10 December 2020
EMA/1767/2021
Committee for Medicinal Products for Human Use (CHMP)

Assessment report

Heplisav B

Common name: Hepatitis B vaccine (recombinant, adjuvanted)

Procedure No. EMEA/H/C/005063/0000

Note

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

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List of abbreviations

AE	Adverse event
AESI	Adverse event of special interest
ALT	Alanine Aminotransferase
Anti-ssDNA	Antibody to single-stranded DNA
Anti-dsDNA	Antibody to double stranded DNA
APTT	Activated Partial Tromboplastin Time
AUC	Area under curve
BMI	Body mass index
BUN	Blood urea nitrogen
CHMP	Committee for Medicinal Product for Human use
CI	Confidence Interval
CKD	Chronic kidney disease
CPV	Continuous Process Verification
ECi	Enhanced chemiluminescence immunoassay
EDTA	Ethylenediaminetetraacetic acid
ELISA	Enzyme-Linked Immunosorbent Assay
EMA	European Medicines Agency
EOP	End-of-production
ESRD	End-stage renal disease
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GFR	Glomerular filtration rate
GMC	Geometric mean antibody concentration
GMP	Good manufacturing practice
HBsAg	Hepatitis B surface antigen
HBV	Hepatitis B virus
HCC	Hepatocellular carcinoma
INF α	Interferon alpha
IPC	In-process control
IPS	In-process specification
ISS	Immunostimulatory sequence
IV	Intravenous
IVRP	In vitro relative potency
LC-MS	reversed-phase high-performance liquid chromatography with tandem ultraviolet/mass spectroscopy detection
LIM	Liquid injection moulded
MAA	Marketing authorisation application
MACE	Major adverse cardiovascular event
MAE	Medically attended adverse event
MCB	Master Cell Bank
MI	Myocardial infarction
MIA	Manufacturing and Import Authorisation
mIU	milli international units
mITT	modified Intent-to-Treat Population

NOAEL	No observed adverse effect level
NOR	Normal Operating Range
PAMP	Pathogen-associated molecular pattern
PAR	Proven Acceptance Range
PBMCs	Peripheral blood mononuclear cell
PD	Pharmacodynamic
pDC	Plasmacytoid dendritic cells
PEC	Process Evaluation Criteria
PEG	Polyethylene glycol
PFS	Pre-filled syringe
PIR	Post-injection reactions
PK	Pharmacokinetic
PO	Phosphodiester
PP	Per protocol population
PPQ	Process Performance Qualification
PS ODN	Phosphorothioate oligodexoynucleotide
PSP	Primary safety population
RH	Relative Humidity
RP-HPLC	Reversed-Phase High Performance Liquid Chromatography
RR	Relative Risk
QA	Quality Assurance
QC	Quality Control
SAE	Serious adverse event
SDS-PAGE	sodium dodecyl sulfate polyacrylamide gel electrophoresis
SC	Subcutaneous
SPR	Seroprotection rate
THS	Tolosa Hunt syndrome
TLR 9	Toll-like receptor 9
TK	Toxicokinetic
TSE	Transmissible Spongiform Encephalopathy
TSP	Total safety population
UC	Ultracentrifugation
UK	United Kingdom
WCB	Working Cell Bank
WHO	World Health Organisation

1. Background information on the procedure

1.1. Submission of the dossier

The applicant Dynavax GmbH submitted on 11 March 2019 an application for marketing authorisation to the European Medicines Agency (EMA) for Heplisav B, through the centralised procedure falling within the Article 3(1) and point 1 of Annex of Regulation (EC) No 726/2004.

The applicant applied for the following indication:

"Heplisav B is indicated for prevention of infection caused by all known subtypes of hepatitis B virus in adults 18 years of age and older."

The legal basis for this application refers to:

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

The application submitted is 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 tests or studies.

Information on Paediatric requirements

Pursuant to Article 7 of Regulation (EC) No 1901/2006, the application included an EMA Decision P/0051/2012 on the agreement of a paediatric investigation plan (PIP).

At the time of submission of the application, the PIP P/0051/2012 was not yet completed as some measures were deferred.

Information relating to orphan market exclusivity

Similarity

Pursuant to Article 8 of Regulation (EC) No. 141/2000 and Article 3 of Commission Regulation (EC) No 847/2000, the applicant did not submit a critical report addressing the possible similarity with authorised orphan medicinal products because there is no authorised orphan medicinal product for a condition related to the proposed indication.

Applicant's request(s) for consideration

New active Substance status

The applicant indicated the active substance hepatitis B surface antigen contained in the above medicinal product to be considered as a known active substance.

Scientific advice

The applicant received the following Scientific advice on the development relevant for the indication subject to the present application:

Date	Reference	SAWP co-ordinators
29 May 2009	EMEA/H/SA/1267/1/2009/SME/III	Dr Jan Mueller-Berghaus, Dr Hans Ovelgönne
19 May 2011	EMEA/H/SA/1267/1/FU/1/2011/II	Dr Jan Mueller-Berghaus, Prof. Dieter

	Corrigendum	Deforce
18 October 2018	EMEA/H/SA/1267/2/2018/SME/II	Dr Jens Reinhardt, Dr Filip Josephson

The Scientific advice pertained to the following clinical aspects:

- Design of phase 3 Study HBV-16 in terms of endpoints, safety assessments, inclusion/exclusion criteria, sample size and statistical analysis plan
- Analysis populations to assess safety
- Assessment of risk of Immune-mediated adverse events and major adverse cardiovascular events (MACE) based on phase 3 clinical studies and the post-marketing vaccine safety surveillance study
- Evidence base for approval and indication statements

1.2. Steps taken for the assessment of the product

The Rapporteur and Co-Rapporteur appointed by the CHMP were:

Rapporteur: Filip Josephson Co-Rapporteur: Christophe Focke

For the appointed rapporteur it was considered exceptionally justified that the individual had previously been acting as coordinator for Scientific advice on the development relevant for the indication subject to the present application. The justification was as follows:

Dr Filip Josephson is a key regulatory expert on vaccines. His involvement as rapporteur, while acting as coordinator in a previous advice, can be exceptionally justified based on his unique expertise.

The appointed co-rapporteur had no such prominent role in Scientific advice relevant for the indication subject to the present application.

The application was received by the EMA on	11 March 2019
The procedure started on	28 March 2019
The Rapporteur's first Assessment Report was circulated to all CHMP members on	18 June 2019
The Co-Rapporteur's first Assessment Report was circulated to all CHMP members on	17 June 2019
The PRAC Rapporteur's first Assessment Report was circulated to all PRAC members on	1 July 2019
The CHMP agreed on the consolidated List of Questions to be sent to the applicant during the meeting on	25 July 2019
The applicant submitted the responses to the CHMP consolidated List of Questions on	16 August 2020
The following GCP inspection was requested by the CHMP and their outcome taken into consideration as part of the Quality/Safety/Efficacy assessment of the product:	

A GCP inspection of study HBV-23, at two investigator sites and at the sponsor site, all located in USA, between 13 August and 2 October 2019. The outcome of the inspection carried out was issued on 13 November 2019.	13 November 2019
The Rapporteurs circulated the Joint Assessment Report on the responses to the List of Questions to all CHMP members on	21 September 2020
The PRAC agreed on the PRAC Assessment Overview and Advice to CHMP during the meeting on	01 October 2020
The CHMP agreed on a list of outstanding issues in writing to be sent to the applicant on	15 October 2020
The applicant submitted the responses to the CHMP List of Outstanding Issues on	06 November 2020
The Rapporteurs circulated the Joint Assessment Report on the responses to the List of Outstanding Issues to all CHMP members on	25 November 2020
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 to Heplisav B on	10 December 2020

During the assessment of this application, a revised timetable had been adopted by the CHMP accounting for a delay from the initially planned timetable due to unforeseeable reasons related to the COVID-19 pandemic. This was done in line with the European Medicines Regulatory Network COVID-19 Business Continuity Plan (EMRN COVID-19 BCP) which describes mitigation measures in case of COVID-19 related delays.

2. Scientific discussion

2.1. Problem statement

2.1.1. Disease or condition

Hepatitis B infection is caused by hepatitis B virus (HBV), a dsDNA hepadna virus. HBV infection causes a broad spectrum of disease from subclinical self-limiting infections to fulminant hepatitis, and some individuals develop chronic hepatitis B infection. Primary HBV infection in susceptible individuals can be either symptomatic or asymptomatic, the latter being often the case. Hepatitis B surface antigen (HBsAg) is the earliest marker of hepatitis B infection, and is widely used in seroprevalence surveys to estimate the number of infected people and as indicator of transmission risk. Although most primary acute infections are self-limiting, their case fatality rate is 0.5–1%. Chronic hepatitis B infection can lead to chronic liver disease and death from liver cirrhosis or hepatocellular carcinoma. Infants infected at birth have the highest risk of developing chronic infection, and the lowest risk of symptoms of acute hepatitis B, while the opposite is true

for older children and adults. The risk of developing chronic HBV infection is dependent on age of infection, and for infants infected during the first year of life, it is estimated at 80–90%.

2.1.2. Epidemiology and risk factors

Disease caused by HBV has a worldwide distribution. HBV is widely prevalent, and it is estimated that approximately one third of the world's population has been exposed to the virus, with 250 million people chronically infected. Every year, more than 780,000 people die worldwide due to complications of hepatitis B, mostly from cirrhosis and liver cancer.

The endemicity of active HBV infection is reflected in the serologic prevalence of the hepatitis B surface antigen (HBsAg) in the general population of a defined geographical area. HBsAg prevalence of ≥8% defines highly endemic areas, prevalence of 5%–7% defines high intermediate, 2%–4% low intermediate, and <2% defines low endemic areas.

The WHO European Region is considered to be an intermediate endemicity region with an HBsAg prevalence of 1.6% ranging from < 0.1% in the United Kingdom (UK) to 10.3% in Kyrgyzstan. Surveillance data suggest a downward trend in the acute HBV infections during 2006–2014 in many EU countries, which is most likely due to the impact of widespread implementation of vaccination programmes.

The major antigenic determinant of the viral envelope is the hepatitis B surface antigen (HBsAg), a 226-amino acid protein. Antibodies directed against a determinant of the hepatitis B surface antigen (anti-HBsAg) confer protection against HBV infection.

Hepatitis B transmission occurs mainly through exposure to infected blood or other body fluids. The main transmission routes are perinatal infection from hepatitis B infected mothers, non-sexual or sexual person to person transmission or percutaneous exposure to infected body fluids. Among adults a number of risk factors for contracting hepatitis B infection have been described: multiple sexual contacts, close family contacts, haemodialysis patients (chronic kidney disease, injecting drugs, and occupational risk of exposure (e.g. health care workers). Persons with diabetes mellitus are at risk of acquiring infection by bloodborne pathogens such as HBV, due to lack of adherence to standard infection control precautions (e.g. inadequate disinfection and cleaning of blood glucose monitors between patients) and failure to implement recommendations against sharing finger-stick devices put diabetes patients (e.g. multi-patient use of finger-stick devices designed for single-patient use). This risk might decline with the introduction of blood glucose monitors without finger pricks.

2.1.3. Management

No specific antiviral treatment is recommended for patients with acute hepatitis B, as approximately 95% of infected immunocompetent adults recover spontaneously with anti-HBs seroconversion. Consequently, supportive care is the mainstay of therapy. Antiviral treatment may, however, be considered in patients with severe acute or fulminant hepatitis B. Antiviral treatment is generally recommended for chronically infected patients on the basis of the presence of active disease (i.e., ALT levels more than twice the upper limit of normal), clinical or histologic evidence of progressive disease and fibrosis, or both. The primary goal of treatment is to minimize progression of liver injury and fibrosis by suppressing viral replication.

Current Immunization Approaches

The main goal of hepatitis B vaccination is to reduce the incidence of chronic hepatitis B, which is mainly achieved through vaccination of infants, with catch-up programs in previously vaccinated older children and adults.

Vaccines against HBV induce antibodies to hepatitis B surface antigen (anti-HBs). An anti-HBs level of greater than or equal to 10 mIU/mL has been shown to correlate with protection against HBV infection. Healthy individuals who develop anti-HBs concentrations greater than or equal to 10 mIU/mL after vaccination are protected against symptomatic HBV infection for decades. Healthy individuals whose anti-HBs concentrations decline to less than 10 mIU/mL typically have strong anamnestic responses to a booster dose of hepatitis B vaccine, indicating persistence of protection against HBV infection.

Among individuals 40 years of age and older, the proportion of individuals who achieve seroprotection after a 3 dose regimen of the currently licensed hepatitis B vaccines declines below 90%, and by age 60, seroprotection develops in only 70% to 75% of those vaccinated. This reduced rate of seroprotection in older adults observed with the current hepatitis B vaccines is likely to contribute to the occurrence of new HBV infections in older adults. In older adults, 45% to 59% of those infected with HBV will develop chronic hepatitis B infection, and up to 40% of chronically infected individuals will develop cirrhosis, liver failure, or hepatocellular carcinoma (HCC). Older adults have higher mortality than younger adults with a case fatality rate of 1.6% to 4.4% among those 40 years of age or older with symptomatic, acute HBV infection.

About the product

Heplisav B is a vaccine against hepatitis B, consisting of recombinant hepatitis B surface antigen (HBsAg) and the novel adjuvant 1018 ISS, which is a synthetic oligodeoxynucleotide (ODN) including CpG motifs. CpG motifs contain an unmethylated cytosine phosphoguanosine (CpG) dinucleotide. The desired biological activity of 1018 ISS adjuvant is to stimulate the natural immune response to an infectious agent by activating the innate immune system via Toll-like receptor 9 (TLR9), an intracellular, pathogen-associated molecular pattern (PAMP)-recognition receptor.

The aim of the clinical development programme for Heplisav was to develop a hepatitis B vaccine with safety and tolerability similar to the currently licensed vaccines, which induces superior peak seroprotection and antibody concentrations, earlier seroprotection, and requires fewer doses than currently licensed hepatitis B vaccines.

Of note, a marketing authorisation application for Heplisav was initially submitted on 20 July 2012. Based on the review of the data, the CHMP had had some concerns and was of the provisional opinion that Heplisav could not have been approved for the prevention of hepatitis B. The marketing authorisation application was therefore withdrawn at that time.

The Committee considered that the way in which the study in patients with kidney disease had been carried out and documented was not satisfactory. This followed an inspection of some of the sites involved in the study, to ensure GCP standards for medicines studies had been followed. The nature of the findings from the inspection also raised questions about the other main studies. Therefore, there were serious uncertainties at that point about the reliability of the data submitted in support of the application. Furthermore, the number of patients in whom the safety of the medicine had been tested was insufficient to rule out an unacceptable level of risk for less common but serious side effects.

Therefore, at the time of the withdrawal, the CHMP was of the opinion that the medicine could not have been approved based on the data presented by the company.

2.2. Quality aspects

2.2.1. Introduction

The finished product is presented as a sterile, preservative-free solution that is administered as an intramuscular injection. The product is clear to slightly opalescent, colourless to slightly yellow and essentially free of visible particles. An administered dose of 0.5 mL Heplisav B contains 20 µg of HBsAg active substance (AS) and 3000 mcg of Dynavax's toll-like receptor 9 agonist adjuvant, cytidine phosphoguanosine immunostimulatory sequence (ISS) 1018 (a 22-mer sequence oligonucleotide) which is a novel excipient.

Other ingredients are: sodium chloride, disodium phosphate dodecahydrate, sodium dihydrogen phosphate dihydrate, polysorbate 80, and water for injections.

The product is available as a 0.5 ml of solution in a 1 mL prefilled syringe (Type I glass) with tip cap (synthetic isoprene-bromobutyl rubber blend) and plunger stopper (chlorobutyl rubber). Syringes are provided without needles in packages of 5 syringes.

2.2.2. Active substance

General information

The hepatitis B surface antigen (HBsAg) component of Heplisav B is a recombinant protein of known sequence produced in *Hansenula polymorpha* yeast and encoded by the S region of the HBV genome. The purified recombinant protein and associated lipids form a particle containing the subtype of the HBsAg. These lipoprotein particles resemble natural HBsAg-containing particles. The desired biological activity of HBsAg is to generate antibodies to the alpha-determinant.

HBsAg is purified as globular protein/lipid particles. Several analytical methods have confirmed the average particle size. The lipid component of the particles is host cell derived and consists mainly of fatty acids and phospholipids, such as phosphatidylserine, phosphatidylethanolamine, phosphatidylcholine, and lysophosphatidylcholine. The HBsAg protein monomer consists of 226 amino acids and has a theoretical molecular weight of 25.4 kDa. The amino acid sequence is identified as the adw2 subtype.

The active substance is classified as a known active substance.

Manufacture, process controls and characterisation

The active substance manufacturing, testing, and storage sites all have suitable GMP authorisations. Primary manufacturing occurs at Dynavax GmbH, Duesseldorf, Germany.

Description of manufacturing process and process controls

The HBsAg active substance manufacturing process has been adequately described. The batch scale is targeted to a specified final working volume in the main fermentor, but may vary within predetermined ranges depending on the range limits for media stock solutions and media additives, and according to the amounts of solutions used during the phases of the upstream manufacturing process. The manufacturing process is divided into Upstream Manufacturing, Downstream Manufacturing, and Filtration and Filling. The Upstream Manufacturing process includes the following steps: Seed Fermentation, Main Fermentation, Cell Recovery and Disruption, PEG Precipitation, Aerosil Treatment, and Aerosil Desorption. The Downstream Manufacturing process includes the following steps: Ion Exchange Chromatography, Concentration and Ultracentrifugation, and Gel Filtration Chromatography. The Filtration and Filling process adjusts the product concentration by ultrafiltration and filter sterilizes the HBsAg active substance into containers. After filling, the HBsAg active substance is stored at 2°C to 8°C.

Reprocessing is not claimed at any stage of the active substance process.

The ranges of critical process parameters and the routine in-process controls along with acceptance criteria, including controls for microbial purity and endotoxin, are described for each step. The active substance manufacturing process is considered acceptable.

Control of materials

Sufficient information on raw materials, resins, filters, membranes, and container and closures used in the active substance manufacturing process has been submitted. Compendial raw materials are tested in accordance with the corresponding monograph, while specifications (including test methods) for non-compendial raw materials are presented. The only animal-derived raw material is deoxycholic acid sodium salt, which is a non-compendial raw material manufactured from bovine bile sourced from healthy animals from countries considered to have a negligible BSE risk.

Sufficient information regarding development genetics has been given. The producer cell line and the establishment of the master cell bank (MCB)/working cell bank (WCB), including future WCBs, is described. The immunogenic component, HBsAg, is produced in a recombinant *Hansenula polymorpha* yeast strain. The coding sequence for human HBsAg (subtype adw) was isolated from the serum of an asymptomatic chronic carrier of hepatitis B virus. An HBsAg expression vector was used to transform *H. polymorpha* host strain RB11 and stable integrants (clones with the expression vector DNA integrated into the host genome) were selected. A suitable strain was selected and a master seed bank was produced and characterized.

To date, a MCB and three WCBs have been generated. All cell banks have been extensively tested and characterized (viability, microbial purity, plasmid copy number, plasmid identity, correct sequence, proof of mitotic stability, amount and quality of recombinant protein) and met the acceptance criteria to demonstrate that they are suitable for manufacturing HBsAg active substance. End-of-production (EOP) cells, derived from fermentation batches representative of the commercial manufacturing process using the WCBs, were also characterized. All cell banks have demonstrated mitotic stability beyond the limit of in vitro cell age used for production of HBsAg active substance. MCB and WCB stability has also been affirmed by analysis of viability and strain identity. Acceptance criteria for future WCBs have been presented. The cell bank inventory will be

stored in at least 2 separate locations, as the current WCB is. The stability of MCB is assessed by routine monitoring of the viability of the cells and checking strain identity.

Control of critical steps and intermediates

A comprehensive overview of critical in-process controls and critical in-process tests performed throughout the HBsAg active substance manufacturing process is given. Acceptable information has been provided on the control system in place to monitor and control the active substance manufacturing process with regard to critical, as well as non-critical operational parameters and in-process tests. Actions taken if limits are exceeded are specified.

Process validation

The HBsAg active substance manufacturing process has been validated adequately at production scale at the commercial manufacturing site. During the development of the HBsAg active substance manufacturing process, studies were performed to provide an understanding of the process and unit operations. Normal operating ranges (NORs) and proven acceptance ranges (PARs) for all parameters that could potentially impact product quality were established. Manufacturing support studies were performed to support the process validation program. The validation support studies include residual and impurity profiling, chromatography resin reuse studies, membrane reuse studies, hold-time studies, final filtration validation, and shipping validation. All analysed process residuals and impurities are removed as shown by intermediate product and active substance testing or controlled to acceptable or low levels during the manufacture of HBsAg active substance.

Consistency in production has been shown on batches manufactured with process 3 (the proposed commercial process of the 2012 application[1]). Data from release testing of three consecutive batches manufactured with process 4b (the proposed commercial process of Heplisav B active substance) were also presented. All acceptance criteria for the critical operational parameters and likewise acceptance criteria for the in-process tests are fulfilled demonstrating that the purification process consistently produces HBsAg active substance of reproducible quality that complies with the predetermined specification and in-process acceptance criteria.

Manufacturing process development

The development of HBsAg active substance has encompassed two production sites, Rhein Biotech GmbH (now called Dynavax GmbH, commercial active substance site) and a second site. Several manufacturing process changes were introduced. The process technology was developed at Rhein Biotech GmbH and then transferred to the second site for GMP production in order to produce the first clinical material. There were 4 process variations used for the manufacture of HBsAg active substance throughout development, designated as Process 1 through Process 4. The majority of the process changes have been associated with equipment changes and optimisation of the process to increase the purity of the HBsAg AS. The final process is referred to as Process 4b and is the current proposed commercial process for HBsAg.

Vaccine containing HBsAg manufactured with process 3 has been used in all phase III trials except one. HBsAg manufactured according to process 4b, the proposed commercial process, has not been used in any clinical trial. Sufficient comparability is shown between the HBsAg processes 1, 2 and 3. The presented data from release testing of active substance from processes 3 and 4b also supports comparability between these processes.

[1] A previous MAA for Heplisav-B was submitted in July 2012 by Dynavax but subsequently withdrawn in February 2014.

Characterisation

The HBsAg active substance has been sufficiently characterised by physicochemical and biological state-of-the-art methods in order to verify the primary and higher order structure and HBsAg particle composition and size. Primary structure was confirmed. The HBsAg lipid component was found to be principally composed of free fatty acids and phospholipids.

The immunodominant epitope in HBsAg is a conformational epitope called the alpha determinant. Antigenicity of the HBsAg has been determined by using an *in vitro* assay.

Characterisation data of the active substance is obtained using clinically relevant batches from process 2 and 3.

The ability of the active substance manufacturing process to remove process-related impurities was assessed using process-stream material collected during production of batches produced at commercial scale. Since the process used for the clinical material is highly similar to the commercial process, the clinical material is representative of the commercial vaccine in terms of impurity profile. As such, the levels of impurities are considered as clinically qualified. Active substance purity is also routinely tested.

In summary, the characterization is considered appropriate for this type of molecule.

Specification

The specification includes suitable physicochemical tests and appropriate tests for identity, purity and potency.

The HBsAg active substance release and stability specifications are defined based on manufacturing experience, pharmacopoeial standards, and statistical analysis of lot release and stability data. Justifications for the commercial HBsAg active substance release and stability specifications are provided. Initially, a major objection was raised regarding the active substance specifications, as these were not considered to fulfil the requirements for "purified antigen" included in the Ph. Eur. monograph "Hepatitis B Vaccine (rDNA). These issues were all resolved.

Analytical methods

The analytical methods used have been adequately described and (non-compendial methods) appropriately validated in accordance with ICH guidelines.

Batch analysis

Batch analysis data of the active substance were provided. Active substance batches were manufactured using the final commercial scale process (process 4B). The results are within the specifications in place at the time of release and confirm consistency of the manufacturing process.

Container closure system

The container closure system for HBsAg active substance is a transparent bottle with an injection moulded (LIM) closure insert that has 2 holes. A risk assessment was performed to evaluate the potential of packaging components used in the final HBsAg active substance container-closure system to contribute extractable or leachable compounds into the product that could impact patient safety, product quality, or stability. Extraction studies were performed to identify the analytes that could potentially be extracted from the media

bottle with the cap, the closure insert, and the cap alone. The container-closure system did not yield any extractables with either water or the HBsAg buffer system, it was concluded that no leachables studies were required and that the container-closure system is compatible with HBsAg active substance. Sufficient information regarding the container has been provided.

Stability

Real time, real condition stability data were provided on batches of active substance from the commercial manufacturing process stored in a container which is considered representative for the commercial container and data on batches stored under accelerated and stress conditions according to the ICH guidelines. Appropriate stability-indicating tests have been applied.

Photostability testing following the ICH guideline Q1B was performed. Given the results from this study and the duration and intensity of light, which HBsAg active substance is exposed to during the manufacturing process and storage, no additional precautionary measures to protect HBsAg active substance from light are warranted.

In summary, the stability results indicate that the active substance is sufficiently stable and justify the proposed shelf life in the proposed container when stored at the proposed storage conditions.

1018 Immunostimulatory sequence (ISS) adjuvant excipient

The 1018 ISS oligonucleotide is a 22-mer phosphorothioate molecule.

ISS Adjuvant contains an immunostimulatory sequence (unmethylated cytosine and phosphoguanosine [CpG]) recognized by Toll-like receptor 9, resulting in the activation of innate immune responses that subsequently amplifies the adaptive-immune response. The synthesis of the molecule is stereo-random. Accordingly, there are 2^{21} possible diasteromeric forms of the 1018 ISS oligonucleotide.

There is no INN for the molecule.

1018 ISS adjuvant manufacture, process controls and characterisation

The solid phase synthesis of the 1018 ISS Adjuvant is performed with a qualified solid phase deoxyribonucleic acid (DNA) synthesizer connected to a packed synthesis column. After freeze-drying, the 1018 powder is harvested and distributed into bottles, which are sealed with a bag sealer in tamper-evident foil pouches.

Compendial and non-compendial materials were described. Specifications were provided for the non-compendial materials including for each starting material. In addition, a risk assessment has been completed to assess the criticality of raw materials, components, resins, filters, membranes, and containers and their impact on the final quality of released 1018 ISS adjuvant.

Flow diagrams list the process parameters, in-process controls (IPCs), process evaluation criteria (PECs), and in-process specifications (IPSs) for each step in the upstream and downstream manufacturing processes. The process control strategy, which determined the NORs and PARs for the parameters that could potentially impact product quality, is described.

A series of manufacturing support studies was performed to support the PPQ. These studies evaluated: residual and impurity profiling; solution mixing studies, in-process (product) mixing studies; hold time studies; final filtration validation; shipping validation.

PPQ for 1018 was initially conducted at lower manufacturing scale and subsequently at routine commercial manufacturing scales. The PPQ results of three lots indicate that all unit operations and the entire process to manufacture 1018 are qualified to ensure the quality of the product produced. The steps performed in the manufacture of 1018 reproducibly generate product that consistently meets IPCs, IPSs and release specifications, and is of appropriate quality. A number of lots were manufactured and monitored in the CPV program.

The development of 1018 has encompassed various scales and manufacturing process changes at a single site. The majority of process changes have been associated with scale-up (equipment and batch size), raw materials, and improvements to unit operations methodology as part of a program to increase the purity and yield of 1018. Comparability studies have been performed demonstrating that material from all different processes is comparable and equivalent.

Analytical characterisation studies have been performed to determine the structure of the 1018 molecule. The chemical structure was established through a combination of state-of-the analytical techniques.

Process-related impurities are removed during the process as demonstrated during process validation studies. Product-related impurities of 1018 include synthesis failure sequences (deletion or addition of nucleotides), by-products from incomplete sulfurisation, depurination, n-1 terminal thio-monophosphates, covalent addition of acrylonitrile or chloral, and incomplete removal of protection groups. The toxicity profile of phosphorothioate oligonucleotides is largely defined by the chemical class, phosphorothioate, and is generally independent of sequence-related effects. Therefore, it is claimed that product-related impurities of 1018 are expected to have a comparable toxicological profile to 1018. Since the process used for the clinical material is highly similar to the commercial process, the clinical material is representative of the commercial vaccine in terms of impurity profile. As such, these levels of impurities are considered as clinically qualified.

1018 ISS adjuvant specification

During the evaluation further justification was requested for the specifications, in particular regarding the control of impurities. The levels of impurities, as justified by process capability, clinical use, toxicology studies and a theoretical discussion on the probable low risk of toxicity from the related substances, was accepted. The 1018 release specifications were sufficiently justified.

Presented production batches, including three PPQ lots, complied with the specifications in place at the time of release.

Several container-closure systems can be used based on the amount of 1018 to be transferred.

All containers are sealed in tamper-evident foil pouches prior to storage. 1018 is shipped to the fill-finish facility on dry ice. Data in support of the suitability of the container-closure system is provided and comprise microbial assessment, particulate assessment, stability data, as well as data to support compatibility (extractables and leachables assessment).

1018 ISS adjuvant stability

Stability studies for 1018 were performed according to ICH Q1A(R2). Stability data for 1018 lots manufactured at the commercial scale using the validated manufacturing process are included as primary stability data. All lots have completed the long-term stability studies, and lots have also completed stability studies at accelerated and stress conditions. All stability lots were tested for stability indicating 1018

attributes. For all primary stability lots no significant change in any of the tested attributes was observed after storage for the study duration. All results comply with the specifications.

In conclusion, the proposed re-test period is agreed.

2.2.3. Finished medicinal product

Description of the product and Pharmaceutical development

Heplisav B pre-filled syringe (PFS) is a sterile, preservative-free solution that is administered as an intramuscular injection. The product is clear to slightly opalescent, colourless to slightly yellow and essentially free of visible particles.

Table 1: Composition of finished productIngredient	Function	Quantity per mL	Quantity per dose (0,5 mL)
HBsAg active substance	Active	40 µg	20 µg
1018	Adjuvant	6000 µg	3000 µg
Disodium phosphate dodecahydrate	Buffer		
Sodium dihydrogen phosphate dihydrate	Buffer		
Polysorbate 80	Surfactant		
Sodium chloride	Isotonicity		
Water for injection (QS)	Diluent		

QS = quantum sufficit

All excipients are tested per compendial (Ph. Eur.) requirements except for the 1018 ISS adjuvant. All excipients are from non-human or non-animal sources. Apart from the 1018 ISS adjuvant (see 1018 ISS adjuvant section) there are no novel excipients.

The primary packaging is a 1 mL prefilled syringe (Type I glass) with tip cap (synthetic isoprene-bromobutyl rubber blend) and plunger stopper (chlorobutyl rubber). Syringes are provided without needles in packages of 5 syringes. The tip caps and stoppers of the prefilled syringes do not contain natural rubber latex. The material complies with Ph. Eur. and EC requirements. The choice of the container closure system has been validated by stability data and is adequate for the intended use of the product.

The finished product contains a slight overfill but no overage.

Heplisav B was initially developed as a vial presentation and subsequently developed as a PFS presentation. The PFS presentation is the intended commercial process. The development of the Heplisav B formulation matrix was performed for the vial presentation and applies to the PFS presentation as there was no change in dose composition with the introduction of the PFS.

Formulation process development performed in the transition to the PFS presentation included the removal of the active substance overage in the formulation, change in scale, change of contract manufacturing

organisation and the implementation of a streamlined formulation process. Development studies and formal comparability studies demonstrated no impact on the strength, identity, safety, purity or quality of the product as a result of these improvements.

During the manufacturing history for Heplisav B, three similar but distinct formulation matrices for the finished product were developed that are referred to as formulations 1, 2, and 3. The three different HBsAg active substance formulation matrices used by the three different HBsAg manufacturers differ only slightly in composition. Formulation 3 (vial presentation) was used in all phase III clinical trials except one, and also in one phase I and two phase II studies.

The PFS presentation has not been used in any clinical trials presented in the MA dossier. However, there are no major differences in the manufacturing methods between the vial and PFS finished products. Sufficient comparability between the vial and PFS presentations has been shown. Thus, the lack of clinical experience of the PFS presentation is acceptable.

Manufacture of the product and process controls

Heplisav B is tested and released by Dynavax GmbH (a wholly owned subsidiary of Dynavax Technologies Corporation) Eichsfelder Strasse 11, 40595 Düsseldorf Germany.

Initially, several issues related to the MIA and GMP status of the finished product manufacturing/ testing sites were raised. All issues were resolved and all sites for finished product manufacture and testing carry suitable GMP authorisations and appropriate Manufacturing and Import Authorisation (MIA) are available.

The finished product manufacturing process has been adequately described and comprises the preparation of a buffer solution and a polysorbate 80 stock solution. The required amount of 1018 and HbsAg are mixed and formulation buffer is added to the target batch weight. The solution is mixed to ensure homogeneity. The formulated bulk is filtered using a 0.22 micron sterilizing-grade filters (initial and second filtration) into a glass receiving vessel, which is connected to the syringe filling and stoppering machine in the Grade A filling area. After stoppering and inspection, the plunger rod is inserted prior to syringe labelling and backstop mounting. The finished product is then packaged and labelled and stored at 5°C ± 3°C.

Sufficient explanation for the classification of the process parameters as critical or non-critical has been provided, and the proposed ranges for process parameters have been sufficiently justified.

The manufacturing process has been validated. It has been demonstrated that the manufacturing process is capable of producing the finished product of intended quality in a reproducible manner. The in-process controls are adequate. Maximum process times and temperature conditions for the formulation process, hold times, mixing steps and filling homogeneity were properly validated. The bioburden retention capacity of the 0.22-micron sterilizing-grade filters has been validated. The syringe filling line was successfully validated for aseptic filling operations by a media fill PQ. Routine validation confirmations are performed semi-annually via routine media fills. Shipping of finished product has not been validated yet, and the applicant is requested to provide the shipping validation data (Performance Qualification) for the finished product. In case these data are not immediately available, it is acceptable to provide these post-approval, but in any case before launch of the product in the EU (see recommendation).

The PPQ for the Heplisav B manufacturing process at the formulated bulk batch scale was successfully demonstrated using three consecutive batches. The results indicate that all unit operations performed during Heplisav B manufacturing are validated to ensure the safety, efficacy, and quality of the product. The steps

performed in the manufacture of Heplisav B reproducibly operates within the ranges of the CPPs and consistently meets IPCs, IPACs, as well as the release specification, and is therefore of appropriate quality. All deviations during the execution of PPQ batches have been assessed as not to impact product quality and not to affect the conclusion that the process is validated. Impurities in the finished product are discussed in the active substance and 1018 ISS adjuvant sections and respective impurity levels present in product were studied in clinical trials. No new impurities are added during finished product manufacture.

A risk evaluation concerning the presence of nitrosamine impurities in the finished product has been performed (as requested) considering all suspected and actual root causes in line with the "Questions and answers for marketing authorisation holders/applicants on the CHMP Opinion for the Article 5(3) of Regulation (EC) No 726/2004 referral on nitrosamine impurities in human medicinal products" (EMA/409815/2020) and the "Assessment report- Procedure under Article 5(3) of Regulation EC (No) 726/2004- Nitrosamine impurities in human medicinal products" (EMA/369136/2020). Based on the information provided it is accepted that no risk was identified on the possible presence of nitrosamine impurities in the active substance or the related finished product. Therefore, no additional control measures are deemed necessary.

Product specification

The finished product specifications specification includes appearance, HBsAg Identity and size, potency, adjuvant identity, integrity and content, particulate contamination, extractable volume, pH, endotoxin and sterility.

During the evaluation, further justification was requested on the proposed specifications, in particular regarding the inclusion of a test for antigenicity. The vaccine complies with the relevant Ph. Eur. monograph (1056), and the requirement for Assay is covered by the Heplisav B lots and the results show that the manufacturing process consistently yields lots of comparable quality. Therefore, the omission of a test for antigenicity was accepted. Some specifications were tightened upon request and a release specification for particle size was introduced.

The potential presence of elemental impurities in the finished product has been assessed on a risk-based approach in line with the ICH Q3D Guideline for Elemental Impurities. It was confirmed that the risk for any elemental impurities being present in the final vaccine is negligible.

Analytical methods

The analytical methods used have been adequately described and (non-compendial methods) appropriately validated in accordance with ICH guidelines.

The potency and HBsAg identity of Heplisav B is determined using a relative potency of the test article.

Batch analysis

Presented Heplisav B PFS batches have been manufactured using the commercial scale process, including three PPQ batches and two verification batches. All batches complied with the proposed commercial release specification. The results from the batch analyses indicate consistent quality of the finished product.

Reference materials

No compendial reference standard is available. An in-house reference standard has been developed and qualified.

Stability of the product

A shelf life of 3 years at 2°C to 8°C is proposed for the finished product. Stability studies for long-term, accelerated and photo-stability were performed to establish the stability profile of HEPLISAV-B according to ICH Guideline Q1A(R2).

Primary stability data from Heplisav B batches manufactured at commercial scale using the validated commercial manufacturing process are included in this application. All batches use the container and closure system proposed for commercial Heplisav B.

Based on the available stability data, the shelf-life of 3 years and storage conditions (i.e. stored in a refrigerator (2°C to 8°C)), as stated in the SmPC are acceptable.

Adventitious agents

All raw materials used in the manufacture of the MCB, WCB, and HBsAg active substance (with the exception of deoxycholic acid, sodium salt), are of non-animal origin, confirmed by supplier certificates.

The microbial host for production of the recombinant HBsAg protein is the methylotrophic yeast *Hansenula polymorpha*. Yeast cell lines are considered highly unlikely to be capable of propagation of adventitious mammalian viral contaminants or mycoplasma that might constitute a safety concern for the patient; therefore, in compliance with ICH guidelines Q5A and Q5D, viral clearance studies were not performed.

The only animal-derived raw material used directly in the manufacture of the HBsAg active substance is deoxycholic acid, sodium salt, which is a non-compendial raw material manufactured from bovine bile sourced from healthy animals. The animals are sourced from countries considered to have a negligible BSE risk. No animal-derived materials are used during the manufacture of the adjuvant 1018.

2.2.4. Discussion on chemical, and pharmaceutical aspects

Information on development, manufacture and control of the HBsAg active substance, the 1018 ISS adjuvant and Heplisav B vaccine finished product has been presented in a satisfactory manner. The results of tests carried out indicate consistency and uniformity of important product quality characteristics, and these in turn lead to the conclusion that the product should have a satisfactory and uniform performance in clinical use.

During the evaluation procedure, a number of major objections were raised, which could be resolved throughout the procedure, the details of which are summarized below.

Major objections were raised regarding the control of the active substance and specific process related impurities. Further information was also required about the interaction between the active substance and the ISS adjuvant.

Also, major objections were raised as regards the GMP status of manufacturing/testing facilities. With updated information, all sites for finished product manufacture and testing carry suitable GMP authorizations and appropriate Manufacturing and Import Authorization (MIA) is available.

At the time of the CHMP opinion, there were no unresolved quality issues having impact on the Benefit/Risk ratio of the product.

One recommendation has been agreed in relation to the provision of shipping validation data (Performance Qualification) for the finished product upon availability.

2.2.5. Conclusions on the chemical, pharmaceutical and biological aspects

The quality of this product is considered acceptable when used in accordance with the conditions defined in the SmPC. Physicochemical and biological aspects relevant to the uniform clinical performance of the product have been investigated and are controlled in a satisfactory way. Data have been presented to give reassurance on viral/TSE safety.

2.2.6. Recommendation(s) for future quality development

In the context of the obligation of the MAHs to take due account of technical and scientific progress, the CHMP recommends the following points for investigation:

1. The applicant is requested to provide as a post-approval measure the shipping validation data (Performance Qualification) for the finished product upon availability, but in any case, before launch of the product in the EU.

2.3. Non-clinical aspects

2.3.1. Introduction

Nonclinical pharmacological, pharmacokinetics (PK) and toxicity studies were undertaken on the vaccine, CpG 1018 + HBsAg (adjuvant and antigen) and CpG 1018 (adjuvant) alone.

2.3.2. Pharmacology

Primary pharmacodynamic studies

Immunogenicity of the 1018 ISS adjuvanted vaccine

The immunogenicity of HBsAg adjuvanted with 1018 ISS has been evaluated in a total of 7 different non-GLP studies. An overview of these studies is included in Table 8. The immunogenicity of HBsAg adjuvanted with 1018 ISS has been evaluated in mice, rat and baboons. These studies demonstrated that despite some known differences in TLR9 expression pattern between rodent and primate hematopoietic cells, the 1018 ISS adjuvanted vaccine is immunogenic in all three animal species. This provides support to the choice of mice, rat and monkeys (baboons and cynomolgus monkeys) for the safety studies.

The titres of anti-HBsAg have been directly compared between the vaccine candidate and a licensed comparator in the mice and baboon immunogenicity studies. The adjuvant has shown to provide higher

antibody responses to HBsAg, compared to HBsAg alone or HBsAg plus aluminium adjuvant. The anti-HBV antibody response after two administrations of the vaccine candidate was much higher in magnitude in comparison to injections with an equal dose of HBsAg alone or a licensed comparator (HBsAg antigen plus aluminium). This indicates that the 1018 ISS is a potent adjuvant.

The immunogenicity studies also demonstrated that the anti-HBV antibody response and more importantly the frequency of animals responding with antibody levels ≥10 mIU/ml (the defined threshold for seroprotection in humans) is depending on the 1018 ISS dose. In baboons 3000 µg of the 1018 ISS adjuvant was required for a 100% seroprotection rate. The optimal dose-ratio of antigen and adjuvant in baboons was 1/150, which also yielded a good response in rats. 3000 µg 1018 ISS and the 1/150 antigen/adjuvant ratio were subsequently selected for the clinical formulation.

An antibody isotype analysis in mice showed that HBsAg + 1018 ISS induced an antibody response dominated by IgG2a while HBsAg alone and Engerix-B induced IgG1 production. This indicates that 1018 ISS skews the immune response towards a more Th1 type of response.

Table 2: Nonclinical immunogenicity Studies of CpG 1018 + HBsAg and CpG 1018 Alone

Type of Study	Descriptive Study Title	Study Number
CpG 1018 + HBsAg Immunogenicity	Immunogenicity of CpG 1018 + HBsAg in Mice after 2 IM Injections Weeks 0 and 2 – Including Evaluation of Serum DNA Antibody 99-0086	99-0086
	Immunogenicity of CpG 1018 + HBsAg in Rats after 2 IM Injections Weeks 0 and 2	05-446
	Immunogenicity of CpG 1018 + HBsAg in Baboons after 2 IM Injections Weeks 0 and 8 – Including Evaluation of Serum DNA Antibody	99-0089
	Immunogenicity of CpG 1018 + HBsAg in Baboons after 2 IM Injections Weeks 0 and 8 – Dose Response of CpG 1018 ISS	00-104
	Immunogenicity of CpG 1018 + HBsAg in Baboons after 2 IM Injections Weeks 0 and 8 – Comparability of HBsAg adr and adw	03-322
	Immunogenicity of CpG 1018 + HBsAg in Baboons after 2 IM Injections Weeks 0 and 8 – Single-Vial Presentation	04-416
	Immunogenicity of CpG 1018 + HBsAg in Baboons after 2 IM Injections Weeks 0 and 4 – Comparability of HBsAg subtypes adr and adw from Different Manufacturing Facilities	06-505

<u>The immunostimulatory activity of the 1018 ISS adjuvant</u>

It is well known that CpG-oligodeoxynucleotides acts as adjuvants via activating TLR9. Therefore, additional studies were conducted in rats and mice to verify the TLR9 mediated immunostimulatory activity of 1018 ISS (CpG-oligodeoxynucleotide adjuvant). In vitro studies on human peripheral blood mononuclear cell (PMBCs) and purified B cells were also done to confirm that the adjuvant stimulates human immune cells. An overview

of the studies is included in Table 9. These studies demonstrated the induction of known down-stream targets of TLR9 activation such as induction of IL-12, IL6 and IFNα and the mitogenic activity of the adjuvant on human PBMCs and purified B-cells.

In an effort to address the potential of 1018 ISS to enhance autoimmunity, the applicant (as part of studies 99-0086 in mice and 99-0089 in baboons) analysed potential induction of anti-ssDNA and/or anti-dsDNA antibodies in these two species injected twice with 1018 ISS adjuvanted HBsAg.

In addition to the known function of TLR9 in hematopoietic cells, TLR9 has also been shown to be expressed in different non-haematopoietic cells such as cardiomyocytes (Boyd et al. 2006, Nishimura and Naito 2005). Although literature data are limited it appears as TLR9 could have a different role in immune cells and non-hematopoietic cells such as cardiomyocytes. The function in cardiomyocytes seems complex with activation of TLR9 by ODNs resulting in pro-inflammatory cytokine production and loss of contractility by cardiomyocytes, but also protection of heart tissue from injury and inflammation (induced by pressure overload, ischemic injury and trauma-haemorrhage) in mice and reduced stress tolerance in heart organ cultures. Data on similarities in expression pattern of TLR9 in non-hematopoietic cells between animals and humans is also sparse. However, given the known similarities in expression and function of TLR9 in the immune system of rats, mice, and in particular between non-human primates and humans, it seems reasonable to assume that the overall expression pattern is also similar. It is thus concluded that the chosen animal models were relevant for assessing the safety of the product. Importantly, if TLR9 would have a significant physiological role in non-haematopoietic cell that has not been covered in the repeated dose toxicity studies in rodents and monkeys, the effects of such an interaction would likely be limited considering the very low and transient (<8 h) systemic exposures to 1018 ISS observed in subjects at the proposed dose regiment of Heplisav B.

Lastly, no significant homologies of the 1018 ISS sequence were found in the human genome that would imply that the adjuvant could disturb function of any gene. This indicates a low potential for off-target effects and furthermore that 1018 is unlikely to induce site-directed mutagenesis, which has been implied as a potential risk associated with this class of compounds.

Table 3: Nonclinical pharmacology Studies of CpG 1018 + HBsAg and CpG 1018 Alone

Type of Study	Descriptive Study Title	Study Number
CpG 1018 Pharmacology (In Vivo)	Serum Cytokine Responses of Mice after a Single SC Injection with CpG 1018	99-0039
	Serum Cytokine Responses of Rats after a Single SC Injection with CpG 1018	04-365
CpG 1018 Pharmacology (In Vitro)	CpG 1018 Activity on Human PBMC In Vitro: Evaluation of Cell Proliferation and IL-6 Production	hPBMC-1
	CpG 1018 Activity on Human PBMC In Vitro: Evaluation of IFN-gamma and IFN-alpha Production	182/183
	CpG 1018 Activity on Human PBMC In Vitro: Evaluation of Expression of Interferon-gamma, Interferon-alpha and Interferon-alpha-Inducible Genes	194

	CpG 1018 Activity on Human B Cells In Vitro: Evaluation of Cell Proliferation	172/176/181/191
	CpG 1018 Activity on Human B Cells In Vitro: Evaluation of IL-6 and TNF Production	176/181

Secondary pharmacodynamic studies

No studies addressing secondary pharmacodynamic of the combined vaccine or 1018 ISS were conducted. This was considered acceptable by the CHMP.

Safety pharmacology programme

Safety pharmacology of 1018 ISS was evaluated as part of single dose-toxicity studies in rabbit and baboon and the repeated dose toxicity study in cynomolgus monkey. No adverse acute effects on vital organ function were observed in these studies.

Pharmacodynamic drug interactions

Pharmacodynamic drug interaction studies were not performed with 1018 ISS Adjuvant + HBsAg or with 1018 ISS adjuvant alone. This was considered acceptable by the CHMP.

2.3.3. Pharmacokinetics

Pharmacokinetic data are generally not required for a vaccine. Pharmacokinetics/toxicokinetics of the antigen was therefore not studied. However, limited PK/TK data on the new adjuvant was provided to support the product. The presented PK/TK documentation for 1018 ISS is acceptable, considering that the applicant has also referred to several peer-reviewed scientific publications, which describes the absorption, distribution, metabolism, and excretion of phosphorothioate oligodeoxynucleotides.

The applicant has evaluated absorption of the 1018 ISS oligodeoxynucleotide after single SC administration in the rat (study number 01-191) and after repeated 1018 ISS administration to rats (study number 00-158) and cynomolgus monkeys (study number 00-157) as part of the 8-week repeated-dose toxicity studies. The overview of these studies is included in the Table 10 below.

Table 4: Overview of nonclinical pharmacokinetic and toxicokinetic studies with CpG 1018

Test Article	Type of Study	Species (Strain)	Route	Testing Facility	Study Number
CpG 1018	Absorption after single-dose	Rat (Sprague-Dawley)	SC	Covance, Richmond, CA	01-191
	Absorption after repeat-dose	Rat (Sprague-Dawley)	SC	MPI, Mattawan, MI	00-158
	Absorption after repeat-dose	Cynomolgus monkey	SC	MPI, Mattawan, MI	00-157

In rat, detectable levels of 1018 ISS in plasma were observed from 0.5 mg/kg = 0.8 µg/ml (single dose). In repeated dose studies in rat and cynomolgus 1018 ISS was only detected from dose levels of 2.5 mg/kg (reaching up to 188 µg/ml in rat) after the 8th weekly dose of 12.5 mg/kg/w. The absorption and plasma kinetic data obtained with 1018 ISS is in line with the published data of PS ODNs. Peak levels after SC administration of 1018 ISS was reached within a few hours post dosing followed by a rapid decrease in 1018 ISS plasma levels. According to published data the rapid decline is due to initial low affinity binding to plasma proteins (95%) and to significant distribution to kidney (up to 20 % of the dose), liver, and to a minor extent spleen. In general, there is a dose-proportional relationship between blood levels of PS ODNs (i.e., plasma Cmax, AUC values) and tissue concentrations. PS ODNs do not cross the blood brain barrier and poorly distributes to skeletal muscle, heart and lung (Geary et al. 2001; Geary 2009).

PS-ODNs are primarily catabolised by exonucleases in the blood compartment and tissues. This elimination is slow partly because the phosphorothioate backbone increases the resistance to exonucleases and tissue half-life for PS ODN can range from a few days to several weeks. At higher doses there is increased risk for accumulation of PS ODN in kidney and liver. This is likely part of the explanation why these organs are targets for 1018 ISS toxicity at frequent and high doses (see also overall conclusions on toxicology). Mass balance studies have demonstrated that up to 40-50% of the nuclease metabolites of ODNs (short-chained ODN) are excreted through the urine (Geary 2009).

A published report on the impact of kidney damage on the PK of an antisense PS ODNs (Masarjian et al, 2004) indicated that kidney damage (either tubular or glomerular) may reduce the uptake to the kidney tissue but do not lead to altered plasma kinetics or tissue distribution. Data available from patients with chronic kidney disease vaccinated with Heplisav B do not indicate any alteration of human PK.

2.3.4. Toxicology

A summary of all safety studies with HBsAg + 1018 ISS or 1018 ISS alone is provided in the Table 11 below.

Table 5: Summary of all safety studies with HBsAg + 1018 ISS or 1018 ISS alone

Study no/Type of study/GLP status	Species (strain)	Adm. route	Duration of dosing	1018 ISS dose	HBsAg dose
1018 ISS adjuvant + HBsAg *The HBsAg subtype (adw) that was used in study 00-95 and 05-463 were similar to the HBsAg adw in clinical batches.*					
00-95 Repeated-dose GLP	Mouse (Balb/c)	IM	3 doses: weeks 0, 2, and 4	1, 5, and 50 µg (≈ 0.04, 0.2 and 2 mg/kg)	0.5 µg (≈0.02 mg/kg)
12-728 Repeated-dose GLP	Rat (Sprague-Dawley	IM	4 doses: Weeks 0, 2, 4 and 6	600 µg and 3000 µg (≈2 and 10 mg/kg)	4 µg and 20 µg (≈ 0.01 and 0.06 mg/kg)
05-463 Reproductive and developmental toxicity GLP	Rat (Sprague-Dawley)	IM	4 doses: Premating Days 1 and 19; and Gestation Days 6 and 18	1.5, 15, 300, and 3000 µg (≈ 0.005, 0.05, 1.0, and 10 mg/kg)	2.5 µg (≈ 0.008 mg/kg)
1018 ISS adjuvant alone					
98-0034 Single dose Non-GLP	Rabbit (NZW)	IV	Single escalating Doses	0.1, 0.5, and 1.6 mg (≈ 0.05, 0.25, and 0.8 mg/kg)	Not applicable
98-0033 Single dose GLP	Baboon	IV, SC	Single escalating Doses	0.5, 2, 8, and 25 mg (≈ 0.05, 0.2, 0.8,	Not applicable

				and 2.5 mg/kg)	
00-141 Repeated-dose GLP	Mouse (Balb/c)	IM	3 doses: weeks 0, 2, and 4	50 µg (≈2 mg/kg)	Not applicable
00-158 Repeated-dose GLP	Rat (Sprague-Dawley)	SC	8 doses: once weekly for 8 weeks	0.5, 2.5, and 12.5 mg/kg	Not applicable
00-157 Repeated-dose GLP	Cynomolgus monkey	SC	8 doses: once weekly for 8 weeks	0.5, 2.5, and 12.5 mg/kg	Not applicable
01-GT1 Genotoxicity (Bacterial mutagenicity) GLP	Bacteria *(Salmonella typhimurium)*	*In vitro*		50, 158, 500, 1580, and 5000 µg / plate	Not applicable
01-GT2 (chromosomal aberrations) GLP	Human (peripheral blood lymphocytes)	*In vitro*		625, 1250, 2500, and 5000 µg/mL	Not applicable
04-413 Bone marrow micronucleus assay GLP	Mouse (ICR)	IP	Single dose	100, 200, and 400 mg/kg	Not applicable

Single dose toxicity

Single dose toxicity studies with 1018 ISS only were conducted in rabbit (study number: 98-0034) and baboon (study number: 98-0033). In both studies individual animals received escalating doses every third week of 1018 ISS. In rabbits 3 doses up to ≈0.8 mg/kg, IV and in baboons at total of 4 doses up to ≈2.5 mg/kg, IV or SC. Studies were designed to evaluate acute effects of 1018 ISS administration and included evaluations of body temperature, body weight, weight gain and vital signs (heart rate, systolic blood pressure, respiratory rate, ophthalmic examinations and gross examination of injection site). There were no treatment related changes in any of these parameters in rabbits or baboons. A minor swelling at the injection site was observed in one baboon at day 42 and 105 (correlating with administration of 3rd and 4th subcutaneous dose of the adjuvant (8 and 25 mg respectively), which was likely due to its immunostimulatory activity. No other treatment related change was observed in baboons or rabbits.

In addition, in vivo cytokine responses to 1018 ISS were measured in baboons at 0, 1 and 6 h post-dose (this assay was non-GLP compliant). No significant induction of any of the cytokines measured (IL-12, IL-6, IL-8, IFN gamma and TNF alfa) was observed.

Repeat dose toxicity

General toxicity of 1018 ISS adjuvanted vaccine

The general safety of the 1018 ISS adjuvanted HBsAg vaccine was investigated in two pivotal studies were the vaccine was administrated by the IM injection, the clinical route of administration: a repeated-dose toxicity study in mice (study 00-95) where 3 IM doses were given (at day 0, 14 and 28) and a repeated dose toxicity study in rats (study 12-728) where 4 IM doses were given (at day 0, 14, 28 and 42). The number of doses were comparable to/or exceeded the 2 doses intended for humans. In mice, the doses of 1018 ISS spanned from clinically comparable doses (0.046 mg/kg based on a 65 kg human) up to 43-fold the clinical

dose while the HBsAg dose was fixed at 0.5 µg (≈67-fold the clinical dose of 0.31 µg/kg). In rats, 2 doses of HBsAg+1018 ISS representing ≈32- and 194-fold the clinical dose with respect to HBsAg and ≈43- and 217-fold the clinical dose with respect to 1018 ISS were administered.

Injection site reactions, an expected response to this type of product, were observed from the lowest 1018 ISS dose administered (0.04 mg/kg in mice). Additional organs affected by higher doses of HBsAg + adjuvant were the hematopoietic system, spleen, and liver and in mice also kidney and heart to a minor extent. Findings were similar to the adjuvant only study except for in rats where there were no kidney findings at the dose levels studied. Not all organs affected at the high dose in mice (i.e. kidney and heart) were investigated in lower dose groups for this pivotal toxicity study. This made it difficult to evaluate whether the addition of HBsAg could exacerbate the effects of the adjuvant. The level of 1018 ISS without adverse effects on the heart is thus not known. However, the incidence of both the heart and kidney findings in mice were low and/or of low grade and the cases of epicarditis most likely a spontaneous finding common for the mice strain use. Of importance, no kidney or heart findings were observed in monkeys or in the rat study of 1018 ISS adjuvanted HBsAg administered with a dosing regimen (4 IM every second week) more similar to the clinical dosing regimen of 2 IM doses 1 month apart than in the rat study of 1018 ISS alone (8 SC doses every week).

The optimal dose-ratio of adjuvant and antigen was not explored in mice where the maximum ratio of Ag/adjuvant tested was 1/100, in comparison to 1/150 for the final clinical formulation. However, since there is no physical association between the antigen and adjuvant, and given that the doses were sufficiently high, it was concluded that the ratio of antigen/adjuvant would not have a significant effect on the immune response in this study. The doses used in the rat study corresponds to an Ag/adjuvant ratio of 1/150.

A NOAEL at the high dose of 3000 µg 1018 ISS + 20 µg HBsAg (i.e. approximately 10 and 0.06 mg/kg, respectively) in study 12-728 in rats was accepted as there were no severe vaccine-related toxic or harmful effects with impairment of growth, function or life span of the animals, or deemed to affect the overall well-being of the animals. All findings observed were considered well understood (e.g. expected due to mode of action and class, dose-related in incidence and partly or fully reversed at recovery) and without clinically relevant severe toxicity. With exception of lymphoid hyperplasia, which was mild to moderate in all dose groups and severe in inguinal or iliac lymph nodes of individual rats in the high-dose group, the observed systemic effects and changes were of minimal to mild severity or of a small magnitude. As the lymphoid hyperplasia is an expected response to the immunostimulatory effect of 1018 ISS and showed complete recovery all findings observed, including separate findings of severe lymphoid hyperplasia, can be considered non-adverse.

General toxicity of 1018 ISS adjuvant alone

The safety of IV administration of 1018 ISS adjuvant alone was evaluated in escalating dose tolerability studies in rabbits (98-0034) and baboons (98-0033), followed by a repeated dose toxicity study in mice (00-141) where 3 doses of 1018 ISS adjuvant were administered IM every other week.

In addition, reports from two 8-week repeated dose toxicity studies of 1018 ISS in rat (00-158) and cynomolgus monkey (00-157) were submitted. However, in these studies, which was originally conducted to support safety of another application and indication, 1018 ISS was administered subcutaneously once a week and the dose range tested and systemic exposure were much higher compared to the intended clinical dose schedule (ranging from 11x to 270x the clinical 1018 ISS dose). According to EMA guidelines on adjuvants the study designs were not representative for the clinical dose regime of Heplisav B. This complicated the assessment of potential safety issues related to Heplisav B administration, in particular the risk for systemic

effects of 1018 ISS at clinical dose levels. However, by taking all safety data into account (as discussed along this report) these deficiencies of the 8-week repeated dose toxicity studies were considered acceptable.

As stated above, the findings observed in the studies with 1018 ISS adjuvant alone were similar to the effects observed with HBsAg + 1018 ISS and related to 1018 ISS dose. The findings are also at large consistent with published non-clinical data describing class effects of immunostimulatory phosphorothioate oligodeoxynucleotides.

A NOAEL could not easily be defined for any of the repeated dose toxicity studies of 1018 ISS alone as signs of immunostimulation i.e. injections site reaction were present at the lowest doses tested, in mice 0.04 mg/kg (approximating clinical dose) and 0.5 mg/kg in rat and cynomolgus monkey (at 8x the clinical dose using a conservative mg/kg dose for a 50 kg human). In rat, additional findings at the 0.5 mg/kg level were peripheral reductions in erythrocytes and platelets, bone marrow hyperplasia and signs of kidney toxicity. At higher adjuvant doses the local and systemic effects of 1018 ISS stimulation were more pronounced and included hematopoietic alterations and inflammatory changes in spleen, lymph nodes and liver (with Kupffer cell hyperplasia and in rodents also cell necrosis), transient increases in ATPP, signs of complement activations (monkeys only) and epicardial mineralization/chronic inflammation in the heart (mice studies only).

Almost all changes in the high dose groups were either completely or partially resolved after a 3 or 4 weeks long treatment free period. However, kidney alterations observed in the rat study were still prominent in recovery animals although BUN levels were diminished.

Key findings in the repeated-dose toxicity studies and their potential clinical relevance

Kidney toxicity i.e. dose-dependent increases in tubular and interstitial inflammation/ degeneration of the kidney and biomarkers of kidney toxicity were observed at the lowest dose tested (0.5 mg/kg) in the rat study (8 doses: once weekly for 8 weeks). The kidney findings were still present in recovery animals and could potentially be a concern as the product is intended to be administered to patients with chronic kidney disease. However, taking into consideration that these effects were presented at a dose schedule that was more frequent than the intended clinical dose regimen, the lack of renal toxicity in monkeys in the 8-week repeated dose toxicity study, lack of significant kidney toxicity in mice and rats at >40 fold and 217-fold the clinical dose in the more clinically relevant toxicity studies with 1018 ISS adjuvanted HBsAg, and lastly, PK data in monkeys and humans (including patients with renal impairment) demonstrating lack of 1018 ISS accumulation at the proposed clinical dose regiment, it is concluded that a clinical relevance of the kidney findings is unlikely.

Bone marrow hyperplasia (increased erythro- and thrombocytopoiesis) possibly related to peripheral reductions in erythrocytes and platelets, was observed at the lowest dose levels in 8-week study in rat with a similar tendency regarding peripheral changes noted in monkeys. These effects were reversible. There is sufficient support in the scientific literature that this effect is manifested in rodents but not monkeys due to a stronger systemic proinflammatory response in rodents (Campbell, Cho et al 2012). This was also supported by the lack of bone marrow alterations in the 8-week repeated dose toxicity study in monkeys. It should be noted that other systemic manifestations of TLR9 immunostimulation were seen in monkeys (i.e. spleen hyperplasia from 2.5 mg/kg/w and lymph node hyperplasia and liver inflammation at 12.5 mg/kg/w). Of importance, a NOAEL of 0.5 mg/kg 1018 ISS (>10-fold the clinical dose on mg/kg basis) for systemic immunostimulatory effects was identified in this study. Given published data which supports that humans are similar to monkeys in their sensitivity to TLR9 immunostimulation there are thus reassuring margins regarding systemic consequences of 1018 ISS administration at the clinical dose range.

Complement activation: In cynomolgus monkeys, treatment related complement activation was observed at the 12.5 mg/kg dose (at 270 x the clinical dose). Complement activation has been shown to be a common property of phosphorothioate ODNs. It was concluded that the effects to the complement system seen in the cynomolgus monkey study are not to be expected in humans at the proposed concentration of the adjuvant.

Transient increases in Activated Partial Tromboplastin Time (APTT), a known plasma concentration dependent class effect of PS ODNs, were observed in rats and non-human primates at high doses in the 8-week repeated dose toxicity studies. The lack of clinical relevance of this finding was adequately justified by the fact that increases of this parameter was detected only at the maximum dose of 12.5 mg/kg/w in rats and monkeys (at >200-fold the maximum clinical plasma levels of 1018 ISS measured), and that this effect was mild and transient in both species: at most a < 2-fold increase in bleeding time.

Epicardial and myocardial findings: In the repeated dose-toxicity studies in mice of 1018 ISS alone (study 00-141) and HBsAg + 1018 ISS (study 00-95) a notable increased incidence of low-grade epicardial mineralization of mainly the right ventricle was observed at the highest dose level of 1018 ISS (50 µg; 2 mg/kg). At this dose level a low incidence (2/10 animals) of chronic inflammation or mononuclear cell infiltration of the myocardium was also noted. Given that TLR9 has been shown to have a functional role in cardiomyocytes and the clinical observations of a potential increase of myocardial infarcts in Heplisav B vaccinated subjects in clinical study HBV-23, these nonclinical observations are considered of interest. As discussed by the Applicant, epicardial mineralisation is a common spontaneous finding in this strain of mice. However, it is possible that the systemic immunostimulatory effect at the high dose of 1018 ISS in mice contributed to the high incidence of epicardial findings and to the infiltration of inflammatory cells in the myocardium. However, taking together the low-severity grade of these findings, a large margin (43-fold the human dose of 1018 ISS on a body weight basis) and the lack of cardiac findings in safety studies in rats and monkeys, nonclinical data do not point to a risk for cardiac toxicity at the intended clinical dose regime.

The apparent general toxicity profile of the candidate vaccine was clearly related to 1018 ISS dose and at large consistent with published data on class effects of immunostimulatory phosphorothioate oligodeoxynucleotides that manifests at high systemic levels. Most if not all effects can be linked to either the TLR9 mediated immunostimulatory activity or the polyanionic characteristics of oligodeoxunucleotides. As expected, the studies indicate that administration of the candidate vaccine will be associated with local injection site reactions. After a thorough assessment of available data, it can be concluded that non-clinical data indicate that the risk for adverse systemic immunostimulation of the product is low at clinically relevant doses.

A potential increased risk of myocardial infarction has however been noted with Heplisav B and it is not clear whether this association is causal or not. In a recent review, it was concluded that the role of TLR9 in the development of atherosclerotic lesions remains controversial (Roshan et al., 2016). An association between 1018 ISS (CpG 1018) and cardiovascular disease therefore seems biologically plausible. An in-depth discussion on e.g. the role of TLR9 in thrombosis, atherosclerosis, ischemia, TLR9 expression patterns in different tissues, off-target effects and influences on the coagulation system was provided.

Non-clinical literature data show contrasting results. Proinflammatory mechanisms are in this context possibly induced via the possibility of TLR9 to bind mitochondria DNA released from necrotic and apoptotic cells, as occurring due to ischemia. Some articles indicate that TLR9 has proinflammatory effects with putative impact especially on the course of the myocardial infarction rather than the event itself. On the other hand, several studies demonstrate that CpG-ODN treatment before induction of myocardial ischemia/reperfusion or after induction of ischemia has protective effects by being able to enhance stress tolerance, angiogenesis and by reducing infarct size through TLR9 dependent pathways. Of note, the concentrations reached with the vaccine

in humans are up to 1000 times lower than concentrations used in some of the animal experiments described above.

Literature data is currently also inconclusive regarding the question whether TLR9 may have proatherogenic or antiatherogenic effects. In a mouse model low doses of CpG ODN resulted in reduction of atherogenic lesions whereas more frequent and higher dosing was shown to induce atherosclerotic plaque formation. Furthermore, plasmacytoid dendritic cells (pDCs) in plaque tissue have been shown to produce modest levels of interferon-α (IFN-α) upon in vitro incubation with high levels of CpG-ODN (100 µg/mL). The cumulative dose of CpG-ODN which enhanced atherosclerotic plaque formation in the mouse model was 680-fold higher than the cumulative dose presented by 2 doses of Heplisav B and the CpG-ODN concentration which showed induction of IFN-α in atherosclerotic plaque in vitro was more than 1000 times higher than the maximum plasma concentration obtained after Heplisav B IM injection.

In humans TLR9 appears to be mainly expressed in B-lymphocytes, pDCs and to be absent in macrophages, which is a crucial cell type found in atherosclerotic plaques. Additionally, TLR9 is only to a low extent expressed in human plaque and heart tissue. This is in contrast to the more extensive expression pattern of TLR9 in rodents, e.g. including macrophages and all subsets of DCs. However, no heart-related safety concern was indicated based on the studies in rodents and monkeys performed in the frame of the non-clinical program for Heplisav B.

Different TLR9 agonists have been in clinical development for approximately 20 years without identification of significant off-target activity. The stimulation of TLR9 via CpG motif containing ODNs is target specific as shown in knock out rodent models and cell lines expressing TLR-9, with lack of e.g. immune stimulatory effects seen in rodents not expressing TLR-9. Neither have effects been observed that may have been the result of non-specific interactions of the adjuvant with un-identified targets in non-clinical or clinical studies.

PS ODNs can increase the clotting time. This phenomenon is dose dependent, transient and multifactorial. Responsible are especially interactions with thrombin and other clotting factors belonging to the intrinsic tenase complex such as clotting factor IX. A prolongation of aPTT 1.5 above control has been observed in studies with humans and monkeys obtaining very high doses of PS ODNs administered as antisense PS ODNs, however, without observing any bleeding events. Concentrations achieved with antisense ODNs are much higher (16-89-fold) than reached with CpG ODN 1018 used with Heplisav B. Only modest prolongations were observed in cynomolgus monkeys in the non-clinical program of Heplisav B at doses up to 270-fold higher than the clinical dose. Furthermore, no significant effect on platelet function or hypercoagulable effects was seen in rodents and monkeys using up to 270-fold higher doses than applied in the clinical studies of Heplisav B. Additionally, thrombotic events were balanced between study arms in PSP and HBV-23.

It is anticipated that the adjuvant induces cytokine production locally in muscle tissue and draining lymph nodes in humans which is believed to be mirrored in the transient and reversible occurrence of post injection reactions seen within 7 days after injection of the vaccine. This is supported by PK data of CpG 1018 showing that Heplisav B vaccination results in very low and short-lasting plasma concentrations, decreasing the likelihood of a systemic induction of long-lasting inflammatory reactions or an impact on the coagulation system.

Overall, the data and information presented (see also clinical safety section) do not point to a risk for myocardial infarction after administration of Heplisav B. Further non-clinical investigation of a plausible mechanism underlying a potential risk for myocardial infarction in relation to vaccination with Heplisav B is not needed.

Potential risk for enhancement of autoimmunity

A concern with the 1018 ISS adjuvant is whether the TLR9 activation and subsequent immunostimulatory activity of 1018 ISS could increase the risk for the vaccines to develop autoimmunity in particular systemic autoimmune disease (e.g. systemic lupus erythematosis) which are in part caused by generation of anti-dsDNA antibodies. This concern is based on evidence from studies in animal models for autoimmunity where TLR9 stimulation was demonstrated to enhance development of/exacerbate autoimmune disease. There are also some reports that demonstrate that TLR9 may have a protective role against autoimmunity so there is clearly a complex relationship between TLR9 and autoimmunity.

There are no findings indicative of an autoimmune reaction such as glomerular nephritis or vasculitis in the non-clinical safety data of HBsAg+ 1018 ISS or 1018 ISS alone. However, autoimmune reactions are rare events and are unlikely to be detected in standard non-clinical safety evaluations. The applicant has however made an effort to address the concern for autoimmune events non-clinically, by evaluating the potential for development of anti-DNA autoantibodies as part of the immunogenicity studies of HBsAg + 1018 ISS in mice and baboons (studies 99-0086 and 99-0089).

Neither mice nor baboons immunised twice with HBsAg + 1018 ISS showed any significant generation of anti-dsDNA antibodies based on group mean levels although minor elevations in anti-dsDNA titres were noted in single individuals. Based on available non-clinical data one cannot defer or confirm that injection of 1018 ISS would enhance the risk for autoimmunity. Considering the limitations of animal autoimmunity models to predict the risk for induction of autoimmunity in humans, no further non-clinical studies are needed.

Genotoxicity

1018 ISS alone did not show any genotoxic or clastogenic potential in a standard battery of in vitro and in vivo genotoxicity studies. Furthermore, no homologies of concern between 1018 ISS and human DNA was identified and induction of site-directed mutagenesis by the adjuvant is thus not likely.

Carcinogenicity

No studies assessing the carcinogenicity of Heplisav B or 1018 ISS adjuvant have been performed. This is acceptable for this type of product in line with EMAs guidelines for vaccines and adjuvants and compliant with ICH S1A, considering that the exposure to this vaccine will be sporadic.

Reproduction Toxicity

The repro-toxicological safety of HBsAg + 1018 ISS and ISS alone was evaluated in one combined developmental and reproductive toxicity study in rats using a 4-dose schedule (study 05-463, study day 1 and 19 of pre-mating and gestation days 6 and 18). A fixed antigen dose (2.5µg) was used against a range of 1018 ISS doses (0.5x to 200x the clinical dose). The conducted rat developmental and reproductive toxicity study was designed in line with ICH S5(R2) and the FDA guideline Considerations for developmental toxicity studies for preventive and therapeutic vaccines for infectious disease indications.

Relevance of the study design to predict risk pertaining to human pregnancy with 1018 ISS

There is a general concern that adjuvants which induce Th1 type immune responses could potentially interfere with pregnancy by altering the complex maternal immune modulations that occur at early stages to

promote implantation and protect the embryo from rejection (Reviewed in Saito et al, 2010 and Herberts et al 2010).

While there in a general sense may be a theoretical concern of possible adverse effects of strong immunostimulation on pregnancy based on animal data, available clinical data on marketed vaccines that stimulates Th1 type responses (e.g. the AS03 adjuvanted Pandemrix and yellow fever vaccines) do not indicate a risk associated with administration during pregnancy. Although the selected study design for the multi-generation study may not be fully optimal for addressing all consequences of an immunostimulatory agent on all stages of fertility, it is agreed that the conducted study, has given adequate information on the main risks pertaining to female fertility, organogenesis/fetal development and survival and pre/post-natal effects. Importantly, the dams were adequately dosed as indicated by the significant increase in systemic plasma levels of IFN gamma and IL-12p40 cytokines and prominent maternal toxicity at the highest doses tested (3000 µg 1018 ISS +/- 2.5 µg HBsAg). It is also of importance to note that the dose of 1018 ISS in Heplisav B is titrated to mainly act locally and systemic effects manifested as fever and myalgia have been shown to be limited and comparable to the Alum adjuvanted comparator vaccine Engerix B. The large margins to adverse systemic effects of immunostimulation observed in animal safety studies are also reassuring. It is thus concluded that TLR9 stimulation would have no impact on pregnancy at clinically relevant doses.

Findings in the developmental and reproductive toxicity study

Particular findings in this study which were observed at the highest dose level (3000 µg 1018 ISS with/without 2.5 mcg HBsAg) include an increase in mortalities or moribundency of pregnant dams close to or at parturition, a statistically significant increase in a foetal skeletal anomaly- cervical rib at 7th vertebrae- and also an increased number of stillborns in both 3000 adjuvant dose groups (incidence in the adjuvant only group outside the historical control range).

Regarding the mortalities/moribundancies of dams (5 in total in the two groups with 3000 µg 1018 ISS groups out of 95 dams in the study) the prominent signs of adverse systemic immunostimulation of 1018 ISS in these groups (e.g. systemic induction of pro-inflammatory cytokines, lymphoid hyperplasia, liver inflammation similar to the effects in the rat 8-week repeated dose toxicity study) indicate that these deaths were treatment-related. However, taking into account the fact that rats are known to be more sensitive to TLR9 activation compared to humans and that a reassuring NOAEL was established for these adverse effects in dams i.e. 300 µg 1018 ISS + 2.5 HBsAg (> 22- fold the clinical dose on a body weight basis) a clinical relevance seems unlikely.

The increase in cervical rib and stillbirths were observed at the dose levels of 1018 ISS that produced maternal toxicity (3000 µg 1018 ISS +/- 2.5 µg HBsAg). These effects could be secondary to maternal toxicity and induction of stress hormones.

No other significant adverse findings judged to be related to treatment was observed on fertility/ reproduction parameters of F0 females, developmental toxicity and or effects on development of F1 generation.

Toxicokinetic data

TK evaluations were included in the design of the repeat-dose toxicity studies of CpG 1018 in rats and monkeys (studies numbers 00-158 and 00-157).

Local Tolerance

Local toxicity of the adjuvanted vaccine and adjuvant only was evaluated as part of the repeated dose toxicity studies which is acceptable.

2.3.5. Ecotoxicity/environmental risk assessment

HBsAg is a natural substance and both HBsAg and its adjuvant 1018 ISS will be degraded to components which are naturally present in the human body, amino acids, nucleic acids, lipids, etc. Its use will not alter the concentration or distribution of the substance in the environment. Therefore, HBsAg + 1018 ISS are not expected to pose a risk to the environment.

2.3.6. Discussion on non-clinical aspects

The non-clinical immunogenicity studies indicated that the anti-HBV antibody response and the rate of animals responding with antibody levels ≥10 mIU/ml (the defined threshold for seroprotection in humans) depends on the 1018 ISS dose. The pharmacology studies on 1018 ISS provided some indirect evidence of the involvement of a Th1-type immune response.

The non-clinical safety profile of 1018 ISS adjuvanted HBsAg in mice, rats and monkeys showed the exaggerated pharmacological activity of the 1018 adjuvant, manifested at high systemic exposure of the adjuvant. The effects were consistent with known class effects of phosphorothioate oligodeoxynucleotides. At dose levels comparable to the clinical dose, based on mg/kg comparisons, only local injection site reactions were observed, while systemic effects of 1018 ISS immunostimulation appeared to occur at significantly higher doses (11 to 270-fold the clinical dose mg/kg based) and therefore, they were concluded not to be relevant at the intended clinical dose regimen.

The Applicant was requested to discuss the biological plausibility of an association between 1018 ISS and cardiovascular disease and potential mechanisms behind the potential increased risk of myocardial infarction noted with Heplisav B. Overall, the data and information presented did not point to a risk for myocardial infarction after administration of Heplisav B. While there are data indicating that activation of TLR9 can have proinflammatory effects with putative impact on the course of the myocardial infarction and induce development of atherosclerotic lesions in vitro and in mice, the differences in cellular distribution of TLR9 between rodents and humans together with the large margins to the concentrations and doses with proinflammatory and proatherogenic effects did not support a clinically relevant correlation. Further non-clinical investigation of a plausible mechanism underlying a potential risk for myocardial infarction in relation to vaccination with Heplisav B was not considered needed.

In a multigeneration reprotoxicological study in rats, effects on the offspring (stillborn pups and skeletal abnormalities) occurred secondary to maternal toxicity at a dose 200-fold the proposed clinical dose of Heplisav B with respect to 1018 ISS on a body weight basis. Given the large margin to the clinical dose, these effects were not considered clinically relevant.

Regarding a possible link between 1018 ISS mediated immunostimulation and autoimmunity, the CHMP considered that 1018 ISS seems to display potent immunostimulatory activities. However, the non-clinical data could not provide clear evidence for or against this issue. It should also be noted that non-clinical studies are of limited use for predicting autoimmunity in humans. Therefore, the CHMP concluded that the

potential risks for induction or exacerbation of autoimmune diseases after vaccination with Heplisav B should be evaluated based on clinical safety data.

2.3.7. Conclusion on the non-clinical aspects

The CHMP considered the vaccine approvable from a non-clinical perspective.

2.4. Clinical aspects

2.4.1. Introduction

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.

- Tabular overview of clinical studies

Table 6: Overview of clinical studies for the indication development

Phase/ Trial No.	Trial Design	Heplisav B Dose/Schedule/N	Comparator Dose/Schedule/N	Key Immunogenicity Endpoint(s)
Pivotal Trials				
Phase 3 HBV-23	Observer-blinded, randomized, active-controlled, parallel-group, multicenter trial in subjects 18 to 70 years of age conducted in the US	Heplisav B: 20 µg HBsAg, 3000 µg 1018 Schedule: 0, 4 weeks (placebo at 24 weeks) N = 5592	Engerix-B: 20 µg HBsAg, 500 µg alum Schedule = 0, 4, 24 weeks N = 2782	Primary Endpoint: SPR at Week 28 in subjects with type 2 diabetes mellitus
Phase 3 HBV-16	Observer-blind, randomized, active-controlled, parallel-group, multicenter trial in healthy adults 40 to 70 years of age conducted in the US and Canada	Heplisav B: 20 µg HBsAg, 3000 µg 1018 Schedule: 0, 4 weeks (placebo at 24 weeks) N = 1969	Engerix-B: 20 µg HBsAg, 500 µg alum Schedule = 0, 4, 24 weeks N = 483	Primary Endpoints: SPR at Week 12 for Heplisav B and Week 32 for Engerix-B Lot consistency of Heplisav B measured by GMC at Week 8
Phase 3 HBV-10	Observer-blind, randomized, active-controlled, parallel-group, multicenter trial in healthy subjects 11 to 55 years of age conducted in Canada and Germany	Heplisav B: 20 µg HBsAg, 3000 µg 1018 Schedule: 0, 4 weeks (placebo at 24 weeks) N = 1809	Engerix-B: 20 µg HBsAg, 500 µg alum Schedule = 0, 4, 24 weeks N = 606	Primary Endpoint: SPR at Week 12 for Heplisav B and Week 28 for Engerix-B
Supportive Trials				
Phase 1 HBV0001	Observer-blind, randomized, dose-escalation trial of the 1018 and rHBsAg components of vaccine in healthy, seronegative adults	1018 alone or + HBsAg 300 µg, alone or plus HBsAg 650 µg, alone or plus HBsAg	HBsAg alone: 20 µg Schedule: 0, 8 weeks N = 8	Anti-HBs measured after vaccinations

Phase/ Trial No.	Trial Design	Heplisav B Dose/Schedule/N	Comparator Dose/Schedule/N	Key Immunogenicity Endpoint(s)
	18 to 55 years of age conducted in Canada	1000 µg, alone or plus HBsAg 3000 µg, alone or plus HBsAg HBsAg: constant at 20 µg Schedule: 0, 8 weeks N = 32	1018 alone: 300 µg, 650 µg, 1000 µg, 3000 µg Schedule: 0, 8 weeks N = 8	
Phase 2 HBV-08	Double-blind, randomized, parallel-group trial in adults 18 to 39 years of age in Canada	Heplisav B: 20 µg HBsAg, 3000 µg 1018 Schedule: 0, 4 weeks (N = 18) 0, 8 weeks (N = 23) Heplisav B Half Dose (10 µg/1500 mcg) Schedule: 0, 4 weeks N = 20	None	Primary Endpoint: SPR 4 weeks following second injection administered 0, 4 weeks vs 0, 8 weeks Secondary Endpoint: SPR and GMC 4 weeks after each injection of full and half-dose Heplisav B administered at 0, 4 weeks
Supportive Trial				
Phase 3 HBV-17	Observer-blinded, randomized, active-controlled, multicenter trial in adults 18 to 75 years of age with CKD conducted in Germany, the US, and Canada *Note: Due to GCP issues the data from study HBV-017 will not be used to support efficacy claims.*	Heplisav B: 20 mcg/3000 mcg Schedule: 0, 4, 24 weeks (placebo at 8 weeks) N = 258	Engerix-B: 2 doses of 20 mcg HBsAg each Schedule = 0, 4, 8, 24 weeks N = 263	Primary Endpoint Noninferiority of SPR at Week 28
Durability of Seroprotection Trial				
HBV-19	Long-term follow-up trial of adult subjects 18 years of age and older with CKD who previously received a complete series of either Heplisav B or Engerix-B in trial HBV-17	Overall enrollment N = 73 Heplisav B as the second vaccine series in subjects not seroprotected at baseline: • 20 mcg/3000 mcg • Schedule = 0, 4, 24 weeks N = 8	Overall enrollment N = 74 Engerix-B as the second vaccine series in subjects not seroprotected at baseline: • 2 doses of 20 mcg HBsAg each • Schedule = 0, 4, 24 weeks N = 11	Durability of anti-HBs concentrations ≥ 10 mIU/mL subjects who had anti-HBs ≥ 10 mIU/mL at Week 28 in HBV-17

Phase/ Trial No.	Trial Design	Heplisav B Dose/Schedule/N	Comparator Dose/Schedule/N	Key Immunogenicity Endpoint(s)
		Heplisav B single-dose booster (20 mcg/3000 mcg) was administered following an anti-HBs result <10 mIU/mL in subjects who were previously seroprotected N = 10 Seroprotected at enrollment N = 48	Engerix-B booster (2 doses of 20 mcg HBsAg) was administered following an anti-HBs result <10 mIU/mL in subjects who were previously seroprotected N = 13 Seroprotected at enrollment N = 48	
Booster Trial in Patients Receiving Dialysis				
HBV-18	Open-label, randomized, multicenter trial in adults 18 years of age and older with ESRD receiving hemodialysis, and with anti-HBs < 10 mIU/mL at study entry conducted in Germany	Heplisav B: 20 mcg/3000 mcg Schedule: Day 1 (0 Week) N = 54	Engerix-B: 2 doses of 20 mcg HBsAg each Schedule: Day 1 (0 Week) N = 50 Fendrix: 20 mcg HBsAg Schedule: Day 1 (0 Week) N = 51	SPR at Week 4

anti-HBs = antibody against hepatitis B surface antigen; CKD = chronic kidney disease; ESRD = end-stage renal disease; GMC = geometric mean concentration; HBsAg = hepatitis B virus surface antigen; N = number of randomized subjects; a dose of Engerix-B comprises 2 × 20 mcg recombinant HBsAg and 500 mcg aluminum hydroxide; No. = number; SPR = seroprotection rate.

2.4.2. Pharmacokinetics

According to the Guideline on clinical evaluation of new vaccines (EMEA/CHMP/VWP/164653/2005), pharmacokinetic studies are usually not required for vaccines. However, such studies might be applicable when new delivery systems are employed or when the vaccine contains novel adjuvants or excipients.

Heplisav B comprises recombinant hepatitis B surface antigen (HBsAg) and a new phosphorothioate oligodeoxynucleotide CpG 1018 adjuvant (1018 ISS).

No formal clinical PK data are available for the antigen which is acceptable since it is expected that the HBsAg in Heplisav B would exhibit PK properties similar to those of currently licensed vaccines (i.e. distribution from the injection site to the draining lymph node). However, distribution of HBsAg into the plasma has been reported for other currently licensed hepatitis B vaccines, and was also observed with the HBsAg in Heplisav B. One subject included in the HBV-18 trial underwent serological testing (in a Hepatitis B screening program independent of the study) for hepatitis B infection 5 days after vaccination with Heplisav B and showed positive results for HBsAg. The subject had no clinical signs of acute hepatitis. The positive serum HBsAg result was attributed to transient, vaccine-induced antigenemia.

PK properties of CpG 1018 as a component of Heplisav B were assessed in subjects with chronic kidney disease (CKD) in HBV-09 (n=8), and as a single agent in subjects with colorectal cancer in trial ONC-01 (n=14). Because of the known preferential distribution of PS ODNs to the kidneys and the observed renal toxicity findings from nonclinical Study 00-158 (tubular degeneration at doses 50-fold higher on a body-weight basis than the proposed clinical dose of 1018 ISS), there appeared to be potential for changes in the PK of the 1018 ISS component of Heplisav B in individuals with renal disease. There is no data available in a generally healthy population. There is no data on 1018 ISS in special population other than CKD patients.

Due to the limited number of subjects in both studies, statistical analyses were only descriptive. The 1018 ISS levels peak at 1 hour and 2 to 4 hours following administration in HBV-09 and ONC-01 studies respectively. Although PK results of study HBV-09 indicate that there is transient low level circulation of 1018 ISS 2.5 to 3 times higher in the 3000-µg as compared to the 1500-µg for up to 4 hours following vaccination, the safety profile between the two dose levels was similar for treatment-related AEs and SAEs. The mean serum concentration of 1018 ISS was near the LLOQ 8 hours after the vaccine injection in the HBV-09 trial whereas the 1018 ISS level was below the quantifiable level 3 weeks after administration in the ONC-01 trial. The highest 1018 ISS concentrations observed in both studies were lower than the level of 50 to 60 µg/mL associated with transient, clinically nonsignificant aPTT prolongation in clinical trials of other PS ODNs (Kwoh 2008). Therefore, results of both studies suggest that there is no risk of 1018 ISS accumulation since there is one-month interval between vaccine doses.

Distribution, metabolism and excretion have been well characterized for PS ODNs such as 1018 ISS adjuvant and are highly similar among PS ODN of various nucleotide sequences. In consequence, no specific studies of plasma protein binding and of tissue distribution studies have been performed. No studies of the metabolism or excretion of 1018 ISS or of drug-drug interactions with the cytochrome P450 system or with transporters were have been were performed, either. This was deemed acceptable by the CHMP.

2.4.3. Pharmacodynamics

Pharmacodynamic studies are essentially comprised of the immunogenicity studies that characterise the immune response to the vaccine. Clinical PD studies of Heplisav B have assessed the antibody response to HBsAg.

The Applicant enrolled and treated 14,238 generally healthy subjects 18 to 70 years of age in 11 completed clinical trials of Heplisav B, including 10,038 subjects who received Heplisav B and 4200 subjects who received Engerix-B. There were three main pivotal trials. In addition, the Applicant enrolled and treated 758 subjects with CKD, including 385 subjects who received Heplisav B, 322 subjects who received Engerix-B and 51 subjects who received Fendrix.

Mechanism of action

Heplisav B is a hepatitis B vaccine comprised of recombinant HBsAg and the adjuvant 1018 ISS.

Recombinant HBsAg is a 22-nm particle containing the *adw* subtype of the HBV S protein. The desired biological activity of recombinant HBsAg is to generate antibodies to *a*-determinant. The choice of serotype *adw* is deemed acceptable. The *a* determinant contains a neutralizing epitope and is highly immunogenic. Vaccines containing a specific subtype appear to protect against HBV infection of any other subtype. Engerix-B, used as comparator vaccine in the pivotal trials, also contains the *adw*-derived surface antigen.

1018 ISS is a phosphorothioate oligodexoynucleotide (PS ODN) oligomer comprised of 22 nucleotides containing a cytidine phosphoguanosine (CpG) immunostimulatory sequence that is an agonist for Toll-like receptor (TLR) 9. The desired biological activity of 1018 ISS is to enhance the generation of antibodies to HBsAg by (1) activating pDCs through the pattern recognition receptor TLR9, (2) converting pDCs into activated dendritic cells that present the processed HBsAg component of Heplisav B to CD4+ and CD8+ T cells, (3) promoting Th1 T-cell differentiation through the production of IFN-alpha and IL-12, and (4) promoting cytotoxic T cells differentiation and activation. This activation results in a high and sustained antibody response, likely due to the rapid generation of large numbers of anti-HBs-secreting plasmacytes and HBsAg-specific memory B cells. The choice of 1018 ISS as adjuvant is relevant.

The pharmacological activity of 1018 ISS was further confirmed by stimulating human PBMC or isolated B cells with 1018 ISS and by measuring proliferation, secreted in vitro cytokine and interferon (IFN)-regulated gene induction. Results confirmed that 1018 ISS activates PBMC to proliferate and secrete Th1 cytokines.

Microarray results from the HBV-22 study supported the proposed mechanism of immune stimulation of 1018 ISS from the in vitro model, i.e. a rapid and pronounced activation of IFN-regulated genes, reflecting the induction of Type 1 IFNs from pDCs. Nevertheless, the data need to be interpreted with caution since no significant phenotypic changes of B cells, T cells, and DC were observed.

Human DNA sequence databases were searched to evaluate the extent to which 1018 ISS had homology with sequences within the known array of human genes and transcripts. Overall, given the lack of identified targets that could potentially hybridize with 1018 ISS, the data from genome searches indicated that the likelihood for off-target effects by 1018 ISS was low.

Correlates of protection

It is currently admitted that immunocompetent subjects who develop anti-HBs at a concentration ≥10 mIU/mL after vaccination (measured 1 to 3 months after receipt of a complete and adequately administered vaccination course) have virtually complete protection against both acute disease and chronic infection for decades, even if subsequently, over time, anti-HBs concentrations declined to less than 10 mIU/mL. Accordingly, SPR is defined as the percentage of subjects with a serum concentration of antibodies to Hepatitis B surface antigen ≥ 10 mIU/mL. In immunocompromised patients, booster doses are required to maintain anti-HBs concentrations ≥ 10 mIU/ml.

Persistence of anti-HBs over time is correlated with peak level of anti-HBs immediately achieved after primary vaccination. However, long-term protection has been demonstrated in the absence of detectable anti-HBs in immunocompetent persons up to 30 years (observational FU studies, booster studies).

In immunocompromised patients, booster doses are required to maintain anti-HBs concentrations ≥ 10 mIU/mL.

Both seroprotection rates and GMT are informative to assess the added value of vaccinating subpopulations known to have reduced SPR observed with currently licensed hepatitis B vaccines, subpopulations known to have a good SPR with licensed vaccines as well as immunocompromised subjects with Heplisav B compare to Engerix B.

Anti-HBs Assays

The presence of serum anti-HBs can be measured with sensitivity and specificity by radioimmunoassay or enzyme immunoassay (EIA). The clinical development programme for Heplisav B evaluated its clinical

pharmacodynamics in terms of the rise in anti-HBs level after vaccination using commercially available EIA assays for detection of serum anti-HBs.

Two different assays for determination of serum anti-HBs concentrations were used during development of Heplisav B in generally healthy adults. In earlier clinical studies, the AUSAB assay (Abbott Diagnostics Division 2010) was used to measure anti-HBs levels, whereas the OrthoVitros Hep B enhanced chemiluminescence immunoassay (ECi) (Ortho Clinical Diagnostics 2007) was used in trials using the proposed commercial formulation. Both assays are EIAs that quantitatively measure total antibody to HBsAg. Both kits are commercially available and were fully validated (including with calibrators referenced to the World Health Organization (WHO) 1st International Reference Preparation of anti-hepatitis B immunoglobulin). Different laboratories performed the anti-Hbs Ab measurement. No inter-laboratory comparisons were conducted between the different laboratories that performed the experiments. The Applicant confirmed that the laboratories correctly implemented quality assurance and testing procedures according to guidelines.

The AUSAB assay uses paramagnetic microparticles coated with recombinant HBsAg (ad/ay subtypes) as the solid phase and biotin coupled to recombinant HBsAg as the conjugate. The Hep B ECi assay is a solid-phase sandwich, enzyme-labelled immunoassay. This assay uses human HBsAg (ad/ay subtypes) to coat wells as the solid-phase and horseradish peroxidase-labelled human HBsAg as the conjugate.

Both the AUSAB and Hep B ECi assays were designed such that a positive assay result corresponds to the anti-HBs concentration of 10 mIU/mL considered to be protective against HBV infection. Therefore, results from these assays are considered comparable for analysis of SPR. However, quantitative results differ between both kits. Therefore, no pooling of GMC data from the AUSAB and Hep B ECi systems has been performed. This potential difference between assay systems should be taken into account in comparing quantitative data, including GMC.

2.4.4. Discussion on clinical pharmacology

Since Heplisav B comprises 2 components, HBsAg and 1018 ISS, that are not adsorbed or linked together, these two components are expected to exhibit pharmacokinetic properties of the individual components.

HBsAg in Heplisav B is expected to exhibit properties similar to those of currently licensed vaccines (i.e. distribution from the injection site to the draining lymph node) and therefore, no formal clinical PK data on the antigen were considered needed.

Distribution of HBsAg into the plasma has been reported in Heplisav B, similarly to other currently licensed hepatitis B vaccines. No safety concerns were raised.

Results of the two small size studies that assessed the PK properties of the 1018 ISS suggested that there is no risk of 1018 ISS accumulation, since there is a one-month interval between vaccine doses. There was no evidence of accumulation of the adjuvant in the preclinical studies that were conducted. The PK properties of the adjuvant have been reflected in the Product Information.

The choice of a CKD population to study the absorption of 1018 ISS was considered appropriate, since there appeared to be a potential for changes in the PK of the 1018 ISS component of Heplisav B in individuals with renal disease. PK properties of 1018 ISS were assessed in a small number of individuals. There is no data available in healthy volunteers and in special populations other than CKD patients. This was considered acceptable by the CHMP, as PK is not as informative for the overall safety and efficacy profile of the vaccine.

Distribution, metabolism and excretion have been well characterized for PS ODNs such as 1018 ISS and are highly similar among PS ODN of various nucleotide sequences.

No additional bioavailability or bioequivalence studies have been conducted, which is acceptable since the bioavailability of Heplisav B was assessed indirectly through the pharmacodynamic studies.

Pharmacodynamic studies were essentially comprised of the immunogenicity studies that characterise the immune response to the vaccine. Similarly to the development of other hepatitis B vaccines, the Heplisav B clinical development programme used seroprotection, defined as serum concentrations of anti-HBs ≥10 mIU/mL, as the indicator of clinical efficacy and the basis for authorisation.

The choice of 1018 ISS as adjuvant is relevant in that the desired biological activity of 1018 ISS is to enhance the generation of antibodies to HBsAg by mimicking the immunostimulatory activity of single-stranded viral and bacterial DNA. 1018 ISS is thought to activate pDCs through TLR-9, which in turn will promote Th1 CD4+ T cell differentiation. This activation results in a high and sustained antibody response, likely due to the rapid generation of large numbers of anti-HBs-secreting plasmacytes and HBsAg-specific memory B cells. *In vitro* findings confirmed that 1018 ISS activates PBMC to proliferate and secrete Th1 cytokines.

Seroprotection rates can be compared between all studies, since a positive assay result in either the AUSAB or the Hep B ECi assays corresponds to the anti-HBs concentration of 10 mIU/mL. Because of slight variation of Ab concentration, GMC comparison between both kits is not possible. The CHMP considered that the same assay (ECi) was utilized in the 3 pivotal trials (HBV-23, HBV-16, HBV-10) which allows for a pooled analysis.

2.4.5. Conclusions on clinical pharmacology

The CHMP considered that all aspects dealing with clinical pharmacology have been well addressed by the Applicant.

2.5. Clinical efficacy

2.5.1. Dose response studies

Different doses of adjuvant 1018 and dosing schedules of Heplisav B were given in four studies: HBV0001 and HBV-008 in healthy adults, and HBV-009 and HBV-011 in chronic kidney disease patients.

HBV0001 was a phase I study in healthy adults. Two doses of 20 µg HBsAg was given together with 0, 300, 650, 1000 or 3000 µg 1018 ISS to groups of 8 subjects. The sample size was very small, but the immune responses were shown to be higher in the higher doses compared to the lower. Likewise, a higher frequency of local reactions was reported following the highest dose of 1018 ISS compared to the lower doses.

HBV-008 was a phase II study in young healthy adults who received Heplisav (20 µg HBsAg and 3000 µg 1018 ISS) at 0, 4 or 0, 8 weeks, or a half dose of Heplisav at 0, 4 weeks. No significant differences between the 0, 4 and 0, 8 weeks schedule were seen at week 4 after the last dose. The responses to a half dose were significantly lower than the full dose at week 4 after the first dose, but no significant difference at later time points. No differences in safety profile were seen between dose schedules of full and half dose.

HBV-009 was a phase I study in end stage renal failure patients. Three doses of 1018 ISS + HBsAg (10µg HBsAg +1500 µg 1018 ISS, 20 µg HBsAg + 3000 µg 1018 ISS or 40 µg HBsAg + 6000 µg 1018 ISS) were given at 0, 4, 24 weeks and a control group was given double doses of Engerix-B at 0, 4, 8, 24 weeks. The trial was terminated early due to GCP issues at several sites. The highest dose was not given in this study. The results of the two lower doses showed superior immune responses compared to Engerix-B, and an acceptable safety profile.

HBV-011 was a phase II study in adults with CKD. The subjects were given three doses of Heplisav either 20 µgHBsAg + 3000µg 1018 ISS or 40 HBsAg+6000µg 1018 ISS at 0, 4, 24 weeks. A clinical hold of Heplisav interrupted the study and a majority of subjects only received two doses. However, the SPR after the high dose was higher compared to the low dose at all time points. Likewise, the reactogenicity was increased in the high dose group compared to the low dose.

The dose and dose schedule in the pivotal phase III trials was based on the above studies.

2.5.2. Main studies

Methods

Three pivotal studies including generally healthy adults were included in the submission, HBV-010 and HBV-016 and HBV-023.

HBV-010: A Phase III Safety and Efficacy Study to Compare Immune Responses Following Injection with Either Two Doses of Heplisav or Three Doses of Engerix B

HBV-016: An Observer-Blinded, Randomized, Parallel-Group, Multi-Center Study Comparing the Safety and Immunogenicity of Heplisav to Licensed Vaccine (Engerix B) Among Healthy Subjects 40 to 70 Years of Age

HBV-023: A Phase 3, Observer-Blinded, Randomized, Active-Controlled (Engerix B), Multicenter Trial of the Safety and Immunogenicity of Heplisav in Adults 18 to 70 Years of Age

Study Participants

Pivotal studies included generally healthy adults without prior HBV infection or hepatitis B vaccination. HBV-010 included subjects 11-55 years (the results are presented for the 18-55 year olds) and HBV-016 included 40-70 year old subjects. HBV-023 included subjects aged 18-70 years. Subjects with and without type 2 diabetes mellitus were included.

HBV-010 was conducted in Canada and Germany, HBV-016 was conducted in USA and Canada and HBV-023 in USA.

Treatments

In all three studies, the test product Heplisav (20 µgHBsAG combined with 3000 µg 1018 ISS) was administered as a single intramuscular injection (0.5 ml) into the right or left deltoid muscle at Weeks 0 and 4. In all pivotal studies the comparator was Engerix-B (20 mcg recombinant HBsAg combined with 0.5 mg alum adjuvant/mL) given as a single intramuscular injection (1.0 mL) into the right or left deltoid muscle at Weeks 0, 4, and 24.

Objectives

HBV-010

Primary: To compare the seroprotection rate (SPR) at Week 12 following injection at Weeks 0 and 4 to the SPR at Week 28 following injection with Engerix-B at Weeks 0, 4 and 24. SPR was defined as the percentage of subjects achieving seroprotection (antibody to hepatitis B virus surface antigen [anti-HBsAg] ≥ 10 mIU/mL).

Secondary: To compare the SPR for 1018 ISS-HBsAg versus Engerix-B at Week 4.

Exploratory: To compare the SPR for 1018 ISS-HBsAg versus Engerix-B at Weeks 8, 12, 24, and 28; to describe the serum geometric mean concentrations (GMC) for 1018 ISS-HBsAg and Engerix-B at Weeks 4, 8, 12, 24, and 28; and to compare the SPR at Week 8 for 1018 ISS-HBsAg versus Week 28 for Engerix-B.

Safety: To demonstrate the safety and tolerability of 1018 ISS-HBsAg when administered to adolescent and adult subjects.

HBV-016

Primary: To demonstrate the noninferiority of the immune response to Heplisav vaccination as measured by seroprotection rate (SPR) defined as antibody against hepatitis B surface antigen (anti-HBsAg) ≥ 10 mIU/mL at 8 weeks after the last active dose (Week 12) compared to the SPR for Engerix-B vaccination at 8 weeks after the last active dose (Week 32).

To demonstrate lot consistency for immune response as measured by geometric mean concentration (GMC) at 4 weeks after the last active dose (Week 8) among 3 consecutively manufactured lots of Heplisav from the manufacturing process after minor modification

Secondary:

- to demonstrate the safety of Heplisav in healthy subjects 40 to 70 years of age and to compare the safety profile of Heplisav to that of Engerix-B in this population
- to demonstrate the superiority of the immune response after Heplisav vaccination as measured by SPR at 8 weeks after the last active dose ONLY if it is established that Heplisav is noninferior to Engerix-B
- to demonstrate lot consistency for immune response as measured by SPR at 4 weeks after the last active dose (Week 8) among 3 consecutively manufactured lots of Heplisav from the manufacturing process after minor modification
- to demonstrate consistency of immune response at 4 weeks after the last active dose (Week 8) between Heplisav lots prior to and after minor modifications to the manufacturing process
- to evaluate the immune response to Heplisav vaccination as measured by SPR at 8 weeks after the last active dose (Week 12) compared to Engerix-B vaccination at 8 weeks after the last active dose (Week 32) in subjects with a history of type 2 diabetes mellitus on enrollment
- to evaluate the immune response to Heplisav vaccination as measured by the percentage of subjects with anti-HBsAg ≥ 100 mIU/mL compared to Engerix-B vaccination at 8 weeks after the last active dose.

HBV-023

Primary:

- to evaluate the overall safety of Heplisav B with respect to clinically significant events
- to demonstrate the noninferiority of the immune response to Heplisav B to that of Engerix-B as measured by the SPR at 28 weeks in subjects with type 2 diabetes mellitus.

Secondary:

- to demonstrate that the SPR induced by Heplisav B at 28 weeks is statistically significantly higher than the SPR in Engerix-B subjects with type 2 diabetes mellitus, ONLY if it is established that the Heplisav B SPR is noninferior to Engerix-B SPR at 28 weeks;
- to demonstrate that the SPR induced by Heplisav B at 24 weeks is noninferior to the SPR of Engerix-B at 28 weeks in all subjects and by age group, sex, BMI, and smoking status
- to demonstrate that the SPR induced by Heplisav B at 24 weeks is statistically significantly higher than the SPR of Engerix-B at 28 weeks in all subjects by age group, sex, BMI, and smoking status, ONLY if it is established that the Heplisav B SPR is noninferior to the Engerix-B SPR.

Outcomes/endpoints

Immunogenicity assessments: Anti-HBsAg serum concentrations were measured using the Ortho Vitros enhanced chemiluminescence immunoassay (Ortho Vitros Package Insert; Vitros Version 2.1). Seroprotection was defined as anti-HBsAg serum concentration ≥ 10 mIU/mL.

HBV-010

Primary: the SPR measured at Week 12 after vaccination with 1018 ISS-HBsAg was, and measured at Week 28 after vaccination with Engerix-B.

Secondary: The exploratory endpoints were SPR at Weeks 8, 12, 24, and 28 and GMC at Weeks 4, 8, 12, 24, and 28 for both treatment groups.

HBV-016

Primary: the SPR measured at Week 12 after vaccination with 1018 ISS-HBsAg was, and measured at Week 32 after vaccination with Engerix-B. In HBV-016 the other primary endpoint was lot consistency in 3 consecutively manufactured lots of Heplisav from the manufacturing process after minor modification, measured by GMC at 4 weeks after the last active dose of Heplisav (Week 8)

Secondary: SPR, percentage of subjects with HBsAg ≥100 mIU/mL, and GMC measured at Weeks 4, 8, 12, 18, 24, 28, 32, 36, 44, and 52 for subjects treated with Heplisav and subjects treated with Engerix-B. Lot consistency between lot TDG006 and the Heplisav consistency lots measured by GMC at 4 weeks after the last active dose of Heplisav (Week 8)

HBV-023

Primary: Proportion of subjects with new-onset MAEs, SAEs or deaths, AESIs, AESIs + AIAEs. SPR at Week 28 in subjects with type 2 diabetes mellitus

Secondary: SPR at Week 24 in Heplisav subjects and at Week 28 in Engerix-B subjects

Randomisation

Randomisation. In all pivotal studies study personnel used an interactive voice and web response system (IVRS/IWRS) to obtain a screening number, subject number, and kit number for each subject. In study HBV-023, enrollment was stratified by site, age group (18 to 39, 40 to 70 years), and type 2 diabetes mellitus status, and site. Subjects were randomly assigned in a 2:1 ratio to receive Heplisav or Engerix-B. In study HBV-010, the randomization scheme stratified subjects according to their age (11 through 39 years vs 40 through 55 years). Subjects were randomly assigned to study treatment in a 3:1 fashion (1018 ISS-HBsAg: Engerix-B). In study HBV-016, randomization was stratified by age (ages 40 to 49 years, 50 to 59 years, 60 to 70 years), allocation ratio of Heplisav to Engerix-B was 4:1. For the primary objective of noninferiority, the allocation ratio of the 3 consistency lots to Engerix-B was 3:1. For the primary objective of lot consistency, the allocation ratio was 1:1:1.

Blinding (masking)

All pivotal studies used observer-blind study design. To mask the difference in the number of injections, a placebo injection was given in the Heplisav B group at Week 24. The differences in appearance and volumes of the vaccines injected were not blinded.

Statistical methods

HBV-023

Analysis Populations

- *Screened Population*: all subjects who consented to participate in the study and were screened for eligibility
- *Randomized Population*: all subjects who were considered eligible at the time of enrollment and were randomized into the study
- *Safety Population*: all subjects who received at least 1 injection of study drug, excluding subjects who had no on-study safety data
- *Per-Protocol (PP) Population*: all randomized subjects who received all study injections, had no major protocol deviations, and had anti-HBs levels obtained within the protocol-defined study visit window at Week 28. The PP population was the primary analysis population for all immunogenicity analyses
- *modified Intent-to-Treat (mITT) Population*: all randomized subjects who received at least 1 study injection and had at least 1 post-injection immunogenicity evaluation. The mITT population was used for supportive and confirmatory immunogenicity analyses
- *Laboratory Substudy Safety Population*: a subset of the safety population for subjects with at least 1 post-baseline laboratory assessment and who enrolled at the 2 sites that conducted the laboratory substudy

Safety. All safety data were analyzed descriptively based on the Safety Population. Summary statistics were used to describe the incidence of MAEs, AESIs, AESIs + AIAEs, SAEs, and deaths. In addition, concomitant medications (number of patients, non-study vaccinations), thrombotic adverse events, and laboratory measures (renal blood, urine, thrombotic screens, clotting assessment were summarized per treatment group and study visit, respectively.

Immunogenicity. The statistical analysis to compare the randomized treatments regarding the primary immunogenicity endpoint "Seroprotection rate (SPR) at week 28" was conducted in the per-protocol (PP) population in subjects with protocol-defined type 2 diabetes mellitus. The SPR was defined as the percentage of subjects with a serum concentration of anti-HBs ≥10 mIU/mL. Sensitivity analyses based on the modified ITT Population were performed. Heplisav was considered to be non-inferior to Engerix-B if the lower limit of the 95% confidence interval of the difference in SPRs (Heplisav SPR minus Engerix-B SPR) was greater than -10% in the PP population. The difference in SPRs and the 95% confidence limits were calculated under Null hypothesis using the Miettinen and Nurminen method (without stratification). If Heplisav was found to be noninferior in subjects with type 2 diabetes mellitus, then it was to be declared statistically significantly higher than Engerix-B if the lower limit of this confidence interval was greater than 0 (zero). All statistical tests for immunogenicity data were performed at the two-sided 5% level of significance. No adjustments for multiple testing were done for immunogenicity. Dropouts and missing data were assumed to be missing completely at random. No imputations were made for missing data.

Endpoints

- Primary: Proportion of subjects with new-onset MAEs, with new-onset SAEs or deaths, with new-onset AESIs, with new-onset AESIs + AIAEs, respectively, and SPR at Week 28 in subjects with type 2 diabetes mellitus
- Secondary: Proportion of subjects with new-onset GPA or THS, with new-onset thrombotic events, with new-onset abnormal thrombotic screens in the Laboratory Substudy, with new-onset abnormal renal blood or urine tests in the Laboratory Substudy, respectively, and SPR at Week 24 in Heplisav subjects and at Week 28 in Engerix-B subjects

Subgroups analyses were performed on Seroprotection rates (SPR) in several predefined subgroups: Age group, Sex, BMI category, Smoking status, Race.

Sensitivity analyses for the SPR were conducted using logistic regression (or similar) in the PP population including covariates such as duration of diabetes, haemoglobin A1c category, comorbidity as measured by number of diabetes complications, and metformin use, and others.

Amendments. The final Clinical Study Protocol (CSP) as of 2014-05-21 included amendment 1 and two clarification letters. Several minor changes to the Statistical Analysis Plan (SAP, Version 2.0) as of 2015-03-13 were applied. The original and final Clinical Study Report (CSR) was dated as of 2016-03-01 without amendments.

HBV-010

Analysis Populations

- *Immunogenicity, per-protocol (PP) population (primary)*: subjects who met the eligibility criteria, did not violate the protocol in a substantial manner, received all protocol-specified study injections, had anti-HBsAg measurements and all injections within the specified day ranges, and had an anti-HBsAg measurement at their primary endpoint
- *Immunogenicity, modified intent-to-treat (ITT) population*: subjects who received at least 1 study injection and had at least 1 post-baseline anti-HBsAg level.
- *Safety population*: subjects who received at least 1 study injection and had any post-baseline safety data; subjects were included in the treatment group corresponding to the study treatment they actually received for the analysis of safety data; used for safety analysis

General statistical methods for all endpoints (if not stated otherwise)

- Summary statistics included 95% confidence intervals (CIs) for all estimates.
- Differences in categorical variables between treatment groups were tested using a Cochran-Mantel-Haenszel test, which was stratified for categorical age.
- Differences in continuous variables between treatment groups were tested with an analysis of variance model containing fixed effects for treatment and age.

Immunogenicity

The statistical analysis compared the Heplisav and ENGERIX-B groups regarding the difference (ENGERIXB – Heplisav) in the primary endpoint "seroprotection rate after the final active injection" in the PP population. The primary SPR as the proportion of subjects with achieving seroprotection (anti-HBsAg≥ 10 mIU/mL) for Heplisav was measured at Week 12, and for Engerix-B was measured at Week 28. Sensitivity Analysis in the ITT population was performed. The 95% CI on this difference the Miettinen and Nurminen method (Miettinen and Nurminen 1985) to compute the confidence limits was used. Superiority was inferred if the upper limit of 95% confidence interval was less than 0 (ad-hoc test). All immunogenicity analyses used one-sided tests at the 2.5% level of significance. No further adjustment for multiple testing was performed, because only one primary endpoint was studied. Anti-HBsAg levels that were below the lower limit of detection and reported as < 5 mIU/mL were considered as 2.5 mIU/mL. There was no imputation of missing anti-HBsAg data at any visit.

Missing data

In the computation for GMC, anti-HBsAg levels that were below the lower limit of detection and reported as < 5 mIU/mL were considered as 2.5 mIU/mL. There was no imputation of missing anti-HBsAg data at any visit. In computing the SPRs for the ITT population, a subject who had a missing anti-HBsAg result at a given time point was considered as having missing seroprotective immune response and was excluded at that time point. If a subject had a missing anti-HBsAg result at a primary endpoint, then the subject was excluded from the PP population.

Safety

The statistical analysis compared the Heplisav and ENGERIX-B groups regarding safety and tolerability endpoints in the Safety Analysis population. The safety endpoints included: Solicited post-injection local and systemic reactions, AEs, SAEs, Clinical laboratory results including ANA and anti-ds DNA, Vital signs results (systolic blood pressure [mmHg], diastolic blood pressure [mmHg], heart rate [bpm], respiratory rate [breaths per min], and oral temperature [°C]), concomitant medications and vaccinations. Descriptive statistical methods including summary statistics, measures of effect size, and 95% confidence intervals were calculated as appropriate. All statistical tests comparing safety data were two-sided and conducted at the 5% level of significance.

Endpoints

Primary Endpoint: seroprotection rate after the final active injection of Heplisav at Week 12 and of Engerix-B at Week 28.

Secondary Endpoints:

- Immunogenicity:

- The secondary immunogenicity endpoint was the SPR measured 4 weeks after the first injection (onset of response) for both treatment groups.
- SPR at Weeks 8, 12, 24, and 28 and GMC at Weeks 4, 8, 12, 24, and 28 for both treatment groups.
- SPR 4 weeks after the final active injection (Week 8 for the 1018 ISS-HBsAg group and Week 28 for the Engerix-B group).

Subgroups

Descriptive immunogenicity results were summarized by age stratum, by country, and by site for each visit. Solicited post-injection local and systemic reactions and AEs and were summarized separately, each by treatment group and age stratum for subjects.

HBV-016

Analysis Populations

- Screened Population: all subjects who consented to participate in the study and were screened for eligibility assessment.
- Randomized Population: included all subjects who were randomized into the study.
- Safety population: all subjects who received at least 1 study injection, excluding subjects who had no on-study safety data.
- Modified intent-to-treat (mITT) Population for the immunogenicity analysis: all randomized subjects who received at least 1 study injection and had at least 1 post-injection immunogenicity evaluation.
- PP populations were analyzed in this study:
    - Noninferiority PP population: This PP population was used for the primary objective of noninferiority and comprised all randomized subjects who received 1 of the 3 consistency lots of Heplisav or Engerix-B, received all 3 study injections as randomized and within the study visit windows, had no major protocol deviations, and had anti-HBs levels obtained within study visit windows at baseline, Week 12, and Week 32.
    - Lot consistency PP population: This PP population comprised all subjects randomized to 1 of 3 consistency lots of Heplisav (TDG008, TDG009, and TDG010) who received the first 2 study injections within the study visit windows, had no major protocol deviations and had anti-HBs levels obtained within study visit windows at baseline and Week 8.
    - Bridging study PP population: This PP population comprised all subjects randomized to lot TDG006 or to 1 of 3 consistency lots of Heplisav (TDG008, TDG009, and TDG010) concurrently with lot TDG006 who received the first 2 study injections within the study visit windows, had no major protocol deviations and had anti-HBs levels obtained within study visit windows at baseline and Week 8.

Immunogenicity analysis

Noninferiority

The statistical analysis compared the Heplisav group with the ENGERIX-B group regarding the primary immunogenicity endpoint "Seroprotection Rate at 8 week after the last active dose (Heplisav: Week 12,

ENGERIX-B: Week 32)" in the Noninferiority PP population. Seroprotection Rate was defined as the proportion of patients with an anti-HBs serum concentration ≥ 10 mIU/mL. Heplisav was to be considered non-inferior to Engerix-B if the lower limit of the 2-sided 95% confidence interval (CI) of the difference in SPR (Heplisav SPR for the 3 combined consistency lots at Week 12 minus the Engerix-B SPR at Week 32) was greater than -10%. If Heplisav was found to be noninferior and the lower limit of this CI was also greater than 0%, then, and only then, was Heplisav to be declared superior to Engerix-B.

Lot Consistency

For analysis of the primary objective of lot consistency, the 95% CIs for the 3 pair-wise ratios of the GMCs from the consistency lots (TDG008, TDG009, and TDG010) at Week 8 were computed in the lot consistency PP population. Lot consistency was established if all 3 CIs were embedded in the interval between 0.667 and 1.5. For the secondary objective of lot consistency, the 95% CIs for the 3 pair-wise differences in SPRs from the consistency lots at Week 8 were computed. The 95% CIs for the 3 pair-wise differences in SPR were also computed. Lot consistency was established based on SPR if all 3 CIs were embedded entirely between -10% and 10%.

Bridging Analysis

Criteria for comparison of lot TDG006 and the 3 combined consistency lots were based on 2-sided 95% CIs for GMC and SPR in the bridging study PP population, as noted above.

All statistical tests were performed at the 2-sided 5% level of significance. No adjustments for multiple testing were done for immunogenicity. All tests of non-inferiority based on SPRs assumed a non-inferiority margin of 10%. No imputations were made for missing immunogenicity data.

Safety analysis

All safety data were analysed descriptively and were based on the safety population. Summary statistics were used to describe extent of exposure, autoimmune AEs, solicited post-injection reactions, AEs (excluding solicited post-injection reactions), and vital signs. No statistical testing was performed on safety data. Missing AEs or concomitant medication start dates were imputed using conservative rules. Safety endpoints included solicited post-injection reactions (reactogenicity), unsolicited adverse events, deaths, serious adverse events, other significant adverse events and clinical laboratory variables, vital signs, physical findings and other observations related to safety.

Endpoints

Primary: 1) SPR, defined as the percentage of subjects with anti-HBs serum concentration ≥ 10 mIU/mL, measured at 8 weeks after the last active dose of Heplisav or Engerix-B (Week 12 vs Week 32, respectively); 2) Lot consistency in 3 consecutively manufactured lots of Heplisav from the manufacturing process after minor modification, measured by GMC at 4 weeks after the last active dose of Heplisav (Week 8)

Secondary: Incidence of post-injection reactions, Incidence of AEs in subjects treated with Heplisav vs Engerix-B, Incidence of new-onset autoimmune disease in subjects treated with Heplisav vs Engerix-B, Percentage of subjects with anti-HBs serum concentration ≥ 100 mIU/mL, measured at 8 weeks after the last active dose of Heplisav or Engerix-B (Week 12 vs Week 32, respectively), SPR, percentage of subjects with HBsAg ≥100 mIU/mL, and GMC measured at Weeks 4, 8, 12, 18, 24, 28, 32, 36, 44, and 52 for subjects treated with Heplisav and subjects treated with Engerix-B, Lot consistency between lot TDG006 and the Heplisav consistency lots measured by GMC at 4 weeks after the last active dose of Heplisav (Week 8)

Subgroups

Subgroups were defined a-priori "SPR, GMC, and anti-HBs ≥ 100 mIU/mL in subjects with type 2 diabetes mellitus" or post-hoc including SPR, GMC, and anti-HBs ≥ 100 mIU/mL by age stratum, in subjects with HCV, by gender, by BMI, by smoking status.

Interim Analysis

A preliminary analysis was performed to assess immunogenicity and safety, including post-injection local and systemic reactions, AEs, SAEs, and AIAEs when all PP subjects completed their Week 32 visit for the primary immunogenicity endpoint assessment. This analysis was based on the PP population and was final for the evaluation of the primary immunogenicity objectives. Results for individual subjects were not made available to the study monitoring personnel, investigators, or the subjects. Because no additional vaccinations were given after Week 32, the results of these analyses did not alter the course of the trial.

Results

Participant flow

Table 7 Subjects Disposition in the Pivotal Trials

Disposition	HBV-23		HBV-16		HBV-10		Total Heplisav B n (%)	Total Engerix-B n (%)
	Heplisav B n (%)	Engerix-B n (%)	Heplisav B n (%)	Engerix-B n (%)	Heplisav B n (%)	Engerix-B n (%)		
Randomized (N)	5592	2782	1969	483	1809	606	9370	3871
Completed	5092 (91.1)	2567 (92.3)	1818 (92.3)	451 (93.4)	1746 (96.5)	588 (97.0)	8656 (92.4)	3606 (93.2)
Discontinued	500 (8.9)	215 (7.7)	151 (7.7)	32 (6.6)	63 (3.5)	18 (3.0)	714 (7.6)	265 (6.8)
Adverse Event	4 (0.1)	0	1 (0.1)	0	2 (0.1)	2 (0.3)	7 (<0.1)	2 (<0.1)
Subject noncompliance	7 (0.1)	1 (<0.1)	6 (0.3)	3 (0.6)	3 (0.2)	2 (0.3)	16 (0.2)	6 (0.2)
Consent withdrawn	100 (1.8)	39 (1.4)	45 (2.3)	12 (2.5)	18 (1.0)	2 (0.3)	163 (1.7)	53 (1.4)
Lost to follow-up	319 (5.7)	153 (5.5)	81 (4.1)	13 (2.7)	30 (1.7)	10 (1.7)	430 (4.6)	176 (4.5)
Death	25 (0.4)	7 (0.3)	1 (0.1)	1 (0.2)	0	0	26 (0.3)	8 (0.2)
Protocol deviation	1 (<0.1)	0	3 (0.2)	1 (0.2)	2 (0.1)	0	6 (<0.1)	1 (<0.1)
Other	44 (0.8)	15 (0.5)	14 (0.7)	2 (0.4)	8 (0.4)	2 (0.3)	66 (0.7)	19 (0.5)
PP Population	4537 (81.1)	2289 (82.3)	1121 (77.8)[a]	353 (73.1)	1511 (83.5)	521 (86.0)	Not applicable	Not applicable
mITT Population	5278 (94.4)	2635 (94.7)	1947 (98.9)[b]	476 (98.6)	1789 (98.9)	603 (99.5)	9014 (96.2)	3714 (95.9)
mITT Population for HBV-16 noninferiority immunogenicity analysis	-	-	1426 (99.0)	476 (98.6)	-	-	-	-

Baseline data

Baseline characteristics were balanced between vaccine groups within groups. The groups were sufficiently balanced regarding age, sex, race, ethnicity and BMI.

The study population reflected the intended indication. As the pivotal studies included subjects with different age range, the median age of the subjects differs between the studies. Furthermore, as HBV-023 focused on

persons with type 2 diabetes, one can see higher proportion of subjects with this diagnosis than in other studies. Higher BMI and higher proportion of subjects of black race may be result of recruiting subjects with type 2 diabetes or that this study was conducted exclusively in the USA.

Study HBV-023 covered the highest age range (18-70) in comparison to other studies, where investigator focused more on older population (HBV-016; 40-70 years) or younger population (HBV-010; 18-55 years) Higher age of the subjects resulted in HBV-016 having less smokers, higher BMI and higher proportion of subjects with type 2 diabetes in comparison to the HBV-010, where younger population was included.

Numbers analysed

Pivotal studies analysed sufficient numbers of subjects to evaluate efficacy and safety. Per-protocol population was the most important to evaluate efficacy and included altogether 10322 subjects (7169 subjects receiving Heplisav B and 3163 Engerix-B).

Table 8 Number of subjects in each pivotal study

Trial **Subgroup**	**HBV-23** **18 to 70 Years**		**HBV-16** **40 to 70 Years**		**HBV-10** **18 to 55 Years**	
	Heplisav B n (%) (N = 5592)	**Engerix-B n (%) (N = 2782)**	**Heplisav B n (%) (N = 1441)**	**Engerix-B n (%) (N = 483)[a]**	**Heplisav B n (%) (N = 1809)**	**Engerix-B n (%) (N = 606)**
Disposition	**n (%)**	**n (%)**	**n (%)**	**n (%)**	**n (%)**	**n (%)**
Randomized Population	5592	2782	1441	483	1809	606
mITT Population[b]	5278 (94.4)	2635 (94.7)	1426 (99.0)	476 (98.6)	1789 (98.9)	603 (99.5)
PP Population[a]	4537 (81.1)	2289 (82.3)	1121 (77.8)	353 (73.1)	1511 (83.5)	521 (86.0)

mITT = modified Intent-to-Treat; N = number of subjects in that population; n = number of subjects in the group; PP = Per-protocol.
Subjects who discontinued early from the study are included. The denominator for percentages is N.

[a] Data are shown for subjects who were included in Noninferiority PP immunogenicity analysis in HBV-16 and therefore received one of the Heplisav B consistency lots (Lot TGD008, TDG009, or TDG010) or Engerix-B.

[b] The mITT Population comprised subjects who received at least 1 study injection and had at least 1 post-injection immunogenicity evaluation.

Outcomes and estimation

Seroprotection Rates

Seroprotection rates in the pivotal trials are presented in Table 15 and Table 16 below.

Table 9 Seroprotection Rates at the Primary Immunogenicity Endpoint in 2 Pivotal Phase 3 Trials (HBV-16 and HBV-10) (PP Population)

Trial	**HBV-16**			**HBV-10**		
	40 to 70 Years			**18 to 55 Years**		
	Heplisav B[a]	**Engerix-B[b]**	**Difference in SPRs (%) (Heplisav-**	**Heplisav B[a]**	**Engerix-B[b]**	**Difference in SPRs (%) (Heplisav-**
Visit	**SPR (%)**	**SPR (%)**		**SPR (%)**	**SPR (%)**	

Trial	HBV-16			HBV-10		
	40 to 70 Years			18 to 55 Years		
	(95% CI)[c] (N = 1121)	(95% CI)[c] (N = 353)	Engerix-B) (95% CI)[d]	(95% CI)[e] (N = 1511)	(95% CI)[e] (N = 521)	Engerix-B) (95% CI)[f]
Week 12[g]/32[h]	90.1 (88.2, 91.8)	70.5 (65.5, 75.2)	19.6 (14.7, 24.8)	NA	NA	NA
Week 12[g]/28[h]	NA	NA	NA	95.0 (93.9, 96.1)	81.2 (77.8, 84.6)	13.7 (10.4, 17.5)

CI = confidence interval; N = number of subjects in the analysis population in the group; NA = not applicable because Week 28 was not the primary endpoint in HBV-16 and HBV-10 concluded at Week 28; PP = Per-protocol; SPR = seroprotection rate.
The PP Population for the Heplisav B group in HBV-16 is the Noninferiority PP Population excluding subjects who received Lot TDG006.

[a] Study injections were given at Weeks 0, 4, and 24 (placebo).
[b] Study injections were given at Weeks 0, 4, and 24.
[c] 95% CIs were calculated using the Clopper-Pearson method.
[d] Two-sided 95% CIs of the difference in SPRs between the Heplisav B group at 12 weeks and the Engerix-B group at 32 weeks were calculated using the Newcombe score method with continuity correction.
[e] The 95% CIs for the SPR were calculated using the normal approximation to the binomial.
[f] Estimated response (proportion), their differences, and associated CIs are based on a statistical analysis model adjusting for age groups (18 through 39 years versus 40 through 55 years). The stratified Miettinen and Nurminen method was used to calculate the 95% CIs.
[g] Heplisav B.
[h] Engerix-B.

Table 10 Seroprotection Rates in Subjects With Type 2 Diabetes Mellitus at the Primary Immunogenicity Endpoint in the Pivotal Phase 3 Trial HBV-23 (PP Population)

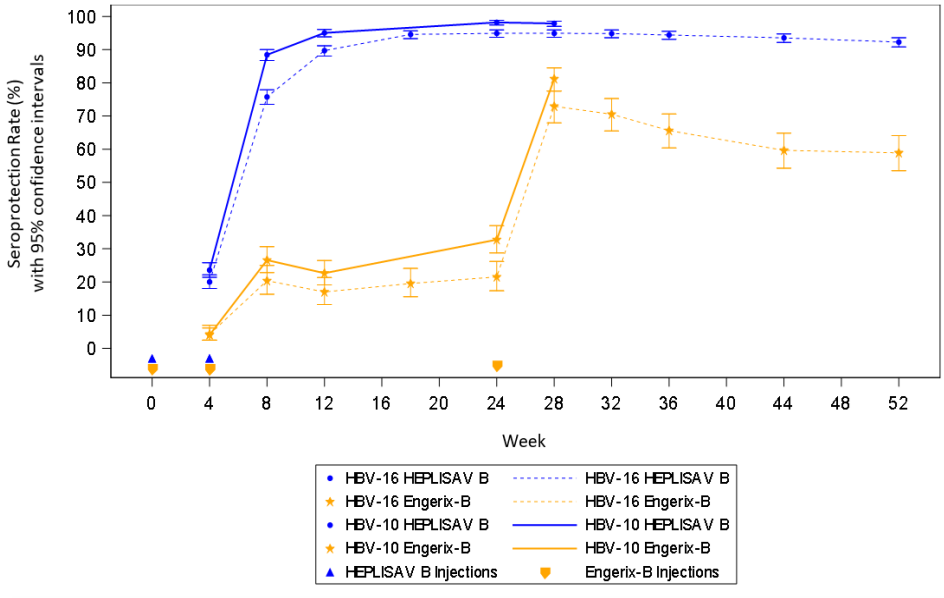
Heplisav B[a]			Engerix-B[b]			Difference
N	n	SPR (%) (95% CI)[c]	N	n	SPR (%) (95% CI)[c]	(Heplisav B - Engerix-B) (95% CI)[d]
640	576	90.0 (87.4 - 92.2)	321	209	65.1 (59.6 – 70.3)	24.9 (19.3 - 30.7)

CI = confidence interval; N = number of subjects in the analysis population in the group; n = number of subjects with post-injection anti-HBs ≥ 10 mIU/mL; PP = Per-protocol; SPR = seroprotection rate.

[a] Study injections were given at Weeks 0, 4, and 24 (placebo).
[b] Study injections were given at Weeks 0, 4, and 24.
[c] 95% CIs were calculated using the Clopper-Pearson method.
[d] 95% CI was calculated using the Miettinen and Nurminen method.

SPR results were collected at each study visit (week 4 to 28) in two of the pivotal trials, HBV-10 and HBV-16. Heplisav B induced significantly higher SPRs than Engerix-B across all study visits in both studies Figure 2). Results from HBV-23 are not included in Figure 1 as results were collected at weeks 24 and 28, but not at weeks 4, 8, nor 12.

Figure 1 Seroprotection Rates by Visit in Trials HBV-16 and HBV-10 (Per Protocol Population)



Geometric Mean Concentration of Anti-HBs in Pivotal Trials

In subjects in the pivotal trials, the antibody response to Heplisav B based on GMC had a similar pattern as that based on SPR. The GMC in the Heplisav B group peaked at Week 24, the same time as the peak SPR. The GMCs in the Heplisav B group increased earlier and peaked at a higher level than the GMCs in the Engerix-B group. Geometric mean concentration (GMC) of anti-HB antibodies are presented in tables x, y and z below.

Summary of main studies

The following tables (Tables 17-19) summarise the efficacy results from the main studies supporting the present application. These summaries should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment.

Table 11 Overview of pivotal trial HBV-023

Title: HBV-023 A Phase 3, Observer-Blinded, Randomized, Active-Controlled (Engerix-B), Multicenter Trial of the Safety and Immunogenicity of Heplisav in Adults 18 to 70 Years of Age		
Study identifier	US IND Number: BB-IND 12,692	
Design	Observer-Blinded, Randomized, Active-Controlled (Engerix-B), Multicenter Trial	
	Duration of main phase: Duration of Run-in phase: Duration of Extension phase:	18 April 2014-16 October 2015 not applicable not applicable
Hypothesis	Non-inferiority (SPR difference % lower limit of 95 % CI more than -10), significance (SPR difference % lower limit of 95 %CI more than 0)	
Treatments groups	Heplisav B	Heplisav B , 28 weeks, 5592 randomized

	Engerix-B		Engerix-B. 28 weeks, 2782 randomized
Endpoints and definitions	Primary endpoint	Safety	All AEs and SAEs (not described in Efficacy section)
	Primary immunogenicity endpoint	Immunogenicity among subjects with type 2 diabetes mellitus	Non-inferior seroprotection rate (SPR) and anti-HepAg antibody geometric mean concentrations (GMC) among Heplisav B in comparison to Engerix-B
	Secondary endpoint	Immunogenicity among entire study population and in subgroups	Non-inferior seroprotection rate, anti-HepAg antibody geometric mean concentrations (GMC) among Heplisav B in comparison to Engerix-B

Results and Analysis

Analysis description	**Primary Analysis**			
Analysis population and time point description	**Per protocol-** all randomized subjects, who had **type 2 diabetes** and who received all study injections, had no major protocol deviations, and had anti-HBs levels obtained within the protocol-defined study visit window at Week 28			
Descriptive statistics and estimate variability	Treatment group	Heplisav B	Engerix-B	
	Number of subject	640	321	
	Seroprotection rate SPR %	90.0	65.1	
	95% CI	87.4-92.2	59.6-70.3	
Effect estimate per comparison	Difference	Heplisav-Engerix		
		SPR%	24.9	
		95% CI	19.3-30.7	
		P-value	Not calculated/presented	
Notes	Heplisav B induced non-inferior and significantly higher seroprotection rate in comparison to Engerix-B			
Geometric mean concentration (GMC) of anti-HB antibodies	Treatment group	Heplisav B	Engerix-B	
	GMC mIU/ml	193.1	75.9	
	95% CI	165.2 - 225.6	54.6 - 105.6	
Effect estimate per comparison	Difference	Heplisav / Engerix		
		GMC ratio	2.5	
		95% CI	1.8 - 3.5	
	Secondary analysis			
Analysis description	**Per protocol-** all randomized subjects who received all study injections, had no major protocol deviations, and had anti-HBs levels obtained within the protocol-defined study visit window at Week 28			
Treatment		Heplisav B	Engerix-B	
Descriptive statistics and estimate variability	Number of subject	4376	2289	
	Seroprotection rate	95.4	81.3	

	95% CI	94.8 – 96.0	79.6 - 82.8	
Effect estimate per comparison	Difference Heplisav-Engerix	SPR%	14.2	
		95% CI	12.5 - 15.9	
		P-value	Not calculated/presented	
Notes	Heplisav B induced non-inferior and significantly higher seroprotection rate in comparison to Engerix-B			
Geometric mean concentration (GMC) of anti-HB antibodies	Treatment group	Heplisav B	Engerix-B	
	GMC mIU/ml	401.0	324.0	
	95% CI	380.0 - 423.2	286.6 - 366.2	
Effect estimate per comparison	Difference	Heplisav / Engerix		
		GMC ratio	1.2	
		95% CI	1.1 - 1.4	

Table 12 Overview of pivotal study HBV-016

Title: HBV-016 An Observer-Blinded, Randomized, Parallel-Group, Multi-Center Study Comparing the Safety and Immunogenicity of Heplisav to Licensed Vaccine (Engerix-B) Among Healthy Subjects 40 to 70 Years of Age			
Study identifier	US IND Number BB-IND 12692 Health Canada CTA Number File no. 9427-D0897-21C; Control No. 135076, 135888, 14405		
Design	Observer-Blinded, Randomized, Active-Controlled (Engerix-B), Multicenter Trial		
	Duration of main phase: Duration of Run-in phase: Duration of Extension phase:	15.02.2010-25.05.2011 not applicable not applicable	
Hypothesis	Non-inferiority (SPR difference % lower limit of 95 % CI more than -10) , superiority, if non-inferiority fulfilled (SPR difference % lower limit of 95 %CI more than 0)		
Treatments groups	Heplisav B, 3 lots		Heplisav B , 52 weeks, randomized 1441
	Engerix-B		Engerix-B. 52 weeks, randomized 483
Endpoints and definitions	Primary endpoint	immunogenicity	Non-inferior seroprotection rate (SPR) among Heplisav B in comparison to Engerix-B
		immunogenicity	Lot consistency of Heplisav (3 lots) to induce antibody titres (GMC)
	Secondary endpoint	safety	Not discussed in this section
Results and Analysis			
Analysis description	**Primary Analysis**		

Analysis population and time point description	**Per protocol-** all randomized subjects, who received all study injections, had no major protocol deviations, and had anti-HBs levels obtained within the protocol-defined study visit window at 8 weeks post active dose.			
Descriptive statistics and estimate variability	Treatment group	Heplisav B Week 12	Engerix-B Week 32	
	Number of subjects	1121	353	
	Seroprotection rate SPR %	90.1	70.5	
	95% CI	88.2-91.8	65.5-75.2	
Effect estimate per comparison	Difference	Heplisav-Engerix		
		SPR%	19.6	
		95% CI	14.7-24.8	
		P-value	Not calculated/presented	
Notes	Heplisav B induced non-inferior and superior seroprotection rate in comparison to Engerix-B			
Lot consistency	Heplisav B 3 lots			
Pre-defined GMC ratio Acceptable if 95% CI is in range of 0.667-1.5	Lot nr.	TDG008	TDG009	TDG010
	Number of subjects	420	424	412
	GMC mIU/ml at week 12	80.3	81.2	89.0
	95% CI	65.4- 98.5	65.8- 100.2	72.0- 109.9
	Difference	008/009	010/008	010/009
	GMC Ratio	0.99	1.11	1.10
	95% CI	0.77- 1.27	0.86- 1.43	0.85- 1.41
Comment	Lot consistency was in acceptable limits since week 12 on, but not earlier			
Exploratory endpoint	Comparison of antibody levels after vaccination with Heplisav or Engerix			
Geometric mean concentration (GMC) of anti-HB antibodies at week 28 (max level)	Treatment group	Heplisav B (N=1120)	Engerix-B (N=350)	
	GMC mIU/ml	230.9	90.9	
	95% CI	208.4- 256.0	60.8- 135.8	
Effect estimate per comparison	Difference	Heplisav / Engerix		
		GMC ratio	2.54	
		95% CI	1.9 - 3.4	

Table 13 Overview of study HBV-010

Title: HBV-010 A Phase III Safety and Efficacy Study to Compare Immune Responses Following Injection with Either Two Doses of Heplisav or Three Doses of Engerix-B	
Study identifier	Health Canada CTA Number File no 9427-D0897-21C/ Control No. 109645 EudraCT 2006-006743-31

Design	Observer-Blinded, Randomized, Active-Controlled (Engerix-B), Multicenter Trial		
	Duration of main phase: Duration of Run-in phase: Duration of Extension phase:		December 2006- March 2008 not applicable not applicable
Hypothesis	Non-inferiority (SPR difference % lower limit of 95 % CI more than -10) superiority, if non-inferiority fulfilled (SPR difference % lower limit of 95 %CI more than 0)		
Treatments groups, duration and N	Heplisav B		Heplisav B , 28 weeks, 1809 randomized
	Engerix-B		Engerix-B. 28 weeks, 606 randomized
Endpoints and definitions	Primary endpoint	Immunogenicity	Seroprotection rate at week 12 (Heplisav) or 28 (Engerix)
	Secondary endpoint	Immunogenicity	Seroprotection rate at week 4
	Exploratory	Immunogenicity	Seroprotection rate, anti-HepAg antibody geometric mean concentrations (GMC) among Heplisav B in comparison to Engerix-B at weeks 4-28
	Safety	Safety	Not described in Efficacy section

Results and Analysis

Analysis description	**Primary Analysis**			
Analysis population and time point description	**Per protocol-** all randomized subjects, who received all study injections and had serology at their primary endpoint			
Descriptive statistics and estimate variability	Treatment group	Heplisav B Week 12	Engerix-B Week 28	
	Number of subject	1511	521	
	Seroprotection rate SPR %	95.0	81.2	
	95% CI	93.9-96.1	77.8-84.6	
Effect estimate per comparison	Difference	Heplisav-Engerix		
		SPR%	13.7	
		95% CI	10.4-17.5	
		P-value	Not calculated/presented	
Notes	Heplisav B induced non-inferior and superior seroprotection rate in comparison to Engerix-B			
Geometric mean concentration (GMC) of anti-HB antibodies at week 28 (max. for Engerix)	Treatment group	Heplisav B	Engerix-B	
	GMC mIU/ml	316.99	352.14	
	95% CI	295.14- 340.45	267.95- 462.78	
Effect estimate per comparison	Difference	Not presented		

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

The SPR and peak geometric mean concentration (GMC) after a 2-dose schedule of Heplisav B were statistically significantly higher than after a 3-dose schedule of Engerix-B (lower bound of the 95% confidence interval of the difference in SPRs between Heplisav B and Engerix-B was greater than 0%; lower bound of the 95% confidence interval of the ratio of GMCs between Heplisav B and Engerix-B was greater than 1.0) in all 3 trials (Table 20, Table 21).

Table 14 Comparison of Seroprotection Rates Between Heplisav B and Engerix-B at Peak Weeks in Pooled Trials HBV-23, HBV-16 and HBV-10 (mITT Population)

Heplisav B			Engerix-B			Difference
N	n	SPR (%) (95% CI)	N	n	SPR (%) (95% CI)	(Heplisav B - Engerix-B) (95% CI)
8701	8327	95.7 (95.3 - 96.1)	3643	2898	79.5 (78.2 - 80.8)	16.2 (14.8 - 17.6)

N = number of evaluable subjects; n = number of seroprotected subjects; SPR = Seroprotection Rate, CI = confidence interval

Table 15 Comparison of Anti-HBs Geometric Mean Concentrations at Peak Weeks Between Heplisav B and Engerix-B in Pooled Trials HBV-23, HBV-16 and HBV-10 (mITT Population)

Heplisav B		Engerix-B		GMC Ratio
N	GMC (95% CI)	N	GMC (95% CI)	(Heplisav B / Engerix-B) (95% CI)
8701	329.1 (317.1 - 341.5)	3642	262.3 (236.4 - 291.1)	1.3 (1.1 - 1.4)

Peak week for Heplisav B is Week 24. Peak week for Engerix-B is Week 28.

Immunogenicity in Selected Populations

Populations that historically have had reduced seroprotection rates from alum-adjuvanted hepatitis B vaccines include older adults, men, obese individuals (BMI greater than or equal to 30 kg/m2), smokers, and subjects with type 2 diabetes mellitus. In all three trials, SPRs induced by Heplisav B were statistically significantly higher than those induced by Engerix-B in each of these populations (Table 22).

Table 16 Comparison of Seroprotection Rates Between Heplisav B and Engerix-B at Peak Weeks by Category in Pooled Trials HBV-23, HBV-16 and HBV-10 (mITT Population)

	Heplisav B			Engerix-B			Difference
Category	N	n	SPR (%) (95% CI)	N	n	SPR (%) (95% CI)	(Heplisav B - Engerix-B) (95% CI)
All subjects	8701	8327	95.7 (95.3 - 96.1)	3643	2898	79.5 (78.2 - 80.8)	16.2 (14.8 - 17.6)
Age Group (years)							
18 - 29	527	526	99.8 (98.9 - 100.0)	211	196	92.9 (88.5 - 96.0)	6.9 (4.1 - 11.2)
30 - 39	1239	1227	99.0 (98.3 - 99.5)	545	483	88.6 (85.7 - 91.2)	10.4 (7.9 - 13.4)
40 - 49	2377	2310	97.2 (96.4 - 97.8)	963	771	80.1 (77.4 - 82.5)	17.1 (14.6 - 19.8)
50 - 59	2712	2578	95.1 (94.2 - 95.8)	1120	872	77.9 (75.3 - 80.3)	17.2 (14.7 - 19.8)
≥ 60	1846	1686	91.3 (90.0 - 92.6)	804	576	71.6 (68.4 - 74.7)	19.7 (16.4 - 23.1)
Sex							
Male	4274	4055	94.9 (94.2 - 95.5)	1765	1361	77.1 (75.1 - 79.1)	17.8 (15.7 - 19.9)
Female	4427	4272	96.5 (95.9 - 97.0)	1878	1537	81.8 (80.0 - 83.6)	14.7 (12.9 - 16.5)
BMI Stratum							
< 30 kg/m^2	4904	4728	96.4 (95.9 - 96.9)	2069	1756	84.9 (83.3 - 86.4)	11.5 (10.0 - 13.2)
≥ 30 kg/m^2	3789	3591	94.8 (94.0 - 95.5)	1570	1140	72.6 (70.3 - 74.8)	22.2 (19.9 - 24.5)
Smoking Status							
Smoker	2634	2538	96.4 (95.6 - 97.0)	1130	852	75.4 (72.8 - 77.9)	21.0 (18.4 - 23.6)
Non-smoker	6067	5789	95.4 (94.9 - 95.9)	2513	2046	81.4 (79.8 - 82.9)	14.0 (12.4 - 15.7)
Type 2 Diabetes Status and Age Group (Years)							
With T2D 20 - 39	38	37	97.4 (86.2 - 99.9)	16	12	75.0 (47.6 - 92.7)	22.4 (5.1 - 47.5)
40 - 49	163	151	92.6 (87.5 - 96.1)	67	49	73.1 (60.9 - 83.2)	19.5 (9.2 - 31.7)
50 - 59	334	303	90.7 (87.1 - 93.6)	160	108	67.5 (59.7 - 74.7)	23.2 (15.6 - 31.4)
≥ 60	377	320	84.9 (80.9 - 88.3)	165	97	58.8 (50.9 - 66.4)	26.1 (17.9 - 34.5)

BMI = body mass index; CI = confidence interval; N = number of evaluable subjects; n = number of seroprotected subjects; SPR = Seroprotection Rate; T2D = type 2 diabetes.
Seroprotection is defined as anti-HBs = 10 mIU/mL.
Peak week comparison is for Heplisav B at Week 24 and Engerix-B at Week 28.
The confidence intervals on seroprotection rates are calculated using the two-sided Clopper-Pearson method.
The confidence interval on the difference between treatment groups is calculated using the Miettinen and Nurminen method without stratification

Haemodialysis

In a phase 3, randomized, open-label, multicentre study of 116 adult subjects with haemodialysis-dependent chronic kidney disease (CKD) who were non-responders to previous hepatitis B vaccination, participants

received a 1-dose booster regimen of Heplisav B or Fendrix, or a double booster dose of Engerix-B.

Week 4 SPR in the Heplisav B group (42.1% n=16/38) was higher than the SPR in the Engerix-B group (18.9%, n=7/37) and the Fendrix group (29.3%, n=12/41). At Week 12, the SPR was 24.3% (n=9/37) in the Heplisav B group, 13.9% (n=11/41) in the Engerix-B group, and 26.8% (n=11/41) in the Fendrix group.

2.5.3. Discussion on clinical efficacy

Design and conduct of clinical studies

The phase 3 pivotal studies (HBV-023, -016 and -010) were adequately designed and randomised, active controlled (Engerix-B) and observer-blind. Double-blind was not possible, as Heplisav B and comparator Engerix-B were visually different (colour and volume). As Engerix-B vaccination requires 3 doses at weeks 0, 4, 24, but Heplisav B two doses at weeks 0 and 4, the subjects receiving Heplisav B were injected with placebo at week 24 to keep blinding.

The selection of study subjects was appropriate considering the intended indication. Generally, the endpoints, timing of samples and the duration of the pivotal studies were considered acceptable by the CHMP. Overall, the pivotal studies were conducted, analysed and reported according to the relevant guidelines.

The efficacy of hepatitis B vaccines based on HBsAg has been shown to correlate to a level of serum antibodies of 10 mIU/mL. Therefore, the clinical development for Heplisav B was designed to demonstrate protective levels of serum antibodies following vaccination. The primary endpoints of the studies were seroprotection rate (proportion of vaccinees, whose antibody levels reach to protective levels at a certain time-point) and Geometric mean concentration (GMC) of antibodies. In each pivotal study, the difference between Heplisav B and comparator along with pre-defined non-inferiority and superiority criteria was presented.

The choice of the time-point when immune response was assessed was not consistent in case of comparator as in two pivotal studies it was week 28 (4 weeks after last dose) and in one study (HBV-016) week 32 (8 weeks after last dose). While there was no rational explanation for this, the CHMP considered that it was acceptable, as it did not influence the overall conclusions. The maximal length of the study in generally healthy population was 52 weeks, which is acceptable. The CHMP considered that the Applicant will conduct longer follow-up studies to investigate durability of the immune response in populations, where immunogenicity might be lower.

In studies including the CKD population, Engerix-B was used as a comparator despite the availability of a more suitable and recommended comparator Fendrix, which is specifically used in the CKD patients. Fendrix was used as a comparator only in the booster study HBV-018 in previously vaccinated subjects. On the other hand, Engerix-B is acceptable as a comparator also in the CKD population. Moreover, it adds to consistency, given that Engerix-B was used in all Heplisav B pivotal trials.

The GCP inspection performed at the time of the previous marketing authorisation application identified concerns with the clinical trial conduct for study HBV-17. Findings included deficiencies in process related study activities such as site selection and monitoring, processes regarding investigational medicinal product (IMP) handling, temperature monitoring, and keeping the study blind. Data from this study were considered to be of questionable quality and the study could not be used for any efficacy claims. The nature of the

findings from the inspection also raised questions about the other main studies. Therefore, there were serious uncertainties at that point in time about the reliability of the data submitted in support of the application.

Due to the GCP concerns found in the previous application, a routine GCP inspection (study HBV-23) was performed in November 2019. Its findings were assessed as having no impact on the completeness, reliability and accuracy of the data reported in this study.

Efficacy data and additional analyses

The total number of subjects that have been vaccinated with Heplisav B (approximately 10,000) provided sufficient evidence of immunogenicity as a correlate of efficacy. This high number of subjects has allowed investigating the efficacy also within sub-groups in the pooled analyses (by age, BMI, smoking status, type 2 diabetes, race, and gender).

The pivotal Phase 3 studies gave consistent results of efficacy. A pooled analysis showed an SPR of 95.7 % (95% CI 95.3-96.1) in 8327 subjects. The SPR of Heplisav B was consistently superior over Engerix-B except in Asians, where only non-inferiority was met. The largest difference of SPR (exceeding 20%) induced by the two vaccines was observed in subjects with obesity, with type 2 diabetes and in smokers, where superiority of Heplisav B had a clear medical benefit.

In addition to the high SPR, rapid occurrence of immune response was shown in the trials. Specifically, 95.7 % of vaccinees have gained protective antibody levels already after two doses (at week 12). The high SPR was durable during the observation time (52 weeks) and was maintained above 90 %. There was no information about durability of SPR after week 52 in generally healthy population. The CHMP considered that a long-term follow-up study in healthy adults would not provide interpretable information, because it is rare for healthy adults who have a protective antibody level post vaccination not to be protected against hepatitis B virus (HBV), even if their blood antibody level drops below the protective level.

GMC of antibodies varied between pivotal studies, but generally showed higher antibody level when Heplisav B was given compared to Engerix-B, which might indicate also longer lasting protection. Although GMC were higher in the Heplisav B group compared to the Engerix-B group, the GMC magnitude distribution (stratified as <10 mIU/ml, 10-99 mIU/ml, 100-1000 mIU/ml, etc) was not systematically in favour of Heplisav B. Higher proportion of high-responders was observed in the Engerix-B group in the 3 studies, which was not expected. The CHMP considered that this observation might be attributed to the difference in the pattern of the immune response due to a different adjuvant and due to the difference in the number of doses administered for Heplisav B and Engerix-B.

Results from studies including CKD population (study HBV-017, durability study 019 (HBV-017 follow-up study) and booster study 018) showed generally improved results in case of Heplisav B in comparison to Engerix-B. However, as there were study conduct issues identified by the GCP inspection in HBV-017, there is some degree of uncertainty as to whether these results can be considered reliable.

The CHMP considered that a larger number of subjects would be needed to confirm the value of vaccinating previous non-responders or poor-responders with Heplisav B over Engerix-B or Fendrix. This consideration also applies to HIV-seropositive subjects, patients undergoing or expecting to receive chemotherapy and subjects with a history of an auto-immune disease. The Applicant presented studies that are ongoing to evaluate the safety and immunogenicity of Heplisav in subjects presenting with factors related to vaccination non-response. These studies focus on various populations, including HIV seropositive subjects, and patients undergoing immunosuppression to treat underlying inflammatory disorder, chemotherapy for cancer, or anti-

rejection therapy for liver transplantation. It was considered that these studies, although with limited sample size, could support the relevance of vaccinating such populations with Heplisav B. However, it was noted that the largest study is in HIV patients who are under HAART and who may not be considered as deeply immunocompromised. It was also noted that the studies performed in patients under immunosuppresion are of a very limited sample size. Therefore, these studies, although supportive, will not address the issue in full.

Only limited data are available in non-naïve subjects, which is a relevant group, as physicians may want to use Heplisav B in non-responders of Engerix-B. The Applicant is further evaluating the immunogenicity and safety of Heplisav B in adult non-responders to hepatitis B vaccination (following 2 rounds of 3 doses of standard vaccination) in an investigator-sponsored post-marketing study. Another study is being conducted in healthcare workers who failed to respond to 2 hepatitis B series with aluminum adjuvant vaccine. Both studies are of limited sample size (n= 50 and 100) and might not fully address the lack of data.

Interaction with other vaccines was not tested. Since Heplisav B induces high Ab GMC in generally healthy subjects, it is not expected that seroprotection rates would be affected by concomitant vaccination. However, in the absence of data, the concomitant use of Heplisav B with other vaccines is currently not recommended. In addition, the CHMP also considered that Heplisav B administration could affect the immune responses induced by the vaccines concomitantly administered.

Amendments of the clinical study protocols, statistical analysis plans or clinical study reports of the three main studies were considered adequately motivated, minor, or not relevant for the benefit-risk assessment.

The statistical analyses in the main studies were in general considered acceptable. To evaluate the immunogenicity endpoint, hierarchical testing of testing non-inferiority and then superiority was performed. This method to control the overall type I error for these tests is acceptable in line with CPMP/EWP/482/99.

Statistical analysis of diabetic and non-diabetic subjects including the age stratification for studies HBV-16 and HBV-023 was presented by the Applicant. Overall, the results suggested an added value of vaccination with Heplisav B for subjects >40 years of age without Type 2 diabetes, and even more for those with Type 2 diabetes, in comparison to Engerix-B. Immunogenicity results in patients with diabetes were reflected in the Product Information.

The CHMP considered that in compliance with the PIP, paediatric studies are deferred and will be conducted after approval in adults.

2.5.4. Conclusions on the clinical efficacy

Clinical studies have shown consistently high seroprotection rates and high antibody levels following a two-dose schedule, including populations of obese subjects, smokers and subjects with type 2 diabetes. The vaccine is adjuvanted with a novel adjuvant CpG 1018, which is considered to result in improved immunogenicity of the antigen, so that only two vaccine doses are needed to achieve acceptable seroprotection rate in 12 weeks. An immune response was elicited fast and the antibody levels were stable after the second dose for at least 52 weeks. The kinetics of antibody persistence is not expected to differ from the currently approved hepatitis B vaccines.

The efficacy profile of Heplisav B is considered to support a positive B/R balance.

2.6. Clinical safety

Patient exposure

Data for generally healthy adults were presented for 2 safety populations, the Primary Safety Population (PSP) and the Total Safety Population (TSP). Those comprised all subjects who received at least 1 dose of study treatment and had follow-up safety information.

The Primary safety Population (PSP) comprised 13,232 adults 18 to 70 years of age in the 3 pivotal phase 3 trials (HBV-23, HBV-16, and HBV-10) who received the intended commercial formulation, dose and number of vaccine of Heplisav B (N = 9365) in the intended vaccination scheme or the active comparator, Engerix-B (N = 3867, licensed hepatitis B vaccine) in a 2.4:1 subject allocation ratio.

The Total safety population (TSP) comprised 14,238 subjects (10,038 subjects in the Heplisav B arm and 4200 subjects in the Engerix-B arm) and includes subjects from the PSP and subjects from 8 additional supportive trials who received different dosages, early formulations of HEPISLAV-B or adjuvant only (8 subjects).

HBV-23 safety population comprised 8,368 subjects derived from 5,587 subjects receiving Heplisav B and 2,781 subjects receiving active comparator Engerix-B and is a subgroup of PSP.

PSP without HBV-23 population (PSP w/o HBV-23) was composed of two randomized studies (HBV-16 and HBV-10) in 3,788 subjects receiving Heplisav B and 1,089 subjects receiving Engerix-B.

Safety data in adults with chronic kidney disease (CKD SP) derived from 331 subjects 18 and older who received at least 1 dose of Heplisav B and 272 subjects ≥ 18 years who received at least 1 dose of Engerix-B. Data were derived from two study pools, i.e. the HBV-17 (trial) Safety Pool (HBV-17 SP), (254 subjects Heplisav B, 262 subjects Engerix) and two smaller supportive studies (77 subjects Heplisav, 10 subjects Engerix). The Phase 3 Study HBV-17 used the proposed commercial formulation, a 3-dose schedule of Heplisav B and used the Engerix-B regimen of 4 double doses (Engerix-B 2 x 20 mcg HBsAg) as a comparator. The two supportive studies used either an earlier formulation of Heplisav B compared to Engerix-B or a different dose of Heplisav B without comparator.

Baseline characteristics were balanced in study arms of the PSP. Specifically, HPV-23 showed higher rates of diabetes type 2 (13,6%), smokers (33%) and BMI (30.97%) than the studies HBV-16 (9.0%, 21,7% and 30,02, respectively) and HBV-10 (2,7%, 36,2% and 27,41%, respectively). In CKD patients (HBV-17 SP) Heplisav vaccinated subjects showed a higher rate of diabetes (68% vs 61%).

Adverse events

General healthy individuals (PSP/TSP)

The overall frequencies of adverse events and medically attended adverse events are presented in Table 23.

Table 17 Summary of AEs and MAEs in Generally Healthy Adults by Treatment Group

Event Type	MAEs	AEs	
	HBV-23	*PSP w/o HBV-23*	Total Safety Population Excluding HBV-23[a]

	Heplisav=B (N=5587) n (%)	Engerix-B (N=2781) n (%)	Heplisav B (N=3778) n (%)	Engerix-B (N=1086) n (%)	Heplisav B (N=4451) n (%)	Engerix-B (N=1419) n (%)
Any AE / MAE	2569 (46.0)	1286 (46.2)	2089 (55.3)	631 (58.1)	2573 (57.8)	869 (61.2)
Any related AE / MAE	58 (1.0)	45 (1.6)	234 (6.2)	65 (6.0)	349 (7.8)	165 (11.6)
Any grade 3 or 4 AE / MAE	887 (15.9)	416 (15.0)	286 (7.6)	112 (10.3)	396 (8.9)	148 (10.4)
Discontinued study treatment due to AE / MAE	32 (0.6)	15 (0.5)	19 (0.5)	4 (0.4)	20 (0.4)	5 (0.4)
Discontinued study treatment due to a related AE / MAE	7 (0.1)	5 (0.2)	6 (0.2)	1 (<0.1)	7 (0.2)	2 (0.1)

MAE- medically attended adverse event; PSP w/o HBV-23 = Primary Safety Population excluding HBV-23 (i.e., HBV-16 and HBV-10 only); SAE = serious adverse event; *TSP w/o HBV-23*
= Total Safety Population excluding HBV-23.
[a] Excludes HBV-23. Includes HBV-16, HBV-10, HBV-14, HBV-22, HBV0001 (all subjects classified as Heplisav B), HBV-02, HBV-03, HBV-04, HBV-05 and HBV-08.

Patients with CKD

The overall frequencies of adverse events in adults with chronic kidney disease are presented in Table 24.

Table 18 Summary of Safety Events in Adults With CKD (HBV-17 Safety Population, CKD Safety Population

Number (%) of Subjects with	HBV-17 SP		CKD SP	
	Heplisav B (N = 254)	Engerix-B (N = 262)	Heplisav B (N = 331)	Engerix-B (N = 272)
Any Post-Injection Reaction (%)[a]	(46.6)	(50.8)	(43.0)	(49.3)
Any Local Post-Injection Reaction (%)	(29.1)	(34.6)	(26.2)	(34.1)
Any Systemic Post-Injection Reaction (%)	(33.9)	(35.0)	(31.1)	(32.2)
Any AE, n (%)	195 (76.8)	198 (75.6)	247 (74.6)	206 (75.7)
Severe AE	59 (23.2)	64 (24.4)	71 (21.5)	65 (23.9)
Related AE	19 (7.5)	26 (9.9)	21 (6.3)	26 (9.6)
Any AESI (AE of Special Interest)	0	0	1 (0.3)	0
Any New-Onset AIAE (Autoimmune AE)	0	0	NA[b]	NA[b]
AE Leading to Discontinuation of Treatment	4 (1.6)	6 (2.3)	6 (1.8)	6 (2.2)
AE Leading to Trial Withdrawal	0	0	1 (0.3)	0
Any SAE	68 (26.8)	76 (29.0)	81 (24.5)	77 (28.3)
Related SAE	1 (0.4)	0	1 (0.3)	0
Death	7 (2.8)	3 (1.1)	9 (2.7)	4 (1.5)
Related Death	0	0	0	0

AE = adverse event; AESI = adverse event of special interest; AIAE = autoimmune adverse event; CKD = chronic kidney disease; CKD SP = chronic kidney disease safety population; CSR = clinical study report; HBV-17 SP = HBV-17 safety population; NA = not applicable; SAE = serious adverse event.
[a] Post-injection reaction rates are presented only as percentages, based on the number of subjects returning a diary card.
[b] Autoimmune AEs were evaluated and adjudicated only in HBV-17.

Post-injection Reactions

Generally healthy individuals (PSP/TSP)

The frequency of local and systemic post-injection reactions was roughly equally distributed between vaccine and control arm in healthy adults. Injection site redness and swelling occurred slightly more often in the

Heplisav B arm compared to the Engerix-B arm. The frequency of severe systemic reactions was lower in the test vaccine arm compared to active comparator (Table 25).

Table 19 Post-injection Reactions by Treatment Group and Safety Population

	PSP w/o HBV-23		***PSP w/o HBV-23* + Supportive Trials**[a]	
	Heplisav B (N = 3777)	Engerix-B (N = 1087)	Heplisav B (N = 4347)	Engerix-B (N = 1344)
Any Post-injection Reaction, n (%)	3762	1084	4332	1341
Subjects with reactions	2071 (55.1)	619 (57.1)	2424 (56.0)	775 (57.8)
Subjects with severe reactions	119 (3.2)	55 (5.1)	134 (3.1)	67 (5.0)
Local Post-injection Reaction, n (%)[b]	3762	1084	4332	1341
Subjects with reactions	1612 (42.8)	445 (41.1)	1863 (43.0)	550 (41.0)
Subjects with severe reactions	21 (0.6)	4 (0.4)	21 (0.5)	7 (0.5)
Systemic Post-injection Reaction, n (%)	3762	1084	4332	1339
Subjects with reactions	1215 (32.3)	405 (37.4)	1459 (33.7)	525 (39.2)
Subjects with severe reactions	106 (2.8)	53 (4.9)	121 (2.8)	62 (4.6)

PSP w/o HBV-23 = Primary Safety Population excluding HBV-23 (i.e., HBV-16 and HBV-10 only).

a Excludes supportive trials HBV-02, HBV-05, and HBV-22.

b Excludes redness and swelling data from HBV-04.

PSP w/o HBV-23 + Supportive Trials population includes supportive trials HBV0001, HBV-03, HBV-04, HBV-08, and HBV-14.

The profile of PIRs from Heplisav B and Engerix-B was consistent for both populations (the PSP w/o HBV-23 + Supportive Trials population and the PSP w/o HBV-23 population). Additional analyses on PIRs stratifying further according to age were presented and no safety concerns were detected.

The frequencies of severe systemic reactions were low (≤1% per dose for each type of systemic reaction). Local and systemic PIRs peaked at day 2 or 3 post-injection. Most PIRs resolved within 7 days. Nevertheless, in both groups, the proportion of subjects still presenting systemic PIR at day 7 is superior to the frequency 30 minutes after injection. Induction of prolonged inflammatory responses could be of concern when vaccinating with adjuvanted vaccines. Detailed data were presented on the kinetics of clinical and laboratory markers measuring inflammatory responses, including PIRs as well as laboratory data such as WBC and CRP. No major differences were detected between Heplisav B and Engerix-B arm.

Patients with CKD

The overall frequency of post-injection reactions was comparable between the test vaccine and control arm in CKD patients and slightly lower compared to healthy adults. The frequency of severe PIRs was twice as high in the Heplisav B arm compared to the ENGERIX-B arm (4.8% vs. 2.7%) and mainly driven by systemic reactions (Malaise, Myalgia, fatigue) (Table 26).

Table 20 Post-Injection Reactions across All Active Injections by Treatment Group in Adults With CKD (HBV-17 Safety Population, CKD Safety Population)

Number (%) of Subjects With	**HBV-17 SP**		**CKD SP**	
	Heplisav B (N = 254)	Engerix-B (N = 262)	Heplisav B (N = 331)	Engerix-B (N = 272)

Any Post-Injection Reaction, N	251	260	328	270
Subjects with Reactions, n (%)	117 (46.6)	132 (50.8)	141 (43.0)	133 (49.3)
Subjects with Severe Reactions, n (%)	12 (4.8)	7 (2.7)	13 (4.0)	6 (2.2)
Any Local Post-Injection Reaction, N	251	260	328	270
Subjects with Reactions, n (%)	73 (29.1)	90 (34.6)	86 (26.2)	92 (34.1)
Subjects with Severe Reactions, n (%)	3 (1.2)	2 (0.8)	4 (1.2)	2 (0.7)

CKD = chronic kidney disease; CKD SP = chronic kidney disease safety population; HBV-17 SP = Trial DV2-HBV-17 safety population; NA = not available.

Other types of Adverse events

Generally healthy individuals (PSP/TSP)

Frequent AEs by PT (frequency of ≥ 2% and ≥1% of subjects in PSP/TSP w/o HBV-23 and HBV-23, respectively) were in general balanced between vaccine and comparator arm in the w/o HBV 23 pool (PSP and TSP w/o HBV 23) and in HBV 23 (Table 27and Table 28).

AE evaluation was primarily based on the PSP. The Applicant considered and re-evaluated all events that have been evaluated as related by the investigator, all events that occurred within 7 days after vaccination (primary focus on a cut off of 1%) and all PIRs (per definition related to the treatment). Additionally, events from the PASS studies HBV 25 and HBV 26 were considered. The pregnancy registry (HBV-27) does not have included yet any cases. Spontaneous AE reports from the US covering the period from initial US commercialization (January 2018) until 08 June 2020 are available. A medical review of these data was conducted to identify any AEs that had not already been observed in clinical trials and for which a causal relationship with Heplisav vaccination was at least a reasonable possibility. Causality was assessed per Section VI.A.2.1.1. 'Causality' of Module VI of the Guideline on Good Pharmacovigilance Practices (GVP). This was endorsed by the CHMP.

Table 21 AEs occurring in ≥ 2% of subjects by frequency of PT in the PSP w/o HBV-23 and in the TSP w/o HBV-23

Preferred Term	**PSP w/o HBV-23**		**TSP Excluding HBV-23[a]**	
	Heplisav B (N = 3778) n (%)	Engerix-B (N = 1086) n (%)	Heplisav (N=4451) n (%)	Engerix-B (N=1419) n (%)
Subjects with any AE	2089 (55.3)	631 (58.0)	2573 (57.8)	869 (61.2)
Nasopharyngitis	383 (10.1)	125 (11.5)	450 (10.1)	176 (12.4)
Headache	260 (6.9)	76 (7.0)	390 (8.8)	138 (9.7)
Back Pain	130 (3.4)	38 (3.5)	164 (3.7)	52 (3.7)
Sinusitis	109 (2.9)	26 (2.4)	127 (2.9)	27 (1.9)
Upper Respiratory Tract Infection	105 (2.8)	38 (3.5)	179 (4.0)	84 (5.9)
Oropharyngeal Pain	102 (2.7)	35 (3.2)	139 (3.1)	47 (3.3)
Arthralgia	92 (2.4)	32 (2.9)	120 (2.7)	45 (3.2)
Cough	89 (2.4)	25 (2.3)	120 (2.7)	57 (4.0)
Diarrhoea	75 (2.0)	22 (2.0)	106 (2.4)	44 (3.1)
Fatigue	53 (1.4)	15 (1.4)	88 (2.0)	42 (3.0)
Hypertension	62 (1.6)	31 (2.9)	77 (1.7)	40 (2.8)
Nausea	55 (1.5)	24 (2.2)	67 (1.5)	28 (2.0)
Pyrexia	30 (0.8)	14 (1.3)	55 (1.2)	30 (2.1)

Malaise	16 (0.4)	13 (1.2)	43 (1.0)	40 (2.8)

AE = adverse event; *PSP w/o HBV-23* = Primary Safety Population excluding HBV-23 (i.e., HBV-16 and HBV-10 only); *TSP w/o HBV-23* = Total Safety Population excluding HBV-23.
[a] Includes HBV-16, HBV-10, HBV-14, HBV-22, HBV0001 (all subjects classified as Heplisav), HBV-02, HBV-03, HBV-04, HBV-05 and HBV-08.
Data Source: SCS Table 6.1.2.1.

Table 22 Treatment-Emergent MAEs by PT in ≥ 1% of Subjects in HBV-23

Preferred Term	Heplisav (N=5587) n (%)	Engerix-B (N=2781) n (%)
Subjects with at least 1 qualifying MAE	2569 (46.0)	1286 (46.2)
Upper Respiratory Tract Infection	192 (3.4)	92 (3.3)
Bronchitis	176 (3.2)	102 (3.7)
Sinusitis	149 (2.7)	84 (3.0)
Hypertension	133 (2.4)	59 (2.1)
Urinary Tract Infection	132 (2.4)	64 (2.3)
Back Pain	116 (2.1)	54 (1.9)
Arthralgia	98 (1.8)	54 (1.9)
Osteoarthritis	77 (1.4)	32 (1.2)
Pain in Extremity	72 (1.3)	28 (1.0)
Type 2 Diabetes Mellitus	67 (1.2)	37 (1.3)
Cough	62 (1.1)	37 (1.3)
Acute Sinusitis	59 (1.1)	37 (1.3)
Laceration	54 (1.0)	19 (0.7)

MAE = medically attended event.
Data Source: HBV-23 CSR Table 14.1.4.5.

Patients with CKD

AEs (by SOCs and PTs) occurred in the majority balanced in test vaccine and comparator arm in the CKD pools. Of note, immune system disorders occurred in slightly higher frequency in the Heplisav B arms compared to Engerix B (6 (2.4%) vs 3 (1.1%) cases). Furthermore, the PTs Rash (2.0 vs 0.8%), Arthralgia (5.2% vs 2.7%), Hyperkaliemia (5.1% vs. 2,3%), hypokalemia (3.5% vs 1.5%) and pulmonary edema (2.4% vs. 1.1%) occurred more often in the Heplisav B arm compared to the Engerix-B arm.

Serious adverse event/deaths/other significant events

General healthy adults (PSP/TSP)

SAEs occurred in general in similar frequency in the vaccination arms of PSP/TSP. A higher rate of myocardial infarctions was seen in the Heplisav B arm (16 vs 2 cases PSP, 17 vs 2 cases TSP) (Table 29).

Table 23 Summary of Serious Adverse Events by System Organ Class and Preferred Term (Heplisav B > 0.1% in the TSP) (PSP, TSP)

	PSP		TSP	
SOC (bold) PT	**Heplisav B (N = 9365) n (%)**	**Engerix-B (N = 3867) n (%)**	**Heplisav B (N = 10038) n (%)**	**Engerix-B (N = 4200) n (%)**
Subjects with at least one treatment-emergent serious adverse event	449 (4.8)	184 (4.8)	466 (4.6)	200 (4.8)
Infections And Infestations	78 (0.8)	35 (0.9)	83 (0.8)	39 (0.9)
Pneumonia	16 (0.2)	8 (0.2)	16 (0.2)	9 (0.2)

Injury, Poisoning And Procedural Complications	64 (0.7)	22 (0.6)	68 (0.7)	25 (0.6)
Cardiac Disorders	59 (0.6)	21 (0.5)	60 (0.6)	22 (0.5)
Acute Myocardial Infarction	16 (0.2)	2 (<0.1)	17 (0.2)	2 (<0.1)
Gastrointestinal Disorders	46 (0.5)	18 (0.5)	47 (0.5)	19 (0.5)
Respiratory, Thoracic And Mediastinal Disorders	45 (0.5)	15 (0.4)	47 (0.5)	16 (0.4)
Neoplasms Benign, Malignant And Unspecified (Includes Cysts And Polyps)	46 (0.5)	19 (0.5)	46 (0.5)	21 (0.5)
Nervous System Disorders	42 (0.4)	17 (0.4)	44 (0.4)	19 (0.5)
Musculoskeletal And Connective Tissue Disorders	42 (0.4)	16 (0.4)	44 (0.4)	17 (0.4)
Osteoarthritis	16 (0.2)	5 (0.1)	17 (0.2)	5 (0.1)
Psychiatric Disorders	26 (0.3)	7 (0.2)	26 (0.3)	7 (0.2)
Metabolism And Nutrition Disorders	20 (0.2)	9 (0.2)	22 (0.2)	9 (0.2)
Vascular Disorders	21 (0.2)	9 (0.2)	21 (0.2)	10 (0.2)
General Disorders And Administration Site Conditions	18 (0.2)	11 (0.3)	19 (0.2)	11 (0.3)
Non-Cardiac Chest Pain	12 (0.1)	8 (0.2)	12 (0.1)	8 (0.2)
Hepatobiliary Disorders	16 (0.2)	10 (0.3)	18 (0.2)	10 (0.2)
Renal And Urinary Disorders	14 (0.1)	9 (0.2)	14 (0.1)	10 (0.2)
Reproductive System And Breast Disorders	5 (<0.1)	7 (0.2)	6 (<0.1)	7 (0.2)
Pregnancy, Puerperium And Perinatal Conditions	6 (<0.1)	3 (<0.1)	6 (<0.1)	3 (<0.1)
Blood And Lymphatic System Disorders	4 (<0.1)	3 (<0.1)	4 (<0.1)	3 (<0.1)
Congenital, Familial And Genetic Disorders	3 (<0.1)	1 (<0.1)	3 (<0.1)	1 (<0.1)
Ear And Labyrinth Disorders	3 (<0.1)	1 (<0.1)	3 (<0.1)	1 (<0.1)
Skin And Subcutaneous Tissue Disorders	2 (<0.1)	2 (<0.1)	2 (<0.1)	2 (<0.1)
Investigations	2 (<0.1)	1 (<0.1)	2 (<0.1)	1 (<0.1)
Immune System Disorders	1 (<0.1)	2 (<0.1)	1 (<0.1)	2 (<0.1)
Endocrine Disorders	1 (<0.1)	2 (<0.1)	1 (<0.1)	2 (<0.1)
Social Circumstances	1 (<0.1)	0	1 (<0.1)	0
Surgical And Medical Procedures	0	0	0	2 (<0.1)
Eye Disorders	0	0	0	0

PSP = Primary Safety Population; TSP = Total Safety Population.
Note: All SOCs are included and PTs occurring in ≥ 0.1% of subjects in the Heplisav B group in the TSP. Table is sorted by SOC from highest to lowest frequency.

The rate of SAEs occurring within 42 days after the last injection was overall low and balanced in the study arms of the PSP. SAEs reported most frequently within 42 days after the last active vaccination represented a wide spectrum of diagnoses. SAEs belonging to the SOCs Nervous System Disorders (6 (<0.1%) vs.2 (<0.1)) and Musculoskeletal and Connective Tissue Disorders 4 (<0.1%) vs. 1 (<0.1%)) occurred in numerically higher frequency in the Heplisav B arm compared to the Engerix-B arm. No imbalances were detected in the analyses on SAEs occurring within 42 days after the last vaccination for HBV-23.

Serious adverse events in CKD

SAEs in the HBV-17 SP (68 (26.8%) vs. 76 (29%)) and the CKD SP (81 (24.5%) vs. 77 (28.3%)) were in both study arms overall higher in frequency compared to the healthy adult safety population. No differences of major concern were observed between treatment arms.

Mortality in general healthy adults (PSP/TSP)

The numerical incidence of deaths of subjects who received Heplisav B was higher (n = 26, 0.26%) compared to Engerix-B (n = 8, 0.19%) in the PSP. 32 of 34 deaths occurred in the HBV-23 study (2 deaths occurred in

study HBV-16). No deaths were assessed by the investigators as related to study treatment. The proportion of cardiovascular caused fatal events was balanced in the PSP and only slightly increased in HBV-23 for Heplisav B (0.14% vs 0.10%). Fatal cases were balanced in the PSP within the first 42 days after the 2nd injection.

Mortality in CKD

Fatal cases occurred almost twice as often in the Heplisav B arm compared to the active comparator (13 deaths: 9/331 (2.7%) in the Heplisav B group and 4/272 (1.5%) in the Engerix-B arm). All cases were judged as unrelated to study drug by the study investigators.

Myocardial Infarction and Major Adverse Cardiovascular Events

In the TSP, 22 (Heplisav B) vs. 5 (Engerix-B) cases belonging to the myocardial Infarction Narrow SMQ were detected. Events of the PT MI/AMI were detected with overall 19 vs 3 cases in the TSP, thereof, 18 and 3 cases in the PSP. Events of MI/AMI occurred mainly in the HBV-23 trial (16), followed by HBV-16 (2) and study HBV-5 (1). 17 vs. 2 cases belonged to the PT "acute myocardial infarction", 2 vs. 1 cases to the PT "myocardial infarction". 7 vs. 2 cases of the MI SMQ occurred in a time frame of < 60 days from the last dose in the PSP. The imbalance of MI cases was specifically caused by events that occurred in the Heplisav B arm of HBV-23. The number of the PT AMIs for studies HBV-23, HBV-16 and HBV-10 is presented in the Table 30.

Table 24 Treatment-Emergent SAEs Reported Coded to the MedDRA Preferred Term *Acute Myocardial Infarction* in Heplisav B Phase 3 Clinical Trials Study

	Heplisav B		**Engerix-B**		**Relative Risk**	**95% CI**
	n/N	**%**	**n/N**	**%**		
HBV-23	14/5587	0.25	1/2781	0.04	6.97	0.92, 52.97
HBV-16	2/1968	0.10	1/481	0.21	0.49	0.04, 5.38
HBV-10	0/1810	0	0/605	0	NA	NA

CI = confidence interval; NA = not applicable
Note: In HBV-23, an acute myocardial infarction occurred in a 54-year-old obese woman during the screening period and 2 weeks prior to receiving an injection of Heplisav B. This event is not included in these analyses. She received both doses of Heplisav B and did not report a treatment-emergent cardiovascular event.

The association of Heplisav B with myocardial infarction in HBV-23 was at the limit of statistical significance (95% CI: 0.92 - 52.97) when computing asymptotic 95% CIs using the Wald method, but was statistically significant (95% CI: 1.00, 184.90) when using the standardized statistic proposed by Miettinen and Nurminen.

The Applicant presented an analysis of the Angina pectoris SMQ with absolute numbers and proportions for all four safety pools detecting overall only very low and equally distributed numbers of events (Table 31). No safety concern is detected here.

Table 25 Treatment Emergent Angina Pectoris Adverse Events by Preferred Term

	HBV-23[a]		HBV-16 and HBV-10		Primary Safety Population[b]		Total Safety Population[c]	
Preferred Term	**Heplisav (N=5587)**	**Engerix-B (N=2781)**	**Heplisav (N=3778)**	**Engerix-B (N=1086)**	**Heplisav (N=9365)**	**Engerix-B (N=3867)**	**Heplisav (N=10038)**	**Engerix-B (N=4200)**
Subjects with at least one qualifying adverse event	4 (<0.1%)	3 (0.1%)	8 (0.2%)	1 (<0.1%)	12 (0.1%)	4 (0.1%)	13 (0.1%)	8 (0.2%)
Angina Pectoris	3 (<0.1%)	3 (0.1%)	8 (0.2%)	1 (<0.1%)	11 (0.1%)	4 (0.1%)	12 (0.1%)	8 (0.2%)
Angina Unstable	1 (<0.1%)	0	0	1 (<0.1%)	1 (<0.1%)	1 (<0.1%)	1 (<0.1%)	2 (<0.1%)

a Medically attended adverse events were collected for HBV-23.
b Includes HBV-23, HBV-16 and HBV-10.
c Includes HBV-23, HBV-16, HBV-10, HBV-14, HBV-22, HBV0001 (all subjects classified as Heplisav), HBV-02, HBV-03, HBV-04, HBV-05 and HBV-08.
Medically attended adverse events and adverse events are combined in this table.Following Preferred terms are summarized here: angina pectoris, angina unstable, anginal equivalent, arteriospasm coronary and Prinzmetal angina

A twice as high risk to experience an event in the MI SMQ (0.22% (21) versus 0.1% (4)) was observed for the PSP. (Table 32)

Table 26 Treatment-Emergent SAEs in the Myocardial Infarction SMQ by PT in the PSP

PT	Heplisav B (N=9365) % (n)	Engerix-B (N=3867) % (n)	Relative Risk (95% Confidence Interval)
Subjects with at least 1 qualifying adverse event	**0.22 (21)**	**0.10 (4)**	2.168 (0.745, 6.311)
Acute Coronary Syndrome	0.01 (1)	0	4.129 (0.006, 2747.487)
Acute Myocardial Infarction	0.17 (16)	0.05 (2)	3.303 (0.760, 14.360)
Angina Unstable	0.01 (1)	0.03 (1)	0.413 (0.026, 6.600)
Coronary Artery Occlusion	0.01 (1)	0.03 (1)	0.413 (0.026, 6.600)
Myocardial Infarction	0.02 (2)	0.03 (1)	0.826 (0.075, 9.105)

Major adverse cardiovascular events (MACE) analysis

The MACE methods included:

(i) a blinded review of clinical annotations and cardiac catheterization data for all reported acute myocardial infarction events in the pivotal trials.

(ii) a broad search of the Heplisav B safety database was conducted for other possibly missed acute myocardial infarctions or strokes, using the myocardial infarction narrow Standardised MedDRA Queries (SMQs).

(iii) a blinded post-hoc central adjudication of all deaths and all potential myocardial infarctions and strokes (potential MACE outcomes). Dynavax engaged the Cleveland Clinic Coordinating Center for Clinical Research (C5Research) to perform this adjudication.

The endpoints assessed in this post-hoc analyses were:

(i) acute myocardial infarction events.

(ii) composite outcome of adjudication-confirmed MACE comprising cardiovascular death, non-fatal myocardial infarction, and non-fatal stroke, and analysis of each independent component of the composite.

MACE events before adjudication

An imbalance in MACE events is specifically seen in study HBV-23 (0.93% (52) vs. 0.47% (13)), which is mainly driven by imbalances in (cardiovascular) deaths (0.20% (11) vs 0.11% (3)) and myocardial infarctions (0.34% (19) vs 0.11% (3)). No imbalance is seen in HBV-16. Frequencies of putative MACE events in the PSP are 0.59% (55) vs 0.39% (15)) (Table 33).

Table 27 Potential Treatment-Emergent, Serious MACE by Treatment Group Sent for Blinded Event Adjudication

	HBV-16		HBV-23		
Treatment	**Heplisav B (N = 1968) % (n)**	**Engerix-B (N = 481) % (n)**	**Heplisav B (N = 5587) % (n)**	**Engerix-B (N = 2781) % (n)**	**Relative Risk (95% Confidence Interval)**
Composite MACE	**0.15 (3)**	**0.42 (2)**	**0.93 (52)**	**0.47 (13)**	**1.991 (1.086 - 3.650)**
Deaths					
All deaths	0.05 (1)	0.21 (1)	0.45 (25)	0.25 (7)	1.778 (0.770 - 4.105)
Death from CVD cause[a]	**0.05 (1)**	**0.21 (1)**	**0.20 (11)**	**0.11 (3)**	**1.825 (0.510 - 6.537)**
Deaths from other causes	0	0	0.25 (14)	0.14 (4)	1.742 (0.574 - 5.288)
Myocardial infarction[b]	**0.10 (2)**	**0.21 (1)**	**0.34 (19)**	**0.11 (3)**	3.152 (0.934 - 10.644)
Acute myocardial infarction	0.10 (2)	0.21 (1)	0.25 (14)	0.04 (1)	6.969 (0.917 - 52.967)
Myocardial infarction	0	0	0.04 (2)	0.04 (1)	0.996 (0.090-10.974)
Acute coronary syndrome	0	0	0.02 (1)	0	
Others[c]	0	0.21 (1)	0.04 (2)	0.04 (1)	0.996 (0.090 - 10.974)
Stroke[d]	**0**	**0**	**0.20 (11)**	**0.18 (5)**	**1.095 (0.381 - 3.149)**

	Primary Safety Population		
Treatment	**Heplisav B (N = 9365) % (n)**	**Engerix-B (N = 3867) % (n)**	**Relative Risk (95% Confidence Interval)**
Composite MACE	**0.59 (55)**	**0.39 (15)**	**1.514 (0.857 - 2.676)**
All deaths	0.28 (26)	0.21 (8)	1.342 (0.608 - 2.961)
Death from CVD cause[a]	**0.13 (12)**	**0.10 (4)**	**1.239 (0.400 - 3.838)**
Deaths from other causes	0.15 (14)	0.10 (4)	1.445 (0.476 - 4.388)
Myocardial infarction[b]	**0.22 (21)**	**0.10 (4)**	2.168 (0.745 - 6.311)
Acute myocardial infarction	0.17 (16)	0.05 (2)	3.303 (0.760 - 14.360)
Myocardial infarction	0.02 (2)	0.03 (1)	0.826 (0.075 - 9.105)
Acute coronary syndrome	0.01 (1)	0	
Others	0.02 (2)	0.05 (2)	0.413 (0.058 - 2.930)
Stroke[c]	**0.12 (11)**	**0.13 (5)**	**0.908 (0.316 - 2.613)**

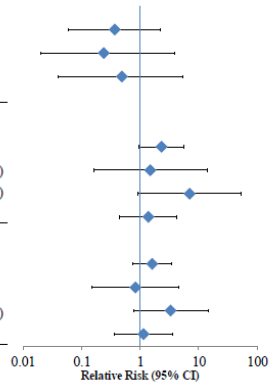
MACE = Major Adverse Cardiovascular Events.
[a] Cardiovascular death includes death due to Acute Coronary Syndrome, Acute Myocardial Infarction, Acute Respiratory Failure, Cardiac Arrest, Cardiac Failure, Cardio-respiratory Arrest, Death, Hypertensive Heart Disease, Myocardial Infarction, or Pulmonary Embolism.
[b] Myocardial infarction includes Acute Coronary Syndrome, Acute Myocardial Infarction, Coronary Artery Embolism, Coronary Artery Thrombosis, Coronary Bypass Thrombosis, Myocardial Infarction, Post Procedural Myocardial Infarction, Silent Myocardial Infarction, Coronary Artery Occlusion, Angina Unstable or Death.
[c] Stroke includes Basal Ganglia Stroke, Brain Stem Stroke, Cerebrovascular Accident, Haemorrhagic Stroke, Haemorrhagic Transformation Stroke, Stroke in Evolution, Basal Ganglia Infarction, Basal Ganglia Stroke, Brain Stem Embolism, Brain Stem Infarction, Brain Stem Stroke, Cerebellar Embolism, Cerebellar Infarction, Cerebral Artery Embolism, Cerebral infarction, Embolic Cerebral Stroke, Embolic Stroke, Ischaemic Cerebral infarction, Ischaemic Stroke, Lacunar Infarction, Lacunar Stroke, Thalamic Infarction, Thrombotic Cerebral Infarction, or Thrombotic Stroke

MACE events after adjudication

Analyses based on MACE cases after adjudication showed increases for MACE and MI in the PSP (RR 1.6; RR 3.3), which was mainly driven by cases in HBV-23 (RR 3.3 and RR 2.32) (Figure 3).

Figure 2: Subjects With Adjudication-Confirmed 3-point MACE by Treatment Group (PSP)

MACE	HEPLISAV-B	Engerix-B	Relative Risk (95% CI)
	N=1968	N=481	
HBV-16	n (%)	n (%)	
Composite 3-point MACE events	3 (0.15)	2 (0.42)	0.37 (0.06, 2.19)
Death from cardiovascular cause	1 (0.05)	1 (0.21)	0.24 (0.02, 3.9)
Myocardial infarction	2 (0.10)	1 (0.21)	0.49 (0.04, 5.38)
Stroke	0	0	N/A N/A
	N=5587	N=2781	
HBV-23	n (%)	n (%)	
Composite 3-point MACE events	28 (0.50)	6 (0.22)	2.32 (0.96, 5.60)
Death from cardiovascular cause	3 (0.05)	1 (0.04)	1.49 (0.16, 14.35)
Myocardial infarction	14 (0.25)	1 (0.04)	6.97 (0.92, 52.97)
Stroke	11 (0.20)	4 (0.14)	1.37 (0.44, 4.30)
	N=9365	N=3867	
PSP (HBV-10, HBV-16, and HBV-23)	n (%)	n (%)	
Composite 3-point MACE events	31 (0.33)	8 (0.21)	1.6 (0.74, 3.48)
Death from cardiovascular cause	4 (0.04)	2 (0.05)	0.83 (0.15, 4.51)
Myocardial infarction	16 (0.17)	2 (0.05)	3.3 (0.76, 14.36)
Stroke	11 (0.12)	4 (0.10)	1.14 (0.36, 3.56)



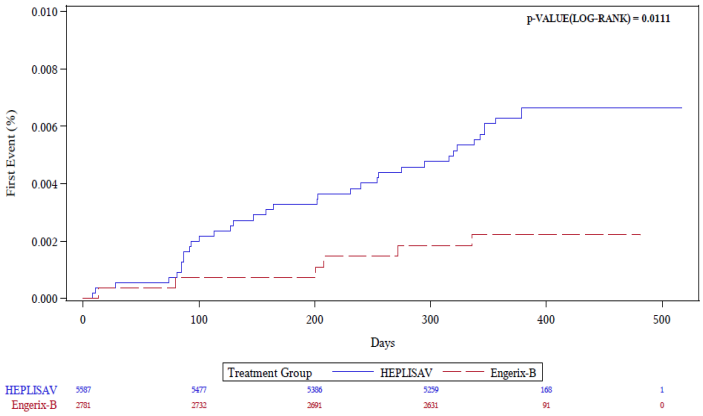
Data Source: Post hoc Tables 30.4.4, 30.4.5, 30.4.6, and 30.4.7; Post hoc Listing 7.2.
CI = confidence interval; MACE = Major Adverse Cardiovascular Events. Composite 3-point MACE comprises death from cardiovascular cause, non-fatal myoca infarction, and non-fatal stroke; N/A= not applicable
Note: Subjects with multiple MACE outcomes are counted once in the composite endpoints.

Through 28 days after the second injection (Study Day 56), the proportion of subjects reporting events was balanced between the 2 treatment groups based on MACE cases after adjudication (Heplisav B: 0.05% [n = 5]; Engerix-B: 0.05% [n = 2]). The imbalance in MACE outcomes occurred late in the studies beginning after Study Day 100. Of note, the incidence in the Heplisav B group in the first 100 days (4.3/1000 person years) was similar to the last 100 days (4.6/1000 person years).

After controlling for risk factors of MACE (age, sex, race, hypertension, BMI, diabetes mellitus, smoking, history of myocardial infarction or stroke), the association between vaccination and MI risk persisted, but was weakened (OR = 1.63; 95% CI: 0.75, 3.55).

The Applicant generated Kaplan-Meier plots for the MI SMQ, MACE events including cases relabeled as undetermined during the adjudication process and for MACE labeled as confirmed cases. While log-rank tests were non-significant (PSP) or borderline significant (HBV-23) for adjudicated cases of MACE and MI, a significant separation of curves was seen when including cases with undetermined cases of deaths specifically in HBV-23 (p=0,011; borderline p-value of 0.0501 for MIs in HBV-23) (Figure 4).

Figure 3 Kaplan-Meier Cumulative Incidence of Treatment-Emergent Serious Confirmed Major Adverse Cardiovascular Events and Undetermined Cause of Death Events (HBV-23)



Interim analysis of the study HBV-25:

To address the observed numerical imbalance in cardiovascular events between vaccine groups in HBV-23 the applicant is performing a observational study (HBV-25).

This study follows up 69,910 hepatitis B vaccine recipients (31,244 subjects in the Heplisav B arm and 38,666 in the Engerix-B arm) for 13 months. The incidence of AMI in patients who receive at least 1 dose of Heplisav B is being compared with that in patients who receive at least 1 dose of the comparator hepatitis B vaccine (Engerix-B).

The study is performed in a non-randomized cluster design, vaccinating subjects with Heplisav B or Engerix-B in 7 and 8 different health centres in the U.S., respectively.

Cases of potential AMI events as identified via health record study are adjudicated by 2 independent cardiologists. The interim analysis was performed on unconfirmed AMIs, corresponding to unadjudicated Type 1 AMI based ICD-10-CM predefined codes. The Applicant claimed that the confirmation rate of early events is 87%.

The interim analysis of HBV-25 was completed on 1 July 2019. The submitted interim results were based on 24,404 person-years with an average of 5.4 months follow-up per person (Heplisav B: 5.2 months, Engerix-B: 5.6months) in 54,020 subjects. The vaccine accrual by number doses is presented in Table 34. The number of subjects with a full vaccination scheme is comparable with the number of Heplisav B recipients evaluated in the phase III programme.

Table 28 Vaccine accrual by number of doses

Table 12-1: Vaccine Accrual by Number of Doses of Vaccine Received (07 August 2018—01 July 2019)

	HEPLISAV-B[a]	Engerix-B[b]	Total
Received one dose	14,841	18,885	33,726
Received two doses	9,076	7,950	17,026
Received three or more doses	102	3,166	3,268
Total number of recipients	24,019	30,001	54,020

Source data: Table 2.1.

[a] HEPLISAV-B doses included are any doses of HEPLISAV-B given to members age 18 or older in the Family Medicine, Internal Medicine or Urgent Care departments at target medical centers starting 07 August 2018 through 01 July 2019. Prior Hepatitis B vaccine doses are not counted.

[b] Comparator vaccine doses included are any doses of comparator Hepatitis B vaccine (non-dialysis formulation) given to members age 18 or older in the Family Medicine, Internal Medicine or Urgent Care departments at non-target medical centers starting 07 August 2018 through 01 July 2019. Prior Hepatitis B vaccine doses are not counted.

Note: Individuals who received both a dose of HEPLISAV-B at a target medical center and a dose of comparator vaccine at comparator medical center during the vaccination period are included only in the arm in which they received their first qualifying dose.

Note: If two hepatitis B vaccine doses were recorded on the same day, then the doses would be counted as one. There were 2 HEPLISAV-B recipients and 60 comparator vaccine recipients who received two doses on the same day.

Based on the interim analysis, between 14795 (62%) and 17965 (75%) of 24019 individuals in the Heplisav B arm have passed 120 days and 80 days after the first dose respectively (Table 35). This roughly covers the time point of interest, i.e. 3 months after the first vaccination, where a clustering of MI events was observed in HBV-23.

Table 29 Number of Patients at Risk for Acute Myocardial Infarction Over Time

Days After Index Dose	HEPLISAV-B	Engerix-B
0	24,019	30,001
40	21,202	26,729
80	17,965	22,940
120	14,795	19,460
160	11,635	15,907
200	8891	13,016
240	5673	9497
280	2435	5565
320	77	1203

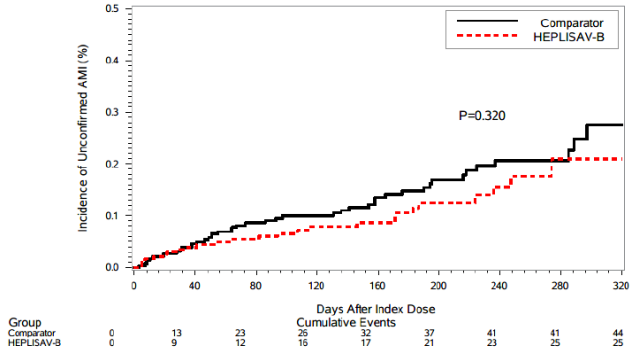
Source data: Figure 2.2c.

In 24,019 Heplisav B recipients, 25 AMIs occurred in 10,318.74 person-years of follow-up, observing an incidence rate of 2.42/1000 person-years. Compared to this observation, in 30,001 Engerix-B recipients, 44 AMIs occurred in 14,085.57 person-years of follow-up for an incidence rate of 3.12/1000 person-years.

The Cumulative Incidence Rate Estimates of Acute Myocardial Infarction are presented in Figure 5. The applicant performed unadjusted and adjusted cox regression analyses calculating a hazard ratio of 0.83 (95% CI=0.50, 1.37, unadjusted HR=0.78) (Table 36).

Figure 4

Cumulative Incidence Rate Estimates of Acute Myocardial Infarction in Heplisav B and Engerix-B Recipients (Kaplan-Meier Method)



Source data: Figure 2.2b.

P value is from log-rank test. Comparator = Engerix-B

Table 30

Acute Myocardial Infarction Hazard Ratio comparing Heplisav B Recipients and Engerix-B Recipients (Cox Proportional Hazards Model)

	Unadjusted HR (95% CI)	Adjusted HR (95% CI)
HEPLISAV-B vs comparator vaccine group	0.78 (0.48-1.28)	0.83 (0.50- 1.37)
Age at index dose (years)	N/A	1.03 (1.01- 1.05)
Sex (male vs. female)	N/A	2.96 (1.66- 5.28)
History of AMI in year prior to index dose	N/A	7.56 (2.92-19.55)
Use of nitrate medication prior to index dose	N/A	5.66 (2.39-13.42)
Race Ethnicity Hispanic vs White	N/A	0.95 (0.53- 1.69)
Race Ethnicity Black vs White	N/A	1.48 (0.65- 3.37)
Race Ethnicity Asian vs White	N/A	0.93 (0.41- 2.15)
Race Ethnicity Other/Unknown vs White	N/A	0.51 (0.12- 2.19)

Source data: Table 2.3.

AMI = acute myocardial infarction; HR = hazard ratio; N/A = not applicable.

Notes: Age, sex and Race/ethnicity are included in the base model, as those are commonly recognized as being associated with AMI risk. To identify other potential confounders that would most strongly modify the risk of AMI, we assessed the association of each factor with vaccine exposure and the absolute percentage change of the estimated hazard ratio for vaccine exposure that would occur by adding potential confounders individually and in combinations to the model.

Adverse Events of Special Interest

Subjects with new onset AESI showed a wide spectrum of diagnoses and occurred in a low frequency in both treatment arms, 0.3% (n=29) vs 0.4% (n=15) subjects in the PSP (Table 37). Most abundant SOCs were neurological disorders with 9 vs 2 cases and endocrine disorders with 5 vs 5 cases in the PSP. The higher number of neurological cases was mainly driven by cases of Bell's palsy (6 vs. 2 cases in the PSP; 7 vs 2 cases in the TSP).

Table 31

Subjects With Potential New-Onset Adverse Events of Special Interest by SOC and Preferred Term (Primary and Total Safety Populations)

Category/Preferred Term n (%)	PSP Heplisav B (N = 9365) n (%)	PSP Engerix-B (N = 3867) n (%)	TSP Heplisav B (N = 10,038) n (%)	TSP Engerix-B (N = 4200) n (%)
Subjects with at least one potential adverse event of special interest	**29 (0.3)**	**15 (0.4)**	**31(0.3)**	**16 (0.4)**

Nervous System Disorders	**9 (<0.1)**	**2 (<0.1)**	**10 (<0.1)**	**2 (<0.1)**
Guillain-Barre Syndrome	1 (<0.1)	0	1 (<0.1)	0
VIth Nerve Paralysis	2 (<0.1)	0	2 (<0.1)	0
VIIth Nerve Paralysis	6 (<0.1)	2 (<0.1)	7 (<0.1)	2 (<0.1)
Endocrine Disorders	**5 (<0.1)**	**5 (0.1)**	**5 (<0.1)**	**5 (0.1)**
Autoimmune Thyroiditis	2 (<0.1)	2 (<0.1)	2 (<0.1)	2 (<0.1)
Basedow's Disease	3 (<0.1)	3 (<0.1)	3 (<0.1)	3 (<0.1)
Skin and Subcutaneous Tissue Disorders	**4 (<0.1)**	**3 (<0.1)**	**4 (<0.1)**	**3 (<0.1)**
Alopecia Areata	1 (<0.1)	0	1 (<0.1)	0
Cutaneous lupus erythematosus	0	1 (<0.1)	0	1 (<0.1)
Erythema Nodosum	1 (<0.1)	0	1 (<0.1)	0
Lichen Planus	1 (<0.1)	2 (<0.1)	1 (<0.1)	2 (<0.1)
Vitiligo	1 (<0.1)	0	1 (<0.1)	0
Musculoskeletal and Connective Tissue Disorders	**4 (<0.1)**	**2 (<0.1)**	**4 (<0.1)**	**3 (<0.1)**
Mixed Connective Tissue Disease	0	1 (<0.1)	0	1 (<0.1)
Polymyalgia Rheumatica	1 (<0.1)	0	1 (<0.1)	0
Rheumatoid Arthritis	1 (<0.1)	0	1 (<0.1)	1 (<0.1)
Scleroderma	0	1 (<0.1)	0	1 (<0.1)
Sjogren's Syndrome	1 (<0.1)	0	1 (<0.1)	0
Systemic Lupus Erythematosus	1 (<0.1)	0	1 (<0.1)	0
Vascular Disorders	**3 (<0.1)**	**1 (<0.1)**	**3 (<0.1)**	**1 (<0.1)**
Granulomatosis With Polyangiitis	1 (<0.1)	0	1 (<0.1)	0
Raynaud's Phenomenon	1 (<0.1)	1 (<0.1)	1 (<0.1)	1 (<0.1)
Takayasu's arteritis[a]	1 (<0.1)	0	1 (<0.1)	0
Gastrointestinal Disorders	**2 (<0.1)**	**2 (<0.1)**	**2 (<0.1)**	**2 (<0.1)**
Coeliac Disease	0	2 (<0.1)	0	2 (<0.1)
Colitis Ulcerative	2 (<0.1)	0	2 (<0.1)	0
Eye Disorders	**0**	**0**	**1 (<0.1)**	**0**
Uveitis	0	0	1 (<0.1)	0
Hepatobiliary Disorders	**1 (<0.1)**	**0**	**1 (<0.1)**	**0**
Biliary Cirrhosis Primary	1 (<0.1)	0	1 (<0.1)	0
Infections and Infestations	**1 (<0.1)**	**0**	**1 (<0.1)**	**0**
Cavernous Sinus Thrombosis[b]	1 (<0.1)	0	1 (<0.1)	0
Metabolism and Nutrition Disorders	**1 (<0.1)**	**0**	**1 (<0.1)**	**0**
Type 1 Diabetes Mellitus	1 (<0.1)	0	1 (<0.1)	0
Immune System Disorders	**0**	**1 (<0.1)**	**0**	**1 (<0.1)**
Anti-Neutrophil Cytoplasmic Antibody Positive Vasculitis	0	1 (<0.1)	0	1 (<0.1)

a Alternative diagnosis of intramural hematoma of the aorta (HBV-23, Section 12.2.2.5).
b Unconfirmed diagnosis of Tolosa Hunt syndrome (HBV-16, Section 12.4.2).

Laboratory findings

Clinical Chemistry and Blood cell count

No clear shifts in liver, creatinine, phosphate of haematology values were observed over time with Heplisav B in HBV-16.

Shifts towards higher values were observed for AST and ALT in a subset of individuals of HBV-23 at the time point 4 weeks post-vaccination with Heplisav B. In the Heplisav B group, 25 subjects reported 31 AEs of TEAEs of liver enzyme disorders (including ALT, AST and transaminase increase), and 7 subjects reported 8 AEs in the Engerix-B group. Because 2 subjects in HBV-16 had elevated liver enzymes on Study Day 1, the correct analysis of treatment-emergent AEs is 0.25% (n = 23) of subjects reported 29 treatment-emergent events in the Heplisav B group compared with 0.18% (n = 7) of subjects in the Engerix-B group, demonstrating only a minor difference between vaccine groups (imbalance not observed in TSP).The Applicant further explained that the numerical imbalances of shifts in liver laboratory values seen at weeks 4

were not associated with any relevant AEs, thus, decreasing the clinical importance of this observation, which was accepted by the CHMP**.**

There was an imbalance of AEs of anaemia observed in the TSP (14 vs 2 cases), which was most probably driven by underlying co-morbid AEs in the Heplisav B arm. No safety concern was detected.

No systematic shifts in WBC are observed in HBV-16 and in the Substudy of HBV-23 (Table 38).

Table 32
Study HBV-23 Shift table of laboratory data

Assay	**Treatment Group**	**Mean Laboratory Value**			
	H (207); E (102)	**Week 4**	**Week 8**	**Week 24**	**Week 56**
AST (U/L) normal to high	Heplisav B	12 (5.8)	3 (1.4)	8 (3.9)	5 (2.4)
	Engerix-B	0	3 (2.9)	1 (1.0)	2 (2.0)
ALT (U/L) normal to high	Heplisav B	8 (3.9)	8 (3.9)	9 (4.3)	5 (2.4)
	Engerix-B	1 (1.0)	2 (2.0)	3(2.9)	3 (2.9)
ALP (U/L) normal to high	Heplisav B	3 (1.4)	0	1 (0.5)	4 (1.9)
	Engerix-B	0	2 (2.0)	2 (2.0)	2 (2.0)
Bilirubin (mg/dl) normal to high	Heplisav B	2 (1.0)	1 (0.5)	3 (1.4)	2 (1.0)
	Engerix-B	1 (1.0)	0	0	2 (2.0)
Creatinine (mg/dl) normal to high	Heplisav B	6 (2.9)	5 (2.4)	2 (1.0)	4 (1.9)
	Engerix-B	4 (3.9)	4 (3.9)	3 (2.9)	3 (2.9)
Urea Nitrogen (mg/dl) normal to high	Heplisav B	1 (0.5)	7 (3.4)	3 (1.4)	3 (1.4)
	Engerix-B	4 (3.9)	1 (1.0)	4 (3.9)	3 (2.9)
RBC (x106/ul)	Heplisav B	10 (4.8)	7 (3.4)	9 (4.3)	14 (6.8)
Normal to low	Engerix-B	2 (2.0)	3 (2.9)	2 (2.0)	9 (8.8)
WBC (x103/ul)	Heplisav B	3 (1.4)	7 (3.4)	3 (1.4)	5 (2.4)
Normal to high	Engerix-B	3 (2.9)	4 (3.9)	5 (4.9)	2 (2.0)

Vital Signs

Safety was further assessed by changes in vital signs after treatment. In the pivotal trials HBV-10 and HBV-16, vital signs were similar between treatment groups.

HBV-23 sub-study of Thrombotic Disease

Overall, new-onset abnormal thrombotic tests occurred in 70 (33.8%) Heplisav B subjects and 33 (32.4%) Engerix-B subjects.

A new-onset increase of beta 2 glycoprotein 1 IgM was more often seen in Heplisav B treated individuals compared to Engerix-B (19 subjects (9.2%) vs. 2 subjects (2.0%) in PSP 7% vs. 1% in HBV-23) Similarly, there was a trend from normal at baseline to elevated levels of lupus anticoagulant screen test in Heplisav B subjects at Week 4 (n = 11 [5.3%]) and Week 8 (n = 30 [14.5%]), followed by a decline at Week 24. No subject with an elevated anti-beta 2 glycoprotein 1 IgM had a co-incident thrombotic or thromboembolic event or developed antiphospholipid syndrome. One subject with slightly elevated lupus anticoagulant and aPTT at baseline developed a myocardial infarction 68 days after vaccination with Heplisav B and a lung embolism 7 months later.

The applicant provided additional data on the kinetics of APL following vaccination. The only elevation of APL antibodies was in the anti-beta 2 glycoprotein 1 IgM. There was no increase in antibodies more specific for antiphospholipid antibody syndrome (APS): anti-cardiolipin antibodies, lupus anticoagulant, and anti-beta 2 glycoprotein 1 IgG. The CHMP concurred that anti-beta 2 glycoprotein 1 IgM is a non-specific antibody that has been reported following Engerix-B vaccination as well as other vaccines and infectious diseases. It is noted however that in HBV-23, the elevation was only seen in the Heplisav B group. Nevertheless, literature questions its value in the diagnosis of antiphospholipid syndrome (APS) (in contrast with anti-beta2 glycoprotein 1 IgG, anti-cardiolipin IgM and IgG, and lupus anticoagulant). In the absence of a thrombotic event, an elevation in anti-beta 2 glycoprotein 1 IgM alone does not constitute a diagnosis of APS, especially if the rise is transient. No subject with elevated anti-beta 2 glycoprotein 1 IgM in the lab subsample had a venous or arterial thrombotic event or MACE. The Applicant confirmed that antiphospholipid (APL) antibodies were not systematically described/available in the subjects who presented MACE, as the evaluation was done post-hoc, and only data collected in routine care were available.

Overall, clinical relevance of the isolated and transient increase of anti-beta 2 glycoprotein 1 IgM in the Heplisav B group is unclear. There is no evidence that Heplisav B induces an anti-phospholipid syndrome. The plausibility that the observed increased risk of AMI in HBV-23 would be due to antiphospholipid antibodies induced by Heplisav B is low.

Clotting assessment by prothrombin time and activated partial thromboplastin time were unremarkable for both groups (HBV-23 CSR).

Safety in special populations

Limited safety data are available for Heplisav B in paediatric subjects 11 to 17 years of age from 1 pivotal phase 3 trial investigating 11 (Heplisav B) and 2 (Engerix-B) individuals. As in adult subjects, the most frequent PIR (Heplisav B) in paediatric subjects was mild to moderate injection site pain. There were no deaths, SAEs, AESIs, related AEs, or discontinuations of study treatment due to an AE in paediatric subjects who received Heplisav B. The small number of subjects limits the ability to evaluate safety of Heplisav B in comparison to Engerix-B in this age subgroup.

No relevant discrepancies were observed in the geriatric patients (>65 years) between the Heplisav and Engerix arm in terms of frequency and severity of AEs.

Women experienced more PIRs compared to men. Of note, a higher rate of deaths was observed in women for Heplisav B (9 (0.2%) vs 1 (<0.1%)) compared to men (17 (0.3%) vs 7 (0.3%)). Furthermore, the difference of new onset AESIS was larger in women (10 (0.2%) vs 2 (<0.1%)) compared to men (8 (0.2%) vs 4 (0.2%)).

PIRs occurred in lower frequency in Blacks compared to White and Asians. The latter group had the highest frequency of PIRs. No other differences were noted in association to race.

Obese individuals experienced less often local or systemic reactions compared to non-obese individuals in both study arms. Of note, a higher slightly rate of deaths occurred with Heplisav B in the TSP in obese individuals. More individuals discontinued study treatment with Heplisav B due to SAEs in the obese group compared to non-obese subjects (8vs 4 in the non-obese group and 16 vs. 3 individuals in the obese group).

For subjects without type 2 diabetes, the overall rate of PIRs was similar between treatment groups. For subjects with type 2 diabetes, Heplisav B subjects had lower overall rate of PIRs compared to Engerix-B subjects. Discontinuation due to AEs and SAEs was higher in the diabetic group compared to non-diabetic subjects. Furthermore, a higher rate of deaths was seen for diabetics (5 (0.5%) vs 1 (0.2%)) compared to non-diabetic individuals (21 (0.2%) vs 7 (0.2%)) in disadvantage for Heplisav B. Of note, the overall difference in discontinuation is clearly driven by diabetic patients in the Heplisav B arm in study HBV-23 (DM: 14 vs 1 cases and subjects without DM: 6 vs 4 in HBV-23). There was a numerical imbalance in deaths between smokers who received Heplisav B, 15 deaths (0.49%) and smokers who received Engerix-B, 2 (0.15%).

40 (Heplisav B) and 20 (Engerix-B) women became pregnant under study treatment in the TSP. Of those, 24 and 11 individuals, respectively, had a healthy term delivery. In the Heplisav B arm, there were 2 premature deliveries, 3 spontaneous abortions and 1 congenital anomaly, 1 still birth, 1 induced abortion and 5 cases lost to FUP. The following cases were observed in the Engerix-B arm: 1 birth with congenital anomaly, 1 subject had a birth with foetal complications leading to SAEs, 2 subjects had spontaneous abortions, 1 induced abortion and 1 lost to FUP. 3 (Heplisav B) and 4 cases (Engerix-B) were elective terminations. Safety during pregnancy will be further evaluated in the pregnancy registry (study HBV 27).

Immunological events

General healthy adults (PSP/TSP)

ANA antibodies

The percentage of subjects converting from a negative to a positive ANA result was slightly higher in the Heplisav B arm (5.7% vs 5.3%). The percentage of subjects with a post-treatment rise in a positive pre-treatment titer was similar between groups (Heplisav B: 15.4%; Engerix-B: 16.8%). In the Heplisav B group, 1 subject with a post-treatment positive ANA had an exacerbation of their lupus and 1 subject had an exacerbation of hyperthyroidism.

Anti-double Stranded Deoxyribonucleic Acid Antibodies

The percentage of subjects converting from a negative to a positive titer was 1.3% in the Heplisav B group and 1.0% in the Engerix-B group. The results in the PSP w/o HBV-23 + supportive trials were similar to those in the PSP. Thus, small differences were seen in the development of anti-dsDNA antibodies in recipients of Heplisav B compared with recipients of Engerix-B.

Anti-neutrophil Cytoplasmic Antibodies

Retrospective pre- and post-vaccination testing for ANCA in 1 pivotal phase 3 trial and 1 supportive trial was performed. Testing did not reveal any additional events of development of ANCA in either treatment group. Positive screening ELISA results were infrequent and none were confirmed as c-ANCA- or p-ANCA-positive.

Patients with CKD

9.0% in the Heplisav B group and 12.3% in the Engerix-B group shifted to positive ANA results post-vaccination in HBV-17 and 5.3% in the Heplisav B group and 4.1% in the Engerix-B group showed a positive post-treatment result regarding anti dsDNA antibodies.

Safety related to drug-drug interactions and other interactions

No analysis of safety with respect to drug interactions was performed.

Discontinuation due to adverse events

Discontinuation rates due to AEs are presented in tables 23 and 24 above. Discontinuation rates due to SAEs were slightly higher in the Heplisav B group compared to Engerix-B (0.3% (24) vs. 0.2% (7)) (Table 39).

Table 33 Summary of Immune-mediated AEs, Deaths, and SAEs (PSP, TSP)

	PSP		TSP	
Event Type	Heplisav B (N=9365) n (%)	Engerix-B (N=3867) n (%)	Heplisav B (N=10038) n (%)	Engerix-B (N=4200) n (%)
Discontinued study treatment due to serious AE	24 (0.3)	7 (0.2)	24 (0.2)	7 (0.2)
Discontinued study treatment due to a serious related AE	1 (<0.1)	1 (<0.1)	1 (<0.1)	1 (<0.1)
Death	26 (0.3)	8 (0.2)	26 (0.3)	8 (0.2)
Related Death	0	0	0	0

AE = adverse event; AESI = adverse event of special interest; PSP = Primary Safety Population; SAE = serious adverse event; TSP = Total Safety Population.

Post marketing experience

Heplisav B was approved by the US FDA in November 2017. Three post-marketing studies are running investigating neurological and cardiac safety as well as safety under pregnancy of Heplisav B in the US. At the time of this marketing authorisation application, the majority of events reported have been injection site reactions, which are expected events, and the outcomes of all the events were reported as resolved. The number of events reported in the post-marketing stage was overall low and did not raise safety concerns at this stage.

2.6.1. Discussion on clinical safety

At the time of this marketing authorisation application, 11 clinical trials of Heplisav B were completed in healthy individuals, comprising 3 pivotal trials (HBV-10, HBV-16, HBV-23) and 8 supportive trials.

The CHMP considered that at the time of the previous submission in 2012, concerns were raised with respect to study HBV-017 and to some extent also to studies HBV-010 and HBV-016. The EMA inspection demonstrated a lack of adequate oversight of the clinical trials conducted by the sponsor. These deficiencies led to a high number of protocol deviations not systematically reported in the CSR and to inconsistencies of the data, and it was concluded that the data were not complete and reliable. The findings detected were related to the inappropriate quality assurance. The inspectors therefore concluded that those findings might also have impacted the previous and ongoing studies performed by the sponsor. The Applicant claimed that following the 2012 EMA inspection (HBV-017), the CSRs for HBV-10 and HBV-16 were amended. However, the CHMP considered that it remained unclear to which extent the issues could be corrected retrospectively. Overall, the inspection outcome was a recommendation not to use data of trial HBV-17 performed in patients with chronic kidney disease. This trial was considered as pivotal at the time of the previous MAA submission. In view of the inspection recommendations, safety data generated in HBV-17 and the smaller supportive trials have only limited value.

In the PSP, the frequency of AEs/MAEs did not differ between study arms (55.3% vs 58.1%). No differences were observed for discontinuation rates.

The most often occurring AEs by PT in the PSP w/o HBV-23 and in HBV 23 were balanced, including nasopharyngitis, headache, back pain and upper respiratory tract infection. In the CKD pool, immune system disorders occurred slightly more often with Heplisav B (6 (2.4%) vs 3 (1.1%)). Hypersensitivity AEs were uncommon and balanced in the different study pools.

Adverse events deemed related by the investigator occurred equally often in PSP w/o HBV-23 (headache, injection site pain and erythema, and fatigue). Rash was observed infrequently (4 (0.1%) vs. 1 (<01%)). A few cases of arthralgia were observed with Heplisav B in PSP w/o HBV-23 (6 (0.2%) vs. 0). AEs deemed related in CKD patients were less frequently seen with Heplisav B (7.5% vs 9.9%). One case of anaphylaxis deemed related to Heplisav B was reported post marketing and anaphylaxis was reflected in the Product Information as a very rare adverse drug reaction.

Severe AEs comprised headache, back pain, and nasopharyngitis in the PSP w/o-HBV23 pool and were balanced (7.6% vs. 10.1%). In the HBV-23 pool, acute myocardial infarction (0.3 vs < 0.1%) was more often observed with Heplisav B. Severe events in CKD occurred in higher frequency in both study arms (59 (23.2%) vs. 64 (24.4%)) and were slightly imbalanced for pulmonary edema (1.6% vs 0.4%), congestive cardiac failure (7 (2.8%) vs 3 (1.1%)), and anemia (5 (2.0%) vs 2 (0.8%)).

The Applicant further investigated disbalanced MAEs in HBV-23. Herpes zoster occurred more often with Heplisav B (0.7% vs 0.3%, RR 2.1). 19 MAEs with large RR and CIs including 1 were detected comprising excoriation (10.45 (0.61 -178.33), wound healing 9.46 (0.55 -162.45), lipoma 8.46 (0.49 -146.57), acute myocardial infarction 6.97 (0.92 - 52.97), and bipolar I disorder 6.47 (0.36 -114.83).

SAEs occurred overall in similar frequency in the study arms (4.8%) of the PSP. A higher number of myocardial infarctions was seen for Heplisav B (16 vs 2 cases PSP, 17 vs 2 cases TSP). SAEs in the CKD pool (HBV-17 SP (68 (26.8%) vs. 76 (29%)) occurred overall in higher frequency compared to healthy individuals. One SAE, end-stage renal disease, was considered as probably related to study treatment according to investigators.

Imbalances in the number of myocardial infarctions have been observed in the pivotal trials, which raised a major concern. A 2.17x higher rate for the MI SMQ (0.22% (21) versus 0.1% (4)) was seen in the PSP and a threefold higher rate specifically in HBV-23. Numbers of MIs in the PSP started to separate between day 80 and 90 after the first injection. Cases within 60 days after the last injection might be slightly unbalanced (7 vs. 2 cases). As a result of MI imbalances, MACE occurred in higher frequency in the Heplisav B arm in the PSP (RR 1.5/1.6 before and after adjudication) which was specifically driven by cases in HBV-23 (RR 3.3 after adjudication). MACE imbalances started after study day 100 in the PSP. When taking relabeled undetermined MACE into consideration, Cox regression analyses become significant for MACE and borderline significant for MI in HBV-23.

Of note, events of stroke are balanced in the PSP. Proportions of cardiac failure, atrial fibrillation or angina pectoris did not show significant differences either. An imbalance in MACE was not observed in the CKD pool.

Further to the CHMP request, the Applicant discussed biological plausibility based on the available literature and on pre-clinical and clinical observations investigating the effect of TLR-9 and the use of TLR-9 agonists on atherogenesis, inflammation and thrombotic/thromboembolic events. Although stimulation of TLR-9 may have the potential to induce inflammatory and proatherogenic effects, these effects are seen at much higher plasmatic concentrations with PS ODNs binding to TLR-9 in animal experiments compared to the

concentrations reached with the adjuvant in humans. Coagulation or platelet function are not to a relevant extent affected by the adjuvant in the doses applied to humans. No significant differences were seen in thrombotic/thromboembolic events. Thus, a plausible mechanism explaining the observed discrepancy in cardiac events in the pivotal trials could not be identified.

A large observational post-marketing surveillance study is currently underway in the United States (HBV-25) to further study cardiac safety with Heplisav B. The study was initiated in the third quarter of 2018 and is expected to be completed in 2021. The applicant performed unadjusted and adjusted cox regression analyses calculating a hazard ratio of 0.83 (95% CI=0.50, 1.37, unadjusted HR=0.78). The Applicant has adjusted for baseline differences between treatment arm in an adjusted Cox PH model (adjusted for sex, age, vaccine exposure, race/ethnicity, history of AMI, use of nitrate medication without a clear description of the variable selection procedure).

In summary, there is no plausible molecular-biological mechanism triggered by the vaccine. The finding of excess cardiovascular risk in the clinical programme was unexpected and largely confined to one study. While some extent of residual confounding cannot be ruled out in the context of a non-randomised study, the available outcomes of the post-marketing surveillance study provided acceptable reassurance that a true increase of cardiovascular risk of patients receiving Heplisav B (vs ENGERIX-B) is sufficiently unlikely. Finally, acute myocardial infarction has been added as an important potential risk in the RMP, and the Applicant has committed to providing the final report of acute MI unconfirmed events and of confirmed events in HBV-25 study.

The frequency of deaths in the PSP was only slightly higher with Heplisav B (n = 26, 0.26%) compared to Engerix-B (n = 8, 0.19%). 32 of 34 deaths occurred in the HBV-23 study. No deaths were assessed by the investigators as related to study treatment. In the CKD SP fatal cases occurred in higher frequency with Heplisav B compared to Engerix-B (13 deaths: 9/331 (2.7%) and 4/272 (1.5%)). All cases were judged as unrelated to vaccination by study investigators.

Acute renal events were comparable in the study pools. AEs of "Liver enzyme elevations" occurred seldomly (0.25% vs. 0.18%) in the PSP. Anaemia (PSP: 17 (0.3%) vs. 7 (0.3%)) was more often seen within 42 days after Heplisav B vaccination (14 vs 2 cases), which was most probably driven by co-morbid AEs.

In the PSP w/o HBV-23, the frequency of local (42.81% vs. 41.1%,) and systemic post-injection reactions (32.3% vs. 37.4%) was roughly equal between the vaccine and control arm. Injection site redness and swelling occurred slightly more often with Heplisav B (3.7% vs 1.1% and 2.4% vs 1.3%). Most PIRs resolved within 7 days. No safety concern was identified. In CKD patients, the frequency of PIRs was comparable in the treatment arms.

New AESIs occurred in an overall low frequency in both treatment arms of the PSP (0.3% (n=29) vs 0.4% (n=15)). Most abundant SOCs were neurological disorders with 9 vs 2 cases and endocrine disorders with 5 vs 5 cases in the PSP. The number of neurological cases was driven by cases of Bell's palsy (6 vs. 2 cases PSP).

No safety concern was raised when looking at the disbalances in Bell's palsy and H. zoster.

AESIs other than Bell's palsy were infrequent and comparable (0.11% / 0.08%). Three rare serious AESIs were reported in the Heplisav B group: cavernous sinus thrombosis, GBS and polyangiitis granulomatosa (possibly related). Guillain-Barré syndrome occurred more than 3.5 months after the last dose and 5 days after an influenza vaccination (HBV-10 trial). Thus, a clear causal association was not observed. In patients

with CKD, two cases of dermatitis (palmoplantar dermatitis and lichenoid keratosis) were considered unrelated.

No imbalances were detected with regards to the thyroid (e.g. hypothyroidism, hyperthyroidism, Basedow disease, autoimmune thyroiditis).

Given the occurrence of two cases of very rare autoimmune vasculitis, the CHMP considered that inclusion of potentially immune mediated events as an important potential risk in the RMP is justified.

Study HBV-26 to evaluate new onset immune mediated diseases, HZ and anaphylaxis with Heplisav B (n=30,000) vs Engerix (n=30,000) is ongoing in the US. No imbalances of AESIs were detected based on the interim data.

Several subjects presented pre-existing autoimmune diseases at baseline in the PSP (106 (Heplisav) and 34 (Engerix). Of those, 4 (3.8%) in the Heplisav B group and 1 (2.9%) subject in the Engerix-B group experienced an exacerbation. In this context, exacerbations of autoimmune disease in persons with a history of autoimmune disease were reflected as an important potential risk in the RMP.

Laboratory value analyses showed new-onset beta 2 glycoprotein 1 IgMs more often with Heplisav B (19 subjects (9.2%) vs. 2 subjects (2.0%)) without co-incident thrombotic events or development of antiphospholipid syndrome. No safety concern was detected. No significant shifts were detected for autoimmunity markers (ANA, ds-DNA, ANCA). Clotting assessments (PT, aPTT) were comparable in both study arms.

No significant safety concerns were detected in special populations.

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

2.6.2. Conclusions on the clinical safety

The safety profile of Heplisav B was considered acceptable. Interim results from an ongoing observational study evaluating the risk of myocardial infarction, which was reported in the primary safety population, were reassuring. The safety profile of Heplisav B is considered to support a positive B/R balance.

2.7. Risk Management Plan

Safety concerns

Summary of Safety Concerns	
Important Identified Risks	None
Important Potential Risks	Acute myocardial infarction Potentially immune-mediated disorders (including inflammatory disorders) Exacerbation of potentially immune-mediated disorders (including inflammatory disorders) in

	individuals with a history of immune-mediated disorder
Missing Information	Safety in pregnancy and lactation Safety in immunocompromised patients including persons living with HIV Concomitant administration with other vaccines

Pharmacovigilance plan

Study	Summary of objectives	Safety concerns addressed	Protocol link Milestones
HBV-25: Post-Marketing Observational Surveillance Study to Evaluate the Occurrence of Acute Myocardial Infarction	The primary objective is to compare the occurrence of AMI in recipients of Heplisav B with recipients of Engerix-B.	Acute myocardial infarction	Protocol HBV-25 HBV-25 IA CSR Interim Study Report: 26/11/2019 Final study report submission: 30/06/2021
HBV-26: Post-Marketing Observational Surveillance Study to Evaluate the Incidence of New-Onset Immune-mediated Diseases, Herpes Zoster, and Anaphylaxis	The primary objective is to describe and compare the incidence of new-onset immune-mediated diseases, herpes zoster, and anaphylaxis in recipients of Heplisav B with recipients of another hepatitis B vaccine.	Potentially immune-mediated disorders (including inflammatory disorders)	Protocol HBV-26 HBV-26 Status Update Report Study Status Report: 06/03/2020 Final study report submission: 30/06/2021
HBV-27: Pregnancy Registry	The objective is to evaluate pregnancy outcomes among women who received a dose of the Heplisav B vaccine	Pregnancy outcomes	Protocol HBV-27 Final Study Report: Not available

	within 28 days prior to conception or at any time during pregnancy.		Final study report submission: 31/12/2023

Risk minimisation measures

Safety concern	Risk minimisation measures	Pharmacovigilance activities
Acute myocardial infarction	No risk minimisation measures planned	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: None Additional pharmacovigilance activities: HBV-25: Study to Evaluate the Occurrence of Acute Myocardial Infarction
Potentially immune-mediated disorders (including inflammatory disorders)	No risk minimisation measures planned	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: None Additional pharmacovigilance activities: HBV-26: Study to Evaluate the Incidence of New-Onset Immune-mediated Diseases
Exacerbation of potentially immune-mediated disorders (including inflammatory disorders) in individuals with a history of immune-mediated disorder	No risk minimisation measures planned	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: None Additional pharmacovigilance activities: None
Safety in pregnancy and lactation	Routine Risk Minimisation Measures: Section 4.6 of the SmPC PL Section 2 regarding lack of data	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: None Additional pharmacovigilance activities:

Safety concern	Risk minimisation measures	Pharmacovigilance activities
	Subject to medical prescription Additional Risk Minimisation Measures: None	HBV-27: Pregnancy Registry
Safety in immunocompromised patients including persons living with HIV	Routine Risk Minimisation Measures: Section 4.4 of the SmPC Refer to PL Section 2 Subject to medical prescription Additional Risk Minimisation Measures: None	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: None Additional pharmacovigilance activities: None
Concomitant administration with other vaccines	Routine Risk Minimisation Measures: See Section 4.5 of the SmPC Subject to medical prescription Additional Risk Minimisation Measures: None	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: None Additional pharmacovigilance activities: None

Conclusion

The CHMP and PRAC considered that the risk management plan version 1.0 is acceptable.

2.8. Pharmacovigilance

Pharmacovigilance system

The CHMP considered that the pharmacovigilance system summary submitted by the applicant fulfils the requirements of Article 8(3) of Directive 2001/83/EC.

Periodic Safety Update Reports submission requirements

Based on the use of a novel adjuvant CpG 1018, the CHMP is of the opinion that a separate entry in the EURD list for Heplisav B is needed, as it cannot follow the already existing entry for hepatitis B vaccine (rDNA). The requirements for submission of periodic safety update reports for this medicinal product are set out in the Annex II, Section C of the CHMP Opinion. The applicant requested alignment of the PSUR cycle with the international birth date (IBD). The IBD is 09.11.2017. The new EURD list entry will therefore use the

IBD to determine the forthcoming Data Lock Points.

2.9. Product information

2.9.1. User consultation

The results of the user consultation with target patient groups on the package leaflet submitted by the applicant show that the package leaflet meets the criteria for readability as set out in the *Guideline on the readability of the label and package leaflet of medicinal products for human use.*

2.9.2. Additional monitoring

Pursuant to Article 23(1) of Regulation No (EU) 726/2004, Heplisav B (hepatitis B surface antigen) is included in the additional monitoring list as it is a biological product authorised after 1 January 2011.

Therefore the summary of product characteristics and the package leaflet includes a statement that this medicinal product is subject to additional monitoring and that this will allow quick identification of new safety information. The statement is preceded by an inverted equilateral black triangle.

3. Benefit-Risk Balance

3.1. Therapeutic Context

3.1.1. Disease or condition

The indication for Heplisav B is prevention of infection caused by all known subtypes of hepatitis B virus (HBV) in adults 18 years of age and older.

Hepatitis B is a potentially life-threatening liver infection caused by HBV. It is a major cause of morbidity and mortality in Europe and worldwide due to its long-term sequelae, including chronic active hepatitis, cirrhosis, liver failure and liver cancer.

HBV infection can occur through several ways of transmission, of which the major modes are sexual exposure, exposure to blood (percutaneous or mucosal) and injection drug use.

Hepatitis B is spread through contact with contaminated body fluids. Most infections occur by contact with infected blood, but semen, saliva and cervical secretions can also be infectious. The virus can live on surfaces for at least seven days which means it can be transmitted via objects that have been contaminated with infected body fluids (e.g. used needles). Sexual transmission and injecting drug use are the most common current routes of transmission in Europe.

Transmission may also occur in healthcare settings due to the reuse or inadequate sterilisation of medical equipment, especially syringes and needles. Transmission via blood transfusion or via plasma-derived products is now rare in Europe due to effective blood safety programmes.

The development of chronic HBV infection is inversely associated with the age at which the individual is infected. Up to 90% of infants who are infected with the virus develop chronic infections but less than 5% of infected adults develop chronic infection. Individuals with chronic HBV infection are at a higher risk of complications including liver cirrhosis (25%) and cancer (5%). In addition, they may pose a risk of transmitting the infection to others.

Thanks to testing programmes during pregnancy and vaccination at birth, perinatal HBV transmission now only occurs rarely in Europe but remains one of the major routes of transmission globally.[2]

3.1.2. Available therapies and unmet medical need

Safe and effective hepatitis B vaccines have been available and recommended for use in adults with risk factors for exposure to HBV since 1992.

Most countries in Europe have implemented a universal vaccination programme in infants through combination vaccines. The implementation of blood safety strategies and safe injection practices, as well as safer sex practices can also prevent HBV transmission.

Standard hepatitis B vaccines (e.g. Engerix B) are typically aluminium adjuvanted. They are given in a 3-dose schedule usually over a six-month period with a higher posology in adults (typically 20 µg) compared to

[2] https://ecdc.europa.eu/en/hepatitis-b/facts

children (10 µg). Three doses of standard hepatitis B vaccine offer long-term protection of at least 30 years to most (>95%) healthy infants, children and young adults.

Only 10-20% of adult vaccinees mount a protective serum antibody titre (10 mIU/mL anti-HBsAg) within one month of the first dose of standard HBV vaccine. The delay in generation of protective antibody responses is of particular importance for individuals at high risk of HBV infection, e.g., health care workers, persons about to begin haemodialysis, travellers to areas where HBV-infection is prevalent, or individuals in high risk behaviour groups.

Vaccine efficacy can be hindered by the occurrence of hypo- or non-responders among some groups such as older individuals and subjects with renal failure or diabetes (30-60% of hypo- or non-responders).

To achieve sufficient protection, patients with renal insufficiency are offered schedules that include >3 doses of the standard vaccine, or vaccine containing a higher dose of HBsAg (e.g. double the usual adult dose) on each occasion, or both.

A more potent adjuvanted vaccine is available (Fendrix) but indicated only in adult patients with renal insufficiency. It contains 20 µg of HBsAg and the potent adjuvant ASO4 (also present in Cervarix) and is given in a 4-dose schedule over a six-month period with additional booster doses recommended in pre-haemodialysis and haemodialysis patients who are at high risk of HBV infection. This vaccine elicits an earlier, higher and more long-lasting antibody response than a corresponding series of 4 double doses (40 µg) of a standard hepatitis B vaccine. The safety and reactogenicity of the adjuvanted vaccine administered as a booster dose in pre-haemodialysis and haemodialysis patients are acceptable, although reactogenicity data suggest an increase in the incidence of local injection site symptoms.

An improved hepatitis B vaccine that can elicit high levels of seroprotection and earlier seroprotection in adults that is also effective in hyporesponsive populations and requires fewer doses over a shorter time than the currently licensed vaccines is considered to reduce the risk of HBV infection and associated morbidity and mortality.

Heplisav B contains the same amount and type of antigen as hepatitis B vaccines licensed previously, but includes a novel adjuvant, 1018 ISS. Heplisav B is designed to give faster protection and requires fewer doses compared to other hepatitis B vaccines.

3.1.3. Main clinical studies

All pivotal phase 3 trials were observer-blinded, randomized, active-controlled, parallel-group, multicentre trials including mainly healthy adult population. In the pivotal studies, Heplisav B was given in a 20 µg HBsAg 3000 µg 1018 dose at 0, 4 weeks (placebo at 24 weeks) in head-to head comparison with licensed Hepatitis B vaccine Engerix- B with its standard composition and schedule. The investigated qualitative measure was seroprotection rate (SPR % of subjects who seroconverted in at least 10 mIU/ml) and the quantitative measure was geometric mean concentration (GMC) of antibodies after vaccination. An anti-HBs concentration of 10 mIU/mL or more measured 1 to 3 months after administration of the last dose of the primary vaccination series is considered a reliable correlate of protection against infection.

3.2. Favourable effects

Adequate immune response has been demonstrated for Heplisav B in generally healthy adults from age 18 to 70. The SPR in pooled pivotal studies was 95.7 % (95% CI 95.3 - 96.1). All clinical studies have consistently

shown non-inferior and mostly superior immune response to Heplisav B compared to Engerix-B. In comparison to available vaccines, fewer doses of Heplisav B are needed (2 vs. 3). Responses are more rapid (after 8 weeks) and occur at a higher level of protection (95% SPR in all subjects and lowest 88% in diabetes 2 subgroup). Higher immune response to Heplisav B compared to Engerix-B was also demonstrated in subgroups where the immunogenicity of Engerix-B has been limited (e.g. persons with type 2 diabetes, obese and smokers).

3.3. Uncertainties and limitations about favourable effects

There are limited data available about immunogenicity and durability of the antibody response induced by Heplisav B in persons with certain medical conditions. The Applicant is conducting clinical studies in following populations: patients receiving hemodialysis, HIV positive subjects, patients receiving immunosuppressive medication, patients with Chronic Lymphocytic Leukemia, non-responders to other Hepatitis vaccines.

Furthermore, there is currently very limited information about Heplisav B interactions with other vaccines. In the ongoing Post-Marketing Observational Surveillance Study, DV2-HBV-25, 45.4% of Heplisav B recipients received another vaccine on the same day as the Heplisav index dose administration. Influenza vaccines and pneumococcal vaccines were the vaccines most frequently administered with hepatitis B vaccines. These are observational studies in which no immunogenicity and reactogenicity data will be generated. In addition, safety data will not be captured using definitions and procedures as in standard interventional co-administration trials. Retrospective studies are not considered adequate to assess vaccines' co-administration.

Since Heplisav B induces high Ab GMC in generally healthy subjects, it is not expected that seroprotection rates would be affected by concomitant vaccination. However, in the absence of data, the concomitant use of Heplisav B with other vaccines is currently not recommended. This has been reflected in the Product Information and in the RMP (as "missing information"). In addition, the CHMP also considered that Heplisav B administration could affect the immune responses induced by the vaccines concomitantly administered.

3.4. Unfavourable effects

Compared to Engerix-B, Heplisav B shows a slightly lower frequency of systemic post-injection reactions and a similar frequency of local post-injection reactions. The most common solicited AEs were injection site pain, headache, malaise, fatigue, and myalgia with frequencies above 10%. The frequency of fever ≥38°C was 1.7% (0.2% had fever ≥39°C).

A few cases of new onset and exacerbations of autoimmune diseases have been observed, some of which had a tight time relationship with vaccine administration in the phase 3 programme. Potentially immune-mediated disorders (including inflammatory disorders) and exacerbation of potentially immune-mediated disorders (including inflammatory disorders) in individuals with a history of immune-mediated disorder have been reflected in the RMP as important potential risks.

However, an overall disbalance in the frequency of immune mediated events has not been observed in the phase 3 programme.

The CHMP considered that a large PASS study is currently performed studying new onset immune mediated events in a prospective manner. The interim study data did not identify critical findings.

3.5. Uncertainties and limitations about unfavourable effects

The CHMP considered that cardiac AEs were more frequent with Heplisav B compared with Engerix-B. An increased RR of 2.17 was observed for the myocardial infarction (MI) SMQ (0.22% (21) versus 0.1% (4)) in the primary safety population (PSP), with a 3-fold higher rate of these events in subjects vaccinated with Heplisav B, specifically in study HBV-2. This included subjects with cardiovascular risk factors. A Kaplan-Meier plot showed that events of the MI SMQ in the PSP started to separate between day 80-90 after the first injection and day 50-60 after the 2nd injection. Through study day 56, (28 days after the 2nd injection), the proportion of subjects reporting a major adverse cardiac event (MACE) was balanced in the PSP when considering cases after confirmation/adjudication. MACE imbalances started after Study Day 100.

Further to the CHMP request, the Applicant presented interim analyses from a large observational, non-randomised study of an MI risk in patients receiving Heplisav B or Engerix B, which is conducted as a post-marketing commitment after the US authorisation. These data showed a hazard ratio of 0.83 (95% CI=0.50, 1.37, unadjusted HR=0.78) favouring Heplisav B, and thus not supporting a cardiac risk with this product. Despite the intrinsic limitations of a non-randomised study, the CHMP considered these analyses, together with the lack of a clear biological rationale, sufficiently reassuring.

3.6. Effects Table

Table 34 Effects Table for Heplisav B - indicated for the active immunisation against HBV infection

Effect	Short Description	Unit	Heplisav B	Egnerix-B	Uncertainties/ Strength of evidence	References
Favourable Effects						
Seroprotection	Seroprotection rate (95%CI) at week 12 (Heplisav) or 28 (Engerix-B)	%	95.0 (93.9 - 96.1)	81.2 (77.8-84.6)	**SoE:** Diff. (95%CI) 13.7 (0.4-17.5) Non-inferior and superior seroprotection rate; **Unc:** GMC ratio's of anti-HB antibodies lower for Heplisav versus Egnerix 317 vs 352.	HBV-010
Seroprotection	Seroprotection rate (95%CI) at week 12 (Heplisav) or 32 (Engerix-B)	%	90.1 (88.2 - 91.8)	70.5 (65.5–75.5)	**SoE:** Diff. (95%CI) 19.6 (14.7-24.8) Non-inferior and superior seroprotection rate; Lot consistency was in acceptable limits since week 12 on, but not earlier; Non-inferior and superior GMC ratio of anti-HB antibodies diff. (95%CI) 2.5 (1.9-3.4)	HBV-016
Seroprotection	Seroprotection rate (95%CI) at week 28 among subjects with DM type 2	%	90.0 (87.4- 92.2)	65.1 (59.6-70.3)	**SoE:** Diff. (95%CI) 24.9 (19.3-30.7) Non-inferior and superior seroprotection rate; findings in line with the total population: 14.2 (12.5-15.9); Non-inferior and superior GMC ratio's of anti-HB antibodies diff. (95%CI) 2.5 (1.8-3.5), 1.2 (1.1-1.4), respectively	HBV-023
Seroprotection	Seroprotection rate (95%CI) at week 24 (Heplisav) and week 28 (Engerix-B)	%	95.7 (95.3 - 96.1)	79.5 (78.2 - 80.8)	**SoE:** Pooled analysis of 3 the pivotal studies Heplisav B induced significantly higher peak SPRs than Engerix B in subpopulations known to have reduced SPRs; older adults, men, subjects with diabetes mellitus, obese subjects, and smokers.	Pooled analysis
Unfavourable Effects						
MACE	Incidence of MACE / (n/N)	%	0.59 (55/9365)	0.39 (15/3867)	**Unc:** RR 1.54; Loose time to event relationship; MACE after adjudication: RR 1.6; differences primarily driven by study HBV-23 (MACE RR 2.0 (1.1-3.7) before adjudication, 2.3 (1.0-5.6) after adj. and by MI (0.2 vs 0.1): RR 2.2 (.8-6.3).	PSP
H. zoster	Incidence of H. Zoster (n/N)	%	0.7 (38/9365)	0.3 (9/3867)	**Unc:** RR 2x increased with Heplisav B	PSP
Hyperthyreosis			n=5	n=0	**Unc:** Numerical imbalance in a few cases	PSP
Bell's palsy			n=6	n=2	**Unc:** Numerical imbalance in a few cases; putatively associated with imbalance in H. zoster?	PSP

Abbreviations: GMC : geometric mean antibody concentration ; SPR: Seroprotection rate; PSP: primary safety population (13,232 adults 18 to 70 years of age in the 3 pivotal phase 3 trials); MACE: Major Adverse Cardiac Event; RR: Relative Risk

3.7. Benefit-risk assessment and discussion

3.7.1. Importance of favourable and unfavourable effects

The immune response to Heplisav B was shown to be superior to those of Engerix-B using SPR (i.e. antibody concentrations ≥10 mIU/mL), which is considered a relevant measure that has been established as a serological correlate of protection. The advantage of Heplisav over Engerix-B lies mainly in the earlier onset of protection (2 months), and the dose schedule: two doses at 0, 4 weeks rather than three doses at 0, 4, 24 weeks.

The overall reactogenicity of Heplisav is considered comparable to Engerix-b and thus acceptable.

Two PASS studies (category 3 studies) are investigating the potential of Heplisav B to induce immune mediated events (HBV-26) and cardiovascular events (HBV-25), respectively. Interim analyses of both studies did not raise a safety concern at this stage.

3.7.2. Balance of benefits and risks

The immunogenicity of Heplisav B was convincingly shown, as the seroprotection rate reached more than 90 % in all subgroups, and duration of protection was at least 52 weeks or longer. The earlier onset of seroprotection and a lower number of doses required for adequate seroprotection compared to Engerix-B were considered important advantages. Overall, the CHMP concluded that benefits of Heplisav B outweighed its risks. The safety profile was considered acceptable and the risks were adequately addressed through the Risk Management Plan and the Product Information.

3.8. Conclusions

The overall B/R of Heplisav B is positive.

4. Recommendations

Outcome

Based on the CHMP review of data on quality, safety and efficacy, the CHMP considers by consensus that the benefit-risk balance of Heplisav B is favourable in the following indication:

Heplisav B is indicated for the active immunisation against hepatitis B virus infection (HBV) caused by all known subtypes of hepatitis B virus in adults 18 years of age and older.

The use of Heplisav B should be in accordance with official recommendations.

It can be expected that hepatitis D will also be prevented by immunisation with Heplisav B as hepatitis D (caused by the delta agent) does not occur in the absence of hepatitis B infection.

The CHMP therefore recommends the granting of the marketing authorisation subject to the following conditions:

Conditions or restrictions regarding supply and use

Medicinal product subject to medical prescription.

Official batch release

In accordance with Article 114 Directive 2001/83/EC, the official batch release will be undertaken by a state laboratory or a laboratory designated for that purpose.

Other conditions and requirements of the marketing authorisation

Periodic Safety Update Reports

The requirements for submission of periodic safety update reports for this medicinal product are set out in the list of Union reference dates (EURD list) provided for under Article 107c(7) of Directive 2001/83/EC and any subsequent updates published on the European medicines web-portal.

The marketing authorisation holder shall submit the first periodic safety update report for this product within 6 months following authorisation.

Conditions or restrictions with regard to the safe and effective use of the medicinal product

Risk Management Plan (RMP)

The MAH shall perform the required pharmacovigilance activities and interventions detailed in the agreed RMP presented in Module 1.8.2 of the marketing authorisation and any agreed subsequent updates of the RMP.

An updated RMP should be submitted:

- At the request of the European Medicines Agency;
- Whenever the risk management system is modified, especially as the result of new information being received that may lead to a significant change to the benefit/risk profile or as the result of an important (pharmacovigilance or risk minimisation) milestone being reached.

Conditions or restrictions with regard to the safe and effective use of the medicinal product to be implemented by the Member States

Not applicable.

These conditions fully reflect the advice received from the PRAC.