements is acceptable.

- *Stability of the AS03 adjuvant*

Data provided from the stability studies for the bulk emulsion support the proposed shelf life of 2 years. For final AS03 container lots a shelf-life of 18 months has been approved.

Drug Product (mixed H5N1 and AS03 vial)

At the time of extemporaneous dispensing, adjuvant is added to antigen vial. Data from 'withdrawable' volume studies conducted to support the required overfill for both antigen and adjuvant vials is provided. Information on long term storage (1 week) of mixed AS03 adjuvanted H5N1 is also provided.

Preservative efficacy of thiomersal concentration after mixing the content of the antigen container with AS03 adjuvant has been shown in line with Ph.Eur.

Sufficient compatibility/stability data has been provided in the dossier.

SDS PAGE and Western blot analysis performed show that profiles of the adjuvanted formulation are comparable to the non-adjuvanted formulation and remain unchanged after a period of 24 hours at 25°C. Interaction between antigen and adjuvant has been shown to be limited by various biophysical methods Uniformity of dose has been demonstrate for the 10-dose product.

In addition, non-clinical studies have shown similar immune responses were observed when antigen and adjuvant were administered separately (one hour apart at the same site, or simultaneously in 2 separate syringes in the same area) or after administration of pre-mixed antigen and adjuvant.

Therefore, it is accepted that there is no need to control antigen/adjuvant interaction for this product. Sufficient evidence has been provided that there is little/no effect of the reconstitution conditions (mixing time and conditions) on the essential characteristics of the antigen/adjuvant combination. The proposed in-use shelf life of 24 hours is considered justified based on stability/characterisation data provided.

Discussion on chemical, pharmaceutical and biological aspects

Pandemrix is a split virion inactivated influenza vaccine. The final formulation is an emulsion for injection and contains 3.8 µg haemagglutinin (HA) of A/VietNam/1194/2004 NIBRG-14 (H5N1) per 0.5 ml dose adjuvanted by AS03. AS03 consists of the oil–in-water emulsion containing DL-α-tocopherol, squalene and tween 80. Thiomersal, 10 µg/ml (5 µg per dose), is added because of the multi-dose presentation.

Pandemrix is presented in multidose vials. After reconstitution prior to use (extemporaneous mixing of the antigen vial (suspension) with the adjuvant vial (emulsion)), the vial contains 10 vaccine doses.

Drug Substance

The reference virus is A/Vietnam/1194/2004 (H5N1) NIBRG-14 which was developed using reverse genetics. The virus is propagated in fertilised hens' eggs. The manufacture of the drug substance, the monovalent antigen bulk, is almost identical to that of the licensed split antigen seasonal influenza vaccine (Fluarix) and is adequately described. The process parameters are generally derived from the Fluarix process. Where needed these have been specifically determined for this H5N1 strain.

The testing of the virus and the eggs is the same as for Fluarix. Critical steps of the drug substance production process have been identified and are generally sufficiently controlled. Process consistency has been demonstrated on three commercial scale batches.

Release specifications are in line with the currently approved specification for Fluarix and include HA content, neuraminidase identity, sterility tests, tests for residual infectious viruses, residual sodium deoxycholate and test for disrupted virus particles (not routine) and are in line with Ph.Eur. monograph <0158>.

Eighteen months of stability data at 2-8°C has been generated for three relevant batches of monovalent bulk.

Drug Product

Manufacture for the H5N1 antigen component consists of formulation of the final bulk with the excipients at SSW (Germany), transport from SSW (Germany) to GSK Biologicals (Belgium: Rixensart or Wavre sites) for filling into final containers and then finally labelling and packaging at GSK Biologicals (Wavre). The H5N1 antigen vial is appropriately controlled. 18 month stability data at 2-8 °C have been generated.

The manufacture and control of the adjuvant vial is adequately controlled. Stability data with the AS03 final containers is available to 18 months at 2-8 °C.

At the time of extemporaneous dispensing, adjuvant is added to antigen vial. It is accepted that there is no need to control antigen/adjuvant interaction for this product: sufficient evidence has been provided that there is little/no effect of the mixing time and conditions on the essential characteristics of the

antigen/adjuvant combination. The proposed in-use shelf life of 24 hours is considered justified based on stability/characterisation data provided.

2.3. Non-clinical aspects

Introduction

Preclinical development of Pandemrix was generally in agreement with current guidelines. The antigen is produced in hen's eggs using the same process as that is applied to the Applicant's own Fluarix brand of seasonal influenza vaccine. This was approved in 1992 and over 200 million doses have been manufactured. As advised in regulatory guidance documents, preclinical testing with Fluarix can be used to support this application, based on the similarity of manufacture.

The safety studies included in the dossier were all compliant with GLP.

Pharmacology

- Primary pharmacodynamics

Primary pharmacodynamic properties were investigated in mice, pigs and ferrets.

The studies demonstrate the ability of the AS03 adjuvant to augment immunological responses to the vaccine in mice and pigs. The data from the pig study show a statistically significant effect of dose of adjuvant in one of the three tested influenza strains, the B/Shangdong strain. A non-significant dose-response effect might be suggested with the A/Panama strain.
Efficacy of the vaccine was tested in a challenge test in ferrets. Survival data are compelling in that, where a sufficient dose of antigen was used, protection from a lethal challenge was obtained.
Consistent changes in serological and pathological parameters were detected and it can be concluded that the vaccine provided protection from virus-associated pathological changes.

- Secondary pharmacodynamics

Secondary pharmacodynamic studies were not performed. This approach is in accordance with the relevant guidelines, note for guidance on preclinical pharmacological and toxicological testing of vaccines (CPMP/SWP/465/95) and the guideline on dossier structure and content for pandemic influenza vaccine marketing authorisation application, CPMP/VEG/4717/03.

- Safety pharmacology programme

No safety pharmacology studies were performed with Pandemrix vaccine. A study was reported in the anaesthetised rat, using trivalent influenza vaccine, adjuvanted with AS03, or saline. Single intramuscular injection of 0.1 ml of this vaccine administered to anaesthetised male Wistar rats (n = 4) did not produce any effects on cardiovascular or respiratory parameters in the 2 hour period following dosing. The vaccine used in this study contains 15 µg HA per strain and AS03. The dose represents 63 fold higher than the human exposure, on a µg/kg bodyweight comparison.

- Pharmacodynamic drug interactions

No studies were performed.

Pharmacokinetics

Experimental studies to demonstrate absorption, distribution, metabolism, and excretion of the active ingredients in Pandemrix have not been performed. This is in line with the relevant guidelines CPMP/SWP/465/95 and CPMP/VEG/4717/03.

Toxicology

- Single dose toxicity

The Applicant refers to the local tolerance study for consideration of single-dose toxicity.

- Repeat dose toxicity (with toxicokinetics)

Two studies in the rabbit using intramuscular injection have been performed. One used four doses of the adjuvanted vaccine and the second used two doses of a trivalent seasonal influenza vaccine, with the AS03 adjuvant.

Pandemic influenza vaccine: toxicity study in the rabbit after 4 injections
10 male and 10 female New Zealand White rabbits were injected with test item by the intramuscular route on Days 1, 15, 29 and 43 and were killed for pathological examination on either Day 46 or Day 71. Periodical assessments included mortality, clinical observations, injection site reactions, body weight, food consumption, ophthalmological examination, body temperature, haematology, clinical chemistry, organ weights and macro- and microscopic examination of tissues, including evaluation of spermatogenesis.
Very slight erythema and/or oedema occurred commonly but abated within 48 hours. On subsequent injections this was no more marked than the control group. There were no other clinical signs noted.
There was evidence of an inflammatory response in haematology, clinical chemistry and pathological parameters. Increases in fibrinogen and white blood cell counts were noted in temporal association with the erythema and oedema noted on observing the rabbits. Relative to body weight, the spleen weight was increased in all groups compared to the control (7 - 41%). This difference was much less marked from rabbits killed on Day 71, 28 days after the last injection, indicating reversibility.
Frequency and severity of fasciitis was higher in rabbits from the vaccine group. This toxicity was attributed to the adjuvant.

Seasonal influenza vaccine: toxicity study in the rabbit after 2 injections
10 male and 10 female New Zealand White rabbits were injected with 0.5 ml of test item by the intramuscular route on Days 1 and 24 and were killed for pathological examination on either Day 27 or Day 52.
There were no deaths and no clinical signs detected in this study, except one instance of very mild erythema shortly after injection of Fluarix. Minor changes indicative of an inflammatory response were noted in clinical chemistry and haematology in rabbits dosed with AS03 or with the trivalent influenza vaccine. These changes reduced over time, indicating recovery.

- Genotoxicity

Genotoxicity of the adjuvant alone was assessed in two *in vitro* tests (reverse mutation test in bacteria; gene mutation in mouse cells) and one *in vivo* test (micronucleus test in the rat after intravenous administration). The vaccine was not tested. No indication of genotoxicity was evident.

- Carcinogenicity

No carcinogenicity studies were conducted which is in line with the Note for Guidance on Preclinical pharmacological and toxicological testing of vaccines (CPMP/SWP/465/95)

- Reproduction Toxicity

Preliminary immunogenicity studies were performed in the rat in which the immunogenic responses of the dams, foetuses and pups were confirmed, demonstrating that the rat is a suitable species for assessing toxicity of the vaccine.

In this study, six groups of 48 female rats were given a single intramuscular dose on Day -30 and paired with males and 44 rats with a positive indication of mating were treated on Days 6, 8, 11 and 15 after mating. 22 were killed at Day 20 and 22 were allowed to deliver and rear their young to Day 25 of age. Measures of reproductive toxicity and maternal health were assessed. The six groups were dosed with:

1 saline, 200 µl
2 AS03 adjuvant, 200 µl
3 saline / split H5N1/AS03, 200 µl
4 split H5N1 / AS03, 200 µl
5 saline /whole H5N1/A1, 100 µl
6 whole H5N1/A1, 100 µl

There was one unexpected death in a maternal rat: however, this was judged unrelated to the vaccine. Treatment of maternal rats did not adversely affect their clinical condition, bodyweight or food consumption throughout the study. Mating performance, fertility of maternal rats, and length of gestation or ability to give birth to a live litter were unaffected. Embryo-foetal survival, growth and development were not affected by vaccination. In neonates, the reflex development was unimpaired, but among offspring from dams treated with AS03 13 offspring from 7 litters did not show the air righting reflect before day 21 of age and this effect may be related to treatment. However, AS03 did not affect the attainment of the surface righting reflex or the ability of the offspring to show startle response reflexes or the pupil reflex. No abnormalities were evident on macro pathological examination of the offspring. This is considered a suitable study to assess the reproductive toxicity of the vaccine.

- Local tolerance

The test items in this study were the adjuvant, AS03, a candidate trivalent influenza vaccine adjuvanted with AS03, and Fluarix, which is the Applicant's approved inactivated influenza vaccine and which is not adjuvanted. Thus, the vaccine intended to be marketed was not used in this test. This study also served as the assessment of single dose toxicity.

Single intramuscular injection of the test items (n = 3) or of saline (n = 2) into the thigh muscle of New Zealand White rabbits was followed by clinical observation for dermal reactions (erythema and oedema were each graded separately on 5 point scales) and clinical signs until Day 4 when rabbits were killed and subject to microscopic examination of the injection sites. Dose volumes of 0.5 ml were used, injected into the upper and the lower thigh. 7.5 and 15 µg of trivalent influenza was used (the clinical dose of the monovalent vaccine is 3.75 µg).

There were no deaths or clinical signs in the study. On visual examination, dermal reactions were unremarkable, with very slight oedema noted in one rabbit 3 hours after injection of the adjuvant AS03, and also in another rabbit injected with the trivalent vaccine. Very slight erythema was noted commonly in all groups. On microscopic examination, the adjuvant was associated with multifocal or diffuse infiltrating low grade sub-acute fasciitis with macrophage infiltration. Fasciitis of the skin was also evident. These effects were more evident than in tissues from rabbits injected with either saline or Fluarix. Such changes were also noted in tissue from rabbits injected with the trivalent vaccine, to a similar degree.

- Other toxicity studies

Immunogenicity described under pharmacology. Other studies were not reported.

Ecotoxicity/environmental risk assessment

No environmental risk assessment is included in this application. According to the guideline EMEA/CHMP/SWP/4447/00 "*Environmental Risk Assessment of Medicinal Products for Human Use"* vaccines due to the nature of their constituents are exempted from the requirement to provide an

environmental risk assessment in the application for a marketing authorisation for a medicinal product for human use.

2.4. Clinical aspects

Introduction

GCP

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

Pharmacokinetics

Pharmacokinetic studies were not performed in accordance with the note for guidance on clinical evaluation of new vaccines (CPMP/EWP/463/97) and the Guideline on dossier structure and content for pandemic influenza vaccine marketing authorisation application, CPMP/VEG/4717/03.

Pharmacodynamics

In relation to vaccines, pharmacodynamic studies are essentially compromised of the immunogenicity studies that characterise the immune response to vaccines. The detailed characterisation of the immunological response to the mock-up vaccines is discussed below.

Clinical efficacy

Clinical trials on protective efficacy for the mock-up vaccine cannot be performed. Therefore the assessment of the potential protective efficacy of Pandemrix has been based on a detailed characterisation of the immunological response to the vaccine.

The available data at the time of the initial filing pertained solely to first series production lots and to administration of 1 ml volumes with a range of HA doses from 3.8 to 30 µg (007) or with 15 µg HA (008). Immunogenicity data were available to D42 i.e. 3 weeks after the second dose.

Study	Primary Objective(s) Countries	Population/ age of subjects	Study vaccines	N safety	N immuno
H5N1-007	Immunogenicity Reactogenicity/safety Belgium only	Unprimed population 18-60 years	Monovalent split vaccine (H5N1). 30 µg, 15 µg, 7.5 µg or 3.8 µg HA * with or without AS03 2-dose schedule 21 days apart	400	394
H5N1-008	Reactogenicity/safety Germany, Estonia, France, Netherlands, Russia, Spain, Sweden	Unprimed population > 18 years	Monovalent split vaccine (H5N1) 15 µg HA with AS03 or Fluarix (first dose), placebo (second dose) 2-dose schedule 21 days apart	3802 1269	455 154

N safety = Total vaccinated cohort;
N immuno = ATP cohort for immunogenicity
*: Vaccine doses were expressed using only one digit throughout the clinical documentation, i.e. 3.75 µg Haemagglutinin [HA] was rounded up to 3.8 µg HA in the dossier.

During the procedure the applicant provided additional data as follows:

I. **H5N1-007 -** Persistence of haemagglutination inhibition (HI) and neutralising antibodies (NA) and cell-mediated immunity data (CMI) at Day 180 after initial vaccination. Also serological and CMI studies of cross-reactivity against several heterologous strains at D42.
II. **H5N1-008 –** NA data in subjects aged < 60 years at D42.
III. **H5N1-008; ext 011**) -Persistence of HI at Day 180 for all subjects and NA at D180 in subjects aged > 60 years.

IV. **H5N1-002**

D42 HI and NA data for 3.8 µg/AS03 in healthy adults aged 18-60 years administered vaccine from Third series (final production process) lots in 0.5 ml dose volumes as below:

Study	Primary Objective(s) Countries	Population/ age of subjects	Study vaccines	N safety	N immuno
H5N1-002	Immunogenicity - lot consistency Reactogenicity/safety Taiwan, Thailand, Singapore and HongKong.	Unprimed population 18-60 years	Monovalent split vaccine (H5N1). 3.8 µg HA with or without AS03 2-dose schedule 21 days apart	954 with AS03 245 without AS03	933 with AS03 236 without AS03

Assays

Haemagglutination inhibition (HI) assay

Anti-haemagglutinin antibody titres were measured using the method described by the World Health Organization Collaborating Centre for Influenza, Centres for Disease Control, Atlanta, USA (1991). All assays were performed in duplicate. The following parameters were assessed:

- Seropositivity rate defined as the percentage of vaccinees with a minimum titre of 1:10.
- Seroconversion factor defined as the ratio of the post-vaccination GMT divided by the pre-vaccination GMT.
- Seroconversion rate defined as the proportion which were either seronegative prior to vaccination and had a post-vaccination titre ≥ 1:40 or who were seropositive prior to vaccination and had at least a 4-fold increase in titre post-vaccination.
- Seroprotection rate defined as the proportion in each group with post-vaccination titres ≥ 1:40.

Virus Neutralisation (NA) Assay

Virus neutralisation was determined in a micro-neutralisation assay. Each serum was tested in triplicate. A standardised amount of virus was mixed with serial dilutions of serum and incubated to allow binding of the antibodies to the virus. Results of neutralising serum antibodies were expressed as follows:

- Seropositivity rate defined as the percentage of vaccinees with a minimum titre of 28 (1/dil).
- Seroconversion rate (with 95% CI) defined as the percentage of vaccinees with a minimum 4-fold increase in titre from pre- to post-vaccination
- GMTs of serum neutralising antibodies pre- and post-vaccination (with 95%CI)

Influenza-specific cellular responses: intracellular cytokine staining assay

The assay involved stimulation of peripheral blood antigen-specific CD4 and CD8 T cells *in vitro* to produce cytokines on incubation with corresponding antigen. Antigen-specific CD4 and CD8 T cells were enumerated by flow cytometry following conventional immunofluorescence labelling of cellular phenotype (using anti CD8 APC Cy7 / anti CD4 PerCP) as well as intracellular cytokine production.

Results were expressed as a frequency of cytokine(s)-positive CD4 or CD8 T cells within the CD4 or CD8 T cell population.

Dose response studies

The dose-finding study **007** employed first series vaccine and compared four dose groups, each with and without adjuvant. Although study **008** was not a dose-finding study the data on immune responses to 15 µg doses of the H5 haemagglutinin (HA) could be considered to add to the rationale for the final choice of dose.

Main studies

Studies H5N1- 002, H5N1-007 and H5N1-008 (extension of study 011) are described together.

METHODS

Study Participants

Studies **H5N1-002 and -007** enrolled healthy males and females aged 18 to 60 years while study **008** also enrolled subjects aged > 60 years. Study groups were as in the tables above.

Treatments

Study groups were as in the tables above.

Objectives

In **study 007** immunogenicity (HI) and safety were co-primary objectives.
In **study 008** the primary objective was to evaluate the safety of a 15 µg/AS03 candidate vaccine but immunogenicity was evaluated in a subset.
In **study 002** the primary objective was to demonstrate the consistency of the HI immune response elicited by four lot groups derived from mixing of 2 lots of HA and 2 lots of AS03.

Outcomes/endpoints

In **study H5N1-007** the primary endpoints were related to the HI titres at days 0, 21, 42. The neutralising antibody (NA) titres were secondary.
In **study H5N1-008** the primary endpoints regarding safety were percentage, intensity and relationship to vaccination of solicited and unsolicited local and general signs and symptoms, occurrence of serious adverse events up to Day 51 and occurrence of new onset chronic diseases up to Day 51 in each group.
In **study H5N1-002** lot consistency based on HI GMTs was the primary endpoint.

Sample size and Randomisation

In **H5N1-007** the randomisation algorithm used a minimisation procedure accounting for centre and age and subjects were stratified according to age (18-30 years old or 31 to 60 years old). The target sample size was 400 (i.e. 50 for each of the eight groups) to provide 360 evaluable subjects.
In **H5N1-008** the randomisation algorithm used a minimisation procedure accounting for centre and age and employed three strata (18-30, 31-60 and > 60 years). There was a 3:1 allocation ratio in favour of the candidate vaccine. The sample size was calculated at 5052 subjects to provide 4800 evaluable taking into account a 5% dropout rate. This number was to include 4526 subjects aged between 18-60 years and 526 aged 61 years or above.
In **H5N1-002**
The target sample size was 1090 enrolled subjects (four groups of 218 subjects, two control groups of 109 subjects) in order to reach 980 evaluable subjects.

Blinding (masking)

Studies were observer-blinded due to differences in the appearances of the vaccines. In order to maintain the blinding study personnel who vaccinated the subjects were not involved in the evaluation of endpoints and access to the vaccines was restricted to the person(s) in charge of accountability, preparation and administration.

Statistical methods

In **study H5N1-007** the analysis of variance (ANOVA) model was used to test "haemagglutinin-dose" (3.8, 7.5, 15 or 30 µg HA) effect and "adjuvantation" effect (with or without AS03).
In **study H5N1-008** the main analysis was performed when all data on the humoral immune response and safety up to D42 were available.
In **study H5N1-002** it was considered that lot consistency was demonstrated if, for all pairs of lots, the two-sided 95% CIs for the ratio of anti-HA GMT at D42 were within [0.5, 2.0].

Study populations

The following populations were defined:
Total Vaccinated cohort: all vaccinated subjects for whom data were available.
According-To-Protocol (ATP) for safety: all vaccinated with sufficient safety data for analysis.
ATP for immunogenicity: all evaluable subjects with immunogenicity data available.

RESULTS

Recruitment

H5N1-007 was initiated at a single site in Belgium (Ghent) in March 2006.
H5N1-008 was initiated at 41 sites in seven countries (6 EU MS plus Russia) in May 2006.
H5N1-002 was initiated on 24 March 2007 in four SE Asian countries.

Numbers analysed / Baseline data / Conduct of the study

In **H5N1-002** 1206 subjects were enrolled into the study and 1190 completed primary immunisation.
In **H5N1-007** 400 subjects were enrolled, with 49-51 randomised to each dose/adjuvant group. Of these 400, 399 were evaluable for safety and 394 met the criteria for the ATP immunogenicity analysis.
H5N1-008 enrolled 5075 subjects of which 5071 subjects were vaccinated and 4904 completed to D51. Immunogenicity was assessed up to D180 in a subset of the total enrolled.

Results

Study H5N1-007

HI titres against the vaccine strain

Pre-vaccination, nine subjects were seropositive for antibody to A/Vietnam/1194/2004 but only three were seroprotected (i.e. HI titre ≥ 1:40). The 70% threshold for seroprotection rates (SPR) was not reached in any group after one dose. After two doses the 70% threshold was exceeded in all four adjuvanted formulation groups (range 84 – 96%) but in none of the non-adjuvanted vaccine groups.

Seroprotection rates against the A/Vietnam/1194/2004 (H5N1) strain (ATP cohort for immunogenicity)

				≥ 40 1/DIL			
				n	%	95%CI	
Antibody	Group	Timing	N			LL	UL
A/Vietnam	H5N1/30	PRE	49	0	0.0	0.0	7.3
		PI(D21)	49	14	28.6	16.6	43.3
		PII(D42)	49	21	42.9	28.8	57.8
	H5N1/15	PRE	49	1	2.0	0.1	10.9
		PI(D21)	49	10	20.4	10.2	34.3
		PII(D42)	49	17	34.7	21.7	49.6
	H5N1/7.5	PRE	49	0	0.0	0.0	7.3

		PI(D21)	49	4	8.2	2.3	19.6
		PII(D42)	49	8	16.3	7.3	29.7
	H5N1/3.8	PRE	50	0	0.0	0.0	7.1
		PI(D21)	50	0	0.0	0.0	7.1
		PII(D42)	50	2	4.0	0.5	13.7
	H5N1/30/AS03	PRE	48	0	0.0	0.0	7.4
		PI(D21)	48	28	58.3	43.2	72.4
		PII(D42)	48	41	85.4	72.2	93.9
	H5N1/15/AS03	PRE	49	0	0.0	0.0	7.3
		PI(D21)	49	24	49.0	34.4	63.7
		PII(D42)	49	47	95.9	86.0	99.5
	H5N1/7.5/AS03	PRE	50	1	2.0	0.1	10.7
		PI(D21)	50	25	50.0	35.5	64.5
		PII(D42)	50	45	90.0	78.2	96.7
	H5N1/3.8/AS03	PRE	50	1	2.0	0.1	10.7
		PI(D21)	50	13	26.0	14.6	40.3
		PII(D42)	50	42	84.0	70.9	92.8

After the second vaccination with all adjuvanted formulations and with non-adjuvanted formulations containing 30 or 15 µg HA the seroconversion factors (SCFs) exceeded 2.5 but ranged from 27.9 to 60.5 for adjuvanted vaccines compared to a maximum value of only 3.9 among non-adjuvanted formulations.

After both the first and second vaccinations the percentage of seroconverted subjects was significantly superior in groups vaccinated with adjuvanted formulations. There was no significant difference in the post-vaccination anti-HA antibody titre between the four groups that received adjuvanted formulations. For each dose of HA, a significant difference was detected between the adjuvanted and non-adjuvanted groups.

Seroprotection rates to A/Vietnam/1194/2004 for the adjuvanted vaccine groups at D180 ranged from 54 - 64 %. In the 3.8 µg HA +AS03 group the seroprotection rates were 84% at D42 and 54% at D180.

Table 4 **Seroprotection rates (SPR) for anti-HA antibody titer against A/Vietnam/1194/2004 and A/Indonesia/5/2005 strains at Day 180 (ATP cohort for persistence)**

			SPR			
					95% CI	
Antibodies against	Group	N	n	%	LL	UL
A/Vietnam	H5N1/30	48	18	37.5	24.0	52.6
	H5N1/15	48	12	25.0	13.6	39.6
	H5N1/7.5	49	7	14.3	5.9	27.2
	H5N1/3.8	50	2	4.0	0.5	13.7
	H5N1/30/AS03	48	30	62.5	47.4	76.0
	H5N1/15/AS03	49	30	61.2	46.2	74.8
	H5N1/7.5/AS03	50	32	64.0	49.2	77.1
	H5N1/3.8/AS03	50	27	54.0	39.3	68.2
A/Indonesia	H5N1/30	48	0	0.0	0.0	7.4
	H5N1/15	48	0	0.0	0.0	7.4
	H5N1/7.5	49	0	0.0	0.0	7.3
	H5N1/3.8	50	0	0.0	0.0	7.1
	H5N1/30/AS03	48	2	4.2	0.5	14.3
	H5N1/15/AS03	49	2	4.1	0.5	14.0
	H5N1/7.5/AS03	50	3	6.0	1.3	16.5
	H5N1/3.8/AS03	50	0	0.0	0.0	7.1

Seroconversion factors against A/Vietnam/1194/2004 were reduced compared to values obtained at Day 42 in both adjuvanted and non-adjuvanted vaccine groups. In the 3.8 µg/AS03 vaccine group the SCFs were 27.9 at D42 but 4.4 at D180.

NA titres against the vaccine strain

Pre-vaccination, between one fifth and one third per group already had NA titres of at least 1:28 while 10 to 25% per group had titres ≥1:40 and 2 to 10% had titres ≥1:80. GMTs increased significantly after the first and second vaccinations with adjuvanted formulations. At D42 most subjects in the adjuvanted groups had titres above the two cut-offs (i.e. 97.9% - 100% at 1:40 and 91.5% - 100% at 1:80). Similarly, seroconversion rates were higher in groups vaccinated with adjuvanted formulations. An HA dose effect was detected in non-adjuvanted groups only.

NA titres against the A/Vietnam/1194/2004 (H5N1) strain (ATP cohort for immunogenicity)

				≥ 28 1/DIL				GMT				
						95% CI			95% CI			
Strain	Group	Timing	N	n	%	LL	UL	value	LL	UL	Min	Max
A/Vietnam/1194/2004	H5N1/30/AS03	PRE	48	9	18.8	8.9	32.6	17.3	15.1	20.0	<28.0	90.0
		PI(D21)	47	45	95.7	85.5	99.5	146.6	113.3	189.8	<28.0	905.0
		PII(D42)	47	47	100	92.5	100	258.2	205.5	324.5	28.0	1420.0
	H5N1/15/AS03	PRE	49	16	32.7	19.9	47.5	22.0	17.9	27.0	<28.0	180.0
		PI(D21)	49	49	100	92.7	100	181.3	144.6	227.3	45.0	905.0
		PII(D42)	49	49	100	92.7	100	400.1	319.3	501.4	113.0	2260.0
	H5N1/7.5/AS03	PRE	50	17	34.0	21.2	48.8	23.3	18.4	29.4	<28.0	284.0
		PI(D21)	49	47	95.9	86.0	99.5	134.6	101.3	178.7	<28.0	1420.0
		PII(D42)	50	49	98.0	89.4	99.9	343.0	260.5	451.5	<28.0	1440.0
	H5N1/3.8/AS03	PRE	50	16	32.0	19.5	46.7	21.7	17.8	26.4	<28.0	113.0
		PI(D21)	50	48	96.0	86.3	99.5	117.9	93.7	148.3	<28.0	905.0
		PII(D42)	49	48	98.0	89.1	99.9	314.7	243.1	407.3	<28.0	1420.0

At D180 all except one of the subjects from the adjuvanted groups were seropositive for NA to the vaccine strain. A rate of 98% was observed at both time points in the group that received the 3.8 µg HA + AS03 vaccine.

Table 7 **Seropositivity rates and GMTs (with 95%CI) for the neutralizing antibodies against the vaccine strain (A/Vietnam/1194/2004 strain) at Day 180 (ATP cohort for Persistence)**

			>= 28 1/DIL				GMT				
					95% CI			95% CI			
Antibodies against	Group	N	n	%	LL	UL	value	LL	UL	Min	Max
A/Vietnam	H5N1/30	49	45	91.8	80.4	97.7	81.7	61.9	107.9	<28.0	905.0
	H5N1/15	49	32	65.3	50.4	78.3	38.3	29.2	50.3	<28.0	453.0
	H5N1/7.5	48	28	58.3	43.2	72.4	32.8	25.6	42.1	<28.0	284.0
	H5N1/3.8	50	21	42.0	28.2	56.8	23.5	19.0	29.0	<28.0	226.0
	H5N1/30/AS03	48	48	100	92.6	100	130.8	109.7	155.9	28.0	569.0
	H5N1/15/AS03	49	49	100	92.7	100	130.0	106.0	159.5	28.0	720.0
	H5N1/7.5/AS03	50	50	100	92.9	100	116.3	97.7	138.4	28.0	360.0
	H5N1/3.8/AS03	50	49	98.0	89.4	99.9	101.8	84.8	122.3	<28.0	453.0

HI and NA titres against heterologous H5N1 strains

Pre-vaccination, no subject was seropositive (HI titre ≥ 10) for A/Indonesia/5/2005 (H5N1) i.e. a clade 2 sub-clade 1 strain.

- By D42 GMTs approximately doubled in the adjuvanted formulation groups and the number of seropositive subjects increased significantly to reach 26.5 to 48% per group. D42 SCFs ranged from 2.0 to 2.8. The percentage of vaccinees that seroconverted after the second vaccination ranged from 20% to 32% per group.
- After both the first and second vaccinations with non-adjuvanted formulations, the seroconversion factors were equal to 1.0 and no subjects seroconverted.

- At D180 seropositivity rates for anti-HA against the A/Indonesia/5/2005 strain were ≤ 10% for both adjuvanted and non-adjuvanted vaccine groups and seroprotection rates were all ≤ 6%.

Cross-reactivity was assessed against two additional H5N1 drifted clade 2 strains - A/Anhui/01/2005/subclade 3 and A/Turkey/Turkey/1/2005 - NIBRG23/subclade 2. Assays were performed on sera from a subset of 40 subjects who received 3.8 µg HA with or without AS03.

- No subject was seropositive for either strain before the first dose. Adjuvanted vaccine mediated a significant increase in GMT at D42 against both strains, although the absolute titres were low, and 55% - 65% were seropositive at this time point.
- At D42, seroconversion and seroprotection rates in the adjuvanted group were both 35% against A/Anhui/01/2005 and 60% against A/Turkey/Turkey/1/2005 but rates were ≤ 5% by D180. In the adjuvanted vaccine group the D42 seroconversion factors were 3.4 and 4.7 for respective strains.

For NA against A/Indonesia/5/05 (H5N1) 0-8.3% in 7/8 groups were seropositive with respect to this strain pre-vaccination but the rate was 21.3% (10/47) in the 7.5/AS03 group. At least 88% were seropositive after two doses and 79 – 83% had a titre ≥ 1:40 while 46 – 58% had ≥ 1:80. Seroconversion rates increased significantly (up to 63-77%) after the second vaccination.

NA seropositivity rates and GMTs against A/Indonesia/5/2005 (H5N1) (ATP)

				≥ 28 1/DIL				GMT				
						95% CI			95% CI			
Antibody	Group	Timing	N	n	%	LL	UL	value	LL	UL	Min	Max
	H5N1/30/AS03	PRE	47	0	0.0	0.0	7.5	14.0	14.0	14.0	<28.0	<28.0
		PI(D21)	46	38	82.6	68.6	92.2	54.6	42.5	70.1	<28.0	284.0
		PII(D42)	46	42	91.3	79.2	97.6	66.8	53.4	83.5	<28.0	226.0
	H5N1/15/AS03	PRE	44	1	2.3	0.1	12.0	14.2	13.8	14.7	<28.0	28.0
		PI(D21)	44	35	79.5	64.7	90.2	38.1	30.0	48.5	<28.0	287.0
		PII(D42)	44	41	93.2	81.3	98.6	72.9	58.5	90.9	<28.0	226.0
	H5N1/7.5/AS03	PRE	47	10	21.3	10.7	35.7	17.3	15.2	19.5	<28.0	57.0
		PI(D21)	47	34	72.3	57.4	84.4	43.7	33.7	56.6	<28.0	284.0
		PII(D42)	46	45	97.8	88.5	99.9	95.7	75.3	121.7	<28.0	453.0
	H5N1/3.8/AS03	PRE	48	4	8.3	2.3	20.0	15.8	13.9	17.9	<28.0	113.0
		PI(D21)	48	32	66.7	51.6	79.6	36.6	28.8	46.5	<28.0	226.0
		PII(D42)	48	42	87.5	74.8	95.3	80.3	62.0	103.9	<28.0	284.0

At D180 the NA seropositivity rates against the A/Indonesia/5/2005 strain in the adjuvanted groups were 82% to 92% compared to 6 – 49% in the non-adjuvanted groups. For the 3.8 µg HA + AS03 vaccine the rates were 87.5% at D42 and 82% at D180. There was no antigen dose effect in the adjuvanted groups for seropositivity rates, GMTs or seroconversion rates.

Table 9 **Seropositivity rates and GMTs (with 95%CI) for the neutralizing antibodies against the A/Indonesia/5/2005 strain at Day 180 (ATP cohort for Persistence)**

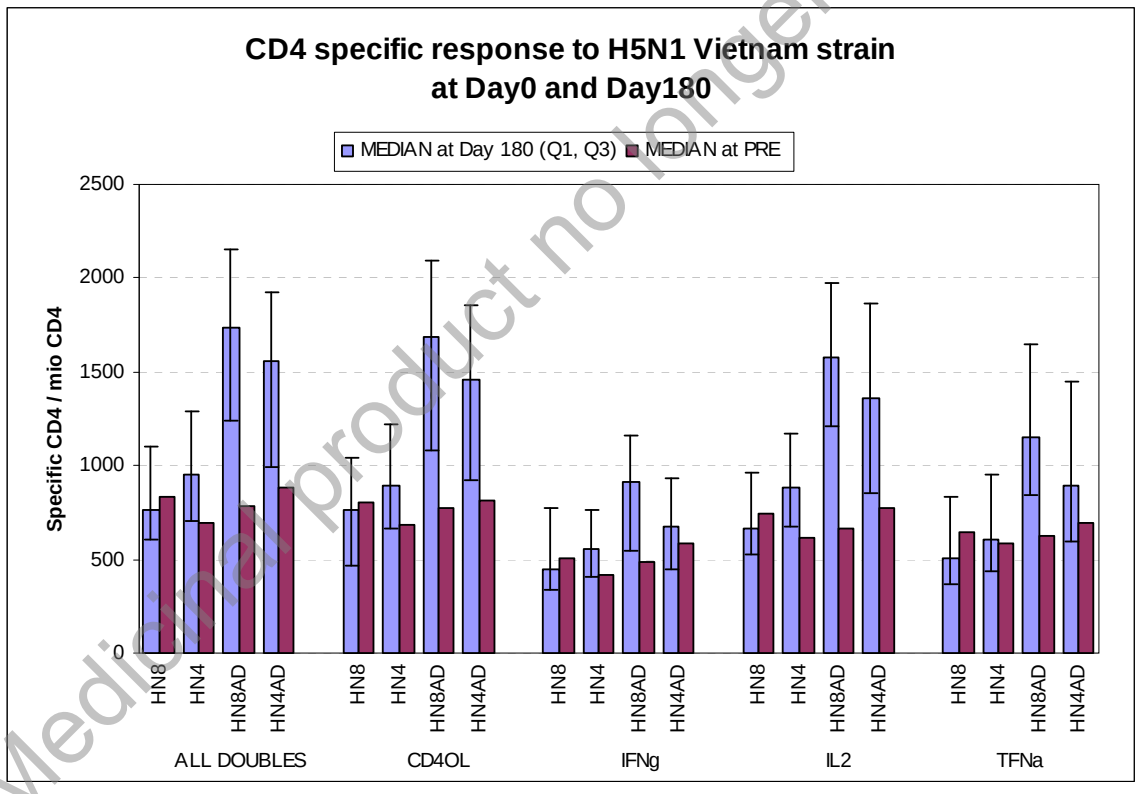
			>= 28 1/DIL				GMT				
					95% CI			95% CI			
Antibodies against	Group	N	n	%	LL	UL	value	LL	UL	Min	Max
A/Indonesia	H5N1/30	49	24	49.0	34.4	63.7	25.4	20.7	31.2	<28.0	226.0
	H5N1/15	49	16	32.7	19.9	47.5	19.9	16.8	23.5	<28.0	113.0
	H5N1/7.5	49	12	24.5	13.3	38.9	18.5	15.7	21.9	<28.0	226.0
	H5N1/3.8	50	3	6.0	1.3	16.5	14.7	13.9	15.5	<28.0	36.0
	H5N1/30/AS03	48	44	91.7	80.0	97.7	48.7	40.1	59.2	<28.0	226.0
	H5N1/15/AS03	49	43	87.8	75.2	95.4	52.9	41.7	67.2	<28.0	284.0
	H5N1/7.5/AS03	50	41	82.0	68.6	91.4	45.8	37.5	55.9	<28.0	180.0
	H5N1/3.8/AS03	50	41	82.0	68.6	91.4	46.1	36.9	57.6	<28.0	180.0

NA assays performed against A/Anhui/01/2005/subclade 3 and A/Turkey/Turkey/1/2005 - NIBRG23/subclade 2 showed that 0-4 subjects per group had detectable antibody before vaccination. By D42 1-2 subjects in the non-adjuvanted groups had become seropositive but none met the criteria for seroconversion and there was no change in GMTs. The adjuvanted vaccine elicited significant increases in GMTs after the first and second doses. At D42 all subjects were seropositive for NA against these strains and 75% and 85% had seroconverted against A/Anhui/01/2005 and A/Turkey/Turkey/1/2005, respectively. At D180 all except one subject in the adjuvanted groups remained seropositive for both strains and 60% and 70% still met the seroconversion criterion although GMTs had declined by about one third.

Influenza-specific T-cells

Pre-vaccination frequencies of influenza-specific CD4 T-cells were similar across groups. On stimulation with split A/Vietnam/1194/2004 frequencies of influenza-specific CD4 T-cells significantly increased in all groups after the first vaccination but essentially remained unchanged after the second dose. This lack of detectable increment after the second dose might have occurred because the sample was taken after the peak response occurred. The frequencies of influenza-specific CD4 T-cells were higher in adjuvanted compared with non-adjuvanted formulation groups. No significant effect of antigen dose was detected. The D0 and D180 results are compared below.

Frequency of positive CD4 T cells at D0 and D180



At D180 frequencies of influenza-specific CD4 T-cells remained high compared to the pre-vaccination levels. Values were higher after vaccination with the AS03 adjuvanted compared to non-adjuvanted formulations. Individual differences between D180 and D0 in CD4 responses showed a statistically significant difference between adjuvanted and non-adjuvanted groups for both 3.8 µg and 7.5 µg formulations for all types of cytokines except IFNγ.

The pre-vaccination frequency of influenza-specific CD8 T-cells was essentially similar in all groups. No significant effect of vaccination was observed on the frequency of influenza-specific CD8 T-cells at D42 or at D180 whatever the formulation received.

T-cell cross-reactivity was examined against the clade 2 subclade 1 A/Indonesia/5/2005 strain for D0 and D42 in the 3.8 µg HA non-adjuvanted and adjuvanted groups from study 007. The 3.8 µg HA/AS03 formulation induced a cross-reactive CD4+ T cell response to the heterologous clade 2 H5N1 A/Indonesia/05/05 strain that was similar to that against the vaccine strain. The response to both strains was higher in the adjuvanted vaccine group. There was also a limited CD4+ T cell response to the heterologous split virions H3N2 New-York (NY) and H1N1 New Caledonia (NC). It is not known whether the cross-recognised domains belong to the split virion backbone (PR8) and/or represent conserved T cell epitopes on haemagglutinin and/or neuraminidase proteins.

T-cell cross-reactivity was also examined using pools of peptides derived from the HA of the vaccine strain, the H5N1 drifted clade 2 subclade1 A/Indonesia/5/2005 and the Anhui clade 2 subclade 3 (A/Anhui/01/2005) strain. Data were generated for D0 and D42 in the 3.8 µg HA non-adjuvanted and adjuvanted groups. There was only a weak response with respect to each strain in the non-adjuvanted group. The adjuvanted vaccine elicited a significant increase in the response against HA peptides from A/Vietnam, A/Indonesia and A/Anhui.

Study H5N1- 008 (and extension to D180 as study 011)

HI titres against the vaccine strain

Pre-vaccination 0-2.5% of subjects aged < 60 years but 10-18% aged > 60 years were already seropositive. Due to the baseline seropositivity rates the data shown below refer only to subjects who were initially seronegative and, therefore, the seroprotection and seroconversion rates are the same.

In adults aged between 18 and 60 years, the seroprotection rate exceeded the 70% seroprotection threshold after the second vaccination. In adults aged above 60 years, the 60% threshold was exceeded after the first vaccination (61.4%). In both age-strata, the seroprotection rates reached 91.4% at D42.

In both age-strata, the SCFs exceeded the relevant CHMP thresholds after the first dose of 15µg/AS03. After the second dose the SCFs significantly increased in both age-strata but the increment and the final GMT were higher in adults aged between 18 and 60 years than in those aged > 60 years. There were no appreciable changes in GMTs after Fluarix was given in either age group.

Initially seronegative subjects: Seroprotection rates for anti-HA (ATP cohort)

Antibody	Group	Timing	N	n	SP with 95%CI			n UNPROT	% UNPROT
					%				
A/Vietnam	H5N1 18-60	PRE	269	0	0.0	0.00	1.36	269	100.0
		PI(D21)	269	147	54.6	48.49	60.70	122	45.4
		PII(D42)	268	245	91.4	87.40	94.48	23	8.6
	H5N1 >60	PRE	146	0	0.0	0.00	2.49	146	100.0
		PI(D21)	145	89	61.4	52.94	69.34	56	38.6
		PII(D42)	140	128	91.4	85.51	95.49	12	8.6
	Fluarix 18-60	PRE	96	0	0.0	0.00	3.77	96	100.0
		PI(D21)	97	5	5.2	1.69	11.62	92	94.8
		PII(D42)	96	3	3.1	0.65	8.86	93	96.9
	Fluarix >60	PRE	50	0	0.0	0.00	7.11	50	100.0
		PI(D21)	49	5	10.2	3.40	22.23	44	89.8
		PII(D42)	50	6	12.0	4.53	24.31	44	88.0

At D180 58% of those aged < 60 years and 79% aged > 60 years in the group that had received 15µg/AS03 were seroprotected. The seroprotection rate at D180 was the same (58%) for the 18-30 and 31-60 years groups. In contrast only 2% of those aged < 60 and 18% of those >60 years who had been primed with Fluarix were seroprotected at D180. The seroconversion rates were 57% and 74% for the younger and older age groups, respectively, who had received 15µg/AS03 compared to 2% and 9% in

the control group. The corresponding SCFs were 5.2 and 8.3 compared to 1.1 and 1.5 in the control group.

Seroprotection rates (SPR) for anti-HA at each time point (ATP cohort for persistence)

					SPR			
							95% CI	
Vaccine strain	Group	Sub-group	Timing	N	n	%	LL	UL
FLU A/VIET/04 AB	H5N1	18-60	PRE	279	4	1.4	0.4	3.6
			PII(D180)	279	161	57.7	51.7	63.6
		>60	PRE	170	18	10.6	6.4	16.2
			PII(D180)	171	135	78.9	72.1	84.8
	Fluarix	18-60	PRE	94	0	0.0	0.0	3.8
			PII(D180)	95	2	2.1	0.3	7.4
		>60	PRE	54	3	5.6	1.2	15.4
			PII(D180)	55	10	18.2	9.1	30.9

H5N1 = H5N1 15µg HA + AS03
Fluarix = Fluarix/Placebo at the 2nd dose

NA titres against the vaccine strain

NA data were provided for subjects aged > 60 years. Data in younger subjects are awaited.

- The majority of these older subjects (82% and 91% per group) were already seropositive (i.e. titres at least 1:28) for NA to the vaccine strain before vaccination. The percentages with pre-vaccination titres ≥ 1:40 or ≥ 1:80 were similar between the 15 µg HA/AS03 and the Fluarix control group (i.e. 69.5% and 67.3% at 1:40, with 44.6% and 40.0% at 1:80).
- After a single dose of the adjuvanted H5N1 vaccine all except one subject was seropositive. After a second dose there were further and significant increments in GMT and SCR.
- In the 15 µg HA/AS03 group the percentages with ≥ 1:40 and ≥ 1:80 at D21 were 99.4% and 93.8%, respectively. At D42 these percentages were 100% and 99.4%. At D21 and D42 there was a statistically higher immune response in the 15 µg HA/AS03 group.

Percentage with NA titres ≥ 1:40 and ≥ 1:80 at each time point against vaccine strain H5N1 A/Vietnam/1194/2004 in H5N1-008 (ATP cohort for Immunogenicity)

			≥40 1/DIL				≥80 1/DIL			
		N	n	%	95%CI		n	%	95%CI	
Group	Timing				LL	UL			LL	UL
H5N1 15/AS03	PRE	177	123	69.5	62.1	76.2	79	44.6	37.2	52.3
	PI(D21)	176	175	99.4	96.9	100.0	165	93.8	89.1	96.8
	PII(D42)	170	170	100	97.9	100.0	169	99.4	96.8	100.0
Fluarix/Placebo	PRE	55	37	67.3	53.3	79.3	22	40.0	27.0	54.1
	PI(D21)	53	48	90.6	79.3	96.9	35	66.0	51.7	78.5
	PII(D42)	53	46	86.8	74.7	94.5	31	58.5	44.1	71.9

At D180 all except one subject aged > 60 years who had received the 15µg + AS03 vaccine and 82% in the control group had NA titres to the vaccine strain ≥ 1:28. The seroconversion rates at D180 in this age group were 43% for the 15µg/AS03 group compared with 6% for the control group.

NA titres at D180 in elderly

				≥28 1/DIL				GMT				
						95% CI			95% CI			
Antibodies against	Group	Timing	N	n	%	LL	UL	value	LL	UL	Min	Max
A/Vietnam	H5N1	PRE	152	127	83.6	76.7	89.1	63.0	54.2	73.3	<28.0	905.0
		PI(D21)	152	152	100	97.6	100	248.4	220.4	279.9	36.0	2260.0
		PII(D42)	147	147	100	97.5	100	362.0	329.8	397.3	71.0	1420.0
		PII (D180)	153	152	99.3	96.4	100	219.8	194.3	248.7	<28.0	905.0
	Fluarix/ Placebo	PRE	50	45	90.0	78.2	96.7	62.0	47.9	80.1	<28.0	360.0
		PI(D21)	48	47	97.9	88.9	99.9	114.8	89.0	148.2	<28.0	900.0
		PII(D42)	48	47	97.9	88.9	99.9	104.8	81.7	134.4	<28.0	720.0
		PII (D180)	51	42	82.4	69.1	91.6	61.8	46.1	82.8	<28.0	905.0

Study H5N1-002

HI titres

Consistency among the four adjuvanted vaccine lots based on pre-defined criteria applied to D42 data was demonstrated. GMTs for anti-HA antibody were very similar between the four adjuvanted groups on D0, D21 and D42 for the homologous vaccine strain and were also similar between groups but much lower against A/Indonesia/5/2005 (next table). Results for the two non-adjuvanted groups were similar to each other but showed a very small anti-HA response to the vaccine strain and no discernible response to the heterologous strain.

The SCF threshold of ≥ 2.5 was reached in the pooled H5N1 adjuvanted AS03 vaccine group after the first dose (4.1) and the second dose (39.8) for the A/Vietnam strain but only after the second dose (4.9) for the A/Indonesia strain.

Similar findings applied to the seroconversion rates for anti-HA antibody for each vaccine group and for pooled adjuvanted and non-adjuvanted groups. The SCR in the pooled adjuvanted group at D42 was 94% for the vaccine strain and 50% for the heterologous strain and the required 40% threshold rate was reached after a single dose.

Geometric mean titres (GMTs) of Anti-HA antibody titres at days 0, 21 and 42 by H5N1 strain Pooled vaccine groups (ATP cohort for immunogenicity)

				>= 10 1/DIL				GMT				
						95% CI			95% CI			
Antigen	Group	Timing	N	n	%	LL	UL	value	LL	UL	Min	Max
H5N1 (A/VIET)	HN-AS03	PRE	933	59	6.3	4.8	8.1	5.5	5.4	5.7	<10.0	320.0
		PI(D21)	925	544	58.8	55.6	62.0	22.8	20.7	25.0	<10.0	1280.0
		PII(D42)	924	881	95.3	93.8	96.6	219.4	203.3	236.9	<10.0	5120.0
	HN DIL	PRE	236	17	7.2	4.3	11.3	5.6	5.3	5.9	<10.0	57.0
		PI(D21)	234	33	14.1	9.9	19.2	6.7	6.0	7.4	<10.0	320.0
		PII(D42)	234	50	21.4	16.3	27.2	7.5	6.7	8.3	<10.0	320.0
H5N1 (A/IND)	HN-AS03	PRE	933	8	0.9	0.4	1.7	5.1	5.0	5.1	<10.0	40.0
		PI(D21)	925	111	12.0	10.0	14.3	6.0	5.8	6.2	<10.0	453.0
		PII(D42)	924	587	63.5	60.3	66.6	24.9	22.8	27.3	<10.0	640.0
	HN DIL	PRE	236	2	0.8	0.1	3.0	5.0	5.0	5.1	<10.0	20.0
		PI(D21)	234	7	3.0	1.2	6.1	5.2	5.0	5.3	<10.0	28.0
		PII(D42)	234	7	3.0	1.2	6.1	5.2	5.0	5.4	<10.0	40.0

HN-AS03 = pooled adjuvanted group (H5N1_AX, H5N1_AY, H5N1_BX, H5N1_BY)
HN DIL = pooled un-adjuvanted group (H5N1_AD, H5N1_BD)

A small proportion (15; 1.6%) had seroprotective anti-HA antibody before vaccination. The threshold of 70% seroprotected was reached in the pooled H5N1 adjuvanted group (94.3%) after the second

dose (D42) for the A/Vietnam strain but the D42 seroprotection rate against the heterologous strain was 50.2%.

Seroprotection rates for anti-HA antibody – Pooled vaccine groups (ATP cohort for immunogenicity)

				SPR			
						95% CI	
Antigen	Group	Timing	N	n	%	LL	UL
H5N1 (A/VIET)	HN-AS03	PRE	933	15	1.6	0.9	2.6
		PI(D21)	925	412	44.5	41.3	47.8
		PII(D42)	924	871	94.3	92.6	95.7
	HN DIL	PRE	236	5	2.1	0.7	4.9
		PI(D21)	234	16	6.8	4.0	10.9
		PII(D42)	234	24	10.3	6.7	14.9
H5N1 (A/IND)	HN-AS03	PRE	933	1	0.1	0.0	0.6
		PI(D21)	925	27	2.9	1.9	4.2
		PII(D42)	924	464	50.2	46.9	53.5
	HN DIL	PRE	236	0	0.0	0.0	1.6
		PI(D21)	234	0	0.0	0.0	1.6
		PII(D42)	234	1	0.4	0.0	2.4

HN-AS03 = pooled adjuvanted group (H5N1_AX, H5N1_AY, H5N1_BX, H5N1_BY)
HN DIL = pooled un-adjuvanted group (H5N1_AD, H5N1_BD)

NA titres

Before vaccination 17 - 20% of subjects were seropositive for NA against the vaccine strain while 5 - 11% of subjects were seropositive for NA against A/Indonesia/2005.

For the individual adjuvanted groups the percentages with titres ≥1:28 at D42 ranged from 98-100% for the vaccine strain and 92-100% for the heterologous strain (next table). In contrast, percentages with titres ≥1:28 at D42 in the two non-adjuvanted groups were 42% and 54% for the vaccine strain and 17% and 11% for the heterologous strain.

For the pooled adjuvanted vaccine groups the seroconversion rate was 96% for the H5N1 A/Vietnam strain and 91.4% for A/Indonesia/2005.

Seropositivity rates and GMTs for NA at D0 and D42 (ATP cohort for immunogenicity)

				>= 28 1/DIL				GMT				
						95% CI			95% CI			
Antibody	Group	Timing	N	n	%	LL	UL	value	LL	UL	Min	Max
FLU A/VIET/04 AB	HN-AS03	PRE	279	56	20.1	15.5	25.3	17.5	16.5	18.7	<28.0	360.0
		PII(D42)	277	276	99.6	98.0	100	308.4	283.1	336.1	<28.0	4530.0
	HN DIL	PRE	71	12	16.9	9.0	27.7	17.7	15.4	20.4	<28.0	226.0
		PII(D42)	71	34	47.9	35.9	60.1	29.0	23.4	35.9	<28.0	226.0
FLU A/IND/05 AB	HN-AS03	PRE	279	15	5.4	3.0	8.7	14.9	14.3	15.4	<28.0	569.0
		PII(D42)	279	266	95.3	92.2	97.5	84.0	77.1	91.4	<28.0	720.0
	HN DIL	PRE	71	8	11.3	5.0	21.0	15.6	14.4	16.7	<28.0	57.0
		PII(D42)	71	10	14.1	7.0	24.4	16.2	14.8	17.8	<28.0	71.0

Ancillary analyses

Comparison of results with adjuvanted vaccine in studies 002, 007 and 008

On comparing D42 data for the 3.8 µg/AS03 vaccine the HI titres against the vaccine strain tended to be higher for the 924 subjects tested in study 002 compared to the 50 tested in study 007. On comparing study 002 with the results for the 15 µg HA/AS03 vaccine used in study 008 the GMT and SCF were higher (95% CI do not overlap) with the higher dose vaccine but the seroconversion rates and seroprotection rates were similar between the two doses of adjuvanted HA. There is clearly a need

for two doses of Pandemrix to meet all three CHMP criteria with respect to HI against the vaccine strain in healthy adults aged 18-60 years.

D42 HI against vaccine strain in adults aged 18-60 years from H5N1-007, H5N1-008 and H5N1-002 (ATP immunogenicity cohort) compared to CHMP criteria

Study	HA (µg per dose)	N	GMT			SCF >2.5 <60 years of age >2.0 > 60 years of age			SCR >40% <60 years of age >30% > 60 years of age			SPR >70% <60 years of age >60% > 60 years of age		
			Value	95% CI		GMR	95% CI		%	95% CI		%	95% CI	
				LL	UL		LL	UL		LL	UL		LL	UL
H5N1-007	3.8	50	149.3	93.2	239.1	27.9	17.2	45.2	82.0	68.6	91.4	84.0	70.9	92.8
H5N1-008	15	275	312.8	264.2	370.4	58.6	49.4	69.5	91.6	87.7	94.6	91.6	87.7	94.6
H5N1-002	3.8	924	219.4	203.3	236.9	39.8	36.8	43.1	93.7	92.0	95.2	94.3	92.6	95.7

The seroprotection rates in subjects aged 18-60 years at D180 were 54% after priming with 3.8 µg/AS03 in study 007 compared to 58% primed with 15 µg/AS03 in study 008. The respective HI GMTs at D180 were 23.3 and 27.2.

Based on these comparisons of D42 data between studies the D180 immunogenicity data from studies 007 and 008 (i.e. 3.8 µg/AS03 or 15 µg/AS03) should be indicative of the D180 status of subjects who have received two doses of the final production process 3.8 µg/AS03 vaccine in study 002. Thus the CHMP requirement for provision of 6-month post-primary vaccination immunogenicity data was considered to be fulfilled.

The D42 NA response against the vaccine strain was similar between 002 and 007 (all 95% CI overlap). At least 98% were seropositive with respect to the vaccine strain while the seropositivity rate with respect to the heterologous strain was numerically higher in 002. The seroconversion rates were numerically or (borderline) significantly greater with respect to homologous and heterologous strains, respectively, in study 002. However, the GMTs were the same between studies for each of the homologous and heterologous strains and about 4-fold higher for the former than for the latter strain.

At D180 in 007 all except one subject was seropositive for NA against the vaccine strain in the 3.8 µg HA/AS03 vaccine group with a seropositivity rate of 98% and seroconversion rate of 72%. The corresponding rates for the heterologous strain were 82% and 40%.

NA against vaccine strain and H5N1 A/Indonesia/5/2005 in adults aged 18-60 years in H5N1-002 and H5N1-007 (ATP immunogenicity cohort)

Study	HA (µg per dose)	N	≥ 1:28			SCR			GMT		
			%	95% CI		%	95% CI		Value	95% CI	
				LL	UL		LL	UL		LL	UL
Vietnam											
H5N1-002	3.8	277	99.6	98.0	100.0	96.0	93.0	98.0	308.4	283.1	336.1
H5N1-007	3.8	49	98.0	89.1	99.9	85.7	72.8	94.1	314.7	243.1	407.3
Indonesia											
H5N1-002	3.8	279	95.3	92.2	97.5	91.4	87.5	94.4	84.0	77.1	91.4
H5N1-007	3.8	48	87.5	74.8	95.3	77.1	62.7	88.0	80.3	62.0	103.9

NA data in the age group 18-60 years are not yet available from study 008 but in subjects aged > 60 years the D42 NA data for the vaccine strain showed that all those tested were seropositive and 67% had seroconverted with a GMT of 384, which compares well with the value of 308 in the younger subjects in study 002 despite the difference in pre-vaccination seropositivity rates.

These data strongly suggest that most or all subjects in study 002 will still be seropositive for NA with respect to the vaccine strain at D180 and probably around 80% will be seropositive for the

heterologous strain. However proportions with titres of at least 1:40 or 1:80 will be lower. As for HI, it is not known how having NA titres at these levels may correlate with protection against a pandemic strain at or beyond D180.

Clinical safety

Solicited symptoms were recorded during the 7-day follow-up period after each dose together with any analgesics and/or antipyretics taken. Unsolicited symptoms occurring during a 21-day follow-up period after the first vaccination and 30 days after the second one were also recorded in the CRF.

- Patient exposure

Overall, 4,002 healthy subjects (minimum age 18 years) were exposed to H5N1 AS03 adjuvanted vaccine in studies 007 and 008 i.e. total across all doses. More than 3800 of these subjects received an HA dose ≥15 µg i.e. at least 4-fold the HA dose in the intended marketed formulation. Another 961 subjects aged 18-60 years received at least one dose of Third series 3.8 µg/AS03 vaccine in study 002 and were included in the safety evaluation.

- Adverse events

AEs in study H5N1-007

The incidence of symptoms, and in particular local symptoms, was higher in the groups vaccinated with adjuvanted formulations. No significant effect of the antigen dose or consistent trend by dose was observed on the overall incidence of symptoms among the adjuvanted vaccines.

Solicited AEs (any severity) within 7 days after each dose and overall

		General symptoms					Local symptoms				
					95% CI					95% CI	
	Group	N	n	%	LL	UL	N	n	%	LL	UL
Dose 1	H5N1/30	50	25	50.0	35.5	64.5	50	28	56.0	41.3	70.0
	H5N1/15	50	27	54.0	39.3	68.2	50	20	40.0	26.4	54.8
	H5N1/7.5	50	31	62.0	47.2	75.3	50	17	34.0	21.2	48.8
	H5N1/3.8	50	24	48.0	33.7	62.6	50	18	36.0	22.9	50.8
	H5N1/30/AS03	49	40	81.6	68.0	91.2	49	46	93.9	83.1	98.7
	H5N1/15/AS03	50	41	82.0	68.6	91.4	50	46	92.0	80.8	97.8
	H5N1/7.5/AS03	50	32	64.0	49.2	77.1	50	44	88.0	75.7	95.5
	H5N1/3.8/AS03	51	29	56.9	42.2	70.7	51	46	90.2	78.6	96.7
Dose 2	H5N1/30	50	21	42.0	28.2	56.8	50	23	46.0	31.8	60.7
	H5N1/15	50	19	38.0	24.7	52.8	50	14	28.0	16.2	42.5
	H5N1/7.5	50	14	28.0	16.2	42.5	50	14	28.0	16.2	42.5
	H5N1/3.8	50	13	26.0	14.6	40.3	50	16	32.0	19.5	46.7
	H5N1/30/AS03	49	26	53.1	38.3	67.5	49	39	79.6	65.7	89.8
	H5N1/15/AS03	50	36	72.0	57.5	83.8	50	40	80.0	66.3	90.0
	H5N1/7.5/AS03	50	27	54.0	39.3	68.2	50	41	82.0	68.6	91.4
	H5N1/3.8/AS03	51	30	58.8	44.2	72.4	51	42	82.4	69.1	91.6
Overall/ dose	H5N1/30	100	46	46.0	36.0	56.3	100	51	51.0	40.8	61.1
	H5N1/15	100	46	46.0	36.0	56.3	100	34	34.0	24.8	44.2
	H5N1/7.5	100	45	45.0	35.0	55.3	100	31	31.0	22.1	41.0
	H5N1/3.8	100	37	37.0	27.6	47.2	100	34	34.0	24.8	44.2
	H5N1/30/AS03	98	66	67.3	57.1	76.5	98	85	86.7	78.4	92.7
	H5N1/15/AS03	100	77	77.0	67.5	84.8	100	86	86.0	77.6	92.1
	H5N1/7.5/AS03	100	59	59.0	48.7	68.7	100	85	85.0	76.5	91.4
	H5N1/3.8/AS03	102	59	57.8	47.7	67.6	102	88	86.3	78.0	92.3
Overall/ subject	H5N1/30	50	32	64.0	49.2	77.1	50	38	76.0	61.8	86.9
	H5N1/15	50	32	64.0	49.2	77.1	50	24	48.0	33.7	62.6

	H5N1/7.5	50	32	64.0	49.2	77.1	50	24	48.0	33.7	62.6
	H5N1/3.8	50	27	54.0	39.3	68.2	50	24	48.0	33.7	62.6
	H5N1/30/AS03	49	41	83.7	70.3	92.7	49	47	95.9	86.0	99.5
	H5N1/15/AS03	50	45	90.0	78.2	96.7	50	48	96.0	86.3	99.5
	H5N1/7.5/AS03	50	39	78.0	64.0	88.5	50	48	96.0	86.3	99.5
	H5N1/3.8/AS03	51	38	74.5	60.4	85.7	51	48	94.1	83.8	98.8

In contrast, the incidences of severe symptoms were low and variable across groups with no obvious dose trends. However, the highest rates were seen with one or more of the adjuvanted formulations.

Solicited severe AEs within 7 days overall by subject

		General symptoms					Local symptoms				
					95% CI					95% CI	
	Group	N	n	%	LL	UL	N	n	%	LL	UL
Overall/ subject	H5N1/30	50	1	2.0	0.1	10.6	50	0	0.0	0.0	7.1
	H5N1/15	50	2	4.0	0.5	13.7	50	1	2.0	0.1	10.6
	H5N1/7.5	50	1	2.0	0.1	10.6	50	1	2.0	0.1	10.6
	H5N1/3.8	50	2	4.0	0.5	13.7	50	0	0.0	0.0	7.1
	H5N1/30/AS03	49	0	0.0	0.0	7.3	49	3	6.1	1.3	16.9
	H5N1/15/AS03	50	7	14.0	5.8	26.7	50	10	20.0	10.0	33.7
	H5N1/7.5/AS03	50	2	4.0	0.5	13.7	50	8	16.0	7.2	29.1
	H5N1/3.8/AS03	51	6	11.8	4.4	23.9	51	3	5.9	1.2	16.2

Pain at the injection site was the most frequently reported local symptom in all groups (e.g. 86% in the 3.8 µg/AS03 group). The incidence was significantly higher in groups given adjuvanted vaccines (86-92% after dose 1 and 69-80% after dose 2 compared to 28-50% and 22-40% after respective doses in the groups given non-adjuvanted vaccines). There was also a trend for higher incidences of swelling and redness in groups with adjuvanted vaccines and the rate of induration was significantly higher in adjuvanted groups. However, severe swelling, redness and induration were all reported at low rates.

The most frequently reported general symptoms were fatigue and headache. For all of the solicited general symptoms there was at least a trend for higher incidence in groups that received adjuvanted vaccines. For example, myalgia reported as being vaccine-related was reported for 10-20% in the non-adjuvanted vaccine groups but 36-44.9% with adjuvanted vaccines. Fever was reported with a very low incidence in all groups and no fever above 39°C was reported.

Lymphadenopathy followed administration of 7.5 µg HA (3 cases), 15 µg HA (3 cases) and 30 µg HA (1 case) adjuvanted vaccines. Six of these 7 cases were considered to be related to vaccination. No reports of local lymph node swelling were of grade 3 severity and all subjects recovered without sequelae.

AEs in study H5N1-008

The highest incidence of any severe symptom after vaccination with 15 µg/AS03 vaccine (16.3%) was reported by subjects aged 18 to 60 years after the first dose. After the first injection, severe symptoms were reported more frequently in subjects aged 18-60 years than in those > 60 years but after the second injection there was little difference between the age strata. Severe symptoms were reported with a significantly lower incidence in subjects vaccinated with Fluarix and placebo.

Among solicited local symptoms pain was the most frequently reported AE in both treatment groups and age strata but was reported with a significantly higher incidence in subjects vaccinated with 15 µg/AS03 than in subjects vaccinated with Fluarix or placebo. Pain was predominantly reported by subjects aged between 18 and 60 years.

In the 15 µg/AS03 group general symptoms were reported more frequently in those aged 18-60 years than in those aged > 60 years. The most common symptoms were fatigue, headache and myalgia in both treatment groups and age strata.

Solicited and unsolicited symptoms reported during the 7-days post-vaccination (Total vaccinated cohort)

		General symptoms					Local symptoms				
					95% CI					95% CI	
	Group	N	n	%	LL	UL	N	n	%	LL	UL
Dose 1	H5N1 18-60	3341	2285	68.4	66.8	70.0	3342	3007	90.0	88.9	91.0
	H5N1 >60	403	169	41.9	37.1	46.9	403	281	69.7	65.0	74.2
	Fluarix 18-60	1123	560	49.9	46.9	52.8	1123	808	72.0	69.2	74.6
	Fluarix >60	133	39	29.3	21.8	37.8	133	57	42.9	34.3	51.7
Dose 2	H5N1 18-60	3246	1766	54.4	52.7	56.1	3247	2590	79.8	78.3	81.1
	H5N1 >60	395	157	39.7	34.9	44.8	395	242	61.3	56.3	66.1
	Placebo 18-60	1102	297	27.0	24.4	29.7	1102	256	23.2	20.8	25.8
	Placebo >60	132	32	24.2	17.2	32.5	132	25	18.9	12.6	26.7
Overall/subject	H5N1 18-60	3342	2569	76.9	75.4	78.3	3343	3107	92.9	92.0	93.8
	H5N1 >60	403	225	55.8	50.8	60.7	403	316	78.4	74.1	82.3
	Fluarix/Placebo 18-60	1123	651	58.0	55.0	60.9	1123	833	74.2	71.5	76.7
	Fluarix/Placebo >60	133	47	35.3	27.3	44.1	133	63	47.4	38.7	56.2

In both vaccine groups subjects aged 18 - 60 years more frequently reported unsolicited symptoms than those aged > 60 years. The most common symptoms were injection site pruritus and warmth, influenza like illness and related symptoms and gastrointestinal symptoms (diarrhoea, nausea). There were 95 AE reports coded as the MedDRA preferred terms lymphadenopathy, lymph node pain or lymphadenitis, of which 84/95 occurred in the 15µg/AS03 group. These subjects were more likely than others to report other general symptoms.

Based on the applicant's assessment from D51 to D180 there were 16 subjects in the 15µg/AS03 group and three in the Fluarix group who reported 17 and 3 unsolicited symptoms, respectively, classified as new onset chronic diseases (NOCD). There was no clear disease pattern identified.

Summary of new onset chronic diseases (GSK assessment) reported (Total vaccinated cohort)

	Group				
	HN AS03		Fluarix	All	
	s18	s61	s18	s18	s61
N with at least one unsolicited symptom reported	9	7	3	12	7
N doses followed by at least one unsolicited symptom	9	7	3	12	7
N unsolicited symptoms classified by MEDDRA Term*	9	7	3	12	7
N unsolicited symptoms reported	10	7	3	13	7

HN AS03 = H5N1 15µg HA + AS03, Fluarix = Fluarix/ (Placebo at the 2nd dose)
s18 = 18-60 years, s61 = 61-120 years
* Symptoms reported by a subject after a given dose and classified by the same Preferred Term are counted once

Also, 64 subjects in the 15µg/AS03 group and 14 in the Fluarix group reported 75 and 17 unsolicited symptoms, respectively, classified as medically significant conditions.

Summary of medically significant conditions (GSK assessment) reported (Total vaccinated cohort)

	Group					
	HN AS03		Fluarix		All	
	s18	s61	s18	s61	s18	s61
N with at least one unsolicited symptom reported	56	8	11	3	67	11
N doses followed by at least one unsolicited	56	8	11	3	67	11

	Group					
	HN AS03		Fluarix		All	
	s18	s61	s18	s61	s18	s61
symptom						
N unsolicited symptoms classified by MEDDRA	65	9	14	3	79	12
Number of unsolicited symptoms reported	66	9	14	3	80	12

AEs in study H5N1-002

These are the safety data most relevant to the use of the commercial product in adults aged 18-60 years. However, there were no consistent differences in AEs reported in this study compared to those reported with the 3.8 µg/AS03 vaccine used in study 007.

The next table shows the incidence of solicited local symptoms over the 7-day follow-up period after vaccination overall by subject. Subjects in the adjuvanted groups frequently reported pain, swelling and redness with generally similar rates across the four groups. Rates were much lower in the non-adjuvanted vaccine groups in which no grade 3 local symptoms were reported.

Incidence of solicited local symptoms reported during the 7-day (Days 0-6) post-vaccination period following each dose and overall (Total vaccinated cohort)

		H5N1_AX					H5N1_AY					H5N1_BX				
					95 % CI					95 % CI					95 % CI	
Symptom	Type	N	n	%	LL	UL	N	n	%	LL	UL	N	n	%	LL	UL
Overall/subject																
Ecchymosis (mm)	All	238	14	5.9	3.3	9.7	237	15	6.3	3.6	10.2	240	14	5.8	3.2	9.6
	> 50 mm	238	0	0.0	0.0	1.5	237	2	0.8	0.1	3.0	240	0	0.0	0.0	1.5
Induration (mm)	All	238	57	23.9	18.7	29.9	237	55	23.2	18.0	29.1	240	54	22.5	17.4	28.3
	> 50 mm	238	5	2.1	0.7	4.8	237	4	1.7	0.5	4.3	240	2	0.8	0.1	3.0
Pain	All	238	210	88.2	83.4	92.0	237	199	84.0	78.7	88.4	240	208	86.7	81.7	90.7
	Grade 3	238	12	5.0	2.6	8.6	237	12	5.1	2.6	8.7	240	11	4.6	2.3	8.1
Redness (mm)	All	238	88	37.0	30.8	43.4	237	79	33.3	27.4	39.7	240	77	32.1	26.2	38.4
	> 50 mm	238	6	2.5	0.9	5.4	237	9	3.8	1.8	7.1	240	3	1.3	0.3	3.6
Swelling (mm)	All	238	95	39.9	33.6	46.4	237	80	33.8	27.8	40.2	240	86	35.8	29.8	42.3
	> 50 mm	238	8	3.4	1.5	6.5	237	5	2.1	0.7	4.9	240	10	4.2	2.0	7.5

		H5N1_BY					H5N1_AD					H5N1_BD				
					95 % CI					95 % CI					95 % CI	
Symptom	Type	N	n	%	LL	UL	N	n	%	LL	UL	N	n	%	LL	UL
Overall/subject																
Ecchymosis (mm)	All	239	24	10.0	6.5	14.6	122	4	3.3	0.9	8.2	123	6	4.9	1.8	10.3
	> 50 mm	239	1	0.4	0.0	2.3	122	0	0.0	0.0	3.0	123	0	0.0	0.0	3.0
Induration (mm)	All	239	80	33.5	27.5	39.8	122	3	2.5	0.5	7.0	123	8	6.5	2.8	12.4
	> 50 mm	239	6	2.5	0.9	5.4	122	0	0.0	0.0	3.0	123	0	0.0	0.0	3.0
Pain	All	239	213	89.1	84.5	92.8	122	26	21.3	14.4	29.6	123	36	29.3	21.4	38.1
	Grade 3	239	14	5.9	3.2	9.6	122	0	0.0	0.0	3.0	123	0	0.0	0.0	3.0
Redness (mm)	All	239	77	32.2	26.3	38.5	122	27	22.1	15.1	30.5	123	21	17.1	10.9	24.9
	> 50 mm	239	1	0.4	0.0	2.3	122	0	0.0	0.0	3.0	123	0	0.0	0.0	3.0
Swelling (mm)	All	239	104	43.5	37.1	50.1	122	8	6.6	2.9	12.5	123	15	12.2	7.0	19.3
	> 50 mm	239	14	5.9	3.2	9.6	122	0	0.0	0.0	3.0	123	0	0.0	0.0	3.0

For solicited general symptoms myalgia and fatigue were reported frequently by subjects in the adjuvanted vaccine groups (myalgia in > 60% in adjuvanted groups but 20-30% in non-adjuvanted; fatigue in 50-60% compared to 30-40%).

Unsolicited symptoms were reported by 383 subjects with rates across all six groups in the range 28-35%. The most frequently reported unsolicited symptoms tended to be associated with likely

intercurrent illnesses affecting the respiratory and gastrointestinal tracts and so were very varied in nature.

- Serious adverse event/deaths/other significant events

In **study H5N1-007** there were no deaths or non-fatal SAEs up to D51. From D51 to D180 seven subjects reported a SAE, ranging from 0-2 per dose group but none was considered to be related to vaccination.
In **study H5N1-008** there were 11/3802 (0.3%) subjects in the 15µg/AS03 vaccine group and 6/1269 (0.5%) in the Fluarix/placebo group who reported SAEs. All SAEs were considered as not related to vaccination by the investigator and all resolved. There were no deaths.
Between D51 and D180 3.8% of those aged > 60 years in the 15µg/AS03 group reported a SAE compared to 3.9% in this age group from the Fluarix group. Rates in subjects aged 18-60 years were 0.8% and 0.9% in the two vaccine groups. There was no discernible pattern in SAEs observed. In the 15µg/AS03 vaccine group none of the SAEs seemed likely to be in any way related to vaccination based on nature or date of onset.
In **study H5N1-002** there were seven subjects who reported SAEs during the study. Review of these SAEs indicates that none was related to vaccine. One subject died but this was unrelated to vaccine.

- Safety in special populations

There is no experience in children. For further information, see SPC section 5.1.

- Safety related to drug-drug interactions and other interactions

There are no data on co-administration of Pandemrix with other vaccines.

- Discontinuation due to adverse events

There were no discontinuations due to AEs in study 007.
In 008 three subjects who received 15µg/AS03 vaccine dropped out due to SAEs assessed as not related to vaccination. In addition, 22 subjects reported non serious AEs that led to premature discontinuation from the study. Of these 21 were from the 15µg/AS03 group (9 by day 21) and there was only one withdrawal from the Fluarix group (by Day 42). After review the applicant concluded that there was an excess of AEs associated with withdrawal in the adjuvanted vaccine group but no pattern of AEs associated with withdrawals could be discerned.
In 002 the only early discontinuation was due to the death that was unrelated to vaccine.

- Post marketing experience

There is no post-marketing experience at present.

2.5. Pharmacovigilance

Detailed description of the Pharmacovigilance system

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

Risk Management Plan

The MAA submitted a risk management plan that was drafted in accordance with the CHMP core RMP for vaccines intended for use in a declared pandemic situation. .

The CHMP, having considered the data submitted in the application was of the opinion that it was not appropriate to consider risk minimisation activities at this time.

2.6. Overall conclusions, risk/benefit assessment and recommendation

Quality

The manufacture of the H5N1 antigen, the H5N1 formulated vial and the AS03 (adjuvant) vial are appropriately controlled. Adequate release and shelf life specifications have been set. Commitments are made by the applicant to update some missing information, which does not impact on the risk/benefit assessment of this vaccine.

Non-clinical pharmacology and toxicology

The ability to induce protection against homologous and heterologous vaccine strains was assessed non-clinically using ferret challenge models.
Of animals receiving adjuvanted vaccine 87% and 96% were protected against the lethal homologous or heterologous challenge, respectively. Viral shedding into the upper respiratory tract was also reduced in vaccinated animals relative to controls, suggesting a reduced risk of viral transmission. In the unadjuvanted control group, as well as in the adjuvant control group, all animals died or had to be euthanized as they were moribund, three to four days after the start of challenge.

Non-clinical safety data reveal no special hazard for humans based on conventional studies of safety pharmacology, acute and repeated dose toxicity, local tolerance, embryo-foetal and postnatal toxicity (up to the end of the lactation period).

Efficacy

Clinical trials on protective efficacy for the mock-up vaccine are not possible and so a detailed characterisation of the immunological response has been performed.

Study 007 supported the choice of an adjuvanted formulation and the use of the lowest tested dose of HA antigen for further assessment. The immunogenicity data in subjects aged 18-60 years were similar between studies 007 (using vaccine from a preliminary production process) and 002 (using vaccine from the final production process). The CHMP criteria for HI responses are fulfilled following administration of two doses of Pandemrix administered 21 days apart to subjects aged 18-60 years. Based on the data from study 008 and the lack of a significant effect of dose on responses to the adjuvanted formulations tested in 007 it is anticipated that responses in subjects aged > 60 years would be at least as good as those in younger subjects. The NA data are supportive of the HI data.

The available data at D180 indicate as expected a waning of antibody titres. However, more than half were still seroprotected based on HI titres and almost all were seropositive based on NA.

The influenza-specific T-cell data are supportive of the immunogenicity of the vaccine.

Antibody and T-cell responses to clade 2 strains were lower than to the vaccine strain. In any case this application concerns a vaccine into which an appropriate final pandemic strain will be inserted by variation.

Safety

In study 007 addition of the AS03 adjuvant clearly increased the reactogenicity of the various HA doses tested but there was no significant effect of the antigen dose on the overall incidence of symptoms among the adjuvanted vaccines.

Across the studies pain at the injection site was the most frequently reported local symptom. There was also a trend for higher incidences of swelling and redness in groups with adjuvanted vaccines and the rate of induration was significantly higher in adjuvanted groups. However, severe swelling, redness and induration were all reported at low rates. Regional lymphadenopathy may also occur.
The most frequently reported general symptoms were fatigue and headache.

All SAEs were considered as not related to vaccination by the investigator. There was one unrelated death across all studies.

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

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 has committed to perform a further round of user testing with the final approved PL in the post-authorisation period.

Risk-benefit assessment

Clinical context

It is not known which strain (in terms of H and N type) will trigger the next human influenza pandemic.Pandemrix is a mock-up influenza vaccine, whose scientific development is based on the guideline on dossier structure and content for pandemic influenza vaccine marketing authorisation application (CPMP/VEG/4717/03) and the guideline on submission of marketing authorisation applications for pandemic influenza vaccines through the centralised procedure (CPMP/VEG/4986/03). .

Benefits

The benefit of Pandemrix can only be assessed during a pandemic and following insertion of an appropriate final pandemic strain into the vaccine. At present the potential benefit can only be evaluated based on detailed characterisation of immunological responses to vaccination.

After two doses administered 21 days apart the three CHMP criteria as laid down in CPMP/VEG/4717/03 have been fulfilled. Therefore the expected benefit of Pandemrix is to provide some protection against clinically-apparent infection and/or possibly against development of severe disease in case of an influenza pandemic. It is highly unlikely that Pandemrix containing the antigens from the strain derived from A/Vietnam /1194/2004 would provide adequate protection if used during a pandemic. In line with the developed core dossier concept, a variation would therefore have to be submitted to introduce the WHO/EU recommended strain, prepared from the influenza virus causing the pandemic, prior to use of Pandemrix.

Risks

Pandemrix is commonly or very commonly associated with a range of local and systemic adverse reactions but these are not often of severe intensity and the safety profile would not preclude the use of the vaccine in healthy adults aged 18-60 years or > 60 years.

The current safety database is considered to be sufficient to describe adverse reactions that occur uncommonly and to give an indication of any rare events. However, there are some adverse reactions known to be very rarely associated with influenza vaccines and it is currently not possible to predict if higher rates might be observed with Pandemrix compared with, for example, seasonal influenza vaccines.

Balance

The overall B/R of Pandemrix is positive.

A risk management plan was submitted in accordance with the CHMP-recommended core RMP for these types of vaccines when intended only for use during an actual pandemic.

Recommendation

Based on the CHMP review of data on quality, safety and efficacy, the CHMP considered by consensus that the risk-benefit balance of Pandemrix for the prophylaxis of influenza in an officially declared pandemic situation, in accordance with official guidance, was favourable and therefore recommended the granting of the marketing authorisation under exceptional circumstances.