30 May 2024
EMA/323453/2024
Committee for Medicinal Products for Human Use (CHMP)

Assessment report

Durveqtix

International non-proprietary name: fidanacogene elaparvovec

Procedure No. EMEA/H/C/004774/0000

Note

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

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List of abbreviations

AAV	Adeno-associated virus
AE	Adverse event
AIDS	Acquired immunodeficiency syndrome
ALP	Alkaline phosphatase
ALT	Alanine transaminase
aPTT	Activated prothrombin thromboelastin time
AS	Active substance
ARMM	Additional risk minimisation measure
AST	Aspartate transaminase
ATHN	American Thrombosis and Hemostasis Network
BU	Bethesda unit
CI	Confidence interval
CMA	Critical material attributes
CO_2	Carbon dioxide
CPV	Continued Process verification
CQA	Critical quality attributes
CpG	Cytosine-phosphate-guanine dinucleotides
CPP	Critical Process Parameter
CPV	Continued process verification
CSR	Clinical study report
DOE	design-of-experiments
DNA	deoxyribonucleic acid
DP	drug product
DRG	dorsal root ganglion
DS	drug substance
EEA	European Economic Area
EMA	European Medicines Agency
EPAR	European Public Assessment Report
ESI	Electrospray ionization
EU	European Union
FIX	Factor IX
FIX:C	Factor IX activity in circulation
FMEA	Failure mode and effects analysis
GMO	Genetically modified organism
HCP	healthcare provider
HEK293	human embryonic kidney cell line
IFN	Interferon
IPT	In-process test
IV	Intravenous
kg	kilogram
LFT	liver function test
LM-PCR	Ligation-mediated PCR
LoQ	Limit of quantitation
MCB	Master cell bank

MedDRA	Medical Dictionary for Regulatory Activities
MHC	major histocompatibility complex
mL	Millilitre
MS	Mass spectrometry
nAb	Neutralizing antibody
Non-CPP	Non-Critical Process Parameter
NHP	Nonhuman primate
NOAEL	No-observed adverse effect-level
OOS	Out of specification
PBMC	Peripheral blood mononuclear cells
PD	pharmacodynamics
Ph. Eur.	European Pharmacopoeia
PL	package leaflet
PPA	Process performance attribute
PPQ	Process performance qualification
PoC	Proof of concept
PSUR	Periodic Safety Update Report
PT	Preferred Term
QA	Quality Assurance or Quality Attribute
QC	Quality Control
RA	Risk assessment
rAAV	Recombinant Adeno-Associated Virus
RB	Roller bottle
RM	risk management
RMM	risk minimisation measure
RMP	Risk Management Plan
RP	Reverse phase
RSD	Relative Standard Deviation
S-EPTS	shearing extension primer tag selection
SD	standard deviation
SmPC	Summary of Product Characteristics
SMQ	Standardised MedDRA Query
SMR	standardized mortality ratio
SOC	system organ class
TAT	thrombin-antithrombin
TEG	Thromboelastography
TES	Targeted enrichment sequencing
ULN	upper limit of normal
U/ml	Units per milliliter
US	United States
USP	United States Pharmacopeia
VF	Viral filtration
vg	Vector genomes
vg/kg	Vector genomes per kilogram
WBCT	Whole blood clotting time
WCB	Working cell bank

WFH	World Federation of Hemophilia
WT	Wildtype

1. Background information on the procedure

1.1. Submission of the dossier

The applicant Pfizer Europe MA EEIG submitted on 20 April 2023 an application for marketing authorisation to the European Medicines Agency (EMA) for Durveqtix, through the centralised procedure falling within the Article 3(1) and point 1 of Annex of Regulation (EC) No 726/2004.

Durveqtix was initially designated as an orphan medicinal product EU/3/18/2090 on 19 November 2018 in the following condition: Treatment of haemophilia B. The orphan designation was later withdrawn on 13 December 2023.

The applicant applied for the following indication: for the treatment of severe and moderately severe haemophilia B (congenital factor IX deficiency) in adult patients without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74.

1.2. Legal basis, dossier content

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 test(s) or study(ies).

1.3. Information on paediatric requirements

Pursuant to Article 7 of Regulation (EC) No 1901/2006, the application included EMA Decisions P/0222/2020, P/0382/2021 and P/0277/2022 on the agreement of a paediatric investigation plan (PIP).

At the time of submission of the application, the PIP EMEA-002362-PIP02-19 was not yet completed as some measures were deferred.

The PDCO issued an opinion on compliance for the PIP EMA/776360/2022 and EMA/434594/2023.

1.4. Information relating to orphan market exclusivity

1.4.1. Similarity

Pursuant to Article 8 of Regulation (EC) No 141/2000 and Article 3 of Commission Regulation (EC) No 847/2000, the applicant did submit a critical report addressing the possible similarity with authorised orphan medicinal products.

A third-party intervention was received during the evaluation of the MAA, claiming that fidanacogene elaparvovec is to be considered similar to etranacogene dezaparvovec (Hemgenix).

1.5. Applicant's requests for consideration

1.5.1. Conditional marketing authorisation

The applicant applied for a full marketing authorisation, but during the assessment, in response to CAT and CHMP concerns on the comprehensiveness of the data, requested consideration of its application for a conditional marketing authorisation in accordance with Article 14-a of Regulation (EC) No 726/2004.

1.5.2. New active substance status

The applicant requested the active substance fidanacogene elaparvovec contained in the above medicinal product to be considered as a new active substance, as the applicant claims that it is not a constituent of a medicinal product previously authorised within the European Union.

1.6. PRIME

Durveqtix was granted eligibility to PRIME on 23 February 2017 in the following indication: treatment of haemophilia B.

Eligibility to PRIME was granted at the time in view of the following:

• The unmet need can be acknowledged in particular on the basis of breakthrough bleeds in prophylactic treatment settings and the development of bleeding sequelae such as haemophilic arthropathy.

• The potential to address the need can be accepted on the basis of preliminary clinical data showing that a single IV administration of the product results in a sustained increase in factor IX activity within the mild haemophilia levels.

Upon granting of eligibility to PRIME, Jan Mueller-Berghaus was appointed by the CHMP as rapporteur.

A kick-off meeting was held on 13 June 2017. The objective of the meeting was to discuss the development programme and regulatory strategy for the product. The applicant was recommended to address the following key issues through relevant regulatory procedures:

During the Kick-off Meeting, the sponsor was advised to seek further Scientific Advice on Quality (with an emphasis on the plan to demonstrate comparability regarding changes in manufacturing site, testing sites and scale-up), as well as on Clinical Development issues (in particular the changes to the Phase 3 development program). The need to submit a PIP and to consider a Protocol Assistance in case of a potential future Orphan Medicinal Product Designation was also conveyed to the applicant.

1.7. 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
13 October 2013	EMEA/H/SA/3416/1/2016/ADT/III	Dr Alexandre Moreau and Dr Rune Kjeken

18 May 2017	EMEA/H/SA/3416/2/2017/SME/ADT/PR/I and EMEA/H/SA/3416/1/FU/1/2017/SME/ADT/PR/II	Dr Olli Tenhunen and Dr Rune Kjeken
28 June 2018	EMEA/H/SA/3416/1/FU/2/2018/ADT/PR/HTA/SME/II	Dr Alexandre Moreau and Dr Jan Mueller-Berghaus
28 March 2019	EMEA/H/SA/3416/3/2019/ADT/PR/III	Dr Alexandre Moreau and Dr Jan Mueller-Berghaus
14 November 2019	EMEA/H/SA/3416/3/FU/1/2019/PA/ADT/II	Dr Jens Reinhardt and Dr Stephan Lehr
15 October 2020	EMEA/H/SA/3416/3/FU/2/2020/PA/ADT/PR/III	Dr Jens Reinhardt and Dr Johanna Lähteenvuo
19 May 2022	EMA/SA/0000081009	Dr Elena Wolff-Holz, Bruno Delafont and Dr Rune Kjeken

The Scientific advice pertained to the following pertained to quality, nonclinical and clinical aspects briefly summarised below:

EMEA/H/SA/3416/1/2016/ADT/III Pre-clinical and clinical development

- The proposed non-clinical package to support registration; the toxicology studies to support dosing of paediatric patients and the need for juvenile animal studies.
- The proposed strategy for paediatric development; the overall design of the Phase 1/2a study SPK-9001-101 and in particular the population, primary/secondary endpoints, dosing, long-term follow up; the proposed strategy to monitor immunogenicity; the dosing strategy for patients >12 years.

EMEA/H/SA/3416/1/FU/1/2017/SME/ADT/PR/II Clinical development

- Whether the dose used in the ongoing Phase 1/2a study represents the appropriate dose for use in Phase 3; the proposed Phase 3 study design, in particular the primary endpoint, within-patient comparison, and statistical strategy including the treatment effect size assumptions and sample size calculation; the proposed data package for MAA; the proposed long-term safety monitoring.

EMEA/H/SA/3416/2/2017/SME/ADT/PR/I Quality development

- The proposed changes of the manufacturing process for clinical trials and commercial supply; the proposed potency assay; orphan similarity.

EMEA/H/SA/3416/1/FU/2/2018/ADT/PR/HTA/SME/II Clinical development

- The proposed data on duration of effect at the time of an initial conditional marketing authorisation and later full marketing authorisation to establish long-term efficacy; the subject eligibility criteria of the Phase 3 studies in adults (studies C0371004 and C0371002); the proposed plans to generate comparator data in clinical study C0371004; the proposed efficacy measures of co-primary endpoints; the anticipated data set at the time of the proposed interim analysis for CMA; whether the proposed pivotal study supports the target indication; whether the proposed secondary endpoints are relevant to demonstrating efficacy and safety of the product; the proposed QoL tool to support clinical benefit.

EMEA/H/SA/3416/3/2019/ADT/PR/III Quality and clinical development

- The approach to assess fidanacogene elaparvovec potency; the sampling strategy for the microbiological tests; analytical comparability to support the introduction of a new manufacturing process into ongoing clinical studies; the approach for viral clearance study plans; the approach in sterilising filter validation studies; the adventitious agent testing strategy; the plans for centralised assay methods for FIX:C.
- Demonstration of clinical bridging between manufacturing processes.
- Introducing potential bias in study design by splitting the pivotal Phase 3 trial in a separate lead-in observation study to generate intra-patient baseline data and the pivotal study with drug administration and subsequent follow-up.
- Adequacy of the proposed analysis plan, in particular the non-inferiority margin selection.
- The overall safety monitoring plan.
- The possibility for full marketing authorisation based on the results of an interim analysis.

EMEA/H/SA/3416/3/FU/1/2019/PA/ADT/II Clinical development

- The approach to a field study to assess variability of FIX coagulant activity (FIX:C) assay results.

EMEA/H/SA/3416/3/FU/2/2020/PA/ADT/PR/III Quality and clinical development

- Whether the proposed type of information is the appropriate framework to allow the determination that two gene therapy medicinal products are non-similar.
- The appropriateness of the proposed non-interventional, observational, cohort study design using data from hemophilia registries globally (Study C0371007), including the study population, duration, sample size, and core data set to describe the long-term safety and effectiveness of fidanacogene elaparvovec after approval.
- The approach of longer-term follow-up on clinical trial participants in Study C0371017, including the clinical assessments to support ongoing evaluation of liver abnormalities to assess for malignancy.
- The proposed decentralization of data collection during long-term follow-up in the Phase 2a (C0371003) and Phase 3 (C0371002) studies, including physical exam and vital sign assessment, liver ultrasound acquisition and review, data collection for patient reported outcomes (PROs), the HJHS assessments and the proposed method of imputing potentially missing gait analysis scores, and the acquisition of joint X-ray.
- The proposal to combine bleeding data collected in the lead-in study C0371004 with data collected prior to dosing in C0371002 for use as the prior prophylaxis comparison group.

EMA/SA/0000081009 Quality and clinical development

- The analytical comparability strategy to support the comparability across Processes 1, 2 and 3 for registration; a new cell-based release assay for commercialisation; the content of the Control of Materials section regarding the three plasmids used in the manufacturing process of the AAV vector.
- The methodology to evaluate the association between ABR and FIX:C.
- The approach to investigate the discrepancies in FIX activity assay measurements and describe the identified assays variability to the prescriber.

1.8. Steps taken for the assessment of the product

The appointed CAT Rapporteur and Co-Rapporteur were:

CAT Rapporteur: Jan Mueller-Berghaus CAT Co-Rapporteur: Silke Dorner

The application was received by the EMA on	20 April 2023
The procedure started on	18 May 2023
The CAT Rapporteur's first Assessment Report was circulated to all CAT and CHMP members on	8 August 2023
The CAT Co-Rapporteur's Assessment was circulated to all CAT and CHMP members on	17 August 2023
The PRAC Rapporteur's first Assessment Report was circulated to all PRAC members on	21 August 2023
The PRAC agreed on the PRAC Assessment Overview and Advice to CAT during the meeting on	31 August 2023
The CAT agreed on the consolidated List of Questions to be sent to the applicant during the meeting on	8 September 2023
The applicant submitted the responses to the CAT consolidated List of Questions on	11 January 2024
The CAT Rapporteur circulated the Joint Assessment Report on the responses to the List of Questions to all CAT and CHMP members on	22 February 2024
The PRAC agreed on the PRAC Assessment Overview and Advice to CHMP during the meeting on	7 March 2024
The CAT agreed on a list of outstanding issues in writing and/or in an oral explanation to be sent to the applicant on	15 March 2024
The applicant submitted the responses to the CAT List of Outstanding Issues on	24 April 2024
The CAT Rapporteurs circulated the Joint Assessment Report on the responses to the List of Outstanding Issues to all CAT and CHMP members on	8 May 2024
The CAT, 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 Durveqtix on	24 May 2024
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 Durveqtix on	30 May 2024
The CAT and CHMP adopted a report on similarity of Durveqtix with Alprolix, Idelvion and Hemgenix	24 May 2024 30 May 2024
Furthermore, the CAT and CHMP adopted a report on New Active Substance (NAS) status of the active substance contained in the medicinal product	24 May 2024 30 May 2024

2. Scientific discussion

2.1. Problem statement

2.1.1. Disease or condition

Severe and moderately severe haemophilia B (congenital factor IX deficiency).

2.1.2. Epidemiology

Hemophilia B, or Christmas disease, is a rare X-chromosome-linked congenital bleeding disorder, characterized by deficiency in blood factor IX clotting activity. The factor deficiencies are the result of pathogenic variants of the FIX clotting factor gene, which leads to a loss of FIX activity in the hemostatic system either due to the absence or the expression of a non-functional FIX protein. Individuals affected are predominantly males; females may be (mostly asymptomatic) carriers.

The final diagnosis is achieved by genetic testing of patients with laboratory values of low factor IX clotting activity and elevated partial thromboplastin time. The result of hemizygous F9 pathogenic variant on molecular genetic testing in male patients confirms the diagnosis, and the result of heterozygous F9 pathogenic variant on molecular genetic testing in a symptomatic female confirms the diagnosis. Approximately 30% of heterozygous females have factor IX clotting activity lower than 40% and are at risk for bleeding, although symptoms are usually mild.

Individuals with moderate hemophilia B seldom have spontaneous bleeding (Konkle et al, Haemophilia B, GeneReviews 2023), however, they do have prolonged or delayed oozing after a relatively minor trauma and are usually diagnosed before age five to six years. The frequency of bleeding episodes varies from once a month to once a year. Individuals with severe hemophilia B are usually diagnosed during the first two years of life. Without prophylactic treatment, they may have two to five spontaneous bleeding episodes each month, including joint or muscle bleeds.

Adult patients with moderate and/or severe haemophilia B are the intended target indication for treatment with fidanacogene elaparvovec.

Incidence

Global

Haemophilia B occurs at a rate of approximately 3.33 to 5.0 per 100,000 male live births per year.

Europe

In the EU-28, approximately 2.55 million male live births were reported in 2018. Applying an incidence rate of 3.33 to 5.0 per 100,000 male live births, approximately 85 to 128 cases of haemophilia B were diagnosed in 2018 in the EEA.

Prevalence

Global

The overall prevalence of haemophilia B among the male population of Western European countries, the United States (US), and Canada is estimated to be between 0.5 and 8.1 per 100,000 men. The estimated prevalence of haemophilia B from registry data from six countries was 3.8/100,000 men.

Europe

Based on the WFH Annual Global Surveys (2013 to 2021), there is an estimated 8499 persons with haemophilia B in the EU-28 in the EEA.

Morbidity and mortality

According to several publications reporting on the severity of haemophilia B cases, 21.0-63.9% were severe, 10.7 – 47.6% were moderate, and 8.7-61.9% were mild; the median life expectancy of men with severe haemophilia is approximately 63 years, and with mild or moderate haemophilia approximately 75 years.

2.1.3. Clinical presentation, diagnosis and prognosis

Patients suffer from acute spontaneous bleeding [intracranial haemorrhage (ICH) included] or prolonged bleeding after injuries, tooth extractions, or surgery, or delayed and recurrent bleeding prior to complete wound healing. The most common sites for spontaneous bleeding are intraarticular and intramuscular, oral cavity, and soft tissue. Irreversible haemophilia B induced joint destruction (arthropathies, chronic synovitis) from recurrent bleeding, leading to immobility and need for joint replacement is the largest source of morbidity for patients with haemophilia B. According to Zanon et al, Intracranial haemorrhage in children and adults with haemophilia A and B: A literature review of the last 20 years, Blood Transfus, 2019, although the use of prophylactic regimens has improved outcomes, the mortality caused by ICH is still around 20%, whereby for adults, presence of severe haemophilia, inhibitors and older age play the major role.

Normal plasma levels of FIX range from 50% to 150%; levels below 50%, or half of what is needed to form a clot, determine a person's symptoms. According to the National Hemophila Foundation, stages of haemophilia B are defined as follows:

Mild hemophilia B. 6% up to 49% of FIX in the blood. People with mild hemophilia B typically experience bleeding only after serious injury, trauma or surgery. In many cases, mild hemophilia is not diagnosed until an injury, surgery or tooth extraction results in prolonged bleeding. The first episode may not occur until adulthood. Women with mild hemophilia often experience menorrhagia, heavy menstrual periods, and can hemorrhage after childbirth.

Moderate hemophilia B. 1% up to 5% of FIX in the blood. People with moderate hemophilia B tend to have bleeding episodes after injuries. Bleeds that occur without obvious cause are called spontaneous bleeding episodes.

Severe hemophilia B. <1% of FIX in the blood. People with severe hemophilia B experience bleeding following an injury and may have frequent spontaneous bleeding episodes, often into their joints and muscles.

2.1.4. Management

Considering Srivastava et al, WFH guidelines for the management of hemophilia, 3rd edition World Federation of Haemophilia 2020, the treatment is based on IV administration of either plasma-derived or recombinant FIX protein replacement therapy to raise the FIX:C activity level to the lowest effective level to achieve either resolution of bleeding (on-demand treatment) or prevention of bleeding (prophylaxis treatment). The development of inhibitory antibodies to infused FIX may occur rarely but often manifest as potentially severe anaphylaxis to infused concentrate and later as nephrotic syndrome. As per A. L Dunn, Transfusion Medicine and Hemostasis, 2019, there appears to be no difference in the rate of inhibitor development between plasma derived and recombinant products.

In November 2022, the FDA approved the gene therapy product Hemgenix, followed by the EMA approval in February 2023 for treatment of adult patients with moderate to severe haemophilia B. Gene therapy for hemophilia works by packaging a functional form of the FIX gene into a virus, most commonly an AAV. Using hepatotropic AAV serotypes allows the transgene to be delivered to hepatocytes in an anticipated safe and efficient manner. Once delivered to hepatocytes, the transgene is expressed and FIX entered to the blood circulation contributing to the hemostatic processes.

2.2. About the product

Fidanacogene elaparvovec is a gene therapy designed to introduce a functional copy of the high activity Padua variant of the factor IX gene (FIX-R338L) in the transduced cells to address the monogenic root cause of haemophilia B.

The intended indication is for the treatment of severe and moderately severe haemophilia B (congenital factor IX deficiency) in adult patients without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74.

The recommended dose of fidanacogene elaparvovec is a single-dose of 5×10^{11} vector genomes per kg (vg/kg) of body weight.

2.3. Type of application and aspects on development

During the procedure, the applicant requested consideration of its application for a Conditional Marketing Authorisation in accordance with Article 14-a of Regulation (EC) No 726/2004, based on the following criteria:

- The benefit-risk balance is positive.
- It is likely that the applicant will be able to provide comprehensive data. The applicant proposed to provide the following follow-up data from the pivotal trial:
    - Study C0371002: 6-year data from subjects with dose calculated using actual batch concentration, and at least 34-month post-infusion data for all participants dosed using the fixed nominal concentration, both cutoff date June 2028.
    - In addition, for study C0371003 5-year data from subjects dosed with 5×10^{11} vector genomes per kg (vg/kg) of body weight will be provided.
    - The applicant also agreed to categorize studies C0371007 and C0371017 as post-authorization efficacy studies. The results from studies C0371002, C0371003, and C0371017 will be able to elucidate long-term efficacy and safety outcomes.
- Unmet medical needs will be addressed, as disease and treatment burden are not addressed by currently authorised FIX products with standard or extended half-life such as nonacog alfa (BeneFIX), nonacog gamma (Rixubis), eftrenonacog alfa (Alprolix), albutrepenonacog alfa (Idelvion), nonacog beta pegol (Refixia) and human plasma-derived factor IX products.

  In addition, the applicant provided a justification why Durveqtix should be regarded to address the existing unmet medical need to a similar or greater extent than what is understood for the already conditionally authorised product Hemgenix in the form of an Indirect Treatment Comparison (ITC) with Hemgenix. This was based on an unanchored matching-adjusted indirect comparison (MAIC) for bleed-related endpoints, resulting in an effective sample size of 15 subjects. For parameters where a MAIC was not feasible, a side-by-side comparison was provided. The applicant concluded that Durveqtix addresses the current unmet medical need

associated with SOC FIX prophylaxis at least to a similar extent as Hemgenix. This occurs with a different safety profile, a 40-fold lower vector dose, and a longer total duration of long-term follow-up than that of Hemgenix. Based on absence of IRRs, lower vector dose exposure, different capsid serotype/seropositivity, improvement in joint health and quality of life, the applicant claimed that there may be patients or clinical situations for which Durveqtix would be the preferred gene therapy to address remaining unmet needs.

- The benefits to public health of the immediate availability outweigh the risks inherent in the fact that additional data are still required. The applicant claimed that, in view of the favourable benefit-risk profile and the demonstrated efficacy in a life-threatening disease, the immediate availability of fidanacogene elaparvovec on the market outweighs the risk inherent in the fact that additional data are still required. Durveqtix would provide moderately severe and severe haemophilia B patients with an important therapeutic option whist additional data are being generated to confirm safety and efficacy of fidanacogene elaparvovec in the Phase 3 study (C0371002).

2.4. Quality aspects

2.4.1. Introduction

Durveqtix is a gene therapy medicinal product. The finished product (FP) is presented as concentrate for solution for infusion containing 0.79 - 1.21 × 10^{13} vector genomes/mL of fidanacogene elaparvovec as active substance (AS).

Other ingredients are: sodium dihydrogen phosphate monohydrate (E339), disodium hydrogen phosphate heptahydrate (E339), sodium chloride, poloxamer 188, and water for injections.

The product is available in a plastic vial with an elastomeric stopper and plastic snap fit cap. Each vial contains 1 mL of extractable volume.

The exact dose volume and number of vials required for each patient are calculated according to the patient-specific dose required.

The total number of vials in each finished pack corresponds to the dosing requirement of the individual patient, depending on the body weight and actual concentration, and is provided on the package.

2.4.2. Active substance

2.4.2.1. General information

Fidanacogene elaparvovec is produced in human embryonic kidney cells (HEK293) by recombinant DNA technology and consists of a recombinant viral capsid derived from a naturally occurring adeno-associated viral serotype Rh74 (AAVRh74var) packaging genome containing the human coagulation factor IX (FIX) transgene modified to be a high factor IX activity (Padua) variant known as FIX-R338L (also referred to as SPK-9001, SPKFIX Padua, PF 06838435).

The AAV-Spark100 capsid was derived from the naturally occurring Rh74 AAV serotype. The capsid consists of three capsid proteins, VP1, VP2 and VP3. The amino acid (aa) sequence of the therapeutic gene product (R338L variant of human coagulation factor IX) is included in the dossier. Regarding the capsid, the amino acid sequences and information on the capsid structure were provided. Directed

mutagenesis was applied to evolve the parental capsid (Rh74) towards specific amino acid residues more conserved among AAV serotypes traditionally considered hepatotropic.

Fidanacogene elaparvovec contains a single-stranded DNA vector genome containing the therapeutic gene flanked by inverted terminal repeats (ITRs). The rep gene, which encodes proteins involved in DNA replication, transcription and packaging, and cap gene, which encodes the capsid proteins, have been removed and replaced with the therapeutic gene expression cassette, rendering fidanacogene elaparvovec replication incompetent. The ITR are retained in the vector genome. In fidanacogene elaparvovec the ITR are required for vector genome packaging and stable expression of the therapeutic gene following *in vivo* delivery of the vector.

The fidanacogene elaparvovec vector DNA cassette is designed to express the R338L variant of the human coagulation factor IX. The expressed variant, which contains the naturally occurring mutation R384L, is associated with an increase in specific coagulation activity compared to the wildtype factor IX protein. R384L refers to the mutated residue number pre-processing – after processing (removal of the activation peptide), that residue is number 338 in the mature, active Factor IX transgene-encoded polypeptide and the variant is referred to as R338L.

In addition to the hepatotropic nature of the AAV capsid, additional specificity is granted by the DNA vector. The fidanacogene elaparvovec DNA vector utilises a liver specific promoter and hepatic control centre. The single stranded DNA sequence (5'-3') of the genome is provided in the dossier.

2.4.2.2. 1Manufacture, characterisation and process controls

The name, address and general responsibility of each manufacturer involved in the active substance manufacturing process stream and testing has been provided.

The AS will be manufactured at Wyeth Holdings LLC, Sanford, USA. The GMP compliance certificate initially provided by the applicant for this site did not cover gene therapy medicinal products. To address this issue, during the assessment, a pre-approval GMP inspection has taken place and the inspection report has been received and concluded that the site is GMP compliant. A valid GMP compliance certificate, covering Gene therapy medicinal products, has been issued by Spanish competent authority (ES/055HV/24).

All sites involved in manufacture and control of the active substance operate in accordance with EU GMP.

Description of manufacturing process and process controls

The manufacturing process of fidanacogene elaparvovec starts with the upstream process consisting of working cell bank (WCB (HEK293 cells)) vial thaw and culture followed by culture expansion steps, transfection and cell harvest.

The subsequent downstream process begins with recovery of the AAV particles, followed by several purification steps. Viral filtration is performed. Subsequently, filtration (0.2 µm) is performed. The bulk AS is filled into bottles, frozen, and stored.

Overall, the manufacturing process steps have been sufficiently described. In Process Controls (IPCs) (parameters and tests) are included in the detailed flow charts.

Control of materials

The starting materials for manufacture of fidanacogene elaparvovec AS include a HEK 293 master cell bank (MCB) and WCB and the three plasmids.

A description of the source, history, and generation of the HEK293 MCB and derived WCBs has been provided. In general, testing of the MCB and WCBs complies with the recommendations given in Ph. Eur. 5.2.3 and respective ICH guidance. The provided data support the proposed limit of an *in vitro* cell age. The procedure for the preparation and qualification of a renewal HEK293 WCB has been described. Comparability of laboratory and commercial scale has been addressed in process development. Acceptance criteria for culture performance parameters and specifications have been defined.

For each of the plasmids, adequate information has been provided. Control of the bacterial cell banks largely complies with Ph. Eur. 5.14 and with the EMA guideline on gene transfer medicinal products (EMA/CAT/80183/2014). An overview of the plasmid manufacturing process has been provided. Plasmid testing in general complies with Ph. Eur. 5.14. Information about plasmid reference materials including specifications and stability data has been provided.

Raw materials

A list of materials used in AS manufacturing indicating the process stage they are used in, and their quality grade has been provided. For non-compendial raw materials, specifications including acceptance criteria are available. The qualitative composition of cell culture and purification solutions and their intended use have been indicated and information on the vendors for critical raw materials have been provided. Materials of biological origin used in the manufacturing process have been declared. For assessment of TSE risk and viral safety, information can be found in adventitious agents safety evaluation section.

Control of critical steps and intermediates

Acceptance criteria and analytical procedures applied for IPCs of the upstream and downstream process have been summarised and described. The analytical methods used for biochemical in-process tests are sufficiently described. Method description and suitability data were provided to demonstrate compliance with Ph. Eur. 2.6.7 requirements.

Process validation

Consecutive fidanacogene elaparvovec AS process performance qualification (PPQ) batches using the commercial manufacturing process have been manufactured at the Sanford, NC facility.

Prior to the PPQ campaign, a risk assessment was conducted to identify the CPPs, CMAs, and relevant Process Performance Attributes (PPAs). Importantly, after the PPQ runs, the risk and criticality assessments were updated and used to support the final process parameter criticality selection. Some of the parameters that were added due to this iterative risk assessment were not included in the initial validation protocol. However, all parameters were previously included in PPQ process description, and the data was gathered during the PPQ runs.

All PPQ runs were evaluated to meet validation requirements. The acceptance criteria consisted of all CPPs, select non-CPPs, and quality attributes of the process, selected IPTs, process-related impurities, relevant PPAs, demonstration of sanitary control throughout the process and final AS release testing per specification. All release test results for the PPQ AS batches met acceptance criteria in place at the time of process validation and additionally meet the commercial specifications.

In-process pool holds were validated.

Impurities were evaluated in PPQ studies. For some impurities, all PPQ and commercial acceptance criteria were met. All other impurities were consistently removed below assay quantitation limits. It could be shown that the purification process is able to significantly deplete these impurities. The analytical procedures used to test impurity removal are adequately described.

Furthermore, for all resins used in columns during the purification process, resin lifetimes were established and confirmed. Column performance data and data confirming the cleaning efficacy of the column were provided.

Overall, the process validation results support the conclusion that the commercial manufacturing process can be considered validated.

Manufacturing process development

For manufacturing process development, the applicant provided information on development history, critical quality attributes (CQAs), analytical method evolution, process risk assessment strategy, control strategy, comparability and upstream and downstream process development and characterisation. CQAs were identified based on a score calculated by severity and uncertainty of each attribute.

Process development and characterisation of upstream and downstream processes has been performed using a multi-stage risk-based approach including identification and characterisation of important unit operations, process parameters and material attributes that could affect CQAs using one-factor-at-a-time (OFAT) or limited multi-factor design-of-experiments (DOE) designs using scale-down models, followed by a Failure Mode and Effect Analysis (FMEA) to identify critical process parameters and material attributes (CPPs/CMAs).

Several analytical procedures for AS and FP release control have evolved throughout product development. Information on changes of the analytical procedures, time points at which the changes were implemented, and method bridging has been provided. Analytical method evolution has been sufficiently considered, e.g. for comparability or justification of specification. Changes in the analytical procedures have been implemented after the PPQ runs and for commercial manufacturing, and this is considered acceptable.

Comparability

Three processes have been implemented in the manufacturing of fidanacogene elaparvovec. Processes 1 and 2 were used during early clinical development and commercial Process 3 was used in the phase 3/pivotal study.

The main difference between Process 1 and Process 2 was the replacement of the caesium chloride ultracentrifugation with a column purification scheme. Changes from Process 2 to Process 3 included a roller bottle scale-out, the addition of a viral filtration step and increased number of 0.2 µm filtration steps, and a new active substance container closure. Upstream processes are largely similar between the different processes. A summary of downstream process changes and their potential impact to CQAs has been provided. The approach used in the comparability study has been justified. Stability profiles were demonstrated to be similar.

In summary, comparability is considered to be sufficiently demonstrated.

Characterisation

The characterisation strategy for AS is described. Fidanacogene elaparvovec has been extensively characterised using different methods. Furthermore, forced degradation by elevated temperature and light exposure were investigated. Characterisation data from several process 1, 2 and 3 batches were provided, and results were rather consistent for the different batches. No oncogenic adenoviral sequences (E1a, E1b) were detectable. The transcriptional potential of hcDNA impurities has been addressed and is considered to be acceptably low.

AS impurities were analysed in process 1, 2 and 3 material.

Overall, the data provided support the conclusion that characterisation can be considered satisfactory.

2.4.2.3. Specification, analytical methods, batch analysis, reference standards and container closure

Specification

AS release specifications include relevant attributes to assess the quality of fidanacogene elaparvovec including testing of general quality attributes, strength and content, identity, genomic integrity, purity, product and process-related impurities, biological activity and safety. Control of residual reagents is sufficiently addressed. The established release specifications are not fully in line with Eur. Ph. 5.14. This is considered justified by the data provided.

Acceptance criteria of AS specifications were mainly justified based on compendial requirements and a statistical analysis. The applicant commits to re-evaluate the specification acceptance criteria for non-compendial methods for AS and FP after 15 commercial AS batches and 15 commercial FP lots have been manufactured or within five years of approval, whichever comes first (REC9).

Overall, the proposed specifications are considered adequate.

Analytical methods

Compendial methods are used for appearance (clarity), appearance (colour), pH, osmolality, bioburden and endotoxin. In general, the non-compendial analytical methods used for release testing of the AS are sufficiently described. Information on the assay principle, apparatus, equipment, reagents, standard, reference material, procedure description, assay acceptance criteria and calculations has been provided.

Validation summaries as well as full validation reports have been provided for each non-compendial analytical method. Most of the methods have been validated in line with ICH Q2 (R1).

Post-approval, the applicant is recommended to adapt one of the methods used. The adapted method development, validation, and testing will be provided in a post-authorisation recommendation (REC1).

Batch analysis

Altogether, batch analysis data of the active substance were provided. In general, the provided batch data suggests consistent manufacturing for the commercial Process 3.

Reference materials

The fidanacogene elaparvovec reference material is used for several release methods. For commercial manufacturing a two-tiered reference material system has been established which is composed of primary and working reference materials derived from a Process 3 batch. Data have been provided that sufficiently qualify the reference materials. An adequate stability protocol has been provided and provided data support the proposed period of use. A protocol for the qualification of future reference materials has been provided. The qualification protocol in principle includes critical tests covering release and heightened product characterisation tests. Information on the bridging protocol of the primary reference material to future working reference material has been provided.

Container closure system

The AS is stored in bottles specifically designed for controlled freezing, thawing and frozen storage of biopharmaceuticals. The bottles are of adequate microbiological quality, following sterilisation and testing for endotoxins. Materials are compliant with USP VI and Ph. Eur. monographs. Active substance stability studies have been performed in small-volume containers of the same material. Extractables and leachables have been sufficiently addressed.

2.4.2.4. Stability

Stability data of clinical and PPQ Process 3 batches are available at the long-term storage condition and at accelerated conditions. Furthermore, thermal stress studies and thermal cycling studies were performed. Stability samples are stored in bottles made of the same material used for commercial manufacturing.

Test methods used for the stability study are a subset of those used for release testing which is generally in line with the expectations. The proposed testing time points for the long-term storage conditions are in compliance with ICHQ5C.

Overall, long-term stability studies support the proposed shelf-life for fidanacogene elaparvovec when stored at the recommended storage temperature.

Stability data from an additional DS supportive stability study have been provided. The data is considered to support the AS stability claim but the applicant is recommended to initiate an accelerated stability study with an active substance lot from an upcoming active substance manufacture (REC2).

In general, the provided data support the stability of fidanacogene elaparvovec at the chosen thermal cycling conditions. Post approval, the applicant commits to place at least one AS batch on stability under real-time storage condition each year a AS lot is manufactured (REC6).

2.4.3. Finished Medicinal Product

2.4.3.1. Description of the product and pharmaceutical development

The composition of Durveqtix finished product is provided. Durveqtix contains fidanacogene elaparvovec, a recombinant adeno associated-viral vector containing a codon-optimized coding DNA sequence for human coagulation factor IX variant R338L (FIX-Padua) with a labelled range 0.79 - 1.21 × 10^{13} vg/mL and a nominal concentration of 1 x 10^{13} vg per mL. There are no excipients of human or animal origin, and no novel excipients utilised in the formulation. All excipients are pharmacopoeial (sodium phosphate monobasic monohydrate; sodium phosphate dibasic heptahydrate; sodium chloride; poloxamer 188 and water). The FP is supplied in a single-use, clear 2 mL cyclic olefin copolymer vial with elastomeric stopper and plastic, snap-fit cap.

An overfill is included for the finished product.

The clinical trial programme used the actual vector genome concentration of each lot instead of the nominal concentration that is commonly used in the post-marketing setting. The current proposal is to continue actual titre dosing post-marketing. In the SmPC, actual titre dosing is referred to. Actual concentration as well as the calculation of the patient dose is provided in the Lot Information Sheet accompanying the medicinal product.

Pharmaceutical development

The FP formulation is adequately justified. In a formulation robustness study, different formulations were evaluated. Results of this study indicate that overall, the formulation variations did not have a significant impact on quality attributes and FP stability.

Changes between the different FP manufacturing processes have been described and a corresponding rationale has been provided. Process development and characterisation studies have been performed. CQAs and their controls strategy have been described.

Extractable and leachable studies have been performed with the container closure system. Photostability of the secondary container has been demonstrated with regard to visible light and UV light.

Before administration to the patient, the FP is diluted in commercially available 0.9% sodium chloride and 0.25% w/v human serum albumin (HSA) based on the concentration and patient weight. The proposed in-use stability of max. 24 hours at 2 – 30 °C for the diluted solution has been addressed in a verification study. The applicant is recommended to test at least one additional batch at high concentration in an in-use stability study for 24 hours at 30°C (REC8).

2.4.3.2. Manufacture of the product and process controls

All sites involved in manufacture and quality control (QC) testing of the finished product operate in accordance with EU GMP.

The applicant initially requested an exemption of retesting upon importation into the EU based on limited amount of available material, which was not supported. During the assessment, the applicant agreed to implement before CHMP Opinion full retesting upon importation into the EU.

Description of manufacturing process and process controls

A range for the batch size and the batch formula for fidanacogene elaparvovec FP has been provided.

The manufacturing process of the FP consists of AS thawing, followed by filtration steps, aseptic fill, seal and cap vials, visual inspection, bulk packaging and lastly FP storage.

The manufacturing process steps have been described in sufficient detail.

IPCs (parameters and tests) are included in the flow charts. Implemented process parameters together with acceptance criteria are provided.

Process validation and/or evaluation

Validation of the aseptic sterile-filtration-fill process with media simulations is adequately established for initial and routine re-qualification runs with consideration of relevant aspects.

Filter validation studies were successfully performed.

Consecutive PPQ batches were completed, using the commercial process while maintaining process parameters within the defined NORs except where noted and discussed by the applicant. The PPQ batches were evaluated to meet validation requirements and data was provided.

The minimum batch size was challenged in PPQ. No worst-case challenge of the maximum FP batch size was included. This omission was considered justified by the applicant and results from other auxiliary studies/validation activities were included.

The validation activities performed included the evaluation of the performance parameters and in-process tests for each Unit Operation. The process parameters were chosen using a risk-based approach.

Maximum processing and hold times were challenged to validate hold and process time. The applicant committed to evaluate the results of the leachable study (REC3).

Acceptable process validation results were provided for each step of the FP manufacturing process. Additionally, all PPQ runs fulfilled the acceptance criteria of FP release testing. For some parameters, the applicant indicates that they were included in the continued process verification (CPV), the full CPV plan was submitted. Since only a limited number of batches of the AS and FP have been produced so far and relatively wide acceptance ranges have been set for some process parameters, IPT, and specifications,

the applicant is recommended to submit respective CPV data once a year for five years in order to further ensure the consistency of production (REC4).

Sufficient shipping validation studies were provided. In general, the validation of the fidanacogene elaparvovec FP manufacturing and shipping can be considered successfully completed.

2.4.3.3. Finished Product specification, analytical methods, batch analysis, reference standards and container closure

Specifications

The FP specifications have been provided. Relevant FP release specifications including testing of general quality attributes, strength and content, identity, genomic integrity, purity, product-related impurities, biological activity, and safety have been defined. However, specifications are not fully in line with Eur. Ph. 5.14 Gene transfer medicinal products for human use due to some attributes omitted in the FP specifications, that were sufficiently justified by data provided. Overall, the proposed specifications are considered adequate.

A risk evaluation for the presence of nitrosamines was performed in accordance with the EMA CHMP Assessment Report on Nitrosamine impurities in human medical products (EMA/369136/2020). It was concluded that there is no risk for small molecule nitrosamine (cohort of concern) formation or introduction via the active substance, finished product and primary packaging processes. The conclusion of the risk assessment is acceptable and no additional specific control is considered necessary.

A summary of the risk assessment for elemental impurities in accordance with ICH Q3D and Ph. Eur. (2619) is included in the dossier and concludes the observed levels pose negligible risk to patients.

Analytical methods

The analytical procedures for the FP have been sufficiently described and are adequately validated.

Batch analysis

FP release data of Process 1, Process 2 and Process 3 batches have been provided. Variations were observed between the Process 3 batches. In view of the type of assays however, FP release results are rather consistent.

Reference materials

The reference standard used for analysis of FP is the same that is used for AS.

Container closure system

The FP is stored in a 2 mL cyclic olefin copolymer vial body with a thermoplastic elastomeric stopper. Vial body and stopper materials are compliant with Ph. Eur. Sterilization is performed by ionizing radiation (≥ 25 kGy). The secondary container consists of a carton. The container closures system is considered adequate for storage of fidanacogene elaparvovec FP.

2.4.3.4. Stability of the product

A shelf life of 36 months is proposed for unopened FP vials when stored at -60 °C to -90 °C.

Three primary stability batches (two Phase 3 clinical, one Development) and three PPQ Process 3 batches were placed on long-term stability at -60 °C to -90 °C. Accelerated stability was studied on the same clinical and PPQ batches. Furthermore, photostability studies, thermal stress studies and a thermal cycling study were performed.

FP stability samples were stored in the same container closure system as intended for commercial packaging. Vials are stored in an upright position.

Test methods used for the stability study are a subset of those used for release testing. The proposed testing time points for the long-term storage conditions are in line with ICHQ5C.

The proposed in-use stability of maximum 24 hours at 2 – 30 °C for the diluted solution has been addressed.

In the long-term stability studies, the provided long-term stability data support the proposed shelf-life of 36 months when stored at the recommended temperature of -60 °C to -90 °C. An additional supportive stability study was performed and the data support the FP stability claim. The applicant committed to initiate a finished product accelerated stability study with a FP lot from an upcoming FP manufacture (REC5).

The label and the SmPC include instructions to avoid exposure to UV light.

The applicant commits to place at least one FP batch on stability at the real-time storage condition of -60 °C to -90 °C each year a FP batch is manufactured (REC7). The provided post approval stability protocol covers relevant testing parameters and in view of the already provided stability data the proposed testing time points are also considered acceptable.

Overall, the proposed shelf life of 3 years when stored at the recommended storage temperature of -60 °C to -90 °C and the in-use storage conditions of maximum 24 hours at 2 – 30 °C for the diluted solution are acceptable.

2.4.3.5. Adventitious agents

Foetal bovine serum (FBS) has been used and certificates of Suitability have been provided, demonstrating compliance with the Note for Guidance on Minimizing the Risk of Transmitting Animal Spongiform Encephalopathy Agents via Human and Veterinary Medicinal Products (EMEA/410/01 re.3). The cell banking system has been tested and found negative for the presence of adventitious viruses. No materials of human or animal origin, other than the cells and FBS, are used during the fidanacogene elaparvovec manufacturing process.

The viral clearance capacity of the fidanacogene elaparvovec downstream manufacturing process has been assessed. Clearance of model viruses during the purification of the active substance has been demonstrated.

HSA which is used during administration is required to be commercially available (therefore would meet relevant quality standards).

The TSE and adventitious virus safety of fidanacogene elaparvovec has been sufficiently demonstrated.

2.4.3.6. GMO

Fidanacogene elaparvovec is an AAV-based vector with an expression cassette containing a codon-optimized hFIX-R338L transgene variant under the control of a liver-specific promoter.

As fidanacogene elaparvovec is considered a GMO, a separate GMO environmental risk assessment report was submitted to estimate the risk of fidanacogene elaparvovec to third parties and the environment.

The clinical use of fidanacogene elaparvovec provides a negligible risk for the environment and for third parties.

More information under section 2.5.5 on "Ecotoxicity/environmental risk assessment".

2.4.4. Discussion on chemical, pharmaceutical and biological aspects

The applicant has provided a well-structured Module 3, in which all sections have been appropriately addressed. Overall, the manufacturing process of fidanacogene elaparvovec active substance (AS) and finished product (FP) is sufficiently described.

An overall acceptable quality control strategy based on process controls and release specifications for the active substance and the finished product is in place. In addition to the PPQ campaign, the quality control strategy integrates experience gained from process and product characterisation and stability studies. The performed validation of the manufacturing process of the active substance and the finished product together with additionally conducted supportive validation studies indicate that the manufacturing process is capable to consistently deliver material meeting its predetermined specifications and quality attributes. To complete the dataset a number of post-authorisation recommendations have been agreed with the applicant.

Since a limited number of batches of the AS and FP have been produced so far and relatively wide acceptance ranges have been set for several parameters, the applicant is recommended to annually provide CPV data for active substance and/or finished product in the first five years in which AS and/or FP are manufactured, in order to further ensure the consistency of production (REC4).

In addition, the applicant will re-evaluate the specification acceptance criteria for non-compendial methods for AS and FP after 15 commercial active substance batches and 15 commercial finished product lots have been manufactured or within five years of approval, whichever comes first (REC9).

Taking into account the overall risk-benefit ratio, the applicant has agreed to the recommendation to adapt one of the methods used. The adapted method development, validation, and testing will be provided after granting of the marketing authorisation (REC1).

The applicant has further agreed to conduct a post-approval evaluation of the final results of the ongoing container closure long term leachable study (REC3).

The proposed shelf life and the in-use storage conditions are acceptable, and additional post-approval stability data will be provided for AS and FP (REC2, REC5, REC6, REC7, REC8).

The TSE and adventitious virus safety of fidanacogene elaparvovec has been sufficiently demonstrated.

2.4.5. Conclusions on the chemical, pharmaceutical and biological aspects

The quality of this product is considered to be 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 has been presented to give reassurance on viral/TSE safety.

The CHMP endorses the CAT assessment regarding the conclusions on the chemical, pharmaceutical and biological aspects as described above.

2.4.6. Recommendation(s) for future quality development

In the context of the obligation of MAHs to take due account of technical and scientific progress, the

CAT recommends the following points for investigation:

Proposed list of recommendations:

Description of post-authorisation measure(s)
1. The applicant is recommended to adapt one of the methods used. The adapted method development, validation, and testing will be provided after granting of the marketing authorisation.
2. The applicant is recommended to initiate an active substance accelerated stability study with an active substance lot from an upcoming active substance manufacture.
3. The applicant is recommended to provide the final results of the leachable study.
4. The applicant is recommended to annually provide CPV data for active substance and/or finished product in the first five years in which drug substance and/or drug product are manufactured.
5. The applicant is recommended to initiate a finished product accelerated stability study with a finished product lot from an upcoming drug product manufacture.
6. The applicants is recommended that post-approval, a minimum of one active substance batch will be enrolled in the commercial stability program at the long-term storage condition each year that active substance is manufactured.
7. The applicants is recommended that post-approval, a minimum of one finished product lot will be enrolled in the commercial stability program at the long-term storage condition of -60 to -90 °C each year that finished product is manufactured.
8. The applicant is recommended to test at least one additional batch at high concentration in an in-use stability study for 24 hours at 30°C. Inclusion of testing at interim timepoints is highly recommended, as in-use stability might need to be limited in case a downward trend will be observed.
9. The applicant is recommended to re-evaluate the specification acceptance criteria for non-compendial methods for active substance and finished product after 15 commercial active substance batches and 15 commercial finished product lots have been manufactured or within five years of approval, whichever comes first.

The CHMP endorses the CAT assessment regarding the recommendation(s) for future quality development as described above.

2.5. Non-clinical aspects

2.5.1. Introduction

A comprehensive set of pharmacology, biodistribution, and toxicology studies has been performed in mice, dogs, rabbits, and non-human primates (NHPs).

Various PD *in vivo* studies were performed both in haemostatically normal C57BL/6 and diseased HB-mice, which present a targeted deletion of murine coagulation factor 9 (HB). FIX protein amount in

plasma and the respective biological activity was assessed considering both safety and activity. Excessive coagulation was determined through the analysis of thrombin-antithrombin (TAT) complexes and D-dimers. Since humans might have pre-existing humoral immune responses, immune resistance of AAVRh74var capsid to naturally occurring nAbs was evaluated using haemostatically normal C57BL/6 mice. Further proof-of-concept (PoC) as well as immunogenicity studies were performed applying a canine FIX-R338L variant in adult, juvenile, and neonatal haemophilia B dogs. PoC studies in non-human primates (NHP) were performed to compare expression levels and biological activity resulting from AAVRh74var-hFIX-R338L and AAV8-hFIX-R338L upon intravenous infusion.

The biodistribution of fidanacogene elaparvovec was evaluated using the expression of vector DNA in mice and monkey, covering gene expression levels, hFIX protein concentrations, anti-hFIX antibody levels, and anti-AAV neutralizing antibody titers.

Fidanacogene elaparvovec was assessed in a series of single-dose nonclinical toxicity studies, with all toxicity studies conducted using the IV route of administration. Repeat-dose toxicity studies were not performed because fidanacogene elaparvovec is intended for single administration to patients. The definitive 3-month GLP toxicity study in cynomolgus monkeys used AAVRh74var-hFIX19-R338L, while a 2-year genetic toxicity vector integration study in monkeys used fidanacogene elaparvovec. Furthermore, a 12-month male mouse study was conducted, using AAVRh74var-FIX-WT or AAVRh74var-FIX19-R338L. Moreover, specialized genotoxicity studies regarding vector integration were conducted to assess the risk of hepatocellular carcinoma and were evaluated in adult NHP as well as neonatal and juvenile haemophilia B dogs.

Stand-alone reproductive and developmental toxicity studies were not conducted for fidanacogene elaparvovec. Local tolerance was evaluated as part of the 3-month general toxicity and the 2-year vector integration study in monkeys.

The fidanacogene elaparvovec batches used in the pivotal nonclinical safety testing regarding biodistribution and toxicity/genotoxicity were considered representative of batches used in the clinic.

2.5.2. Pharmacology

2.5.2.1. Primary pharmacodynamic studies

FIX transgene expression, immunogenicity, and long-term safety of expressing the FIX-R338L variant from an AAV8 vector were evaluated in both haemophilia B and haemostatically normal mice. Haemostatically normal C57BL/6 male mice received increasing doses of AAV8-hFIX16-WT or AAV8 hFIX16 R338L and were followed for 18 months. It was confirmed that the hFIX-R338L is more effective, presenting a 7-10-fold increased specific activity (applying various doses ranging from $1x10^{10}$ to $2x10^{12}$ vg/kg). Compared to the original Padua-patient, from whom the FIX variant has been derived, the observed increased activity did not range in exaggerated high levels that might be associated with safety issues such as an increased risk of thrombus formation. FIX protein amount in plasma and the respective biological activity was assessed in view of safety (thrombus formation) and biological activity (coagulation). Excessive coagulation (thrombosis) activity was analysed via ELISA checking for thrombin-antithromin (TAT) complexes and D-dimers. The risk for abnormal activation of coagulation was studied in haemostatically normal C57BL/6 mice, which received increasing amounts of AAV8-hFIX16-WT or AAV8-hFIX16 R338L. No increase in plasma TAT has been detected in mice, showing up to 2020% specific FIX activity. The D-dimer values are slightly increased in a low-activity mouse cohort (100-230% FIX-activity) over time (8 month after administration) in mice treated with hFIX-R338L or hFIX-WT group, compared to the control group treated with empty AAV8 capsids only. However, there was no correlation between the increased D-dimer value and the TAT-value. Of note, as the transduction

rate in mice might often be higher compared to humans (up to 30-fold), the mouse data might be considered as worst-case scenario in view of an exaggerated coagulation.

These observations were confirmed in adult haemophilia B mice that were injected with AAV8 vectors encoding either WT hFIX (hFIX16-WT) or hFIX16 R338L at a dose of 4 x 10^{10} vg/kg. FIX activity levels were determined using an activated partial thromboplastin time assay. Similar circulating hFIX levels were observed comparing hFIX16-WT and hFIX16 R338L (850 ± 96 ng/ml compared to 724 ±116 ng/mL, respectively). However, the specific activity of the FIX-R338L protein was approximately 6× higher than that of the FIX-WT protein (3109 versus 518 U/mg, respectively).

The FIX-R338L Padua variant was also studied in haemophilia B dogs of the Lhasa Apso colony, whereby the canine variant (cFIX-R338L) was used at a dose of 1x10^{12} vg/kg and 3x10^{12} vg/kg. Plateau levels of FIX antigen and FIX activity for the low dose group were 3.3% and 39.9% of normal, respectively, while the dog that received a 3x higher dose produced similar FIX antigen (3.8% of normal) and lower activity levels (26.8% of normal). In treated adult dogs, an improvement of the coagulation activity has been detected. The improvement of 26-39% of specific activity resulted in a reduced number of bleeds per month and a reduced bleeding time that is with 8-12 minutes (before >60 minutes) comparable to healthy dogs. No elevation of pathological coagulation measured by TAT value has been observed in analysed dogs.

Studies in haemostatically normal mice were performed to determine FIX expression levels following injection of either the novel bioengineered AAVRh74var capsid encoding the hFIX19 WT transgene (AAVRh74var-hFIX19) or AAV8-hFIX19 at a dose of 3×10^{11} or 6×10^{12} vg/kg. FIX antigen levels were measured by ELISA at 2, 4, and 8 weeks post-treatment, with levels being higher at all analysed timepoints upon application of AAVRh74var-hFIX19 compared to AAV8-hFIX19, supporting that the AAVRh74var-hFIX19 variant is slightly more efficient in transducing liver cells.

Since previous clinical trials of AAV-mediated gene transfer to the liver showed pre-existing humoral immune responses to AAV capsids in humans, which might prevent transduction, the AAVRh74var capsid was optimized for enhanced resistance to neutralization. An *in vivo* study in healthy, male C57BL/6 mice was performed that utilized IV Ig to test AAV transduction under conditions of existing nAbs, whereby animals were first injected with 0.75 mg IV Ig to induce a nAb titer against the AAVRh74var and AAV8 capsids of ~1:3. Twenty-four hours later, AAV vectors (AAV8 or AAVRh74var) expressing hFIX19 were injected at a dose of 6×10^{12} vg/kg, and hFIX levels were measured 2 weeks post-dose. The presented data showed only slightly enhanced immune resistance of the AAVRh74var capsid (64% ± 9.8%) with less nAbs compared to the AAV8-capsid (55% ±15.3%). However, no information on the specific hFIX activity was included due to the focus of the study on the vector development and the use of the novel engineered capsid to translate into a lower impact on the immunogenicity. Less immunogenicity of the bioengineered capsid as used in AAVRh74var was also shown in vitro in human hepatocellular Huh7 cells. Additionally, in an *in vitro* killing assay to assess effector T cell responses against the AAVRh74var capsid was confirmed that the AAVRh74var capsid is presented to the immune system to a lesser degree than both AAV8 and AAV2 capsids.

With regard to further optimization, CpG dinucleotides were reduced to minimize potential innate immune responses. Studies in C57BL/6 mice were performed to compare the hFIX levels expressed from AAVRh74var vectors encoding either hFIX19-R338L (non-CpG-reduced) or hFIX39-R338L (CpG-reduced). Haemostatically normal mice were injected at 8 weeks of age with either 1x10^{11} or 1x 10^{12} vg/kg, while both vectors expressed similar levels of FIX antigen, thus supporting the notion that CpG reduction does not impact vector potency.

PoC studies in non-human primates (NHP) were performed to compare expression levels and biological activity resulting from AAVRh74var-hFIX-R338L and AAV8-hFIX-R338L upon intravenous infusion. Cynomolgus monkeys were pre-screened for nAbs, whereby animals with pre-treatment titers of <1:3

were selected for the study. During clinical trials, the presence of neutralizing antibodies above the threshold of 1:1 is an exclusion criterion, which reflects the situation of the pilot study in cynomolgus monkeys. Increased hFIX expression levels were measured in all treated animals, with a significant drop of the hFIX plasma levels over time, starting approximately one month after injection. As a possible explanation, the development of an immune response against the hFIX occurring in approximately 20% of macaques treated with a human FIX vector due to small amino acid differences between the human and macaque proteins was pointed out. The hFIX plasma levels are in the range of 5-25% as compared to normal hFIX levels, which is supportive with regard to the primary clinical efficacy endpoint of achieving a threshold of 5% (which equals to 500 ng/ml) circulating FIX (FIX:C) post-treatment. Overall, the hFIX plasma levels in mice are approximately 10 to 20-fold higher compared to those measured in NHPs. This might be explained by the more efficient transduction of AAV8 in mice (which is between 10-30%, according to data from comparable AAV8 vectors).

Further PoC as well as immunogenicity studies were performed applying a canine FIX-R338L variant in juvenile and neonatal haemophilia B dogs of the Lhasa Apso colony. Study 1 in juvenile animals consists of two age levels including 3-months (reflecting the human age range from 2 to 6 years) and 6-months (reflecting the human age range from 6 to <12 years) old juvenile male haemophilia B dogs, with each group receiving either 5 x 10^{11} vg/kg (2 dogs), 2.5 x 10^{12} vg/kg (3 dogs), or 5 x 10^{12} vg/kg (1 dog). The higher doses are a multiple of the clinical dose in order to potentially compensate for the dilutional impact of growth between 2-12 years of age in the paediatric population.

The first study was conducted in juvenile male hemophilia B dogs to assess the durability of FIX R338L transgene expression over time. The administered vector (AAVRh74var-cFIX-R338L) is identical to the clinical production except that a canine FIX-Padua transgene is used. The AAV-canine FIX-R338L was produced at a development scale consistent with process 3. Prior to treatment, neutralizing antibodies to the AAVRh74var capsid are measured with titers <1:5. The dogs were followed post-treatment for approximately one year, in which liver growth and blood volume expansion occurred and during a time, in which the dogs were fully matured and had an age equivalent to at least a 15-year-old human at study-end. Haemostatic parameters such as whole blood clotting time (WBCT), thromboelastography (TEG), one-stage clotting aPTT assay to evaluate FIX activity, and antigen levels were monitored to determine the persistence of FIX expression and haemostasis against the background of liver growth and weight increase. The treatment was well-tolerated and no spontaneous bleeds occurred post-gene therapy. Treatment with the AAV-canine FIX-R338L resulted in a shortening of the aPTT in all dogs post-treatment, while being slightly less in dogs treated at 3 or 6-months of age in the 5x10^{11} vg/kg treatment groups, compared to the 2.5x10^{12} vg/kg and 5x10^{12} vg/kg groups. Moreover, shortening of the WBCT was observed in all dogs dosed at 3- and 6-months of age in haemophilia dogs treated with AAV-canine FIX R338L and remained decreased over the whole observation period of the study. Baseline TEG-R-values were >60 minutes in all juvenile haemophilia B dogs, while the administration of AAV-canine FIX-R338L resulted in faster clotting onset and faster clotting time in all dogs, which was sustained throughout the study duration. The shortening of the clotting times in the haemostatic assays indicate pharmacodynamic activity and the correction of the haemophilic coagulopathy.

In the study in neonatal haemophilia B dogs, a single IV dose of 2.5x10^{12} vg/kg of AAV-canine FIX-R338L vector (packaged in the AAVrh74var capsid) was administered to two cohorts of dogs, 2-3 days old dogs and 28-31 days old dogs. Haemostatic activity (aPTT, WBCT, TEG), clinical, and cellular chemistries as well as liver growth was assessed over a period of ≥12 months in order to better approximate and understand dosing and long-term efficacy of fidanacogene elaparvovec for patients at <2-year-old child range.

There are uncertainties in treating paediatric patients with regard to dose-response and duration of expression due to the impact of growth and development on transgene stability. In addition, the increase in blood volume is expected to dilute circulating levels of FIX.

Liver growth (evaluated by monthly liver ultrasounds) and blood volume expansion was measured for 12 months, until the dogs reached sexual maturity at an age equivalent to at least a 15 year-old human at study end. A proportional increase in liver growth and body weight was observed: in the 1-month-old cohort, increases of approximately 12-15x and 2-3x at 11-12 months post-treatment, respectively. Similarly, in the neonatal cohort increases of 54-66x and 2-3x were shown, respectively. The estimated blood volume expansion from the time of treatment to 11-12 months in the neonatal cohort and the one-month-old treatment cohort was approximately 0.025 liters to 1.6 liters and 0.1 liter to 1.6 liters, respectively.

In the neonatal cohort as well as in one month-old animal, the aPTT clotting time decreased >50% compared to baseline, while the FIX activity ranged from 1.5-4.4%. Sustained hemostatic activity analyzed via aPTT, WBCT, and TEG-R was observed in growing neonatal hemophilia dogs treated at an age of 2-3 days, with the values remaining stably decreased over the longitudinal assessment >12 months, with the exception of some minor fluctuations. Thus, no dilution effect was observed and the rapid liver growth during this observed period of development did not influence transgene stability, which was an unexpected observation. Overall, the shortening of the clotting times in the hemostatic assays indicate efficacy and pharmacodynamic activity of fidanacogene elaparvovec as well as the associated correction of the hemophilic coagulopathy in neonatal dogs remains stable over time.

The aPTT clotting time in one of the treated juvenile 1-month-old animals decreased from baseline by 20.2% on day 28 and by 47.9% on day 365 with a FIX activity of approximately 0.3%, which might still be protective with no bleeds occurring in this animal requiring treatment, but could be interpreted as insufficient response. In the other treated juvenile 1-month-old hemophilia dog, large fluctuations were observed in the TEG-R and WBCT assays, along with a delay in the decrease in aPTT clotting time, showing a decrease of less than 20% and extremely low FIX activity of less than 0.1%. This animal suffered from a head hematoma, a suspected spinal hematoma with being unable to walk, and several bleeds from approximately 13 months to 18 months post-gene therapy, which required multiple rcFIX-R338L protein treatments. These observations lead to the conclusion that the achieved FIX levels were not protective and resulted in treatment failure in this animal. The animal is thus considered a non-responder. The applicant stated that no inhibitory FIX-R338L antibodies were detected, which was measured by standard Bethesda assay, while a test for non-inhibitory antibodies was not conducted. The dog was euthanized at 18 months. The underlying reason for insufficient response to treatment failure in the early juvenile dogs remains unclear, but might be considered relevant for future procedures.

Overall, the non-clinical assessment of the primary pharmacodynamics of fidanacogene elaparvovec showed that murine, monkey, and dog hepatocytes were efficiently transduced and hFIX transgene was expressed in a dose-dependent manner, thus resulting in sustained hFIX plasma protein and activity levels. A decline in hFIX protein levels and hFIX activity was evident over time in NHP, attributed to the development of antibodies against the hFIX protein in monkeys.

2.5.2.2. Secondary pharmacodynamic studies

No secondary pharmacodynamic studies were performed. Since the pharmacological activity of hFIX is restricted to the human coagulation cascade, other pharmacological effects are not expected, thus dedicated secondary pharmacodynamic studies are not required.

2.5.2.3. Safety pharmacology programme

Safety pharmacology endpoints were included in the GLP-compliant general toxicity study in cynomolgus macaques, whereby no findings on the central nervous system, respiratory, and renal functions were observed.

2.5.2.4. Pharmacodynamic drug interactions

No pharmacodynamic drug interactions studies were conducted based on the intended use of fidanacogene elaparvovec and the known pharmacology of the transgene product.

2.5.3. Pharmacokinetics

Pharmacokinetics assessment of fidanacogene elaparvovec was incorporated in toxicological studies conducted in mice and non-human primate. An additional study on shedding of the vector into semen was conducted in rabbits (for a detailed discussion, please refer to the toxicology section). No dedicated classical ADME studies were performed.

Overall, data on the biodistribution of fidanacogene elaparvovec coming from two pivotal GLP-studies in cynomolgus monkey did not reveal unexpected findings on tissue distribution of the vector DNA or hFIX protein expression (data not shown). The selection of tissues for biodistribution analysis differed between studies, but the following list was collected in all pivotal GLP-compliant studies in cynomolgus monkeys: brain, colon, bone marrow, heart, kidney, liver, lung, lymph nodes, spleen, testes, thymus, and urinary bladder. Blood was collected to determine vector DNA levels in serum. Vector DNA in serum decreased from 910 copies/µg on day 30 post-dose to 262 copies/µg after 92 days (data not shown).

All off-target tissues showed detectable levels of the vector DNA, while liver presented the highest levels in all studies conducted, confirming the liver-specificity of the vector. AAV-Spark100-hFIX19-R338L vector DNA expression pattern was concluded to be similar to that seen with AAV8, with predominant vector genome accumulation in liver and spleen. An exception was the low and transient positive signal in brain upon AAV-Spark100-treatment, which was absent upon treatment with AAV8-vector. Associated DRG toxicity is considered to be low due to the fact that only minimal vector DNA and transgene expression was found in DRG. Associated potential DRG toxicity is further evaluated and discussed under toxicology section 3.2.4.1. Both vector DNA and FIX levels in serum increased with dose and declined over the observation time from 30 and 92 days. Of note, vector DNA was still detectable in liver and spleen until day 542 (data not shown).

Dose-dependency and a decrease of vector DNA levels was also observed for most of the off-target tissues investigated in NHP. Within 90 days of study duration, the vector copies were reduced in brain, colon, kidney, lung, lymph nodes (hepatic and mesenteric), pancreas, testes, and urinary bladder, but maintaining high levels in the liver, which is the target organ as well as spleen, heart, and inguinal lymph node (data not shown). However, increased hFIX DNA levels at day 92 compared to day 30 were detected in the diaphragm and the thymus.

Of note, vector DNA levels in off-target tissues were at least ~100-fold lower than in liver.

Circulating hFIX antigen levels were evaluated both in haemostatically normal C57BL/6 male mice over a period of 12 months (Study 8320414) and in cynomolgus monkeys for either 8 weeks (study 20049402) or 13 weeks (study 20059026). hFIX antigen levels in mice peaked around day 29 and remained stable until day 178, slightly declining until day 360 (data not shown). AAV-Spark100-hFIX treatment resulted in sustained hFIX expression in cynomolgus monkeys with peak levels at approximately week 3, declining to reach plateau levels by week 9, then remaining stable for the whole duration of the study (data not shown). In approximately one third of treated animals, the development of anti-hFIX antibodies following AAV-hFIX hepatic gene transfer was observed, which negatively affected hFIX expression levels in these animals (data not shown). Further kinetics studies of the FIX-R338L transgene protein expression applying LC/MS/MS in plasma from cynomolgus monkeys that received a single intravenous dose of 5×10^{12} vg/kg of fidanacogene elaparvovec showed a progressive slow decline

of FIX-R338L transgene expression from day 30 to day 360, remaining stable until day 741 for some of the treated animals (data not shown).

All animals injected with the AAV-Spark100-hFIX-R338L vector developed anti-capsid neutralizing antibodies.

2.5.4. Toxicology

2.5.4.1. Single dose toxicity

Fidanacogene elaparvovec (and related vectors used in nonclinical studies) were assessed in a series of single dose nonclinical studies. Single dose studies support the intended clinical use of a single administration. The intravenous route of exposure was selected for these studies since it is the intended route of clinical administration.

Toxicity studies conducted in cynomolgus monkeys included a pilot biodistribution study with multiple vectors that included AAV-Spark100-FIX19-Padua, fidanacogene elaparvovec, and AAV-Spark100-FIX103-Padua. The definitive 3-month GLP toxicity study in cynomolgus monkeys used AAV-Spark100-hFIX19-Padua and was intended to support clinical trials and registration.

A 2-year vector integration study to evaluate the integration profile and the risk of hepatocellular carcinoma in monkeys used the final construct fidanacogene elaparvovec. Vector integration was also studied in neonatal as well as juvenile hemophilia B dogs. Vector integration was evaluated in the liver due to the potential for hepatocarcinogenesis as reported in juvenile mice (for further details regarding specialized genotoxicity studies on vector integration, please refer to section 2.5.4.3).

In cynomolgus monkeys, there were no fidanacogene elaparvovec–related clinical signs or changes in food consumption, body weights, haematology, clinical chemistry, or anatomic pathology (gross pathological observations, organ weights, and histopathology) (data not shown). Anatomic pathology of both the liver and the spleen was performed on day 542 to assess long-term toxicity, with no gross or microscopic findings, including gross or microscopic evidence of thrombosis, or organ weight changes being detectable (data not shown). With regard to altered hepatocellular morphology or hepatocyte proliferation, no microscopic or ultrasound-evidenced adverse neoplastic transformation effect could be found over the whole period of study duration (data not shown). Besides liver toxicity, the heart and dorsal root ganglions as well as an increase of inflammatory reactions are described as well-known potentially preferred sites for toxicity due to the tropism of AAV8-based vectors. Since fidanacogene elaparvovec shows a similar tropism as AAV8-based vectors, both liver and heart tissue were examined for putative toxic signs and did not reveal any safety concerns (data not shown). Potential DRG toxicity was evaluated by spinal cord and peripheral nerve histology in the 90-day toxicity study in cynomolgus monkeys. No axonal degeneration was observed, which would be expected upon degeneration of DRG neurons. DRG pathology in the spinal cord and peripheral nerve primarily occurs upon overexpression of the transgene in DRG, whereby only minimal vector DNA and transgene expression was found in DRG in the 2-year toxicity monkey study with fidanacogene elaparvovec. The DNA copy number in DRG samples from terminal necropsies was approximately 3 log lower than in liver samples, and FIX-Padua transgene mRNA expression in the DRG was found in only 1 animal at very low levels at day 92 post-treatment. The average VCN at the 2-year time point for treated animals (10053 copies per µg DNA) was greater than the average at the interim timepoint (5082 copies per µg DNA). However, VCN remained approximately 3 logs lower than the detected VCN in liver samples at each time point, with transgene expression in the DRG at the 2-year time-point remaining low to undetectable in treated animals. Transgene expression was $2.29x10^{-4}$ to $2.21x10^{-5}$ or below LLOQ in 2 of 6 animals necropsied at 2-years (data not shown).

All fidanacogene elaparvovec-treated NHP showed shortened aPTT at all time-points investigated and at all dose regimens, when compared to the control group (data not shown). No thrombus formation was observed in any of the animals, while plasma TAT complexes and D-dimer levels remained mostly unaffected by treatment. No notable histopathological findings were recorded.

Cell-mediated immune response in NHP was assessed by analysing spleen cells collected on day 92 and PBMCs on day 56, while no fidanacogene elaparvovec related FIX-reactive, IFN-γ-producing spleen cells or PBMCs were detected. In one female animal dosed with fidanacogene elaparvovec, higher numbers of IFN-γ-producing spleen cells as well as PBMCs were detected, but could not be definitively attributed to the test article due to only low numbers of spot forming cells (19 SFCs/10^6 spleen cells; 34 SFC/10^6 PBMC). Moreover, there was no indication of a possible cell-mediated immune response in the microscopic examination of the liver and the spleen as well as no organ weight changes at terminal necropsy (data not shown).

In mice, unscheduled deaths occurred in fourteen animals of both the Padua- and WT-vector group, which were associated with pathological findings in the brain and/or skin. Microscopic findings in the brain included minimal to moderate acute haemorrhage, parenchymal loss, necrosis, gliosis, fibroplasia, pigment, vascular thickening/proliferation, and vessel wall degeneration. Among all groups, 20 animals were found dead or were euthanized in a moribund condition during the study. Fourteen of these unscheduled mortalities were considered test article-related based upon anatomic pathology findings in the brain and/or skin. Test article-related adverse brain and skin findings occurred at a higher incidence and/or severity in animals administered 1.04×10^9 vg/animal PD or 3.45×10^9 or 6.44×10^9 vg/animal WT compared with controls or high dose Padua-treated group. According to the applicant the brain findings were likely test article-related exacerbations of sequelae from tissue/vascular injury related to the submandibular blood collection procedure, since animals had at least 18 scheduled submandibular blood collections and deaths could be temporally correlated to this procedure in some animals and interpreted as an exaggerated pharmacological effect after tissue injury (potentially resulting in haemorrhage, thrombosis, necrosis, among others). However, similar microscopic or macroscopic brain findings were also observed in two control animals. Since these were non-haemophilic mice, administration of a single dose of test article resulted in an up to 146% increase in FIX activity and a correlated increase in FIX antigen in hemostatically normal mice (data not shown). Due to the generally unilateral distribution of these findings and the temporal proximity of blood collection using the submandibular technique to the death/euthanasia of these animals, the blood collection procedure likely contributed to the mortality of these animals. Similar blood collection techniques were not employed in the monkey studies nor are they used clinically. Therefore, the adverse findings in mice might not impact the risk to humans.

Pathological skin findings, including scab in the dorsal cervical region presenting mixed cell inflammation with crust, epidermal hyperplasia, and ulcer. Skin findings were considered an exacerbation of the mouse strain-related spontaneous background finding of increased incidence of ulcerative dermatitis, which was potentiated by vascular injury associated with the submandibular bleeding procedure.

A combined pharmacology/toxicology study was conducted in a total of twelve juvenile male haemophilia B dogs of the Lhasa Apso colony. Dogs received a single dose of canine version of AAV-Spark100-hFIX39-R338L vector at 3 months (approx. resembling 2 to 6-years human age range) or 6 months (approx. resembling 6 to <12-year human age range) of age and were followed for a period of ≥12 months. In each age group, dogs received either $5x10^{11}$ vg/kg (2 dogs), $2.5x10^{12}$ vg/kg (3 dogs), or $5x10^{12}$ vg/kg (1 dog) of the AAV-canine FIX-R338L vector. Administration of AAV-canine FIX R338L resulted in the shortening of the WBCT and aPTT clotting time in all dogs dosed at 3- and 6-months of age, indicating FIX activity. TEG measurements show a faster clotting onset and faster clotting time in all treated dogs, while remaining stable and sustained until days 370-547. Clinical chemistry evaluations included liver enzymes ALT and AST post-administration of AAV-canine FIX-R338L. Both body weights and liver growth were monitored upon treatment with the AAV-canine FIX-R338L vector to evaluate the potential dilution

impact through puberty during the first year of the study duration and through natural life thereafter with a special focus on potential development of hepatocellular carcinoma. In the 3-month cohort, an increase of 2.2-3.4-fold in body weights and 1.4-1.8-fold in liver size was observed at 18-24 months post-treatment. Similarly, in the 6-month-old cohort, the body weights increased 1.5-1.7-fold, while liver size augmented 1.4-1.8-fold. Mild elevations of both AST (1/12 dogs) and ALT (3/12 dogs) of less than 5-fold were observed, with all elevations resolving and no overt adverse events being observed. A liver biopsy was taken 1-2 months post-gene therapy for vector copy number and vector integration (discussed below under genotoxicity/integration analysis). The amount of vector copies per µg of DNA retrieved correlated with the vector dose that was administered to the animals, and the same dose-dependent pattern was shown for transgene expression data in liver biopsies 1-2 months post-dose (data not shown).

A combined pharmacology/toxicology study was also conducted in neonatal and early juvenile male haemophilia B dogs of the Lhasa Apso colony receiving a single IV dose of $2.5x10^{12}$ vg/kg of AAV-canine FIX-R338L vector (packaged in the AAVrh74var capsid). Two cohorts of dogs were studied: 2-3 days-old neonatal dogs and 28-31 days-old early juvenile dogs. Haemostatic activity (aPTT, WBCT, TEG), clinical, and cellular chemistries as well as liver growth was assessed over a period of ≥12 months in order to better approximate and understand dosing and long-term efficacy of fidanacogene elaparvovec for patients at <2-year-old child range. To detect potential systemic toxicity of the gene therapy, serum was analysed for clinical chemistry parameters. It was observed that one of the neonatal animals exhibited a mild elevation (1.3x) of ALT above the upper limit of normal at day 362 post-treatment and one of the juvenile dogs exhibited a mild elevation (1.2x) at day 182 post-treatment, whereby it was noted that all elevations resolved. All gene therapy-treated hemophilia B dogs had an elective liver biopsy approximately 3 months post-treatment for the analysis of vector copy number and vector integration (discussed below under genotoxicity/integration analysis).

The overall no-observed-adverse-effect-level (NOAEL) for fidanacogene elaparvovec in NHP was $5x10^{12}$ vg/kg, which is the highest dose tested. At the proposed starting clinical dose of $1x10^{12}$ vg/kg, the peak human FIX activity was within 50-150% of normal FIX activity levels, and the plateau levels were <40% of normal.

2.5.4.2. Repeat dose toxicity

No repeated dose toxicity studies were presented.

2.5.4.3. Genotoxicity

AAV-vectors predominantly reside in episomal concatemeric forms. Low level of vector integration of AAV vectors is considered an identified risk. An integration analysis was conducted with liver tissue obtained in a 2-year cynomolgus monkey study to characterize the integration profile in the liver as well as potential proliferative lesions in the liver, which, in turn, might lead to insertional mutagenesis.

Vector integration was measured by target enrichment sequencing (TES) and using shearing extension primer tag selection ligation-mediated polymerase chain reaction (S-EPTS/LM-PCR) on liver samples from Day 92 and 2-year necropsy. The retrieved integration profile shows a random distribution throughout the host genome with a low frequency that was below the published spontaneous mutation rate estimates for the liver. 41.9% (by S-EPTS/LM-PCR) and a 70.7% (by TES) of integration sites were identified as common integration sites (CIS). A common integration site analysis highlighted albumin (up to 6.4% of CIS measured by S-EPTS/LM-PCR), APOC1 (up to 19.7% measured by TES), and SERPINA1 (up to 15.9% measured by TES) as genes with clusters of recurrent integration. These genes are highly expressed genes in the liver and are likely to represent open chromatin, preferentially being

accessible for integration. Of note, minimal overlap was observed in the otherwise largely random integration profiles between the two applied methods. Moreover, the observed integration profile was not associated with genes previously implicated in clonal outgrowth or malignant transformation (i.e. proto-oncogenes MECOM, LMO2 or HMGA2), with no proliferative changes in the liver following extensive sampling for histopathology nor multiple liver ultrasound exams during the course of the 2-year study in NHP.

However, clusters of recurrent integration were identified for SERPINA1. As analyses with the cancer gene database cBioPortal reveal that SERPINA1 may be associated with a variety of cancer types, including hepatocellular carcinoma. It was further clarified that the observed clusters of recurrent integration proximal to the genes APOC1 and SERPINA1 following administration of fidanacogene elaparvovec were also found in vehicle-treated animals in the same studies as well as in spike-in negative control HepG2 samples used for assay qualification. The applicant pointed out that further examination of these observations identified areas of strong sequence similarity to the vector sequence in both of these putative hotspot loci. Thus, the possibility of false positives exists due to multiple mapping of sites to SERPINA1 and APOC1. As a follow-up measure of these observations, the sponsor improved the insertion detection analysis pipeline in order to reduce false positives by first aligning reads to the vector genome with the aim of improving the specificity of integration detection. Applying this approach, the majority of the reads contributing to the previously identified APOC1 and SERPINA1 insertions were largely or completely masked, indicating that the majority of APOC1 and SERPINA1 apparent integrations were not host-vector true integration sites.

A combined pharmacology/toxicology study was conducted in a total of twelve juvenile (3 months- and 6 months-old) male haemophilia B dogs of the Lhasa Apso colony. A liver biopsy was taken 1-2 months post-gene therapy for vector copy number and vector integration. In the integration sites (IS) data obtained from all samples, 574 IS (20.65% of all IS) were detected in a total of 248 common integration sites (CIS). CIS analysis revealed that 20.65% of IS can be clustered within a threshold of 50 kbp. The Top1 CIS, Alb, consisted of 16 IS (derived from 8 different samples). The Alb locus was previously identified as AAV integration hotspot in dogs and other large as well as small animal models, described in Nguyen et al., 2021. Overall, integration data did not reveal any signs of clonal dominance, no preferred IS loci and no single clones with increased frequencies in the proximity of well-characterized cancer-associated genes.

A further combined pharmacology/toxicology study was conducted in 3 neonatal (2-3 days-old) and 2 early juvenile (1 month-old) male haemophilia B dogs of the Lhasa Apso colony. All gene therapy-treated hemophilia B dogs had an elective liver biopsy approximately 3 months post-treatment. Liver tissue was processed for TES analysis, VCN measurement by qPCR, and integration site analysis by S-EPTS/LM-PCR. Livers from three naïve hemophilia B dogs were included as controls. Treated dogs exhibited a low (0.02%) level of viral integration in liver, which was evaluated by TES, with the majority of the IS being distributed throughout the dog genome, showing recurrent integrations near APOC4. Of note, no evidence of clonal expansion, no preferred integration loci, and no single clones with elevated frequencies in proximity to cancer associated genes was found. Based on the fact that no clear clonal expansion event was identified, the overall low frequency, and the random nature of integration in both neonatal (2-3 days-old) and early juvenile (1 month-old) male haemophilia B dogs, no specific integration event was followed up with alternative methods.

2.5.4.4. Carcinogenicity

Dedicated studies on carcinogenicity were not conducted with fidanacogene elaparvovec. However, rare single integration events may contribute as a predisposing factor for tumour development. Thus, a

theoretical risk of insertional oncogenesis remains, especially in view of potential risk groups like e.g. patients pre-disposed for liver transformation.

2.5.4.5. Reproductive and developmental toxicity

Germline transmission was evaluated in a rabbit semen clearance study, which showed that vector was no longer detectable in the seminal fluid by 5 months after a dose 20x greater than the clinical dose.

As hemophilia B is almost exclusively limited to male patients, and as the current MAA only comprises adult patients, dedicated FEED, EFD, and PPND studies are not required.

A combined pharmacology/toxicology study was conducted in a total of twelve juvenile (3 months- and 6 months-old) as well as two early juvenile (1 month-old) and three neonatal (2-3 days-old) male haemophilia B dogs.

2.5.4.6. Toxicokinetic data

N/A

2.5.4.7. Local tolerance

No dedicated local tolerance studies were conducted, whereby no notable findings were observed at microscopical examination of the infusion site, which was part of the general NHP toxicity studies conducted.

2.5.4.8. Other toxicity studies

Immunotoxicity studies with fidanacogene elaparvovec have not been conducted. However, the immune response to both the AAV capsid and human FIX protein has been extensively characterized. Antigenicity endpoints were included in single dose toxicity studies, with capsid antigens generally causing high titers of anti-AAV antibodies. In addition, anti-hFIX antibodies were observed inconsistently in tested animals. The absence of stand-alone antigenicity studies is acceptable.

2.5.5. Ecotoxicity/environmental risk assessment

As fidanacogene elaparvovec is considered a GMO, a separate GMO environmental risk assessment report was done to estimate the risk of fidanacogene elaparvovec to third parties and the environment.

Fidanacogene elaparvovec is an AAV-based vector with an expression cassette containing a codon-optimized hFIX-R338L transgene variant under the control of a liver-specific promoter. Fidanacogene elaparvovec is replication-incompetent and all viral genes have been removed with exception of the ITRs that remain as the only sequence of viral origin. The transgene and its regulatory sequences are not harmful and non-toxic. The vector genome is packaged within a recombinant viral capsid (AAVRh74var) derived from the naturally occurring AAV serotype Rh74. The genetic modifications introduced in fidanacogene elaparvovec do not affect the host range and tissue tropism of the vector.

Protective measures on handling, preparing and on administration of fidanacogene elaparvovec are in place in the SmPC. In addition, restrictions regarding donation of blood, organs, tissues and cells after treatment with fidanacogene elaparvovec are also in place in the SmPC.

The vector is shed from patients for some time after administration. The amount of vector shed from patients is expected to be too low to cause efficient transduction in any organism. Irrespective of this,

patients treated with fidanacogene elaparvovec are advised to practice good hygiene to limit the spread of the vector. In addition, released viral particles are quickly diluted in the environment to non-detectable levels and will ultimately be rapidly degraded by natural processes.

Overall, the risk of fidanacogene elaparvovec to third parties or to the environment is negligible.

2.5.6. Discussion on the non-clinical aspects

The non-clinical pharmacodynamics program for fidanacogene elaparvovec is comprehensive and includes a panel of in vitro and in vivo studies in haemostatically normal mice and NHP as well as in disease models applying hemophilia B mice and dogs.

Of note, non-clinical efficacy (as well as safety) studies were performed as changes were implemented in order to optimize the vector. Therefore, multiple, highly related constructs were evaluated for optimization of the vector with regards to hFIX-R338L transgene variant and the novel capsid. In vivo pharmacology studies were performed to elucidate both the effects of the FIX-R338L variant and the AAVRh74var capsid in terms of their effects on efficacy. Moreover, for the majority of non-clinical studies, the vectors used were produced by Process 1, while comparability was established across manufacturing processes utilized (Process 1, 2, and 3), with Process 3 being the intended commercial process.

Potential effects of corticosteroid treatment that is applied in patients that develop transaminitis secondary to transfection of hepatocytes, were neither discussed nor addressed by the applicant. Due to the common use and the known clinical effects, this issue will not be followed up from a nonclinical point of view.

Collectively, the employed studies showed efficient liver transduction and expression of hFIX, which is driven by a liver-specific promoter. Circulating hFIX protein levels as well as hFIX activity were determined to be dose-dependent. It was confirmed that the hFIX-R338L is more effective than the WT variant, presenting a 7-10-fold increased specific activity in mice. Efficacy was assessed in view of safety (thrombus formation) with regard to the increased expression of the hFIX-R338L variant as well as biological activity (coagulation). The risk of excessive coagulation (thrombosis) activity was analysed by checking for thrombin-antithrombin (TAT) complexes and D-dimers. Coagulation efficiency was evaluated by activated partial thromboplastin time assay. Overall, the improvement of specific hFIX activity resulted in a reduced number of bleeds per month and a reduced bleeding time, with no elevation of pathological coagulation measured by TAT and D-dimer value.

Since previous clinical trials of AAV-mediated gene transfer to the liver showed pre-existing humoral immune responses to AAV capsids in humans, which might prevent transduction, the AAVRh74var capsid was optimized for enhanced resistance to neutralization by the immune system. Only a slightly enhanced immune resistance of the AAVRh74var capsid (64% ± 9.8%) with less nAb formation compared to the AAV8-capsid (55% ±15.3%) was shown in a murine in vivo study. Less immunogenicity of the bioengineered capsid as used in AAVRh74var was also shown in vitro in human hepatocellular Huh7 cells. Additionally, in an in vitro killing assay to assess effector T cell responses against the AAVRh74var capsid was confirmed that the AAVRh74var capsid may be presented to the immune system to a lesser degree than both AAV8 and AAV2 capsids.

Further proof-of-concept as well as immunogenicity studies applying a canine FIX-R338L variant in juvenile and neonatal haemophilia B dogs of the Lhasa Apso colony were performed. Shortening of the clotting times in the hemostatic assays indicate efficacy and pharmacodynamic activity of fidanacogene elaparvovec as well as the associated correction of the hemophilic coagulopathy in neonatal (2-3 days-old) dogs. However, in early juvenile dogs (1 month-old) one of the treated dogs showed reduced efficacy

and a potential insufficient response, whereby the other early juvenile dog did not achieve protective FIX levels, finally resulting in treatment failure.

Pharmacokinetics assessment of fidanacogene elaparvovec was incorporated in toxicological studies conducted in mice and NHP. An additional study on shedding of the vector into semen was conducted in rabbits. No dedicated classical ADME studies were performed, which is acceptable considering the nature of this gene therapy medicinal product.

While all off-target tissues showed detectable levels of the vector DNA, liver presented the highest levels in all studies conducted, confirming the liver-specificity of the vector. AAV-Spark100-hFIX19-R338L vector DNA expression pattern was concluded to be similar to that seen with AAV8, with predominant vector genome accumulation in liver and spleen, except for the low and transient positive signal in brain upon AAV-Spark100-treatment, which was absent upon treatment with AAV-8 capsid vector. Associated DRG toxicity is considered to be low due to the fact that only minimal vector DNA and transgene expression was found in DRG and no DRG-associated toxic signs were found. Both vector DNA and FIX expression levels in serum increased with dose and declined over the observation time from 30 and 92 days. Of note, vector DNA was still detectable in liver and spleen until day 542. Within 90 days of study duration, the vector copies were reduced in brain, colon, kidney, lung, lymph nodes (hepatic and mesenteric), pancreas, testes, and urinary bladder, but maintaining high levels in the liver, which is the target organ as well as spleen, heart, and inguinal lymph node. In contrast, increased hFIX DNA levels at day 92 compared to day 30 were detected in the diaphragm and the thymus, indicating a potential accumulation over time. However, it is difficult to differentiate if the increase in DNA levels is either related to inter-animal variability in combination with a small number of animals used for distribution studies or a true increase of hFIX DNA levels over time. Moreover, no safety findings were observed in the GLP NHP toxicity study in these organs and to date no reports point to diaphragm and thymus as potential targets for AAV gene therapy.

hFIX antigen levels in mice peaked around day 29 and remained stable until day 178, slightly declining until day 360. AAV-Spark100-hFIX treatment resulted in sustained hFIX expression in cynomolgus monkeys with peak levels at approximately week 3, declining to reach plateau levels by week 9, then remaining stable for the whole duration of the study. In approximately one third of treated animals, the development of anti-hFIX antibodies following AAV-hFIX hepatic gene transfer was also observed, which negatively affected hFIX expression levels in these animals. All animals injected with the AAV-Spark100-hFIX-R338L vector developed anti-capsid neutralizing antibodies.

The non-clinical toxicology program of fidanacogene elaparvovec included single-dose toxicity studies, thus supporting the intended clinical use of a single treatment. The intravenous route of administration was selected for these studies since it is the intended route of clinical administration. Toxicity studies conducted in cynomolgus monkeys included a pilot biodistribution study with multiple vectors (AAV-Spark100-FIX19-Padua, fidanacogene elaparvovec, and AAV-Spark100-FIX103-Padua) and a definitive 3-month GLP toxicity study applying AAV-Spark100-hFIX19-Padua. A combined 2-year toxicity/vector integration study was performed to evaluate the integration profile and the risk of hepatocellular carcinoma in cynomolgus monkeys, using the clinical construct fidanacogene elaparvovec. NHP without pre-existing anti-AAVRh74var antibodies were included in these studies. Liver toxicity as well as vector integration was also evaluated in juvenile hemophilia B dogs in a combined pharmacology/vector integration study. A 12-month GLP male mouse study evaluated toxic signs associated with the treatment of either AAV-Spark100-FIX-WT or AAV-Spark100-FIX19-Padua. A clearance study in semen was performed in rabbits to address germline transmission upon AAV-Spark100-hFIX16-WT and AAV-Spark100-hFIX19-Padua treatment.

The overall no-observed-adverse-effect-level (NOAEL) for fidanacogene elaparvovec in NHP was $5x10^{12}$ vg/kg, which is the highest dose tested in NHP.

In NHP, there were no test article–related clinical signs or changes in food consumption, body weights, haematology, clinical chemistry, or anatomic pathology (gross pathological observations, organ weights, and histopathology). Anatomic pathology of both the liver and the spleen was performed on day 542 to assess long-term toxicity, with no gross or microscopic findings, including gross or microscopic evidence of thrombosis, or organ weight changes being detectable. With regard to altered hepatocellular morphology or hepatocyte proliferation, no microscopic or ultrasound-evidenced adverse neoplastic transformation effect could be found over the whole period of study duration. Since fidanacogene elaparvovec shows a similar tropism as AAV8-based vectors, both liver and heart tissue were examined for putative toxic signs and did not reveal any safety concerns. Potential DRG toxicity was evaluated by spinal cord and peripheral nerve histology in the 90-day toxicity study in cynomolgus monkeys, with no axonal degeneration being observed, and thus, no degeneration of DRG neurons. DRG pathology in the spinal cord and peripheral nerve primarily occurs upon overexpression of the transgene in DRG, whereby only minimal vector DNA and transgene expression was found in DRG in the 2-year toxicity monkey study with fidanacogene elaparvovec.

No thrombus formation was observed in any of the animals, while plasma TAT complexes and D-dimer levels remained mostly unaffected by treatment, thus no signs of exaggerated coagulation were noted.

No notable histopathological findings were recorded. No cell-mediated immune response was detected upon analysis of test-article-related FIX-reactive, IFN-γ-producing spleen cells and PBMC isolated from treated NHP on day 92 and day 56, respectively.

In mice, unscheduled deaths occurred in fourteen animals of both the Padua- and WT-vector group, which were associated with pathological findings in the brain and/or skin. Among all groups, 20 animals were found dead or were euthanized in a moribund condition during the study. Fourteen of these unscheduled mortalities were considered test article-related based upon anatomic pathology findings in the brain and/or skin. Test article-related adverse brain and skin findings occurred at a higher incidence and/or severity in animals administered 1.04×10^{9} vg/animal PD or 3.45×10^{9} or 6.44×10^{9} vg/animal WT compared with controls or high dose Padua-treated group. According to the applicant the brain findings were likely test article-related exacerbations of sequelae from tissue/vascular injury related to the submandibular blood collection procedure since animals on had at least 18 scheduled submandibular blood collections and deaths could be temporally correlated to this procedure in some animals and interpreted as an exaggerated pharmacological effect after tissue injury (potentially resulting in haemorrhage, thrombosis, necrosis, among others). This justification appears generally plausible as treated animals were hemostatically normal mice.

AAV-vectors predominantly reside in episomal concatemeric forms. Low level of vector integration of AAV vectors is considered a potential identified risk. An integration analysis was conducted with liver tissue obtained in a 2-year cynomolgus monkey study to characterize the integration profile in the liver as well as potential proliferative lesions in the liver, which, in turn, might lead to insertional mutagenesis. The retrieved integration profile shows a random distribution throughout the host genome with a low frequency that was below the published spontaneous mutation rate estimates for the liver. Moreover, the observed integration profile was not associated with genes implicated in clonal outgrowth or malignant transformation, with no proliferative changes in the liver following extensive sampling for histopathology nor multiple liver ultrasound exams during the course of the 2-year study in NHP.

Vector integration was measured by the applicant via target enrichment sequencing (TES) and by Protagene using shearing extension primer tag selection ligation-mediated polymerase chain reaction (S-EPTS/LM-PCR) on liver samples from Day 92 and 2-year necropsy. Overall, a low frequency of integration was observed, which is distributed throughout the NHP genome, considering a 41.9% (by S-EPTS/LM-PCR) and a 70.7% (by TES) of integration sites as common integration sites (CIS). A common integration site analysis highlighted albumin (up to 6.4% of CIS measured by S-EPTS/LM-PCR), APOC1

(up to 19.7% measured by TES), and SERPINA1 (up to 15.9% measured by TES) as genes with clusters of recurrent integration. These genes are highly expressed genes in the liver and are likely to represent open chromatin, preferentially being accessible for integration.

It is known from previous studies that actively transcribed genes containing open chromatin are more accessible for integration, thus a higher incidence of rAAV integrations might be expected. However, genes showing high hepatic activity are also often associated with hepatotoxicity. It is established in the literature that CIS with IS <5 often occur by chance, thus are unlikely to have any biological relevance (Wu et al, 2006). TES data presented shows intense clustering of IS for APOC1 and SERPINA1 genes only, with predominance of lower order IS (APOC1: 6 read-counts, SERPINA1: 8 read-counts). If clonal expansion would be present, the 2-year data would be expected to show a dramatic increase in read-counts compared with the 13-week data. The lack of such a shift for both APOC1 and SERPINA1 suggests that integration-related clonal expansion might not be expected. However, analyses of SERPINA1 gene with the cancer gene databases cBioPortal showed that it has been previously associated with carcinogenesis in a variety of cancer subtypes, including hepatocellular carcinoma. However, clusters of recurrent integration proximal to the genes APOC1 and SERPINA1 following administration of fidanacogene elaparvovec were also found in vehicle-treated animals in the same studies as well as in spike-in negative control HepG2 samples used for assay qualification, indicating that the majority of APOC1 and SERPINA1 apparent integrations were not host-vector true integration sites.

An additional combined pharmacology/toxicology study was performed in neonatal (2-3 days-old), early juvenile (1 month-old), and juvenile (3 months- and 6 months-old) haemophilia B dogs. Very few IS were identified in dogs dosed as neonates (14-42 IS) or at 1 month of age (9-14 IS), consistent with overall low VCN, whereby the detected IS levels in the dogs at all treated ages remained several orders of magnitude below previously described spontaneously occurring mutations in the liver. 6-months old dogs presented higher average IS than 3-months old dogs, which was consistent with the VCN levels, resulting in an average of 0.1% of the vector DNA integration in the 3-6 month old dogs, while 99.9% of viral DNA remained in an episomal form. Moreover, the pattern of integration appeared to be similar across ages and doses. Along the same line, common integration site (CIS) analysis revealed recurrent integrations near APOC4 in all treated dogs, regardless of dose or age. Additionally, no clear clonal expansion event was identified and a random integration pattern with an overall low frequency was determined.

Of note, minimal overlap was observed in the otherwise largely random integration profiles between the two applied methods, with no evidence of prominent clonal expansion or preferential integration near known cancer genes.

Dedicated studies on carcinogenicity were not conducted with fidanacogene elaparvovec. Results obtained from integration analyses might not raise any specific carcinogenicity concern so far. Even if the clinical relevance of individual integration events is not known to date, it is acknowledged that rare single integration events may contribute as a predisposing factor to the risk of malignancy.

The risk of germline transmission was evaluated in a rabbit semen clearance study, which showed that vector was no longer detectable in the seminal fluid by 5 months after a dose 20x greater than the clinical dose.

Immunotoxicity studies with fidanacogene elaparvovec have not been conducted. However, the immune response to both the AAV capsid and human FIX protein has been extensively characterized. Antigenicity endpoints were included in single dose toxicity studies, with capsid antigens generally causing high titers of anti-AAV antibodies. In addition, anti-hFIX antibodies were observed inconsistently in tested animals.

A separate GMO ERA assessment report was conducted, since fidanacogene elaparvovec is declared a genetically modified organism, specifically an AAV-based vector. Overall, the risk of fidanacogene elaparvovec to third parties or to the environment is considered to be negligible.

The CHMP endorse the CAT discussion on the non-clinical aspects as described above.

2.5.7. Conclusion on the non-clinical aspects

Overall, the primary pharmacodynamic studies provided adequate evidence that fidanacogene elaparvovec as well as highly related constructs used throughout the nonclinical development efficiently transduce hepatocytes in order to restore FIX protein levels as well as clotting activity in a dose-dependent manner both in haemostatically normal mice and NHP as well as in diseased hemophilia B mice and dogs.

The pharmacokinetic investigation focused on the biodistribution of the vector in cynomolgus monkeys. In addition, FIX antigen and FIX activity were determined in serum samples in mice and cynomolgus monkeys:

- Fidanacogene elaparvovec demonstrated a similar DNA expression pattern to that seen with AAV8, with predominant vector genome accumulation in liver and spleen tissues. Vector DNA was detected in all tissues evaluated out to Day 92 in cynomolgus monkey. For most of the off-target tissues, a decrease in vector DNA copy number was observed from day 30 to 92 (except for thymus and diaphragm). Vector DNA was detected in liver and spleen out to Day 542.
- hFIX antigen levels in mice peaked around day 29 and remained stable until day 178, slightly declining until day 360. AAV-Spark100-hFIX treatment resulted in sustained hFIX expression in cynomolgus monkeys with peak levels at approximately week 3, declining to reach plateau levels by week 9, then remaining stable for the whole observation duration (13 weeks).

GLP-compliant toxicity studies showed that fidanacogene elaparvovec was well tolerated in mice and monkeys. The studies did not reveal any adverse target organ toxicities associated with the administration and biodistribution of the vector, or as a consequence of high-level expression and activity of the hFIX-Padua protein. The overall no-observed-adverse-effect-level (NOAEL) for fidanacogene elaparvovec in NHP was $5x10^{12}$ vg/kg, which is the highest dose tested in NHP.

No repeated dose toxicity studies were presented. This is acceptable, as repeat administration of fidanacogene elaparvovec is not foreseen for clinical application, while the high immunity against the AAV74var vector after the first administration might even complicate subsequent administrations in terms of safety.

Integration analyses were conducted with liver tissue obtained in a 2-year adult haemostatically normal cynomolgus monkey study as well as in a haemophilia B dog study (neonates: 2-3 days-old; early juvenile: 1 month-old; juvenile: 3 months- and 6 months-old) to characterize the integration profile in the liver as well as potential proliferative lesions in the liver, which, in turn, might lead to insertional mutagenesis. Overall, the observed integration profile was not associated with genes previously implicated in clonal outgrowth or malignant transformation (such as proto-oncogenes MECOM, LMO2 or HMGA2), showing a random distribution throughout the host genome with overall low frequency. Moreover, the pattern of integration appeared to be similar across ages and doses.

Fidanacogene elaparvovec can be granted a marketing authorisation from a non-clinical point of view.

The CHMP endorse the CAT conclusions on the non-clinical aspects as described above.

2.6. Clinical aspects

2.6.1. Introduction

GCP aspects

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**

Study/Status	Study Design Number Participants Planned	Study Dose Treatment Duration	Number Participants Enrolled/Dosed/Completed
C0371002 **Ongoing** data cutoff: 16 Nov 2022	• **Phase 3**, open-label, single-arm study to evaluate the efficacy and safety of FIX gene transfer with fidanacogene elaparvovec in adult male participants with moderately severe to severe hemophilia B (FIX:C≤2%). • Total number of participants planned: at least 40	• Single IV infusion of 5×10^{11} vg/kg • Duration of follow-up: 6 years	45 (dosed) 46 (enrolled) 41 (completed 15 months of follow-up) 24 (completed ≥2 years of follow-up) 2 (completed ≥3 years of follow-up)
C0371004 **Ongoing** data cutoff: 02 Nov 2022	• **Phase 3**, Open-label, non-IP, multi-center, lead-in study to evaluate prospective efficacy and select safety data of current FIX prophylaxis replacement therapy in the usual care setting of moderately severe to severe adult hemophilia B participants (FIX:C≤2%) who are negative for nAbs to AAVRh74var prior to Phase 3 study (C0371002).[a] • Total number of participants planned: ~111	• No IP administration • Prospective collection of data while on prophylaxis FIX regimen prior to entering C0371002	102 (enrolled) 59 (completed)
C0371005 **Complete**	• **Phase 1/2a**, open-label, non-randomized, dose-escalation, multi-center study in adult male participants with hemophilia B (FIX:C≤2%). • Total number of participants planned: ~ 20	• Single IV infusion of 5×10^{11} vg/kg • Duration of follow-up: 1 year	15 (dosed) 22 (enrolled) 15 (completed)
C0371003 (also referred to as SPK-9001-LTFU-101) **Ongoing** data cutoff: 02 Nov 2022	• **Phase 2a**, open-label, non-randomized, multi-center, LTFU safety and efficacy study in adult males with hemophilia B who previously received a single infusion of IP in C0371005 or at a higher dose in the C0371005 dose-escalation substudy. • Amendment 2 (Sept 2020) instituted a dose-escalation substudy to include up to [redacted] additional higher doses [redacted].[b]	• Single IV infusion of 5×10^{11} vg/kg dosed in C0371005 • No IP administration in LTFU • Duration of LTFU: 5 years (in addition to the 1 year in C0371005) Dose-escalation substudy:[b] • a single IV infusion of up to [redacted] additional PF 06838435 dose levels [redacted]	5×10^{11} vg/kg dose: 14 (enrolled) 7 (ongoing)5 (completed) Dose-escalation substudy:[b] [redacted] (enrolled) [redacted] (dosed)
C0371017 **Planned**	• **Phase 3**, Non-Investigational Product, Multi-Country, Low Interventional, Cohort Study to Describe Long-Term Safety and Effectiveness of Giroctocogene Fitelparvovec or Fidanacogene Elaparvovec in patients who have received treatment through participation in a Pfizer-sponsored clinical trial.[a] • Total number of participants planned: 145	• Not applicable • Participant duration in the study will vary based upon their date of entry into this study as participants may be followed in this study through 15 years after their infusion of investigational giroctocogene fitelparvovec or fidanacogene elaparvovec	The total number of participants enrolled into this study is dependent upon the total number of eligible participants that receive investigational product in the clinical trials. The number of participants who previously received fidanacogene elaparvovec that can be enrolled into this study would be approximately 145 clinical trial participants from the clinical trials.

Sources: Module 5.3.5.1 C0371004 Interim CSR; Module 5.3.5.1 C0371002 CSR; Module 5.3.5.2 C0371005 CSR; Module 5.3.5.2 C0371003 Interim CSR; Module 5.3.5.4 C0371017 Protocol.

a. Study includes a separate cohort of hemophilia A participants related to a separate clinical development program.

b. Dose-escalation data are limited in quantity and are summarized separately and are provided in Module 2.7.4.5.9.

2.6.2. Clinical pharmacology

2.6.2.1. Pharmacokinetics

No dedicated clinical pharmacology studies were conducted, however, vector shedding data assessed by qPCR is available. Vector DNA concentrations have been measured in plasma in study C0371002, in serum in studies C0371005 and C0371003, in PBMC, saliva, urine and semen in studies C0371002, C0371005 and C0371003. This covers 15 subjects treated in the Phase 1/2a trial C0371005 and 45 patients treated in the pivotal Phase 3 study C0371002 with actual titre dosing.

In study C0371005, vector shedding has been assessed using qPCR analysis of peripheral blood mononuclear cells (PBMC), saliva, urine, semen, and serum. The levels of shed vector were highest during the first two weeks after vector infusion reaching 4.03 x 10^5 (minimum high 4.80 x 10^3) copies/ug in PBMC, 7.13 x 10^5 (minimum high 1.63 x 10^4) copies/mL in saliva, 3.57 x 10^3 (minimum high: Below LOQ) copies/mL in urine, 3.61 x 10^5 (minimum high: Below LOQ) copies/mL in semen and 2.91 x 10^6 (minimum high 6.55 x 10^4) copies/mL in serum. The levels gradually declined in all types of specimens. Full clearance was defined as having 3 consecutive negative samples as measured by qPCR. All but 3 subjects reached full clearance in all specimen types by the end of the study, however all subjects had reached first negative value by the end of the study. All 3 subjects subsequently met criteria for full clearance when tested in the long-term follow-up study (Study C0371003). In general, PBMCs were the slowest sample to clear ranging from 17 weeks to not cleared by end of study. Ranges for clearance of the other samples were: saliva 3-8 weeks, urine 1-7 weeks, semen 1-12 weeks, and serum 3-22 weeks.

In the 45 patients treated as part of the pivotal Phase 3 trial C0371002, assessment of vector shedding showed peak levels of vector DNA were achieved within the first two weeks after infusion and the highest vector DNA concentrations were found in plasma. Vector DNA declined to undetectable levels in plasma, saliva and semen within 1-4 months after infusion and PBMC was slowest to clear to undetectable levels within 7 months. In urine, peak vector DNA concentration was very low and declined to undetectable levels within 3 weeks after infusion. Full clearance, defined as 3 consecutive negative results (below quantification limit) in a particular matrix, was observed in all participants. In general, PBMCs were the slowest matrix to clear with a mean (±SD) and median (min, max) time to last undetectable vector of 163.3 (109.44) and 130 (39.0, 531.0) days in the 45 participants included in the Dosed Analysis Set. Regarding clearing from other samples time to clearance with a mean (±SD) and median (min, max) time to last undetectable vector were in plasma 98.6 (54.69) days and 93.0 (30, 317) days; semen 41.1 (20.09) days and 35.0 (15, 104) days, saliva 42.4 (17.23) days and 37.0 (29, 105) days and urine 20.4 (13.26) days and 21.0 (4, 87) days.

To further characterise the shed material, saliva, semen, and urine samples from a subset of 17 patients in Study C0371002 were tested using nuclease treatment (MNase) prior to DNA extraction. Nuclease treatment digests the free floating vector DNA so it cannot be quantified, ensuring the material being quantified following digestion is only encapsulated viral DNA. After nuclease treatment and subsequent DNA extraction, the amount of fidanacogene elaparvovec was measured by qPCR. In saliva, mean concentrations were similar up to week 2 between the MNase treatment and without MNase treatment subgroups, while all participants had concentrations BQL by week 9. In semen, mean concentrations were approximately 33% lower in the MNase treatment subgroup until week 3, and BQL for all participants by week 11. In urine, mean concentrations were approximately 30% lower in the MNase treatment subgroup until 72 h post infusion and were BQL for all participants by week 2.

Across studies (C0371005/C0371003 and C0371002, total n= 60 patients) highest peak vector DNA concentrations were found in serum/plasma compared to the other liquid matrices (saliva, urine, semen). In plasma (measured only in C0371002), mean peak vector DNA concentration of 2.008 × 10^9 vg/mL

was observed which was the highest concentration observed in all the matrices. Vector DNA fully cleared in serum, plasma, saliva, and semen within a mean of 1 to 4 months after infusion and PBMC was slowest fluid to full clearance within a mean of 12 months. In urine, the peak vector DNA concentration was very low relative to plasma and declined to full clearance within a mean of 4 weeks after infusion. The maximum observed time for vector DNA full clearance in saliva, urine and semen were 105 days, 87 days and 154 days, respectively. Information on vector shedding has been included in the SmPC section 5.2.

Intrinsic factors impacting fidanacogene elaparvovec dosing exist, addressed in the following sub-sections.

Special populations

Impaired renal function

No clinical study was conducted to evaluate the impact of renal impairment on FIX:C upon fidanacogene elaparvovec administration, adequately reflected in the SmPC.

Impaired hepatic function

No clinical study was conducted to evaluate the effect of hepatic impairment on FIX:C upon fidanacogene elaparvovec administration, adequately reflected in the SmPC section 4.2 with cross references to section(s) 4.4 and 4.3.

Based on the mechanism of action, an impact of hepatic impairment is expected on both safety and efficacy. Consequently, absence of significant liver disease is to be confirmed prior to treatment (SmPC section 4.2), and relevant hepatobiliary diseases are listed as contraindication.

Gender

Fidanacogene elaparvovec has not been studied in women. A respective statement is included in SmPC section 4.6.

Weight

Dosing instructing in the SmPC depends on the patient' s BMI, with actual patient's weight only been accounted for up to BMI $\leq$ 30 kg/m^2. For patients with higher BMI, the dose is to be calculated assuming the patient had a BMI of 30 kg/m^2, thus the effective dose is reduced. Extrapolating from clinical study patient population, a relevant number of patients can be expected to have a BMI above 30 kg/m² in the commercial setting. Efficacy and safety findings for this subgroup are included and discussed in the respective section of this document.

Elderly

The inclusion criteria of the C0371005 study did not define an upper age limit, however the C0371002 study defined an upper age limit of 65 years of age. In study C0371005, patients aged 18-61 years, in study C0371002 patients aged 18-62 years were treated.

The SmPC includes the statement that safety and efficacy of fidanacogene elaparvovec in patients $\geq$ 63 years old have not been established.

2.6.2.2. Pharmacodynamics

Fidanacogene elaparvovec aims at endogenous coagulation factor IX (FIX) expression in hepatocytes of patients with inherited deficiency of FIX (haemophilia B). EMA/CAT/80183/2014 clarifies that on a case

by case basis, pharmacokinetics studies need to be carried out depending on the specific GTMPs, e.g. if the gene product is a protein excreted in the blood circulation. Thus, the transgene expression and activity, i.e. kinetics of FIX expression and FIX:C (over time) are relevant PD parameters. Plasma levels of the induced FIX activity are investigated throughout the clinical development programme. Also, immunogenicity of fidanacogene elaparvovec has been addressed in the clinical development programme.

Analytical methods and validation results were provided.

Mechanism of action

Fidanacogene elaparvovec is a gene therapy designed to introduce a functional copy of the high activity Padua variant of the factor IX gene (FIX-R338L) in the transduced cells to address the monogenic root cause of haemophilia B.

Fidanacogene elaparvovec is a non-replicating recombinant AAV vector that utilises AAVRh74var capsid to deliver a stable human factor IX transgene. AAVRh74var capsid is able to transduce hepatocytes, the natural site of factor IX synthesis. The factor IX gene present in fidanacogene elaparvovec is designed to reside predominately as episomal DNA within transduced cells and expression of the transgene is driven by a liver specific promoter, which results in tissue specific, continuous and sustained factor IX protein expression.

Primary and Secondary pharmacology

Factor IX protein concentration is captured under the secondary objective *to characterize the kinetics of PF-06838435* as **FIX antigen** levels in studies C0371005 and C0371003, and under the tertiary/exploratory objective *Pharmacodynamics of PF-06838435* in the pivotal study C0371002, respectively. The data provided is incomplete as exogeneous factor IX substitution pre-gene therapy resulted in invalid measurements. Moreover, the presence of nonfunctioning pre-gene therapy endogenous FIX as cross-reacting material confounds the assessment of expressed transgene FIX.

FIX activity (FIX:C) is captured under the secondary objective to characterize the kinetics of PF-06838435 in study C0371005, under the secondary objective to determine the durability of transgene expression of PF-06838435 in study C0371003, and under the key secondary objective to demonstrate efficacy of PF-06838435 in the pivotal study C0371002, respectively. FIX:C data confirmed an increase post-treatment compared to baseline levels, which are defined as ≤ 2% via eligibility criteria. Thus, FIX:C confirmed a pharmacologic effect of fidanacogene elaparvovec, for both the phase 1/2a and pivotal phase 3 study. The same BMI-dependent fidanacogene elaparvovec target dose of $5x10^{11}$ vg/kg resulted in higher mean steady state FIX:C (measured by ActinFSL OSA) in the phase 1/2a study compared to the pivotal study.

As commonly observed for recombinant FIX products and other approved AAV-based gene therapies for haemophilia, there is a relevant FIX:C assay discrepancy to determine transgene derived FIX:C. An international multi-center field study was conducted to determine the FIX activity levels across different one-stage assay reagents and chromogenic substrate assays. The field study found that transgene derived FIX:C can be measured with the commonly used one-stage and chromogenic assays in clinical laboratories, which is a relevant information also included in the SmPC. Consistently higher FIX activity was observed for the silica-based OSA compared to an ellagic acid-based OSA or the chromogenic one, both in the field study and the pivotal study. In the pivotal study, numerically, the ellagic acid-based OSA and the CSA reported comparable ranges, but patient-level data indicate inconsistent patterns.

The **immunogenicity** evaluation included humoral (FIX inhibitor, ADA and nAb to AAVRh74var capsid) and cellular immune responses (T-cell responses against AAVRh74var capsid, FIX-R338L, wild-type FIX,

FIX-A148T polymorphism, and lambda phage sequences). No development of FIX inhibitors (anti-factor IX antibodies) was observed post-infusion in clinical studies. All participants were ADA/nAb negative at baseline prior to infusion. A sustained increase in nAb was observed in all participants with nAb assessment after administration of fidanacogene elaparvovec that participated in clinical studies, and titers remained high for the duration of follow-up, at least up to 6 years. In the Phase 3 clinical study, ADA were detected in 95.1% of participants at 1 year post-fidanacogene elaparvovec infusion.

2.6.3. Discussion on clinical pharmacology

The Guideline on the quality, non-clinical and clinical aspects of gene therapy medicinal products (EMA/CAT/80183/2014) specifies that classical pharmacokinetic studies based on absorption, distribution, metabolism and excretion (ADME) studies are usually not required for GTMPs.

Due to the nature and administration route of fidanacogene elaparvovec, the lack of dedicated clinical PK studies is therefore acceptable. The release of the vector outside the body via excreta (shedding) is relevant for ERA and discussed there. The applicant proposed a BMI adapted dosing.

The pharmacodynamic effect of fidanacogene elaparvovec is the induction of relevant plasma levels of FIX, which are expected to alleviate the coagulatory deficiency of the haemophilic target population. Quantification of transgene protein (FIX antigen) via a commercially available ELISA is not suited to substantiate an induction of transgene protein expression. No respective warning is included in the SmPC as "FIX antigen quantification for everyday clinical practice may not be relevant".

Monitoring FIX activity (FIX:C) to reflect PK/PD was agreed as acceptable strategy in an EMA scientific advice (EMEA/H/SA/3416/1/2016/ADT/III). Transgene FIX:C was determined using a single one-stage clotting assay (OSA) in the phase 1/2a study, and determined using the same OSA as well as an additional OSA and one chromogenic assay (CSA) in the pivotal trial. Submitted data for FIX:C confirmed an increase post-Fidanacogene elaparvovec in the absence of exogenous FIX as measured by all respectively used assays.

FIX:C is also considered as crucial endpoint for efficacy in the pivotal trial (EMEA/H/SA/3416/1/FU/1/2017/SME/ADT/PR/II). In this context, it is noted that FIX:C means reported for the phase 3 are substantially lower than the respective values reported for the phase 1/2a. The applicant has reviewed the data from multi-factorial aspects including drug substance manufacturing process, study design and population, management of presumed cellular immune responses, and statistical handling of participants resuming prophylaxis in analysis. It was concluded that the underlying reason for the observed differences could be due to inherent differences between the two studies. Consequently, phase 1/2a data is not included in the SmPC.

Previous findings on FIX:C assay discrepancy were also confirmed for fidanacogene elaparvovec transgene protein, further assessed in a global, multicenter field study. Consistently higher FIX activity was observed for the silica-based OSA compared to an ellagic acid-based OSA or the chromogenic one. Disregarding the silica-based OSA for efficacy claims is a conservative approach, however, the relevance of either assay with regard to the assumed bleeding risk is unknown (standard plasmatic coagulation factors do not give these discrepant results) and thus the interpretation of the clinical relevance especially of low FIX:C is severely compromised. Numerically, the ellagic acid-based OSA and the CSA reported comparable ranges, but patient-level data indicate inconsistent patterns, precluding giving a conversion factor in the SmPC.

Neutralizing, pre-existent antibodies are discussed in the context of in vitro biomarker tests for patient selection, and immunogenicity is presented and discussed in the safety section.

2.6.4. Conclusions on clinical pharmacology

The clinical pharmacology as presented in the dossier reveals transgene specific particularities regarding FIX monitoring, i.e. inability of a commonly used FIX antigen assay to quantify the expressed transgene protein, and FIX:C assay discrepancies. The latter is adequately reflected in the SmPC by presenting data from all three assays. The BMI adapted dosing has the potential to impact the benefit risk ratio and is discussed accordingly, see clinical efficacy, clinical safety and B/R sections.

The product can be approved on pharmacology grounds.

The CHMP endorse the CAT assessment regarding the conclusions on the Clinical pharmacology as described above.

2.6.5. Clinical efficacy

2.6.5.1. Dose response studies

At time of MAA, no formal dose finding study has been completed. A dose-escalating substudy is ongoing, and it is included in the dossier to support safety only.

C0371005

This completed Phase 1/2a study was an open label study of fidanacogene elaparvovec in participants with hemophilia B. The primary objective was to evaluate the safety and tolerability of a single IV infusion of PF-06838435 in haemophilia B participants ≥18 years of age with ≤2 IU/dL [≤2%] FIX.

C0371005 was designed as dose-finding study to evaluate up to three ascending doses, but only the lowest dose $5x10^{11}$ vg/kg had been tested in a total of n=15 patients following DMC recommendation to expand the dose cohort based on observed (safety and) efficacy data. The first ten patients received manufacturing process 1 material, the remaining five patients manufacturing process 2 material. The recommended phase 3 and proposed commercial dose is based on this study.

After completion of the 52 weeks on-study follow-up, study participants were invited to enrol into the long-term follow up study C0371003.

C0371003

C0371003 is the respective long-term follow-up study for the phase 1/2a study C0371005, and ongoing. (Partially) updated data with cutoff dates 15-Aug-2023 and 10-Jan-2024 have been submitted. At time of August 2023 data cutoff, 7 out of 14 participants had completed a combined 6 years of follow-up in C0371005/C0371003. 4 out of 14 participants were still ongoing with a minimal follow-up of 5.4 years. 3 participants discontinued (1 lost to follow-up and 2 withdrew by subject).

Per protocol amendment 2, an additional dose-escalation substudy investigating up to four additional dose levels and prophylactic immunosuppression was implemented. The included rationale was "the mean steady-state FIX activity achieved in the C0371005 study at a dose of 5×10^{11} vg/kg was 22.9% which is below the normal range for FIX and suggests there is room to increase the dose to achieve additional efficacy for participants with hemophilia B" (excerpt C0371003 Final Protocol Amendment 3, 15 May 2023).

At time of January 2024 data cutoff, seven previously gene-therapy untreated patients had been treated, with limited data available for [redacted] vg/kg ([redacted]) and [redacted] vg/kg ([redacted]). Two of cohort 2 and one of

cohort 3 patients have completed one year of follow-up and entered the long-term follow-up portion of the trial, while the other are still ongoing in the first year follow-up.

Parameter (Unit)	Observation period		Cohort_1 (5×10^{11} vg/kg)	Cohort_2 ([redacted] vg/kg)	Cohort_3 ([redacted] vg/kg)
Factors IX Activity (%)	Year 1	N	14	[redacted]	[redacted]
		Mean (SD)	23.42 (10.296)		

Mean (SD) FIX activity via central lab Actin-FSL assay using geometric means from each participant during Year 1. Data cut 15-Aug-2023 for cohort 1, and 10-Jan-2024 for cohorts 2 and 3.

The possibility of a dose-response relationship cannot be rejected, but due to the sample size it can also not be confirmed.

2.6.5.2. Main studies

For efficacy assessment, in the pivotal study C0371002 an intra-patient control approach was chosen, with respective baseline data for bleeding frequency and FIX consumption having been generated in the non-interventional study C0371004 by participants recording their use of standard of care FIX replacement therapy and bleeding episodes in their dedicated e-diary.

C0371004: *A Study to Evaluate Prospective Efficacy and Safety Data of Current FIX Prophylaxis Replacement Therapy in Adult Hemophilia B Subjects (FIX:C ≤ 2%) or Current FVIII Prophylaxis Replacement Therapy in Adult Hemophilia A Subjects (FVIII:C ≤ 1%)*

This is an ongoing Phase 3 open label, non-investigational product lead-in study to evaluate prospective efficacy and select safety data of current FIX (Hemophilia B Cohort) or Factor VIII (Hemophilia A Cohort) prophylaxis replacement therapy in the usual care setting of moderately severe to severe adult hemophilia B participants (FIX:C ≤ 2%) who are negative for neutralizing antibodies (nAb) to AAV-Spark100 or moderately severe to severe adult hemophilia A participants (FVIII:C ≤ 1%) who are negative for nAb to SB-525 capsid (AAV6). Only the Hemophilia B Cohort is relevant for MAA.

A total of 316 participants were screened, 204 (64.6%) failed the screening process, for 10 screening was still ongoing. 102 (32.3%) participants completed screening, of which all (100%) entered the lead-in data collection phase. Of them 59 (57.8%) participants completed the study and 3 (2.9%) participants were discontinued. As of the data cutoff date 02 November 2022, 40 (39.2%) participants were ongoing in the non-interventional lead-in study. With the responses to the D120 LoQ, it was clarified that study C0371002 was amended, dosing additional patients to obtain clinical experience using nominal titre dosing. At the time of 26 Oct 2023 cutoff date, 20 participants remained ongoing in C0371004, awaiting a protocol amendment and subsequent screening into C0371002. With the responses to the D180 LoQ the applicant clarified they either already had been dosed in trial C0371002, were screen failures, or were expected to be dosed provided eligibility criteria were met (data cut 10-Apr-2024). The last C0371004 participant was expected to be dosed in trial C0371002 in Q3 2024.

Of 59 patients who completed study C0371004 at November 2022 cutoff, 51 were screened for study C0371002. Of these, 5 were screen failures, and one patient withdrew consent. Based on ABR_{total}, AIR and annualized FIX consumption (IU/kg) data provided for C0371004 completers who did continue to the pivotal study C0371002, and patients who discontinued study C0371004 or completed, but did not continue to study C0371002, a selection bias favoring fidanacogene elaparvovec treatment is not assumed.

C0371002: *A Phase 3, open label, single arm study to evaluate efficacy and safety of FIX gene transfer with PF-06838435 (rAAV-Spark100-hFIX-Padua) in adult male participants*

with moderately severe to severe hemophilia B (FIX:C≤2%) (BeneGene-2), protocol C0371002

Study C0371002 is a single-arm, open-label, multi-site, single-dose phase 3 study in adult male haemophilia B patients with endogenous factor IX activity of ≤2%, comparing the efficacy of a single intravenous infusion of fidanacogene elaparvovec with routine FIX prophylaxis in adult male participants from the lead-in study (C0371004). This study includes a total of 6 years of follow up post fidanacogene elaparvovec infusion. The study is ongoing; at time of initial submission with 16 November 2022 data cut, n=45 patients had been enrolled and treated with the actual titre dosing, among which 41 patients had completed 15 months of follow-up. All 45 patients were included in the primary analysis regardless of their follow-up time.

Updated data was provided with data cut on 30 Aug 2023: 41 out of 45 participants had completed 24 months of follow up. 44 participants had completed 15 months of follow up and the 45th participant was followed for 437 days (over 14 months).

At the cutoff date of 10-Apr-2024, 27 patients have been dosed in C0371002 with nominal titre dosing. As of 26-Oct-2023, none has been available for the primary endpoint. Clinical data is not included in this document due to prematurity.

Methods

- **Study Participants**

Adult male participants with moderately severe to severe hemophilia B (FIX:C≤2%), who must have completed at least 6 months of routine FIX prophylaxis therapy during the lead-in study (C0371004). Anti-AAV-Spark100 neutralizing antibodies (nAb) titer ≥1:1, prior or current history of inhibitor to FIX, and significant liver disease were exclusionary.

- **Treatments**

A single intravenous dose of fidanacogene elaparvovec at a dose of $5x10^{11}$ vg/kg for patients with BMI up to 30 kg/m² (inclusive). For a participant with BMI >30 kg/m^2, dose is to be calculated based on an adjusted body weight determination that assumes a maximum permissible BMI of 30 kg/m².

FIX prophylaxis regimen was to be suspended after infusion of fidanacogene elaparvovec, with FIX replacement therapy being allowed, as needed.

- **Objectives**

Primary

- To demonstrate the efficacy of a single infusion of fidanacogene elaparvovec in male participants ≥18 years of age with moderately severe to severe hemophilia B (FIX:C ≤2%) by demonstrating non-inferiority on **ABR for total bleeds** (treated and untreated) from Week 12 to Month 15 versus standard of care FIX prophylaxis replacement regimen

Key secondary

- To demonstrate the efficacy of fidanacogene elaparvovec in terms of the use of **AIR** of exogenous FIX from Week 12 to Month 15 versus AIR of FIX with standard of care FIX replacement regimen pre-IP infusion, the non-inferiority on ABR for treated bleeds from Week 12 to Month 15 versus standard of care FIX prophylaxis replacement regimen, treated bleeds, and vector-derived **FIX:C** level at steady state (from Week 12 to 15 months) demonstrated to be greater than 5%.

Secondary

- To compare additional efficacy parameters post- fidanacogene elaparvovec infusion to baseline in order to further characterize fidanacogene elaparvovec treatment, including annualized FIX

consumption from Week 12 to Month 15 versus standard of care FIX prophylaxis replacement regimen

- Assess durability of efficacy up to 6 years.

- **Outcomes/endpoints**

Endpoints and definitions	Primary Endpoint	ABR_{total}	Non-inferiority on ABR for total bleeds (treated and untreated) from Week 12 to Month 15 versus usual care FIX prophylaxis replacement regimen, comparing pre- and post-IP infusion.
	Secondary Endpoint	AIR	AIR of exogenous FIX from Week 12 to Month 15 post-IP infusion versus AIR of FIX with usual care FIX replacement regimen pre-IP infusion
	Secondary Endpoint	FIX:C	Vector-derived FIX:C level at steady state (from Week 12 to 15 months post-IP infusion) demonstrated to be greater than 5%. Descriptive summary of FIX:C by study visit.
	Secondary Endpoint	Annualized FIX Consumption	Annualized FIX consumption reported as international units per kilogram (IU/kg) and total units measured from Week 12 to Month 15 post-IP infusion versus pre-IP infusion.
	Secondary Endpoint	Hemophilia Joint Health Score	Improvement of total score at Week 52 and Week 104 post-IP infusion versus baseline.

- **Sample size**

The study sample size is constrained by the non-inferiority analysis of the primary endpoint, ABR. A sample size of n=40 patients having completed at least 15 months of follow-up post-infusion were assumed to provide at least 90% power (one-sided test with alpha=0.025) to demonstrate non-inferiority of gene therapy compared to prophylaxis treatment on the difference in ABR_{total}, using a repeated measure negative binomial regression, with a noninferiority margin of 3.0 bleeds/year. The non-inferiority margin was chosen according to the CHMP guidance for the design of prophylaxis clinical trials and based on a switch from on-demand to prophylaxis in a single arm trial using a paired comparison and calculated via simulation. The assumed background rate of annualized bleeding is a conservative 5.0 and the assumed annualized bleeding rate post-PF-06838435 is 1.5.

- **Randomisation and Blinding (masking)**

Not applicable; this is a non-randomised open-label single arm study.

- **Statistical methods**

Planned methods of analyses were provided in the statistical analysis plan (SAP), which was finalized before performing the data cut for the primary analysis. The SAP was amended 5 times.

Primary Endpoint(s) / Primary Estimand(s)

ABR for total bleeds (treated and untreated bleeds from Week 12 to Month 15) will be compared to pre-infusion of PF-06838435 (under SOC FIX prophylaxis replacement regimen). A repeated measure negative binomial regression model will be used to do the hypothesis test on non-inferiority with one-sided test with the specified α. If the non-inferiority on ABRtotal is established, subsequently testing for superiority will be conducted.

In the original protocol, ABR and FIX from Week 12 to month 12 were defined as co-primary endpoints.

In protocol amendment 2, the Sponsor expanded the analysis period from 12 months post-treatment to 12 months post steady state (corresponding to 15 months post-treatment), clarified that primary endpoint of ABR is for treated bleeds, and added ABR for total bleeds (treated and untreated) as a key secondary objective and endpoint.

In protocol amendment 3, the Sponsor adjusted the ordering of endpoints to align with FDA recommendations (received 28 Mar 2022). The primary endpoint was revised from ABR for treated bleeds to ABR for total bleeds (treated and untreated). ABRtreated and FIX:C were moved from co-primary to key secondary endpoints.

Handling of Intercurrent Events

If prophylaxis FIX regimen is resumed for a participant, then the time period following the resumption of the prophylaxis regimen will be excluded from the ABR endpoint calculation, which means the bleeding events will be excluded and the time period of observation will be deducted as well.

Supplementary estimands were specified were subjects who dropped out/withdrew or resumed prophylaxis were imputed by jump to reference (baseline value of ABR for that subject) or regardless of resumption of prophylaxis. Upon request a tipping point analysis was conducted as well, in which subjects who resumed prophylaxis or withdrew due to a lack of efficacy was imputed after that point with an ABR of X for the remaining observation time. This analysis showed that these patients would need to have an ABR of 17 (18 with updated data) for superiority in ABR to no longer hold true, and 55 (56 with updated data) for non-inferiority to no longer hold true.

Secondary Endpoints / Secondary Estimand(s)

- ABR for treated bleeds from Week 12 to Month 15 will be compared to pre-infusion of PF-06838435 (under SOC FIX prophylaxis replacement regimen) using a repeated measure negative binomial regression, with a non-inferiority margin of 3.0 bleeds/year.
- AIR for FIX from Week 12 to Month 15 versus AIR during SOC FIX replacement regimen will be tested for superiority. Paired T-test will be used to do this hypothesis testing.
- Vector-derived steady state FIX:C (Week 12 to Month 15) will be compared to a threshold of 5% using a one-sided, one sample T-test. FIX:C will be descriptively summarized by study visit.

The steady state FIX:C is calculated for each participant as the geometric mean of all eligible FIX:C measures from 12 weeks [Day 82] to Month 15 [Day 469] inclusive after PF-06838435 infusion. If a participant withdrew consent, dropped out early from the study or resumed FIX prophylaxis, then the assessments at the visits following withdrawal/dropout/resumption will be imputed as 1.9%. With the responses to the D120 list of issues, the applicant provided an additional analysis where subjects with severe hemophilia were imputed as 0.9% and subjects with moderately severe as 1.9%.

Sensitivity analyses were performed to address the impact of intercurrent events, missing data and imputation for primary and key secondary endpoints.

Multiplicity

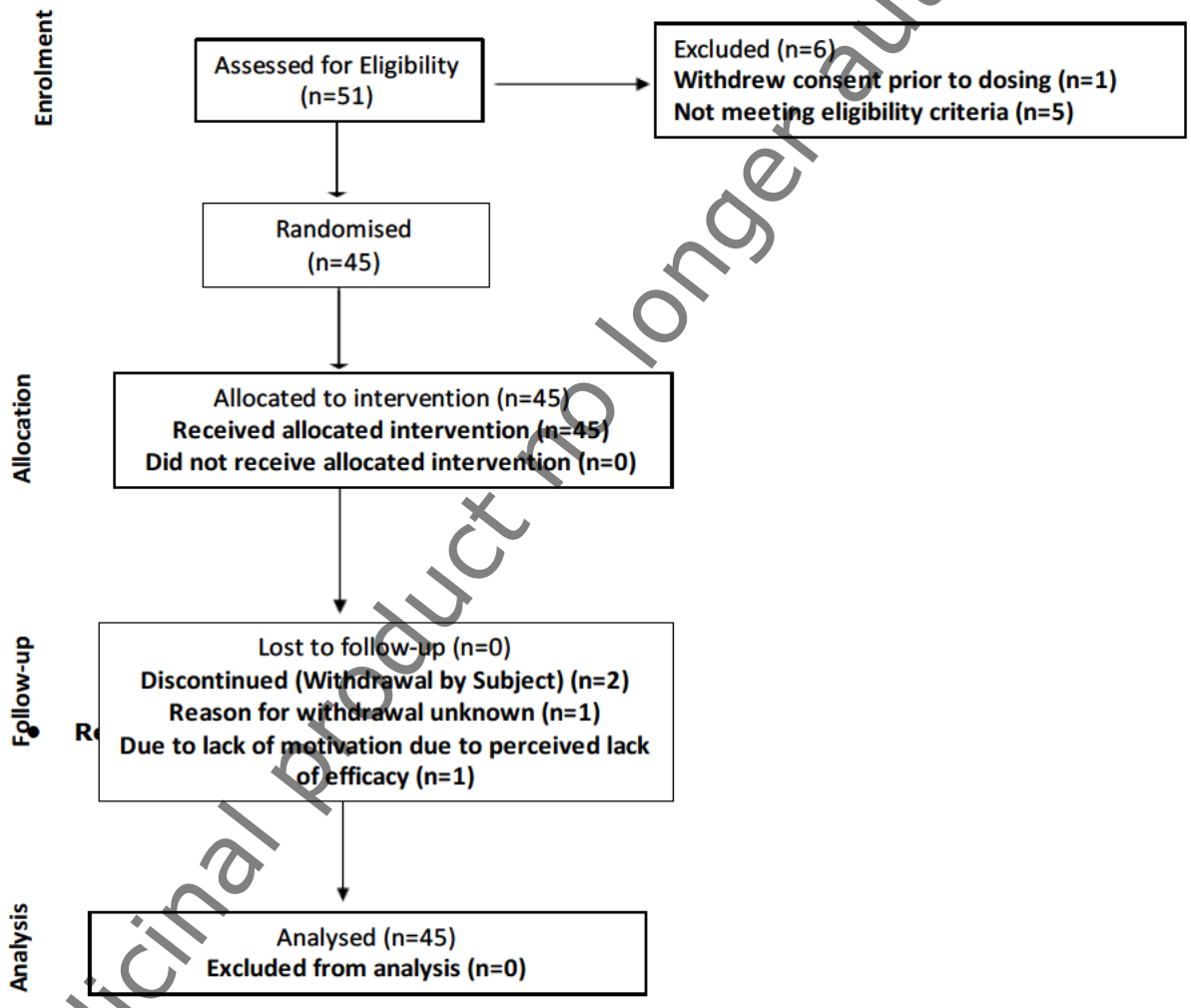
The primary and secondary endpoints were tested hierarchically in a prespecified sequence at two-sided 0.05 alpha level. The sequence was ABR_{total} (non-inferiority to FIX prophylaxis regimen), ABR_{treat} (non-inferiority to FIX prophylaxis regimen), AIR (superiority to FIX prophylaxis regimen), Steady state FIX:C (> 5%), FIX consumption (superiority to FIX prophylaxis regimen), ABR_{treat} (superiority to FIX prophylaxis regimen), ABR_{total} (superiority to FIX prophylaxis regimen), Haem-A-QoL Physical health domain (superiority over baseline), HAL Complex Lower extremity Activities (superiority over baseline), and ABR_{total} of specific types (non-inferiority to FIX prophylaxis regimen and HJHS).

Interim Analyses

One interim analysis was planned in the original protocol with the aim of early regulatory submission in case of compelling efficacy and safety analysis. The interim analysis was planned after 20 of the 40 participants had at least 15 months of follow up (until SAP v3 12 months of FU). An O'Brien-Fleming alpha spending approach was planned to be used. The IA was removed in protocol amendment 2 (SAP version 5). No interim analyses were conducted. However, the applicant planned to conduct unblinded reviews of the data during the course of the study for the purpose of safety assessment.

Results

- **Participant flow**



Patients were recruited from 28 sites in Asia-Pacific, Australia, Europe, Middle East, North America, and South America; 28.9% of the study population was recruited in Europe. Primary completion date was 16 November 2022.

The 20 patients dosed with nominal concentration at data cut 26 October 2023 were recruited in APAC, Australia, Middle East, and South America.

- **Conduct of the study**

There were three global amendments to the study protocol. Several substantial revisions have been made to the protocol impacting interpretation of the generated data, the most important ones the revision of the efficacy evaluation time period in both amendment 2 and 3, introduction of the eventually primary endpoint ABR_{total}, with amendment 2, and the revision of primary and key secondary endpoints with the latest protocol amendment 3. Both revision of evaluation period for the primary and (key) secondary endpoints as well as said endpoints were adjusted based on input from US FDA. Changes were also made repeatedly to reflect the COVID-19 pandemic.

In protocol amendment 4 dated January 2023, the sponsor added approximately 10 patients dosed at nominal concentration, which was increased to 20 in protocol amendment 5 dated May 2023. With the responses to the D180 LoQ the applicant indicated up to 42 patients might be dosed at nominal concentration. The protocol date of the final protocol is later than treatment of the last patient treated with actual titre concentration dosing.

- **Baseline data**

All participants were male and the majority of the study participants dosed with actual titre concentration were <35 years of age, and not Hispanic or Latino or Spanish origin (35 [77.8%] participants). All (45 [100.0%]) participants had a factor mutation. The BMI ranged from 17.6-48.4 kg/m².

- **Numbers analysed**

For the primary analysis, of the 51 participants screened, 45 (88.2%) were included in the **Dosed/Safety Analysis Set**. Of these, 41 (80.4%) had 15 months of follow-up and were included in the **Evaluable Analysis Set**.

- **Outcomes and estimation**

The primary analysis was based on the Dosed Analysis Set (n=45) at the primary completion date of 16 Nov 2022. An updated analysis based on 45 subjects was submitted upon request with an updated cut-off of 30 Aug 2023. The results were in line with the previous results.

ABR_{total}

C0371002 met its primary efficacy objective of non-inferiority of ABR_{total} post infusion of fidanacogene elaparvovec (Week 12 to Month 15) compared to FIX prophylaxis, and in secondary analysis ABR_{total} was demonstrated to be superior post infusion of fidanacogene elaparvovec compared to FIX prophylaxis.

A repeated measures GLM with negative binomial distribution was utilized to model the bleed count data. The model-derived meanABR_{total} was 1.30 (95% CI: 0.59, 2.02) versus 4.43 (95% CI: 1.81, 7.05), treatment difference was -3.13 (95% CI: -5.44, -0.81) and percentage reduction ABR_{total} was 70.63% (95% CI: 49.17%, 83.03%).

The ABR_{total} for participants who were dosed at least 15 months prior to the data cutoff date but discontinued/resumed FIX prophylaxis prior to completing 15 months of follow-up were included in the analysis with the ABR_{total} during the time period between discontinuation/FIX resumption and Month 15 excluded.

At the updated cut-off, the model-derived mean ABR_{total} during the primary efficacy evaluation period (PEP: from Week 12 to Month 15 post fidanacogene elaparvovec infusion) was 1.28 (95% CI: 0.57, 1.98) compared to ABR_{total} in the pre-infusion period (4.42, 95% CI: 1.80, 7.05). The treatment difference in ABRtotal was -3.15 (95% CI: -5.46, - 0.83). This is consistent with the findings at the primary completion date (PCD, 16 Nov 2022).

Regarding longer-term effects, updated data for ABR_{total} (model derived estimate [95% CI]) was 0.40 (0.05, 0.76) for year 2 (n=44), 0.564 (0.04, 1.24) for year 3 (n=40), and 0.32 (-0.04, 0.68) for year 4 (n=15). Over time, 27 out of 45 patients (60%) remained without bleeds in the observation period.

For presentation in the SmPC, presentation of the ABR_{total} including bleeding events which occurred post-resumption of prophylaxis (data cut-off 30 Aug 2023) was chosen, as it is considered to most closely reflect a treatment policy estimand. In addition, in line with the SmPCs of other gene therapy products for treatment of haemophilia the SmPC also details the ABR for treated bleeds only (ABR_{treat}), and the ABR_{total} for spontaneous bleeding, joint bleeding, and target joint bleeding.

AIR

Mean (SD) AIR from Week 12 to Month 15 post fidanacogene elaparvovec infusion was significantly reduced to 4.46 (10.028) from a mean AIR of 58.83 (29.056) in pre-infusion period. Twenty-nine out of 45 (64.4%) participants required no infusions of FIX from Week 12 to Month 15 after infusion with fidanacogene elaparvovec.

The mean AIR from Week 12 to Month 15 post fidanacogene elaparvovec infusion at the updated data-cut was reduced to 4.54. This is consistent with the findings at PCD. The percent reduction in mean was 92.3. Twenty-nine out of 45 (64.4%) participants required no infusions of FIX from Week 12 to Month 15 after infusion with fidanacogene elaparvovec.

Regarding longer-term effects, updated AIR (mean [±SD]) was 6.52 (18.697) for year 2 (n=44), 4.9 (14.871) for year 3 (n=40), and 1.4 (4.691) for year 4 (n=15).

Annualized FIX consumption

The annualized total FIX consumption (mean [±SD]) from Week 12 to Month 15 post-fidanacogene elaparvovec infusion was reduced to 235.04 (538.977) IU/kg compared to 3170.74 (1634.753) IU/kg during FIX prophylaxis in the Dosed Analysis Set. The treatment difference was -2935.70 (95% CI -3403.10, -2468.30) p<0.0001). The percent reduction in mean was 92.6.

The updated annualized total FIX consumption (mean [±SD]) from Week 12 to Month 15 post fidanacogene elaparvovec infusion was reduced to 239.39 (539.617) IU/kg. The treatment difference was -2929.17 (95% CI -3397.49, -2460.85). The percent reduction in mean was 92.4. This is consistent with the findings at PCD.

The updated mean (±SD) annualized FIX consumption reported at year 2 (n=44) post-fidanacogene elaparvovec infusion a reduction to 301.34 (852.206) IU/kg, at year 3 (n=40) a reduction to 2169.01 (570.946) IU/kg, and at year 4 (n=15) a reduction to 230.51 (498.669) IU/kg.

FIX:C

Mean steady-state (geometric mean of all valid measurements from Week 12 to Month 15 post fidanacogene elaparvovec infusion) FIX:C from all 3 assays were assay dependent: Mean [±SD] in the one-stage assay with Actin-FSL reagent: 12.62 [8.92]; in the one-stage assay with SynthAsil reagent: 25.90 [16.89]; and in chromogenic assay: 13.49 [10.40]. The primary analysis for FIX:C was performed with imputation.

Imputations for baseline and postresumption FIX:C activity according to participant's baseline disease severity were implemented with the latest data cut. In addition, change from baseline was summarized and analysed using repeated measures linear mixed effect model (MMRM) with participant as the random effect, and study visit as a fixed effect. The results are presented in the table below.

Table 5. C0371002 study: factor IX activity over time by assay

				Change from baseline[$]		
Visit	**n**	**Mean (SD)**	**Median (min, max)**	**LS mean (SE)[^]**	**95% CI[^]**	**One-sided p-value[^]**
One-stage assay (SynthASil reagent)[*]						
Week 12	44	27.79 (15.226)	26.45 (3.2, 68.6)	26.63 (2.671)	(21.39, 31.87)	< 0.0001
Month 6	39	27.64 (21.373)	23.20 (0.9, 99.7)	26.25 (2.679)	(21.00, 31.51)	< 0.0001
Month 15	39	26.17 (25.100)	22.50 (0.9, 119.0)	24.70 (2.678)	(19.44, 29.95)	< 0.0001
Month 24	39	26.47 (25.092)	22.90 (0.9, 123.4)	24.66 (2.688)	(19.38, 29.93)	< 0.0001
Month 36	13	23.83 (19.165)	21.80 (0.9, 74.8)	25.47 (3.021)	(19.54, 31.40)	< 0.0001
One-stage assay (Actin FSL reagent)						
Week 12	44	13.58 (8.047)	13.58 (1.7, 35.1)	12.53 (1.806)	(8.99, 16.08)	< 0.0001
Month 6	41	13.08 (11.170)	10.10 (0.6, 55.0)	11.93 (1.808)	(8.38, 15.47)	< 0.0001
Month 15	39	13.96 (15.403)	10.20 (0.9, 69.8)	12.57 (1.810)	(9.02, 16.12)	< 0.0001
Month 24	38	15.70 (16.392)	12.85 (0.9, 87.3)	13.81 (1.818)	(10.24, 17.37)	< 0.0001
Month 36	13	14.57 (12.473)	12.50 (0.9, 47.6)	16.88 (2.049)	(12.86, 20.90)	<0.0001
Chromogenic assay						
Week 12	44	13.91 (9.302)	12.05 (1.4, 36.3)	12.78 (1.561)	(9.71, 15.84)	< 0.0001
Month 6	40	14.81 (12.988)	10.30 (0.9, 57.7)	13.04 (1.569)	(9.96, 16.12)	< 0.0001
Month 15	38	15.19 (16.647)	10.00 (0.9, 74.2)	13.60 (1.571)	(10.52, 16.69)	< 0.0001
Month 24	39	14.61 (16.648)	9.60 (0.9, 80.3)	13.07 (1.582)	(9.96, 16.17)	< 0.0001
Month 36	13	11.62 (10.549)	10.10 (0.9, 40.8)	10.45 (1.958)	(6.61, 14.29)	< 0.0001

Any samples taken within 7 days (14 days if extended half-life product was used) of exogeneous FIX replacement therapy were not eligible.
If a participant withdrew consent, dropped out early from the study or resumed FIX prophylaxis, then the assessments at the visits following withdrawal/dropout/resumption were imputed as 1.9% based on their baseline disease severity (0.9% if severe and 1.9% if moderately severe).

Mean steady-state (geometric mean of all valid measurements from Week 12 to Month 15 post fidanacogene elaparvovec infusion) FIX:C as measured using one-stage assay with Actin-FSL reagent for the updated data-cut Aug 2023 is included in the effects table (see section Benefit Risk Assessment). Updated longer term data is available, including n=43 patients with 24 months of follow-up, n=34 with three years of FU, and 10 patients with four years of follow-up. The respective geometric means [±SD]

are in line with the results for weeks 12 to month 15 (2-year Safety Data).

Abbreviated presentation of the FIX:C data as **median (Q1, Q3)** for the one-stage assay with Actin-FSL reagent (OSA) and the chromogenic assay (CA) over time:

Year 2 (day 470 to 744), n=43: OSA 10.818 (3.344, 17.329) and CA 10.001 (3.394, 20.087)

Year 3 (day 745 to 1109), n=34: OSA 11.494 (4.558, 17.991) and CA 9.687 (4.221, 16.850)

Year 4 (day 1110 to 1481), n=10: OSA 14.943 (6.200, 18.300) and CA 11.496 (4.500, 12.700)

The applicant summarized the number of patients with FIX:C ⩾5% levels measured by Actin FSL on weeks 17, 32, 52, 78 and 104 post infusion of fidanacogene elaparvovec were 80.8%, 75%, 70.8%, 72% and 63.6% respectively. Same values were 80.8%, 76.4%, 70.7%, 66.6% and 63.6% when measured by chromogenic assay and 96.1%, 92.2%, 85.4%, 81.3% and 81.8% for Synthasil.

For SmPC section 5.1, based on the Aug 2023 data cut, a presentation of mean (SD) and median (min, max) as well as a summary of FIX:C category (0<5%, 5-<15%, 15-<40%, 40-<150%, <=150%) was implemented upon request. For this analysis, the applicant was asked to update all numbers and tables in the SmPC by imputing FIX:C values after resumption of prophylaxis or withdrawal based on the patient's baseline disease severity.

Resumed prophylaxis

Six out of 45 (13.3%) participants resumed FIX prophylaxis therapy. For patients who resumed prophylaxis, detailed analysis of baseline characteristics and treatment phase characteristics did not reveal a clear trend or factor that might identify patients at increased risk. The SmPC reflects the most current number of patients having resumed prophylaxis and the underlying reasons.

- **Ancillary analyses**

N/A

Summary of main efficacy results

The following table summarises the results from the main studies supporting the present application, i.e. pivotal study C0371002 in relation to baseline data generated in study C0371004, that are deemed most suitable to support the efficacy claim. Feedback from patients' organisations on relevance of outcomes has been considered. These summaries should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment (see later sections).

Table 6. Summary of Efficacy for trial C0371002

Title: Phase 3, Open Label, Single Arm Study to Evaluate Efficacy and Safety of FIX Gene Transfer With PF-06838435 (rAAV-Spark100-hFIX-Padua) in Adult Male Participants With Moderately Severe to Severe Hemophilia B (FIX:C≤2%) (BeneGene-2)		
Study identifier	C0371002, 2018-003086-33, NCT03861273	
Design	Open-Label, Single-Arm	
	Duration of main phase: Duration of Run-in phase: Duration of Extension phase:	Treatment Period = Day 1 (single dose) At least 6 months in lead-in Study C0371004 Total follow-up of 6 years: 52-week post-treatment follow-up phase and 5-year long-term follow-up phase
Hypothesis	Non-inferiority compared to FIX prophylaxis during pre-infusion period	

Treatments groups	Fidanacogene elaparvovec		Single intravenous dose of 5×10^{11} vg/kg fidanacogene elaparvovec 51 participants enrolled and 45 participants were dosed
Endpoints and definitions	Primary Endpoint	ABR_{total}	Non-inferiority on ABR for total bleeds (treated and untreated) from Week 12 to Month 15 versus usual care FIX prophylaxis replacement regimen, comparing pre- and post-IP infusion.
	Secondary Endpoint	AIR	AIR of exogenous FIX from Week 12 to Month 15 post-IP infusion versus AIR of FIX with usual care FIX replacement regimen pre-IP infusion
	Secondary Endpoint	FIX:C	Vector-derived FIX:C level at steady state (from Week 12 to 15 months post-IP infusion) demonstrated to be greater than 5%. Descriptive summary of FIX:C by study visit.
	Secondary Endpoint	Annualized FIX Consumption	Annualized FIX consumption reported as international units per kilogram (IU/kg) and total units measured from Week 12 to Month 15 post-IP infusion versus pre-IP infusion.
	Secondary Endpoint	Hemophilia Joint Health Score	Improvement of total score at Week 52 and Week 104 post-IP infusion versus baseline.
Database lock	20 December 2022		

Results and Analysis

The primary and secondary clinical efficacy endpoints were met, demonstrating the superiority of fidanacogene elaparvovec over usual care in the treatment of hemophilia B. C0371002 met its primary efficacy objective of non-inferiority of ABR_{total} with a one-time infusion of fidanacogene elaparvovec at a dose of 5×10^{11} vg/kg compared to FIX prophylaxis. In addition to non-inferiority, ABR_{total} was further demonstrated to be superior post infusion of fidanacogene elaparvovec compared to FIX prophylaxis.

Analysis description	**Primary Analysis: ABR_{total}**		
Analysis population and time point description	Dosed Analysis Set, pre-infusion period (≥6 months lead-in up to dosing), post fidanacogene elaparvovec infusion (Week 12 to Month 15)		
Descriptive statistics and estimate variability	Treatment group	Fidanacogene elaparvovec	
	Number of subjects	45	
	Model-derived ABR_{total} (95% CI)	Pre-infusion period	Post fidanacogene elaparvovec infusion
		4.43 (1.81, 7.05)	1.30 (0.59, 2.02)
	Treatment difference (95% CI; p-value)	-3.13 (-5.44, -0.81; p-value=0.0081)	
	Percentage reduction (95% CI; p-value)	70.63% (49.17%, 83.03%; p-value<0.0001)	
Effect estimate per comparison	Post fidanacogene elaparvovec infusion versus FIX prophylaxis regimen.		
Analysis description	**Secondary analysis: AIR**		

Analysis population and time point description	Dosed Analysis Set, pre-infusion period (≥6 months lead-in up to dosing), post fidanacogene elaparvovec infusion (Week 12 to Month 15)			
Descriptive statistics and estimate variability	Treatment group	Fidanacogene elaparvovec		
	Number of subjects	45		
	AIR (mean [SD])	Pre-infusion period	Post fidanacogene elaparvovec infusion	
		58.83 (29.056)	4.46 (10.028)	
	Treatment difference (95% CI; p-value)	-54.37 (-63.64, -45.10; p<0.0001)		
	Percentage reduction	92.4%		
Effect estimate per comparison	Post fidanacogene elaparvovec infusion versus FIX prophylaxis regimen			
Analysis description	**Secondary analysis: FIX:C**			
Analysis population and time point description	Dosed Analysis Set, post fidanacogene elaparvovec infusion (Week 12 to Month 15), Month 15, and Month 24			
Descriptive statistics and estimate variability	Treatment group	Fidanacogene elaparvovec		
	Number of subjects	As indicated below		
	FIX activity	Post fidanacogene elaparvovec infusion		
		One-Stage SynthASil Assay	One-stage Actin-FSL	Chromogenic Assay
	From Week 12 to Month 15 (n, geometric mean [SD])	n=45 25.90 (16.89)	n=45 12.62 (8.92)	n=45 13.49 (10.40)
	Month 15 (n, mean [SD])	n=35 27.47 (25.739)	n=34 13.10 (12.792)	n=35 15.82 (16.996)
	Month 24 (n, mean [SD])	n=22 25.00 (22.627)	n=22 12.67 (11.884)	n=22 15.40 (18.829)
Effect estimate per comparison	There are no treatment group comparisons for this study.			
Analysis description	**Secondary analysis: Annualized Total FIX Consumption**			
Analysis population and time point description	Dosed Analysis Set, pre-infusion period, post fidanacogene elaparvovec infusion (Week 12 to Month 15)			
Descriptive statistics and estimate variability	Treatment group	Fidanacogene elaparvovec		
	Number of subjects	45		
		Pre-infusion period	Post fidanacogene elaparvovec infusion	

	Annualized Total FIX Consumption (IU/kg)	3170.74 (1634.753)	235.04 (538.977)
	Treatment difference (95% CI; p-value)	-2935.70 (-3403.10, -2468.30; p<0.0001)	
	Percent reduction in mean	92.6%	
Effect estimate per comparison	Post fidanacogene elaparvovec infusion versus FIX prophylaxis regimen.		
Analysis description	**Secondary analysis: Haemophilia Joint Health Score**		
Analysis population and time point description	Dosed Analysis Set, change from baseline to Week 52 and Week 104 post fidanacogene elaparvovec infusion		
Descriptive statistics and estimate variability	Treatment group	Fidanacogene elaparvovec	
	Number of subjects	As indicated below	
	Mean (SD) change from baseline to Week 52 (n=36)	-2.6 (5.95)	
	Mean (SD) change from baseline to Week 104 (n=20)	-2.3 (4.45)	
Effect estimate per comparison	Post fidanacogene elaparvovec infusion versus baseline.		

2.6.5.3. Clinical studies in special populations

Data on efficacy in elderly patients, in paediatric patients, or with hepatic impairment are not available.

Considering the proposed BMI-dependent dose adjustment, patients having a BMI above 30 kg/m² is considered a special population for this MAA. The data submitted with the responses to the D120 LoQ allowing for assessment of patient level data confirmed, that the BMI adapted dosing in patients up to a BMI of 47.43 kg/m² is not associated with substantially lower FIX:C.

Table 7. Elderly patients included in clinical trials with fidanacogene elaparvovec

	Age 65-74 (Older subjects number /total number)	**Age 75-84 (Older subjects number /total number)**	**Age 85+ (Older subjects number /total number)**
Controlled Trials C0371002	0/45	0/45	0/45
Non Controlled C0371005/C0371003	0/15	0/15	0/15

2.6.5.4. In vitro biomarker test for patient selection for efficacy

Two nAb assays were used in the clinical program for patients' enrollment in the studies: The initial antigen-specific cell-based in vitro assay was developed and validated as a clinical grade assay for the

determination of nAb titer in serum by Spark Therapeutics (Philadelphia, PA 19104 USA) for C0371005 and for a portion of samples in C0371003.

Another qualitative cell-based antibody-mediated neutralization assay using an AAV vector (AAVRh74var CAG lucP) was developed and validated in a CAP/CLIA laboratory at LabCorp/Monogram BioSciences (South San Francisco, CA 94080, USA) as a single site Companion Diagnostic assay. The LabCorp anti-AAVRh74var capsid assay was used in C0371002, C0371003, and C0371004 to determine the level of nAb activity to AAVRh74var capsid.

Bridging studies carried out as part of LabCorp nAb assay validation studies have demonstrated that the results between the Spark and the LabCorp assays are concordant.

The LabCorp nAb assay has been developed in accordance with applicable CDx assay development regulations and standards such as guidelines from the CLSI and ISO. The antiAAVRh74var nAb assay is being submitted for marketing authorization by LabCorp, as pertinent based on applicable regulatory requirements.

Formal evaluation of the CDx is pending.

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

Not applicable

2.6.5.6. Supportive studies

C0371005

This is a Phase 2a long-term follow-up (LTFU) study to evaluate long-term safety, durability of transgene expression, and effect on clinical outcomes in participants completing C0371005 or in participants who enroll and receive fidanacogene elaparvovec in the C0371003 substudy. This study provides an additional 5 years of follow up post vector infusion for participants who enter the LTFU portion from C0371005. For this MAA, only the LTFU portion is of relevance regarding efficacy.

N=14/15 patients treated in study C0371005 enrolled in study C0371003. At updated data cut, 7 out of 14 participants had completed a combined 6 years of follow-up in C0371005/C0371003. 4 out of 14 participants were still ongoing with a minimal follow-up of 5.4 years. 3 participants discontinued (1 lost to follow-up and 2 withdrew by subject).

With regards to durability, mean treated ABR remained lower than 1.0 during each Year 2 through Year 6 post vector infusion with 10 (71.4%) participants having zero treated bleeds during their entire time in the study. AIR generally decreased over the entire follow-up period, with mean AIR from 1.53 during Year 2 to 0.38 during Year 6 post vector infusion. Transgene-derived FIX activity levels were in mild range as measured by central OS Actin-FSL based assay, with geometric mean of 24.67% (n=14) during Year 2 and 24.51% (n=7) during Year 6. No participant had resumed prophylaxis as of the data cutoff.

C0371003

This is a Phase 2a long-term follow-up (LTFU) study to evaluate long-term safety, durability of transgene expression, and effect on clinical outcomes in participants completing C0371005 or in participants who enroll and receive fidanacogene elaparvovec in the C0371003 substudy. This study provides an additional 5 years of follow up post vector infusion for participants who enter the LTFU portion from C0371005. For this MAA, only the LTFU portion is of relevance regarding efficacy.

N=14/15 patients treated in study C0371005 are followed in study C0371003, with follow-up at data cut ranging from 36 to 75 months.

Regarding durability, for the 14 participants in LTFU, mean ABRtreat remained lower than 1.0 from Years 2 through 6 post fidanacogene elaparvovec infusion with 10 (71.4%) participants having zero treated bleeds during their entire time in the study. AIR generally decreased over the entire follow-up period, with mean AIR from 1.53 during Year 2 to 0.38 during Year 6 post vector infusion. Transgene-derived FIX activity levels remained at a clinically meaningful level (>5%) during Years 2 through 6. No participants resumed prophylaxis regimen.

2.6.6. Discussion on clinical efficacy

The clinical development programme presented for MAA consists of a single, ongoing pivotal phase 3 single-arm study (C0371002) with its associated non-interventional lead-in study (C0371004), one supportive, preceding single-arm phase 1/2a study (C0371005), and respective long-term follow-up studies (ongoing C0371003 and planned C0371017). Based on CPMP/EWP/2330/99, in single pivotal trial approaches the study has to be exceptionally compelling, and special attention is to be paid to methodology and clinical relevance.

The primary analysis of a single, ongoing, single-arm pivotal phase 3 study (C0371002) of n=45 patients of which 41 had a minimum follow-up of 65 weeks post vector infusion has been submitted for MAA, supported by a completed phase 1/2a study (C0371005) in n=15 patients and their longer-term follow-up data as generated in the ongoing study C0371003, ranging from 36 to 75 months post vector infusion.

Design and conduct of clinical studies

Acceptability of the single-arm trial with intra-patient comparison derived from a separate run-in study as well as the chosen key efficacy endpoints to support MAA were agreed in EMA scientific advices for the studied patient population (EMEA/H/SA/3416/1/FU/2/2018/ADT/PR/HTA/SME/II, EMEA/H/SA/3416/3/2019/ADT/PR/III).

The study was repeatedly and substantially modified. The protocol was amended 3 times and the SAP even 5 times during the ongoing trial, with final modifications being included after the treatment of the last patient. The amendments addressed, among others, the (co-)primary endpoint(s) and timing of efficacy assessment. Changing the initially co-primary endpoints ABR and FIX:C and timing of efficacy assessment in an ongoing open label study that is intended to support an MAA was not discussed in an EMA scientific advice. However, it is acknowledged the change was based on US FDA recommendations, and the endpoints remained for analysis.

The generated clinical data covers haemophilia B patients with endogenous FIX activity up to 2 IU/dl (inclusive), which includes severe haemophilia B <1 IU/dl and the lower ranges of moderate haemophilia B. The proposed indication refers to severe and moderately severe haemophilia B, with the inclusion criteria of the clinical studies as given in SmPC section 5.1 clarifying the baseline FIX activity levels of patients included in the pivotal study.

Efficacy data and additional analyses

Endpoints covering bleeds and FIX substitution

The main aim of a gene therapy in this clinical setting is to provide patients with the freedom from regular prophylactic and/or therapeutic infusions of external factor IX. Consequently, relevant endpoints are the primary endpoint ABR_{total}, the key secondary endpoint AIR and the secondary endpoint annualized FIX consumption. Provided was the primary analysis of study C0371002 conducted once at least 40 treated patients were evaluable for the primary endpoint.

Model-derived ABR_{total} (95% CI) week 12 to month 15 post-infusion was 1.3 (0.59, 2.02) compared to 4.43 (1.81, 7.05) prior treatment. Non-inferiority of ABR_{total} and subsequently superiority was concluded. The clinical relevance of the primary estimand, where subjects are only considered while they have been on study and while they had not resumed prophylaxis, was, however, discussed controversially. Supplementary estimands where (1) intercurrent events [dropouts, withdrawals or resumption prophylaxis] were imputed by jump to reference and (2) resumption of prophylaxis was ignored and ABR data was analysed regardless of this intercurrent event were provided. While both estimands are considered clinically more relevant than the primary estimand, none of them adequately captures the negative outcome of the 5 subjects (> 10%) who had to resume prophylaxis after gene therapy treatment within the first 15 months after treatment and the total of 6 subjects who resumed prophylaxis until the primary data cutoff. For the presentation in the SmPC, an analysis using data from 30 Aug 2023 cutoff by including the bleeding events which occurred post-resumption of prophylaxis has been chosen upon request.

Model-derived AIR (mean [SD]) week 12 to month 15 post-infusion was 4.46 (10.028) compared to 58.83 (29.056) prior treatment, equal to a 92.4% reduction. 29 out of 45 (64.4%) participants required no infusions of FIX from Week 12 to Month 15 after infusion with fidanacogene elaparvovec.

Annualized Total FIX Consumption (IU/kg) (mean [SD]) week 12 to month 15 post-infusion was 235.04 (538.977) compared to 3170.74 (1634.753) equal to a 92.6% reduction.

Over time, 27 out of 45 patients (60%) remained without bleeds in the observation period.

The key endpoints are clinically relevant, and the observed reduction in bleeding events and associated reduction of exogenous FIX consumption can be directly linked to the mechanism of action, outweighing the methodological concerns. *FIX activity*

In the pivotal study, the geometric mean of FIX:C for the period week 12 to month 15 as determined by ActinFSL OSA was 12.62 (8.92). Median (Q1, Q3) FIX:C was 10.726% (7.415%, 16.632%) in the ActinFSL OSA, while it was 10.236% (5.415%, 19.055%) in the chromogenic assay. As discussed in the clinical pharmacology section, the reported geometric mean of FIX:C compared to baseline levels confirmed a pharmacologic effect.

The clinical relevance of the geometric mean for FIX:C, however, has limitations. It ignores the negative impact of FIX:C dropping below clinically relevant thresholds for shorter periods of time. The data shows that the FIX:C values are variable and declining over time. Hence, it is of primary importance how many patients persistently stayed above the pre-specified threshold of FIX:C > 5% of normal. Furthermore, the pre-specified imputation for subjects who dropped out, withdrew consent or had to resume prophylaxis does not reflect the pre-treatment endogenous FIX:C level but 1.9%, thus assuming a moderately severe instead severe haemophilia. The applicant has provided additional tabulations where subjects with severe haemophilia were imputed with 0.9% FIX:C and subjects with moderately severe haemophilia with 1.9%. While this is still considered rather anti-conservative (as both values are at the upper range of the underlying FIX:C levels for moderately severe and severe subjects) it is preferable to the initial analysis.

From a patient's perspective, the laboratory parameter FIX activity is perceived as more than a surrogate outcome. Better protection against intracranial bleeds has been given as one expectation towards gene therapies, with severity of haemophilia being a related risk factor for intracranial haemorrhage (doi:10.3390/jcm11071969) and severity being defined via FIX:C thresholds.

In the supportive, preceding phase 1/2a study, in which the same target dose was infused, FIX:C levels were substantially higher than in the pivotal study. The data was reviewed from multi-factorial aspects including manufacturing process, study population, management of presumed cellular immune response, and statistical handling. A (temporal) correlation to manufacturing processes exists, but from a CMC

perspective all manufacturing processes are comparable including aspects known to impact immunogenicity of the AAV. Considering the very limited sample size and regional focus in trial C0371005, a population impact cannot be excluded. At the moment, the durability of effect demonstrated by available longer-term FIX expression data generated in the preceding phase 1/2a trial is supported by updated (limited) longer-term FIX:C in the phase 3 trial. Data of the ongoing nominal dosing cohort in study C0371002 enrolling globally but not in North America, might generate data to better inform on the realistically achievable FIX:C.

No formal dose finding study has been completed. Preliminary very limited data of an ongoing dose-escalation substudy might be indicative for a dose-response relationship.

Resumption of prophylaxis

No defined endpoint, but the number of patients resuming prophylaxis post-treatment was six at data-cutoff, five of which resumed due to FIX:C levels.

The updated efficacy data (24 month outcomes) submitted with the D120 responses confirm the results from the initial analyses in terms of ABR, AIR, FIX consumption, and FIX:C levels w12-m15. No additional subject had to resume prophylaxis at the new cut-off.

Durability

To conclude on durability of the transgene effect, data generated in the pivotal study is too immature. Translatability of longer-term data generated in the LTFU of the phase 1/2a study might be limited, due to the substantially higher FIX:C. However, the ongoing pivotal study C0371002 is designed to generate up to 6 years post-treatment data and the planned, subsequent LTFU study C0371017 is expected to generate long-term follow-up data up to 15 years post-treatment. Both are listed in Annex II.

Additional expert consultation

Patients' organisations were invited to comment on any aspects that are of particular importance to patients/carers. Feedback was received from the European Hematology Association (EHA) and the European Haemophilia Consortium (EHC).

The received feedback indicated the most meaningful would be a "haemophilia-free life"; as potential benefits, reduction or even elimination of spontaneous bleeding and the need for long-term regular infusions of factor replacement was stated. Also, greater protection to prevent intracranial bleeds in adults aged 60 or above or those with hypertension was stated as expectation towards gene therapies. Preserving joints or slowing down existing joint damage was also mentioned as reason to join clinical trials.

The organisations acknowledged a large heterogeneity in factor response following gene therapy infusion as well as a lack of predictability of patient response to the therapy with mechanisms predicting patient response to gene therapy not yet fully understood. They feel it was crucial to ensure that management plans for patients with excessive (above the normal range) or suboptimal (below normal/mild range) factor expression are in place.

In terms of expectations relayed by additional experts' consultations, a relevant proportion of achieved FIX:C levels is suboptimal, as they are below normal/mild (haemophilia) range. This applies to approx. 25% of the participants. Clinical relevance of the achieved FIX:C levels, i.e. suitability of the achieved FIX:C levels to provide sustained bleed protection in general and protection against intracranial bleeds in particular, is yet to be determined. Still, the continuous presence of endogenous factor IX is likely to influence the bleeding frequency in a beneficial way, as the peaks and troughs present during SoC factor prophylaxis are avoided by gene therapy.

Assessment of paediatric data on clinical efficacy

No data in paediatric patients is available or currently been generated, which is in line with the agreed PIP.

Additional efficacy data needed in the context of a conditional MA

To address the remaining uncertainties around efficacy, including the lack of comprehensive data on durability of the effect over time, the following specific obligations were agreed:

- An interim clinical study report including 6 years follow-up of Study C0371002 should be submitted no later than December 2028 and is subject to a specific obligation laid down in the MA (SOB-1).

- The final clinical study report of the long-term follow-up study C0371003 including 5 years follow-up data should be submitted no later than January 2025 (SOB-2).

2.6.7. Conclusions on the clinical efficacy

Efficacy has been demonstrated based on the annualised bleeding rate for all, i.e. treated and untreated bleeds, compared to the prophylactic setting (primary endpoint). It is supported by relevant reductions in the key secondary endpoints annualised infusion rate and FIX consumption, addressing the need for regular FIX substitution. Approximately 60% of the patients remained bleeding event free in the individual observation period (ranging 2 to up to 4 years). The results are considered clinically meaningful and indicate a clinical benefit in most but not all patients for the covered time period.

In contrast, six of 45 patients (13.3%) in the ongoing pivotal trial have already resumed prophylaxis. Predictive factors have not been elucidated despite respective analyses by the applicant, with the small sample size being a relevant limitation.

However, the available data in based on a limited number of patients followed for a limited time period. Due to the limitations of the provided dataset, a full MA as initially sought by the applicant was not considered appropriate, and the applicant agreed to request a conditional marketing authorisation.

The CAT considers the following measures necessary to address the missing efficacy data in the context of a conditional MA:

- In order to confirm the efficacy and safety of Durveqtix in adults with severe and moderately severe haemophilia B (congenital factor IX deficiency) without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74, the MAH should submit interim results (6 years of data) of pivotal Study **C0371002** with 45 subjects who received a dose calculated using actual batch concentration and at least 34-month data of patients who received a dose based on nominal concentration dosing.

- In order to confirm the efficacy and safety of Durveqtix in adults with severe and moderately severe haemophilia B (congenital factor IX deficiency) without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74, the MAH should submit the final results (5 years of data) of long-term follow-up study **C0371003** with 14 subjects who received 5×10^{11} vector genomes per kg (vg/kg) of body weight.

The CAT considers the following measures necessary to address additional issues related to efficacy:

- In order to further characterise the long-term efficacy and safety of Durveqtix in adults with severe and moderately severe haemophilia B (congenital factor IX deficiency) without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74, the MAH should conduct and submit the final results of registry-based study **C0371007**, according to an

agreed protocol.

- In order to further characterise the long-term efficacy and safety of Durveqtix in adults with severe and moderately severe haemophilia B, the MAH should submit the final results of Study **C0371017,** which includes patients who have been treated with Durveqtix in the MAH-sponsored clinical trials.

The CHMP endorses the CAT conclusion on clinical efficacy as described above.

2.6.8. Clinical safety

2.6.8.1. Patient exposure

Exposure to fidanacogene elaparvovec

In Study C0371002, n= 45/n=51 enrolled participants received study treatment, and in Study C0371005, n=15/n=22 enrolled participants. For the pooled safety analysis set (n=60 subjects), n=40 subjects had a BMI of <= 30 kg/m^2, and n=20 had a BMI >= 30 kg/m^2.

Study C0371003 was originally planned as long-term follow-up study for participants of Study C0371005 only (= C0371003 Cohort 1), but later modified to include dose escalation cohorts. In the C0371003 sub-study, an open-label, non-randomized dose escalation study for Cohort 2 and Cohort 3, study intervention refers to a single infusion of fidanacogene elaparvovec with an initial dose of [redacted] vg/kg single administration, and with the potential to proceed to one or more of the following doses: [redacted] vg/kg, [redacted] vg/kg single administration and [redacted] vg/kg single administration. In this sub-study, concomitant corticosteroids are allowed to be given prior, during and/or after fidanacogene elaparvovec infusion. As of DCO [redacted] in Cohort 2 ([redacted] vg/kg) and [redacted] in Cohort 3 ([redacted] vg/kg) have completed the one year observation period.

Exposure to *corticosteroids* after infusion of fidanacogene elaparvovec (studies C0371002 and C0371005)

According to the clinical trial protocols, treatment with corticosteroids is recommended after fidanacogene elaparvovec infusion for management of related immunogenicity, apparent by transaminase increase in the blood serum. The regimen described in the clinical trial protocols is in line with SmPC Table 3.

N=31 (51.6%) participants in the pooled analysis set required corticosteroids due to increase of transaminase and/or decrease of FIX activity, both events decisive for initiation of corticosteroids according to the wording in the clinical trial protocols.

2.6.8.2. Adverse events

Safety Assessment Plan and Methods

In the Studies C0371002, C0371005, and C0371003 safety endpoints were analyzed as follows:

- Number of participants and duration of follow-up after fidanacogene elaparvovec infusion
- AEs in the first year after infusion and yearly follow-up
- SAEs

- AESIs including:
- Hypersensitivity reactions (within scope of hypersensitivity SMQ)
- Clinical thrombotic events
- FIX inhibitors
- Hepatic malignancies
- Drug related elevated hepatic transaminases that fail to improve or resolve through
- treatment with immunosuppressive regimens
- Malignancy assessed as having reasonable possibility of being related to study drug
- AEs leading to study discontinuation
- Clinical laboratory results and hepatic evaluations (including LFT, AFP and liver ultrasound)
- Other safety evaluations including vital signs, ECG, and physical examination findings; pregnancies reported for study participants partners; vector shedding

Safety endpoints were analyzed by subgroup including:

- Incidence and severity of AEs for the first-year post fidanacogene elaparvovec infusion
- Incidence of AEs across the total follow-up period
- Incidence of SAEs during the first year of follow-up and the total follow-up period
- Incidence of AESIs across the total follow-up period
- Number and percentage of participants with corticosteroid treatment

Subgroup analyses were done for (if n=5 participants per subgroup):

- Age group (<35 vs ≥35 and <45 vs ≥45 years of age),
- Region,
- BMI (≤30 vs >30 and <25 vs ≥25 kg/m^2), and
- Corticosteroid use (yes vs no).

Reported TEAEs

C0371002: A total of 205 TEAEs were reported in n=38/45 (84.4%) participants during the entire study, in the majority classified as G1 or G2.

Study C0371005: A total of 81 TEAEs were reported in the study in n=15 participants. The most commonly reported TEAEs any grade were in the SOCs of infections and infestation (8 participants, 53.3%), gastrointestinal disorders (7 participants, 46.7%) and musculoskeletal and connective disorders (6 participants, 40.0%). The most frequently reported TEAEs any grade were upper respiratory tract infection (5 participants, 33.3%), nasopharyngitis (3 participants, 20.0%), back pain (3 participants, 20.0%) and muscle strain (3 participants, 20.0%). The majority of TEAEs (53/81) were mild in severity, and the other 28 TEAEs were moderate in severity.

Studies C0371002 and C0371005 (pooled) during the first year of follow-up after fidanacogene elaparvovec infusion: N=52/60 (86.7%) participants had any TEAE any grade. The most frequently reported TEAEs were ALT increased in n=12/60 (20%) participants and nasopharyngitis in n=11/60 (18.3%) participants.

LTFU Study C0371003 Cohort 1: There were n=4 (28.6%) participants with TEAEs. N=3 out of the 14 participants (21%) had clinically significant abnormalities in liver ultrasound test data during LTFU (Years 2 through 6) in addition to abnormal laboratory liver values.

TEAE - analysis by sub-groups (pooled analysis)

Age: percentage of participants with TEAEs was similar across 4 age sub-groups

- 28 (84.8%) participants of <35 years vs 24 (88.9%) participants of ≥35 years
- 42 (89.4%) participants of <45 years vs 10 (76.9%) participants of ≥45 years

BMI: The number of TEAEs reported was numerically higher with rising BMI

Use of Corticosteroids:

- Corticosteroid Use: Yes (N=31)

- 30 (96.8%) participants had TEAEs.

- Most frequent AEs: alanine aminotransferase increased in 12 (38.7%) participants nasopharyngitis 7 (22.6%) participants, arthralgia 7 (22.6%) participants, and hepatic function abnormal in 6 (19.4%) participants.

- Corticosteroid Use: No (N=29)

- 22 (75.9%) participants had TEAEs.

- Most frequent AEs: nasopharyngitis in 4 (13.8%) participants, upper respiratory tract infection in 5 (17.2%) participants, headache in 4 (13.8%) participants.

Reported TEAEs Study C0371003 Cohorts 2 and 3

A total of 14 TEAEs were documented in 2 participants, and mostly considered as hypersensitivity reactions. TEAEs were rash maculo-papular, hypotension, skin laceration, upper respiratory tract infection, myalgia, headache, AST increased, ALT increased, coagulation factor IX level decreased and bradycardia. All these TEAEs were G1 and G2.

G1 and G2 AESIs for hypersensitivity reactions and hepatic transaminases occurred, in the majority of cases reported as G1 and G2, respectively. Study discontinuations due to adverse events have not been documented.

AESIs (as such reported for Study C0371002 and C0371003 cohorts 2 and 3)

Study C0371002: N=6 (13.3%) participants reported AESIs of hepatic function abnormal and 1 (2.2%) participant had abnormal liver function test. N=12 (26.7%) participants had alanine aminotransferase increased, 3 (6.7%) participants had aspartate aminotransferase increased, 3 (6.7%) participants had hepatic enzyme increased, and 3 (6.7%) had transaminase increased. In the majority of cases, the AESIs occurred were classified G1 and G2, respectively.

Study C0371003 cohorts 2 and 3: AESIs of hypotension and rash maculo-papular as possible hypersensitivity reaction were reported in 1 participant in Cohort 2 and hypotension was reported in the participant in Cohort 3.

As of DCO date Aug2023, no AESIs of thrombotic events, development of FIX inhibitors or hepatic malignancies were reported for C0371002, C0371005 and C0371003 all cohorts.

2.6.8.3. Serious adverse event/deaths/other significant events

No deaths were reported in the clinical trials as of DCO date Aug2023.

Serious Adverse Events by Study

Study C0371002: N=7 participants had treatment emergent SAEs (N=11 SAEs). The following terms (n=14) were used: anemia, lymphadenopathy, duodenal ulcer haemorrhage, duodenal ulcer, upper gastrointestinal haemorrhage, asthenia, drug-induced liver injury, hepatotoxicity, Covid-19, Covid-19 pneumonia, alcohol poisoning, coagulations FIX decrease, vascular disorders, haematoma. Two SAEs, duodenal ulcer hemorrhage and associated anemia, occurred in one patient in the setting of corticosteroid use with no concomitant use of gastric acid reducer. These SAEs were assessed as related to treatment and occurred within the first year post infusion of fidanacogene elaparvovec.

Study C0371005: No SAEs reported

Study C0371003 Cohort 1: For n=4 participants (28.6%) n=9 SAEs using n=10 PT have been reported: aggravated/spinal stenosis, appendicitis, accident, joint dislocation, kidney contusion, liver contusion, rib fracture, haemarthrosis, spinal stenosis, aortic dissection. Study C0371005 sub-study cohorts 2 and 3: No SAEs reported as of DCO date.

2.6.8.4. Laboratory findings

For the pooled analysis, apart from the elevations in transaminases up to 2 fold ULN, there were fluctuations in laboratory values for haematology and chemistry parameters during the course of the studies, according to the documents provided in most cases not assessed as clinically important. One participant in Study C0371005 experienced 1 moderate TEAE of normocytic anaemia and 1 mild TEAE of microcytic anaemia with low normal levels of erythrocytes, mean corpuscular hemoglobin, hematocrit, and hemoglobin, as well as high normal levels of erythrocytes distribution width reported. Both TEAEs were considered not related to study drug by the investigator. No action was taken in response to the events. The TEAEs resolved during the study.

LTFU Study C0371003 Cohort 1

Transaminases: N=8/14 (57%) participants experienced ALT increase above ULN during LTFU (Years 2 through 6), and 3 of which had AST increase above ULN. There was 1 participant with ALT within 3-5 × ULN and AST within 3 5 × ULN with normal ALT and AST at baseline respectively, and total bilirubin <2 × ULN and normal alkaline phosphatase at the time of this ALT and AST elevation. One participant had ALT within 2-3 × ULN and AST within 1-2 × ULN. The other 6 participants had ALT within 1-2 × ULN (1 participant had ALT and AST within 1-2 × ULN).

2.6.8.5. In vitro biomarker test for patient selection for safety

Not applicable.

2.6.8.6. Safety in special populations

	Age <35 vs ≥35 years N= (%) Pooled SAS	**Age <45 vs ≥45 years N= (%)** Pooled SAS	**BMI (%) ≤30 vs >30 N=40 (66.7%)** Pooled SAS	**BMI (%) <25 vs ≥25 N=20 (33.3%)** Pooled SAS	**Cortisone use Yes: N=31** Pooled SAS	**Cortisone use No: N=29** Pooled SAS	**Severe Haem B induced joint involvement**
Treatment rel. TEAEs.	18 (54.5%) vs 14 (51.9)	29 (61.7) vs Less than 5	18 (45%) vs 14 (70%)	10 (47.6) vs 22 (56.4)	N=28 (90.3)	N=22 (75.9)	N=22/60 haem B induced target joints at screening. Uncertainty with regard to B/R.
Serious AEs – Total for pooled studies up to 6.3 years: N=11 (18.3%) subjects had n=23 SAEs							
- Fatal	none	none	none	none	none	none	
- Hospitalization/ prolong existing hospitalization	none	none	none	none	none	none	
- Life-threatening	none	none	none	none	none	none	
- Disability/incapacity	none	none	none	none	none	none	
AE leading to drop-out	none	none	none	none	none	none	
Infections and infestations Liver enzyme increase	No age dependency	No age dependency			Indication for cortisone initiation		
Anticholinergic syndrome	none	none	none	none	none	none	
Quality of life decreased	none	none	none	none	none	none	
other AE appearing more frequently in older patients	none	none	none	none	none	none	

2.6.8.7. Immunological events

In Study C0371002, participants were tested prior to fidanacogene elaparvovec infusion and when corticosteroid treatment was given (for presumed T-cell response, based on transaminase increase

and/or FIX:C decrease). N=28/45 (62.2%) participants were treated with corticosteroids for presumed cellular immune response. At baseline, n=12/23 (52.2%) participants with corticosteroid use and ELISPOT assessment tested positive in the overall capsid pool and 11/23 (47.8%) tested positive in the overall FIX pool, which includes WT, FIX polymorphism, and Padua pools. As of the data cutoff date, 10/28 (35.7%) participants with corticosteroid use for presumed T-cell response had ELISPOT assessment prior to or within 24 hours of corticosteroid initiation (protocol-specified time window considered most indicative of a T-cell response). Prior to corticosteroid use, n=4/10 (40.0%) participants with ELISPOT assessment tested positive in the overall capsid pool and n=3/10 (30.0%) participants tested positive in the overall FIX pool. Approximately 3 weeks after corticosteroid treatment, n=4/8 (50.0%) participants with ELISPOT assessment tested positive in the overall capsid pool and n=2/8 (25.0%) participants tested positive in the overall FIX pool.

FIX Inhibitor results

As of the DCO date Aug2023, no FIX inhibitor has been detected in any participant who received fidanacogene elaparvovec infusion in Study C0371002 or C0371005 and its LTFU study C0371003, Cohort 1, monitoring participants up to 6 years post-infusion.

2.6.8.8. Safety related to drug-drug interactions and other interactions

Not applicable.

2.6.8.9. Discontinuation due to adverse events

None.

2.6.8.10. Post marketing experience

Not applicable.

2.6.9. Discussion on clinical safety

The safety evaluation of Durveqtix to support registration in adult patients with severe and moderately severe haemophilia B (congenital factor IX deficiency) without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74 is based on the results from 3 open-label clinical trials submitted until the respective cut-off dates:

- Ongoing Phase 3 registrational Study C0371002 (cutoff date Aug2023)
- Completed Phase 1/2a dose exploration/dose expansion Study C0371005
- Ongoing Phase 2a long-term follow-up Study C0371003 (cutoff date Aug2023)

Results submitted for participants in Cohort 2 and Cohort 3 of the dose escalation sub-study of Study C0371003 are presented separately, which is acceptable as these subjects received higher doses of fidanacogene elaparvovec under prophylactic/concomitant corticosteroid treatment.

The safety results were provided for both the individual studies and as an integrated safety analysis (pooled across studies). The analysis population defined as the safety analysis set consists of all participants from applicable studies in the study pools who received an infusion of fidanacogene elaparvovec (n=60).

Total numbers of adverse Events as of DCO date Aug2023

A total of 283 adverse events (205 in study -002 and 78 in study -005/-003) were reported across the total follow-up period. N= 52/60 individuals had any adverse events, the majority of which were mild or moderate in severity; n=11/60 (18.3%) were severe. A total of 58 treatment-related preferred-term events were reported across the total follow-up period in 32/60 (53.3%) of individuals. The vast majority of these events occurred in the first year after treatment (noting that follow-up times are limited). Across the entire follow-up period, 11/60 (18.3%) participants experienced 23 treatment-emergent SAEs (PT events) of any cause.

No death cases, no discontinuations due to adverse events, no treatment-related clinical thrombotic events, no development of FIX inhibitors, and no hepatic or treatment-related malignancies were documented for the clinical studies during the reporting period.

Hepatotoxicity

Across clinical studies a total of 43/60 (71.7%) individuals had elevations in ALT and 44/60 (73.3%) individuals had elevations in AST (total number of ALT and AST episodes was 101 and 102, respectively).

The mean duration of elevated ALT elevation was 104 days (range: 4 to 1373 days) and the mean duration of AST elevation was 66 days (range: 1 to 719 days). A total of 31/60 (51.7%) patients had increased ALT levels that required corticosteroid treatment. The mean time to corticosteroid initiation was 46 days. The mean duration of corticosteroid treatment was 112 days (range: 41 to 276 days). The majority of transaminase elevations resolved over time. Reoccurrence of transaminase elevations up to G3 were observed in the majority of participants in Cohort 1 of the LTFU study.

Hepatotoxicity in context with BMI and/or COVID-19

The percentage of participants with treatment-related TEAEs across the total follow-up period was numerically higher when comparing BMI >30 (14/20; 70%) to BMI ≤ 30 (18/40; 45%). This difference was also noted for treatment-emergent SAEs (5/20; 25% in participants with BMI >30 vs. 6/40; 15%, in participants with BMI ≤ 30). Trial participants with a BMI >30 also required higher doses of corticosteroids for the management of AST and/or AST elevations, whereas there was no relevant difference in the fidanacogene elaparvovec dose applied between subjects with BMI >30 and BMI ≤ 30. The applicant provided a literature-based scientific discussion on the increased risk for liver related adverse events potentially associated with a BMI >30 due to metabolic particularities in obese patients. According to literature data, the association between (abdominal) obesity and increase of variable liver enzyme levels may be strong for gamma-glutamyl transferase (GGT). With regard to the association with increased levels of ALT, AST and AP, results reported seem to be not that clear, and the authors outline the complexity of the clinical diagnosis overweight and obesity, respectively (e. g. *Obesity, insulin resistance and their interaction on liver enzymes, Liu et al, 2021 Abstract,).* With respect to the reported persistence and/or re-occurrence of increased liver enzymes and abnormal liver ultrasound findings (liver parenchyma damage) after treatment with fidanacogene elaparvovec despite normal results at baseline in at least 3 subjects in the pivotal trial, the applicant discussed COVID-19 infection/vaccination as possible underlying reasons, scientific publications on this topic included. According to scientific publications, elevations in ALT and AST have been observed in patients with long-term COVID-19, particularly in those hospitalized during the acute phase (*Int J Environ Res Public Health, de Lima et al., 2023*). The authors emphazise the significance of accompanying factors other than a COVID-19 infection as such as underlying causes for liver enzyme increase. Cases reported on COVID-19 vaccination related liver enzyme increase are rare.

Immunology

Humoral immunity

The majority of patients (95.1%) treated with fidanacogene elaparvovec developed antibodies against AAVRh74var by year 1 post-infusion that persisted with high titers up to 6 years post-infusion. All study participants with nAb assessment developed anti-AAVRh74var capsid nAb, with high titers persisting up to 6 years post-infusion. The data show that a humoral immune response against fidanacogene elaparvovec is induced in (almost) all treated individuals.

Hypersensitivity events of G1/G2 were reported in 11/60 (18.3%) of individuals and documented as not treatment related.

FIX inhibitors (nAbs against FIX) were not detected in any subject hitherto.

Cellular immunity

Approximately half of the participants with evaluable ELISPOT had detectable circulating T cells that responded to capsid peptides likely contributing to elevation in transaminase, and approximately one-quarter of participants with evaluable ELISPOT assessment were positive to FIX peptide pools. The clinical relevance remains unknown particularly in the absence of FIX inhibitor formation.

Safety in special populations

Overall, no differences in the Durveqtix safety profile for relevant subgroups (age, race, and region) became apparent across the clinical studies. However, formal statistical analysis was not performed, and numbers per subgroups are generally very limited.

Vector biodistribution and shedding

In summary, the biodistribution profile has been characterized adequately. See clinical pharmacology section for details.

Pregnancy and newborns

At the time of the data cut-off 2 pregnancies in partners of Durveqtix treated subjects have been reported. While no clinical sequelae were reported in the mothers or newborns, neither pregnancy studies in humans receiving Durveqtix nor animal reproduction studies have been conducted with Durveqtix. Thus, it is not known whether Durveqtix can cause foetal harm when administered to a pregnant woman or can affect reproduction capacity.

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

Additional safety data needed in the context of a conditional MA

Additional long-term safety information is needed, and therefore the following specific obligations were agreed:

- An interim clinical study report including 6 years follow-up of Study C0371002 should be submitted no later than December 2028 (SOB-1).
- The final clinical study report including 5 years follow-up data of the long-term follow-up study C0371003 should be submitted no later than January 2025 (SOB-2).

2.6.10. Conclusions on the clinical safety

Based on the submitted safety data set as of DCO date August 2023, the safety profile of fidanacogene elaparvovec appears to be acceptable. The most relevant short to medium-term safety concern is elevated transaminases that may require treatment with corticosteroids.

Diligent post marketing surveillance is of utmost importance to detect potential rare adverse events and to investigate the potential risk of malignancy (due to vector integration, see Non Clinical section) on the longer term. Patients must be well-informed about this before receiving fidanacogene elaparvovec. In this regard, a warning has been added to the SmPC and package leaflet to inform on the potential risk of malignancy as a result of vector integration in liver cells and in other body cells.

The CAT considers the following measures necessary to address the missing safety data in the context of a conditional MA:

- In order to confirm the efficacy and safety of Durveqtix in adults with severe and moderately severe haemophilia B (congenital factor IX deficiency) without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74, the MAH should submit interim results (6 years of data) of pivotal Study **C0371002** with 45 subjects who received a dose calculated using actual batch concentration and at least 34-month data of patients who received a dose based on nominal concentration dosing.
- In order to confirm the efficacy and safety of Durveqtix in adults with severe and moderately severe haemophilia B (congenital factor IX deficiency) without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74, the MAH should submit the final results (5 years of data) of long-term follow-up study **C0371003** with 14 subjects who received 5×10^{11} vector genomes per kg (vg/kg) of body weight.

The CAT considers the following measures necessary to address additional issues related to safety:

- In order to further characterise the long-term efficacy and safety of Durveqtix in adults with severe and moderately severe haemophilia B (congenital factor IX deficiency) without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74, the MAH should conduct and submit the final results of registry-based study **C0371007**, according to an agreed protocol.
- In order to further characterise the long-term efficacy and safety of Durveqtix in adults with severe and moderately severe haemophilia B, the MAH should submit the final results of Study **C0371017,** which includes patients who have been treated with Durveqtix in all MAH-sponsored clinical trials.

The CHMP endorses the CAT conclusion on clinical safety as described above.

2.7. Risk Management Plan

2.7.1. Safety concerns

Important identified risks	Hepatotoxicity
Important potential risks	Development of FIX inhibitors Thromboembolic events Risk of malignancy in relation to vector integration in the DNA of body cells Transmission to third parties (horizontal transmission) Germline transmission
Missing information	Long-term safety

2.7.2. Pharmacovigilance plan

No additional pharmacovigilance activities.

Post authorisation efficacy studies included in RMP PART IV, will also address safety concerns for

fidanacogene elaparvovec.

2.7.3. Risk minimisation measures

Safety Concern	Risk Minimisation Measures	Pharmacovigilance Activities
Important Identified Risk		
Hepatotoxicity	Routine risk minimisation measures: SmPC Section 4.4, Special warnings and precautions for use SmPC Section 4.5, Interaction with other medicinal products and other forms of interaction SmPC Section 4.8, Undesirable effects PL Sections 2 and 4 Legal status: Medicinal product subject to restricted medical prescription. Treatment should be administered in a qualified treatment centre by a physician experienced in the treatment of haemophilia. Additional risk minimisation measures: Guide for Healthcare Professionals Patient Guide Patient Card	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: Hepatic Events Questionnaire Additional pharmacovigilance activities: None
Important Potential Risks		
Development of FIX inhibitors	Routine risk minimisation measures: SmPC Section 4.2, Posology and method of administration SmPC Section 4.4, Special warnings and precautions for use PL Section 2 Legal status: Medicinal product subject to restricted medical prescription. Treatment should be administered in a qualified treatment centre by a physician experienced in the treatment of haemophilia. Additional risk minimisation measures: Guide for Healthcare Professionals Patient Guide Patient Card	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: None Additional pharmacovigilance activities: None
Thromboembolic events	Routine risk minimisation measures: SmPC Section 4.4, Special warnings and precautions for use PL Section 2 Legal status: Medicinal product subject to restricted medical prescription. Treatment should be administered in a qualified treatment centre by a physician experienced in the treatment of haemophilia. Additional risk minimisation measures: Guide for Healthcare Professionals Patient Guide Patient Card	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: Thromboembolic Events Questionnaire Additional pharmacovigilance activities: None
Risk of malignancy in relation to	Routine risk minimisation measures: SmPC Section 4.4, Special warnings and precautions for use	Routine pharmacovigilance activities beyond adverse

Safety Concern	Risk Minimisation Measures	Pharmacovigilance Activities
vector integration in the DNA of body cells	PL Section 2 Legal status: Medicinal product subject to restricted medical prescription. Treatment should be administered in a qualified treatment centre by a physician experienced in the treatment of haemophilia. Additional risk minimisation measures: Guide for Healthcare Professionals Patient Guide Patient Card	reactions reporting and signal detection: Malignancy Questionnaire Additional pharmacovigilance activities: None
Transmission to third parties (horizontal transmission)	Routine risk minimisation measures: SmPC Section 4.4, Special warnings and precautions for use SmPC Section 4.6, Fertility, pregnancy and lactation SmPC Section 5.2, Pharmacokinetic properties SmPC Section 6.6, Special precautions for disposal and other handling PL Section 2 Legal status: Medicinal product subject to restricted medical prescription. Treatment should be administered in a qualified treatment centre by a physician experienced in the treatment of haemophilia. Additional risk minimisation measures: Guide for Healthcare Professionals Patient Guide Patient Card	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: None Additional pharmacovigilance activities: None
Germline transmission	Routine risk minimisation measures: SmPC Section 4.4, Special warnings and precautions for use SmPC Section 4.6, Fertility, pregnancy and lactation SmPC Section 5.2, Pharmacokinetic properties PL Section 2 Legal status: Medicinal product subject to restricted medical prescription. Treatment should be administered in a qualified treatment centre by a physician experienced in the treatment of haemophilia. Additional risk minimisation measures: Guide for Healthcare Professionals Patient Guide Patient Card	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: Pregnancy Questionnaire Additional pharmacovigilance activities: None

Safety Concern	Risk Minimisation Measures	Pharmacovigilance Activities
Missing Information		
Long-term safety	Routine risk minimisation measures: SmPC Section 4.4, Special warnings and precautions for use PL Section 2 Legal status: Medicinal product subject to restricted medical prescription. Treatment should be administered in a qualified treatment centre by a physician experienced in the treatment of haemophilia. Additional risk minimisation measures: Guide for Healthcare Professionals Patient Guide	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: None Additional pharmacovigilance activities: None

2.7.4. Conclusion

The CAT considers that the risk management plan version 1.0 is acceptable.

The CHMP endorses the CAT conclusion on the RMP as described above.

2.8. Pharmacovigilance

2.8.1. Pharmacovigilance system

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

2.8.2. Periodic Safety Update Reports submission requirements

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 27.12.2023. 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, Durveqtix (Fidanacogene elaparvovec) is included in the additional monitoring list for the following reasons: New biological, new active substance and Conditional Marketing Authorisation.

Therefore, the summary of product characteristics and the package leaflet include 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

Durveqtix is indicated for the treatment of severe and moderately severe haemophilia B (congenital factor IX deficiency) in adult patients without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74.

3.1.2. Available therapies and unmet medical need

The treatment of haemophilia B is based on IV administration of either plasma-derived or recombinant FIX protein replacement therapy to raise the FIX:C activity level in order to achieve either resolution of bleeding (on-demand treatment) or prevention of bleeding (prophylaxis treatment).

These treatment options for haemophilia B have several limitations. Treatment with prophylactic regular IV injections of FIX is not curative and very demanding due to the need for frequent IV infusions and concomitant risk for infection and thromboses related to the placement of indwelling catheters. Periodic or regular FIX infusion results in peaks and troughs in plasma factor levels allowing for breakthrough bleeding episodes. Due to these factors, poor adherence to treatment is a concern and a major contributing factor to failure of prophylaxis, associated with increased risk of bleeding and subsequent joint damage, thereby adding to the all-cause morbidity and mortality rate.

There is also a risk of developing neutralizing antibodies (nAbs) against the administered FIX. The burden of the disease is high, both for the individual subject and their families, and for society. Due to (long-term) impairments in mobility and functional status, subjects may not be able to fully participate in social activities, such as sports, school, or work. Living with haemophilia can have a substantial effect on mental wellbeing, particularly among young people and signs of major depressive disorder are not uncommon. The economic burden for the society is significant.

In February 2023, the gene therapy product etranacogene dezaparvovec was conditionally approved in the EU for the treatment of adult patients with moderate to severe haemophilia B irrespective of detectable antibodies against AAV5 vector.

There remains an unmet medical need in haemophilia B.

3.1.3. Main clinical studies

The main evidence for efficacy derives from the ongoing single pivotal study (C0371002). For efficacy assessment, in the pivotal study C0371002 an intra-patient control approach was chosen, with respective baseline data having been generated in the non-interventional study C0371004, thus both C0371002 and C0371004 are considered main clinical studies.

Study **C0371004** is an open label, non-investigational product lead-in study to prospectively evaluate efficacy and select safety data of current FIX (Haemophilia B Cohort) or Factor VIII (Haemophilia A Cohort) prophylaxis replacement therapy in the usual care setting of moderately severe to severe adult haemophilia B participants (FIX:C ≤ 2%) who are negative for neutralizing antibodies (nAb) to AAV-Spark100 or moderately severe to severe adult haemophilia A participants (FVIII:C ≤ 1%) who are negative for nAb to SB-525 capsid (AAV6). Only the Haemophilia B Cohort is relevant for this MAA.

Of 59 patients who completed study C0371004, eight (8) did not continue to C0371002 for "missing reason", the remaining 51 were screened for study C0371002. Of these, 5 were screen failures, and one patient withdrew consent. Based on ABR_{total}, AIR and annualized FIX consumption (IU/kg) data provided for C0371004 completers who did continue to the pivotal trial, and patients who discontinued study C0371004 or completed, but did not continue to the pivotal trial, a selection bias favouring fidanacogene elaparvovec treatment is not assumed.

Study **C0371002** is a single-arm, open-label, multi-site, single-dose phase 3 trial in adult male haemophilia B patients with endogenous factor IX activity of ≤2%, comparing the efficacy of a single intravenous infusion of fidanacogene elaparvovec with routine FIX prophylaxis in adult male participants from the lead-in study (C0371004). This study includes a total of 6 years of follow up post fidanacogene elaparvovec infusion. The trial is ongoing; at time of updated data cut 30-Aug-2023, n=45 patients have been enrolled and treated with the actual titre concentration, and were evaluable for the key endpoints. The favourable effects section reflects the updated data.

3.2. Favourable effects

Efficacy of fidanacogene elaparvovec in the treatment of haemophilia B was demonstrated by annualised bleeding rate for all, i.e. treated and untreated bleeds (ABR_{total}). In the updated data with cut-off date 30-Aug-2023, the model-derived mean ABR_{total} was 1.28 (95% CI: 0.57, 1.98) for fidanacogene elaparvovec versus 4.42 (95% CI: 1.80, 7.05) for prophylaxis, treatment difference was -3.15 (95% CI: -5.46, -0.83); p-value=0.0081) and percentage reduction ABR_{total} was 71.12% (95% CI: 50.09%, 83.029%; p-value< 0.0001). ABR_{total} (model derived estimate [95% CI]) was 0.40 (0.05, 0.76) for year 2 (n=44), 0.564 (0.04, 1.24) for year 3 (n=40), and 0.32 (-0.04, 0.68) for year 4 (n=15). Over time, 27 out of 45 patients (60%) remained without bleeds in the observation period.

Mean AIR from Week 12 to Month 15 post fidanacogene elaparvovec infusion was significantly reduced to 4.54 from a mean AIR of 58.83 in pre-infusion period (p<0.0001). AIR (mean [±SD]) was 6.52 (18.697) for year 2 (n=44), 4.9 (14.871) for year 3 (n=40), and 1.4 (4.691) for year 4 (n=15).

The annualized total FIX consumption (mean [±SD]) from Week 12 to Month 15 post-fidanacogene elaparvovec infusion was reduced to 239.039 (539.617) IU/kg compared to 3168.56 (1635.545) IU/kg during FIX prophylaxis in the Dosed Analysis Set. The treatment difference was -2929.17 (95% CI -3397.49, -2460.85). The mean (±SD) annualized FIX consumption at year 2 (n=44) post-fidanacogene elaparvovec infusion was reduced to 301.34 (852.206) IU/kg, at year 3 (n=40) was reduced to 219.01 (570.946) IU/kg, and at year 4 (n=15) to 230.51 (498.669) IU/kg.

Mean steady-state (geometric mean of all valid measurements from Week 12 to Month 15 post fidanacogene elaparvovec infusion) FIX:C from all 3 assays were significantly higher than the fixed

threshold of 5% (mean [±SD] and p-value for the 3 assays: one-stage assay with Actin-FSL reagent: 12.65 [9.05], p<0.0001; one-stage assay with SynthAsil reagent: 25.986 [16.92], p<0.0001; and chromogenic assay: 13.42 [10.33], p<0.0001]).

3.3. Uncertainties and limitations about favourable effects

The trial is a single pivotal trial with multiple protocol and SAP amendments including changes to the primary and (key) secondary endpoints and timing of their assessment.

The primary estimand for ABR_{total} only considers subjects while they have been on study and while they had not resumed prophylaxis.

Six out of 45 (13.3%) participants resumed FIX prophylaxis therapy. These participants were between 18 to 47 years old, were not geographically clustered, received corticosteroid treatment and showed peak FIX:C >5% in all three assays. Except for receiving at least one course of corticosteroid treatment, this population appeared comparable to the rest of the study participants. No clear trend has emerged that would suggest a population/characteristic that would increase the probability of a participant to **resume prophylaxis**, however, the small sample size precludes a definitive conclusion.

As commonly observed for recombinant FIX products and other approved AAV-based gene therapies for haemophilia, there is a relevant **FIX:C assay discrepancy** to determine transgene derived FIX:C. Patient level data indicate a patient-individual and inconsistent pattern regarding ActinFSL OSA and chromogenic assay results.

In terms of expectations relayed by additional experts' consultations, clinical relevance of the achieved FIX:C levels, i.e. suitability of the achieved FIX:C levels to provide **sustained bleed protection** in general and **protection against intracranial bleeds** in particular, is yet to be determined.

Updated data on (geometric) mean FIX:C indicates a slight decrease over time in all assays, with an increase of patients with FIX:C below 5% from approx. 29% at w52 to approx. 30% at w104 in both ActinFSL OSA and chromogenic assay.

3.4. Unfavourable effects

The most important unfavourable effects of fidanacogene elaparvovec documented in the clinical trial program were G1/G2 increase of ALT and AST in the majority of participants after infusion.

3.5. Uncertainties and limitations about unfavourable effects

There are uncertainties resulting from the lack of a control arm in the pivotal study and the lack of a proper clinical dose finding study for evaluation of the safe and effective dose of fidanacogene elaparvovec to be applied.

The documented high number of liver enzyme increases in the clinical trials, requiring treatment with oral and/or systemic corticosteroids, and the ultrasound findings of liver parenchyma damage in at least two trial participants during the 2 to 6 years observation period, reported as clinically relevant, trigger some uncertainty with respect to safety.

Vector integration was observed in nonclinical studies with cynomolgus macaques and haemophilia B dogs. Integration site analysis studies performed on liver biopsies of both cynomolgus monkeys and haemophilia B dogs showed low level of vector integration, a random nature of integration, and no

evidence of clonal expansion. While recombinant AAV are not expected to integrate their genome in host cells at high frequency, all integration events could still potentially contribute to tumoral transformation.

3.6. Effects Table

Table 8. Effects Table for Durveqtix, treatment of haemophilia B (data cut August 2023).

Effect	Short Description	Unit	Treatment w12 to m15	Control (lead-in)	Uncertainties/ Strength of evidence	References
Favourable Effects						
ABR_{total}	Annualised rates of all bleeds (mean [95% CI])	Bleeds/ year	1.28 (0.57, 1.98)	4.42 (1.80, 7.05)	SoE: relevant endpoint Unc: Clinical relevance of estimand in the light of 13.3% resuming prophylaxis	Studies C0371002 and C0371004
FIX activity	FIX:C (mean [SD])	% of normal	12.65 (9.05)	≤2	Unc: FIX:C assay discrepancy, use of geometric mean, clinical relevance	Study C0371002

Effect	Short Description	Unit	Treatment	Control	Uncertainties/ Strength of evidence	References
Unfavourable Effects						
Summary of all AEs		N	283 in N=52 (86.7%) subjects	N/A		Studies C0371002, C0371005 and C0371003
Severe AEs		N (%)	11 (18.3)	N/A		Studies C0371002, C0371005 and C0371003
TEAEs		N	58 in N=32 (53.3%) subjects	N/A	TEAE increase by BMI despite comparable dose of fidanacogene elaparvovec and higher doses of corticosteroids	Studies C0371002 and C0371005 Pivotal study
New/progress of liver parenchyma damage	Ultrasound scan	N=3				

Effect	Short Description	Unit	Treatment	Control	Uncertainties/ Strength of evidence	References
ALT increase AST increase			43 in N=60 (71.7%) subjects 44 in N=60 (73.3%) subjects	N/A	Corticosteroid treatment of elevated transaminases in 31 patients	Studies C0371002, C0371005 and C0371003

Abbreviations: Annualised bleeding rate (ABR), Adverse Events (AEs), confidence interval (CI), factor IX (FIX), factor IX activity (FIX:C), standard deviation (SD), Treatment Emergent Adverse Events (TEAEs)

3.7. Benefit-risk assessment and discussion

3.7.1. Importance of favourable and unfavourable effects

Submitted data of the pivotal trial demonstrated non-inferiority of fidanacogene elaparvovec over FIX prophylaxis regarding ABR_{total}, with significant reductions of AIR and annualized FIX consumption. Significant reduction or even absence of bleeding events with a corresponding reduction of FIX infusions in the absence of regular prophylactic FIX infusions meets the expectations of haemophilia patients for disease amelioration by gene therapies.

Stable, clinically relevant levels of FIX activity are expected to provide sustained bleed protection in general and better protection against intracranial bleeds. Available data reports FIX:C constantly above 5% of normal for approx. 2/3 of the pivotal trials study population. There are currently no major concerns about the identified unfavourable effects of fidanacogene elaparvovec.

3.7.2. Balance of benefits and risks

The updated efficacy data provided during the assessment confirm the results from the initial analyses in terms of ABR, AIR, FIX consumption, and FIX:C levels w12-m15. No additional subject had to resume prophylaxis at the new cut-off. ABR and FIX activity were comparable for the two BMI cohorts; the safety profile shows more incidents of higher transaminases in the higher BMI cohort, supporting the cautious approach limiting the vector dose for higher BMI patients, but is still considered acceptable.

The reported liver enzyme increases as an indicator of potential FIX activity decreases due to suggested immune response are considered as fidanacogene elaparvovec related hepatotoxicity. According to the SmPC, patients with severe hepato-biliary-renal impairment are excluded from therapy with Durveqtix. Patients who received treatment will be adequately monitored as per the SmPC provided instructions.

The beneficial treatment effect was sustained at the 2-year analysis. That uncertainties persist for novel therapies in general and for gene therapy approaches to haemophilia in particular is to be expected and one of the reasons for granting a conditional MA.

The favourable effects of fidanacogene elaparvovec are considered able to outweigh the identified unfavourable effects.

3.7.3. Additional considerations on the benefit-risk balance

Quality of evidence

The main body of evidence for efficacy is derived from an ongoing, single pivotal trial with a limited number of patients enrolled and shortcomings in the conduct as discussed in detail previously.

Precision of effect size

The lower and upper bound of the 95% confidence interval of ABR for total bleeds (treated and untreated) was -5.44 and -0.81. This estimated precision is supported by a number of additional analyses and therefore considered reliable. Moreover, the study was planned to demonstrate non-inferiority over FIX prophylaxis.

Clinical meaningfulness of the endpoint

The key endpoints are clinically relevant, and the observed reduction in bleeding events and associated reduction of exogenous FIX consumption can be directly linked to the mechanism of action.

However, clinical relevance of the achieved FIX:C levels, i.e. suitability of the achieved FIX:C levels to provide sustained bleed protection in general and protection against intracranial bleeds in particular, is yet to be determined. In addition, (as expected from the nature of the gene therapy) a decline of FIX levels over time was observed. Thus, the durability of the observed beneficial haemostatic effects is yet to be determined.

Duration of efficacy

Durability of the observed effects can currently not be concluded due to limited and too immature data, and has to be further substantiated by long-term data from the ongoing and planned clinical trials as well as from post-marketing observations.

Safety: Exposure

Based on the mechanism of action of fidanacogene elaparvovec the liver health in the intended target population is of particular importance. Given the available results from Cohort 1 of the LTFU study, uncertainty with regard to safety and effectiveness of the recommended corticosteroid therapy regimen for drug related immunogenicity, measured by elevation of hepatic transaminases, remains.

Safety: Length of follow-up

This initial marketing application includes safety data from 41/45 of participants who have accrued over 2 years of follow-up. As the effects of AAV-gene therapy are not reversible and life-long, this dataset is only able to characterise the short and mid-term safety.

Target population vs study population

Only subjects who are generally healthy and have no history of FIX inhibitors were eligible for the clinical trial programme. The age distribution is skewed towards younger participants with only limited data in older subjects. Most participants of the pivotal trial (84.4%) were <45 years of age.

Pharmacologic rationale

The pharmacologic rationale of fidanacogene elaparvovec, i.e. AAV based transduction of liver cells with subsequent transgene derived haemostatic effect, is strong.

Natural history/course of the disease

Severe or moderately severe haemophilia B is characterised by a predisposition to spontaneous and traumatic bleeding events, which can lead to considerable morbidity and also, especially in case of

intracerebral bleeds, mortality. It is a lifelong condition that can be ameliorated by the substitution of exogenous coagulation factor IX, which is standard of care in the EU. The natural history renders the freedom of bleeds in the absence of prophylaxis that has been experienced by part of the participants most likely to be due to fidanacogene elaparvovec treatment.

Based on the issues listed above, comprehensive efficacy and safety data are lacking in the current MAA. The long-term durability of the treatment effect and long-term safety are still unknown factors.

Conditional marketing authorisation

As comprehensive data on the product are not available, a conditional marketing authorisation was proposed by the CAT during the assessment and agreed by the applicant.

The product falls within the scope of Article 14-a of Regulation (EC) No 726/2004 concerning conditional marketing authorisations, as it aims at the treatment of a seriously debilitating disease.

Furthermore, the CAT considers that the product fulfils the requirements for a conditional marketing authorisation:

- The benefit-risk balance of the product is positive, as discussed.

- It is likely that the applicant will be able to provide comprehensive data
  For a fully comprehensive data set, clinical data from more patients treated with the commercial process are required. The applicant has agreed to provide the following follow-up data from the pivotal trial:

  Study C0371002: 6-year data from subjects with dose calculated using actual batch concentration, and at least 34-month post-infusion data for all participants dosed using the fixed nominal concentration, both cutoff date June 2028. In addition, for study C0371003 5-year data from subjects dosed with 5×10^{11} vector genomes per kg (vg/kg) of body weight will be provided. In addition, the applicant also agreed to conduct studies C0371007 and C0371017 as post-authorization efficacy studies. The results from studies C0371002, C0371003, and C0371017 will be able to elucidate long-term efficacy and safety outcomes.

- Fulfilment of unmet medical need
  The applicant has adequately discussed the unmet medical need with regard to disease and treatment burden which are not addressed by currently authorised FIX products with standard or extended half-life such as nonacog alfa (BeneFIX), nonacog gamma (Rixubis), eftrenonacog alfa (Alprolix), albutrepenonacog alfa (Idelvion), nonacog beta pegol (Refixia) and human plasma-derived factor IX products. Therefore, it is agreed that Durveqtix would bring a major therapeutic advantage over these products.

  Hemgenix, another gene therapy medicinal product in the treatment of haemophilia B is currently approved under a conditional MA. In line with Section 4.1.2 c) of the 'Guideline on the scientific application and the practical arrangements necessary to implement Commission Regulation (EC) No 507/2006 on the conditional marketing authorisation for medicinal products for human use falling within the scope of Regulation (EC) No 726/2004' and upon request, the applicant provided a justification why Durveqtix should be regarded as addressing the existing unmet medical need to a similar or greater extent than what is understood for the already conditionally authorised product Hemgenix in the form of an Indirect Treatment Comparison (ITC) with Hemgenix. Where feasible, this was based on an unanchored matching-adjusted indirect comparison (MAIC) for bleed-related endpoints, resulting in an effective sample size of 15 subjects. For parameters where a MAIC was not feasible, a side-by-side comparison was provided. The methodological difficulties in the conduct of an ITC using less-comprehensive data and resulting uncertainties are acknowledged.

Despite a *"~10-15% absolute difference between both products […] maintained over time […] through 3 years post-infusion for both products"* regarding transgene FIX:C levels as measured with one-stage SynthASil assay, similarity in ABR and other bleed related endpoints is agreed.

With regard to the safety outcomes, protocol specifications required more stringent liver function monitoring and had lower thresholds for corticosteroid triggers, hence a meaningful comparison is hampered. However, the proportion of subjects reporting ALT or AST elevations as TEAEs are similar in both programmes, and Durveqtix caused no infusion related reactions in contrast to a significant proportion observed with Hemgenix administration.

In conclusion and interpreting the ITC with caution, Durveqtix is considered able to address the unmet medical need for the treatment of haemophilia B to a similar extent as Hemgenix.

- The benefits to public health of the immediate availability of the medicinal product outweigh the risks inherent in the fact that additional data are still required.
  The observed beneficial effects on the bleeding frequency and the benign medium term safety profile are able to offset the need for additional data confirming the long-term efficacy and safety.

The CHMP endorsed the CAT conclusion on conditional marketing authorisation as described above.

3.8. Conclusions

The overall benefit/risk balance of Durveqtix is positive, subject to the conditions stated in section 'Recommendations'.

The CHMP endorsed the CAT conclusion on Benefit Risk balance as described above.

Divergent position(s) are appended to this report.

4. Recommendations

Similarity with authorised orphan medicinal products

The CAT by consensus is of the opinion that Durveqtix is not similar to Alprolix, Idelvion and Hemgenix within the meaning of Article 3 of Commission Regulation (EC) No. 847/2000.

A 3rd party intervention was received during the evaluation of the MAA, claiming that fidanacogene elaparvovec is to be considered similar to etranacogene dezaparvovec (Hemgenix). CAT/CHMP considered this intervention and concluded that the arguments put forward do not alter the conclusion that fidanacogene elaparvovec is not similar to etranacogene dezaparvovec.

The CHMP endorsed the CAT conclusion on similarity as described above.

Outcome

Based on the CAT review of data on quality, safety and efficacy, the CAT considers by majority decision that the benefit- risk balance of Durveqtix is favourable in the following indication:

Durveqtix is indicated for the treatment of severe and moderately severe haemophilia B (congenital factor IX deficiency) in adult patients without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74.

The CAT therefore recommended the granting of the conditional marketing authorisation subject to conditions; based on that draft opinion adopted by the CAT and the review of data on quality, safety and efficacy, the CHMP also considers by majority decision that the benefit- risk balance of Durveqtix in the treatment of severe and moderately severe haemophilia B (congenital factor IX deficiency) in adult patients without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74 is favourable and therefore recommends the granting of the conditional marketing authorisation subject to the following conditions:

Conditions or restrictions regarding supply and use

Medicinal product subject to restricted medical prescription (see Annex I: Summary of Product Characteristics, section 4.2).

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 marketing authorisation holder (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.

- **Additional risk minimisation measures**

Prior to the launch of Durveqtix in each Member State the Marketing Authorisation Holder (MAH) must agree about the content and format of the educational programme, including communication media, distribution modalities, and any other aspects of the programme, with the National Competent Authority.

The educational programme is aimed at providing information on the safe use of Durveqtix and to inform about important risks associated with Durveqtix.

The MAH shall ensure that in each Member State where Durveqtix is marketed, all healthcare professionals and patients/carers who are expected to prescribe, use or oversee the administration of Durveqtix have access to/are provided with the following educational package. These documents will be translated in the local language to ensure understanding of proposed mitigation measures by physicians and patients:

- Physician Educational Material

- Patient Information Pack

The Physician Educational Material consists of:

- The Summary of Product Characteristics
- The Guide for Healthcare Professionals
- The Patient Guide
- The Patient Card

The Guide for Healthcare Professionals

- Patients should be selected for treatment with Durveqtix based on the absence of pre-existing antibodies to AAVRh74var using a validated assay and status of liver health based on laboratory and imaging data.
- To inform of the important identified risks of hepatotoxicity and the important potential risks of development of factor IX inhibitors, thromboembolic events, risk of malignancy in relation to vector integration in the DNA of body cells, transmission to third parties (horizontal transmission) and germline transmission, and missing information of long-term safety and details on how these risks can be minimised.
- Before a treatment decision is made, the healthcare professional should discuss the risks, benefits, and uncertainties of Durveqtix with the patient when presenting Durveqtix as a treatment option, including:
    - That no predictive factors for no or low responders have been identified. Patients who do not respond are still exposed to long-term risks.
    - That the long-term treatment effects cannot be predicted.
    - That there would be no plans to re-administer the medicinal product for patients who do not respond or have lost the response.
    - Reminding patients about the importance to enrol in a registry for follow-up of long-term effects.
    - That Durveqtix use will require in some cases co-administration of corticosteroids to manage the liver damage that this medicinal product might induce. This requires adequate monitoring of patients and careful consideration of other co-medications, herbal supplements, and/or alcohol to minimise the risk of hepatoxicity and a potential reduced therapeutic effect of Durveqtix.
    - That the patient should be routinely tested for factor IX inhibitors development after Durveqtix treatment.
    - That the patient will be provided the patient guide and the patient card by the healthcare professional.

The Patient Information Pack consists of

- The Patient Information Leaflet
- The Patient Guide
- The Patient Card

The Patient Guide:

- Importance of fully understanding the benefits and risks of Durveqtix treatment, what is known and not yet known about the long-term effects, related to safety and efficacy.
- Therefore, before a decision is made about starting on the therapy the doctor will discuss with the patient the following:
    - That Durveqtix will, in some cases, require co-treatment with corticosteroids to overcome the liver damage that this medicine may produce, and that the doctor will ensure that patients are available for regular blood tests to check responses to Durveqtix and assess liver health. Patients should inform the healthcare professional about current use of corticosteroids or other immunosuppressants. If the patient cannot take corticosteroids, the doctor may recommend alternative medicines to manage problems with the liver.
    - That not all patients may benefit from treatment with Durveqtix and the reasons for this have not been established. Patients not responding to treatment will still be exposed to long-term risks of Durveqtix.
    - Details how the important potential risks of development of factor IX inhibitors, thromboembolic events, risk of malignancy in relation to vector integration in the DNA of body cells, transmission to third parties (horizontal transmission) and germline transmission can be recognised and minimised by regular monitoring as recommended by doctors.
        - The patient should seek immediate medical advice for any symptoms suggestive of a thromboembolic event.
        - Male patients or their female partners should use barrier contraception for six months after administration of Durveqtix.
        - That Durveqtix has a viral vector component, and it may be associated with an increased risk of malignant tumour. Regular liver monitoring for at least 5 years after Durveqtix treatment is needed in patients with pre-existing risk factors for hepatocellular carcinoma.
        - Patients must not donate blood, semen, or organs, tissues, and cells for transplantation.
    - That the Patient Card should be carried by the patient at any time and shared with any doctor or nurse whenever the patient has a medical appointment.
    - The importance to participate in the patients' registry for long-term surveillance of 15 years.

The Patient Card:

- This card is to inform healthcare professionals that the patient has received Durveqtix for haemophilia B.
- The patient should show the patient card to a doctor or a nurse whenever they have an appointment.
- The patient should seek medical advice for any symptoms suggestive of a thromboembolic event.
- That the patient should have regular blood tests and examinations as directed by their doctor.
- The card should warn healthcare professionals that the patient may undergo treatment with corticosteroids for minimising the risk of hepatotoxicity with Durveqtix.
- The patient must not donate blood, semen, organs, tissues and cells for transplantation.
- Male patients should ensure that they use a barrier method of contraception for 6 months after receiving Durveqtix.

- **Obligation to conduct post-authorisation measures**

The MAH shall complete, within the stated timeframe, the below measures:

Description	Due date
Post-authorisation efficacy study (PAES): In order to further characterise the long-term efficacy and safety of Durveqtix in adults with severe and moderately severe haemophilia B (congenital factor IX deficiency) without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74, the MAH should conduct and submit the final results of registry-based study C0371007, according to an agreed protocol.	31 Dec 2045
Post-authorisation efficacy study (PAES): In order to further characterise the long-term efficacy and safety of Durveqtix in adults with severe and moderately severe haemophilia B, the MAH should submit the final results of Study C0371017, which includes patients who have been treated with Durveqtix in all MAH-sponsored clinical trials.	31 Mar 2040

Specific Obligations to complete post-authorisation measures for the conditional marketing authorisation

This being a conditional marketing authorisation and pursuant to Article 14-a of Regulation (EC) No 726/2004, the MAH shall complete, within the stated timeframe, the following measures:

Description	**Due date**
In order to confirm the efficacy and safety of Durveqtix in adults with severe and moderately severe haemophilia B (congenital factor IX deficiency) without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74, the MAH should submit interim results (6 years of data) of pivotal Study C0371002 with 45 subjects who received a dose calculated using actual batch concentration and at least 34-month data of patients who received a dose based on nominal concentration dosing.	31 Dec 2028
In order to confirm the efficacy and safety of Durveqtix in adults with severe and moderately severe haemophilia B (congenital factor IX deficiency) without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74, the MAH should submit the final results (5 years of data) of long-term follow-up Study C0371003 with 14 subjects who received 5 × 1011 vector genomes per kg (vg/kg) of body weight.	31 Jan 2025

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

Not applicable.

New Active Substance Status

Based on the review of available data on the active substance, the CAT considers that Fidanacogene elaparvovec is to be qualified as a new active substance in itself as it is not a constituent of a medicinal product previously authorised within the European Union.

The CHMP endorses the CAT conclusion on the new active substance status claim.

Divergent positions

CAT Divergent position to the majority recommendation is appended to this report.

CHMP Divergent position to the majority recommendation is appended to this report.

5. Appendices

5.1. CAT Divergent position to the majority recommendation

DIVERGENT POSITION DATED 24 May 2024

Durveqtix EMEA/H/C/004774/0000

The undersigned members of the CAT did not agree with the CAT's positive draft opinion recommending the granting of the conditional marketing authorisation of Durveqtix (fidanacogene elaparvovec) for the following indication:

Durveqtix is indicated for the treatment of severe and moderately severe haemophilia B (congenital factor IX deficiency) in adult patients without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74.

The reasons for the divergent opinion were the following:

Although it is acknowledged that fidanacogene elaparvovec shows efficacy as determined by the annualised bleeding rate (ABR), the data and justifications provided by the applicant to demonstrate that Durveqtix addresses the unmet medical need at least to a similar extent as an already conditionally authorized ATMP in the same patient population are not considered sufficient. This particularly pertains to observed FIX:C levels which are considered inferior and lack robust information on durability. The inherent risks of the lower FIX:C based on reported non-linear relationship between FIX:C and bleed rate are not outweighed by the observed similarity in ABR in the very limited follow-up period.

CAT Members expressing a divergent opinion:

Jan Müller-Berghaus

Heli Suila

Kerstin Sollerbrant

5.2. CHMP Divergent position to the majority recommendation

DIVERGENT POSITION DATED 30 May 2024

Durveqtix EMEA/H/C/004774/0000

The undersigned members of the CHMP did not agree with the CHMP's positive opinion recommending the granting of the conditional marketing authorisation of Durveqtix (fidanacogene elaparvovec) for the following indication:

Durveqtix is indicated for the treatment of severe and moderately severe haemophilia B (congenital factor IX deficiency) in adult patients without a history of factor IX inhibitors and without detectable antibodies to variant AAV serotype Rh74.

The reasons for the divergent opinion were the following:

Although it is acknowledged that fidanacogene elaparvovec shows efficacy as determined by the annualised bleeding rate (ABR), the data and justifications provided by the applicant to demonstrate that Durveqtix addresses the unmet medical need at least to a similar extent as an already conditionally authorized ATMP in the same patient population are not considered sufficient. This particularly pertains to observed FIX:C levels which are considered inferior and lack robust information on durability. The inherent risks of the lower FIX:C based on reported non-linear relationship between FIX:C and bleed rate are not outweighed by the observed similarity in ABR in the very limited follow-up period.

CHMP Members expressing a divergent opinion:

Jan Müller-Berghaus

Simona Badoi

Outi Mäki-Ikola

Martina Weise

Hrefna Gudmundsdottir