EUROPEAN MEDICINES AGENCY
SCIENCE MEDICINES HEALTH

6 July 2012
EMA/CHMP/413121/2012

Assessment report

NovoThirteen

International non-proprietary name: **catridecacog**

Procedure No. **EMEA/H/C/002284**

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

7 Westferry Circus • Canary Wharf • London E14 4HB • United Kingdom
Telephone +44 (0)20 7418 8400 **Facsimile** +44 (0)20 7523 7455
E-mail info@ema.europa.eu **Website** www.ema.europa.eu

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List of abbreviations

AE	adverse event
aPTT	activated partial thromboplastin time
ASAT	aspartate aminotransferase
AUC	area under the plasma activity/concentration-time curve
$AUC_{0\text{-}24h}$	area under the plasma activity/concentration-time curve from zero to 24 hours
$AUC_{0\text{-}28days}$	area under the plasma activity/concentration-time curve from zero to 28 days
$AUC_{0\text{-}\infty}$/$AUC_{0\text{-}INF}$	area under the plasma activity/concentration-time curve from zero to infinity
C_0	initial concentration
CD	congenital FXIII deficiency
CI	confidence interval
CL	clearance
C_{max}	the maximum drug concentration in serum/plasma
CNS	central nervous system
CV	coefficient of variation
CVS	cardio vascular system
ELISA	enzyme-linked immunosorbent assay
EU	Endotoxin Unit
FFP	fresh frozen plasma
FAS	Full Analysis Set
FXIII	coagulation factor XIII
GLP	good laboratory practice
HCP	Host Cell Proteins
IASMS	Initial active substance manufacturing site
ICH	international Conference of Harmonization
IE-HPLC	Ion-exchange High Performance Liquid Chromatography
INR	International normalized ratio
IU	international units
IV	intravenous(ly)
JP	Japanese Pharmacopoeia
LAL	Limulus Amoebocyte Lysate
MCB	Master Cell Bank
MedDRA	Medical Dictionary for Regulatory Activities
MRT	mean residence time
PD	pharmacodynamic(s)
Ph Eur	European Pharmacopoeia
PK	pharmacokinetic
PP	per protocol
ppm	parts per million
PT	prothrombin time
rFXIII	recombinant human coagulation factor XIII
$rFXIII_{IASMS}$	rFXIII active substance produced at the initial manufacturing site
$rFXIII_{NN}$	active substance produced at Novo Nordisk
rFXIIIa°	Non-proteolytically activated rFXIII
rFXIIIa*	thrombin-activated rFXIII
SAE	serious adverse event
SAP	statistical analysis plan
SD	standard deviation
SE-HPLC	Size Exclusion High Performance Liquid Chromatography
SOC	system organ class
$t_{½}$	terminal half-life
TEG	thromboelastography
USP	United States Pharmacopoeia
V_{ss}	volume of distribution at steady state
WCB	Working Cell Bank
WFI	Water for Injections

1. Background information on the procedure

1.1. Submission of the dossier

The Applicant Novo Nordisk A/S submitted on 3 May 2011 an application for Marketing Authorisation to the European Medicines Agency (EMA) for NovoThirteen, through the centralised procedure falling within the Article 3(1) and point 1 of Annex of Regulation (EC) No 726/2004. The eligibility to the centralised procedure was agreed upon by the EMA/CHMP on 19 November 2009.

NovoThirteen, was designated as an orphan medicinal product EU/3/03/179 on 12 December 2003 in the following indication: treatment of hereditary factor XIII deficiency.

The Applicant applied for the following indication: *prophylactic treatment of bleeding in patients 6 years and above with congenital factor XIII A-subunit deficiency*.

In connection with the review of the orphan designation criteria by the Committee on Orphan Medicinal Products (COMP) at its meeting of 10-11 July 2012, the Applicant requested the Commission to remove the product from the Community Register of Orphan Medicinal Products on 11 July 2012

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 bibliographic literature supporting certain tests or studies.

Information on Paediatric requirements

Pursuant to Article 7 of Regulation (EC) No 1901/2006, the application included an EMA Decision(s) P/273/2010 on the agreement of a paediatric investigation plan (PIP)

At the time of submission of the application, the PIP P/273/2010 was not yet completed as some measures were deferred.

Information relating to orphan market exclusivity

Similarity

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

New active Substance status

The Applicant requested the active substance catridecacog contained in the above medicinal product to be considered as a new active substance in itself.

Protocol Assistance

The Applicant received Protocol Assistance from the CHMP on 25 of January 2007; 21 of January 2010; 22 of April 2010. The Protocol Assistance pertained to quality, non-clinical and clinical aspects of the dossier.

Licensing status

A new application was filed in the United States of America in February 2011.

1.2. Steps taken for the assessment of the product

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

Rapporteur: **Philippe Lechat**

Co-Rapporteur: **Jan Mueller-Berghaus**

- The application was received by the EMA on 3 May 2011.
- The procedure started on 25 May 2011.
- The Rapporteur's first Assessment Report was circulated to all CHMP members on 16 August 2011. The Co-Rapporteur's first Assessment Report was circulated to all CHMP members on 15 August 2011.
- During the meeting on 22 September 2011, the CHMP agreed on the consolidated List of Questions to be sent to the Applicant. The final consolidated List of Questions was sent to the Applicant on 23 September 2011.
- The Applicant submitted the responses to the CHMP consolidated List of Questions on 15 December 2011.
- The Rapporteurs circulated the Joint Assessment Report on the Applicant’s responses to the List of Questions to all CHMP members on 02 February 2012.
- During the CHMP meeting on 16 February 2012, the CHMP agreed on a List of Outstanding Issues to be addressed by the Applicant.
- The Applicant submitted the responses to the CHMP List of Outstanding Issues on 19 March 2012.
- The Rapporteurs circulated the Joint Assessment Report on the Applicant’s responses to the List of Outstanding Issues to all CHMP members on 02 April 2012.
- During the BWP meeting on 10 April 2012, outstanding issues were addressed by the Applicant during an oral explanation before the BWP.
- During the CHMP meeting on 19 April 2012, the CHMP agreed on a second List of Outstanding Issues to be addressed by the Applicant.
- The Applicant submitted the responses to the second CHMP List of Outstanding Issues on 27 April 2012.
- The Rapporteurs circulated the Joint Assessment Report on the Applicant’s responses to the second List of Outstanding Issues to all CHMP members on 02 May 2012 and the updated version on 18 May 2012.

- During the meeting on 21-24 May 2012, 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 NovoThirteen on 24 May 2012.

2. Scientific discussion

2.1. Introduction

Congenital coagulation factor XIII (FXIII) deficiency is a rare autosomal recessive bleeding disorder with an estimated prevalence of 1 per 2 to 5 million individuals. Congenital FXIII deficiency is usually caused by mutations in the F13A1 gene (6p24.2-p23) encoding the catalytic A subunit, but mutations have also been found in the F13B gene (1q31-q32.1) encoding the B subunit. The phenotype is less severe when the F13B gene is mutated. In patients with FXIII A-subunit deficiency the FXIII activity level is lower than 3 to 5% (or approximately 0.04 to 0.06 IU/mL), compared with a wide range of approximately 50% to 220% (0.60 to 2.60 IU/mL) in the normal population.

Diagnosis is based on quantitative FXIII activity measurement and antigen assays. Common clotting assays such as activated Partial Thromboplastin Time (aPTT) and Prothrombin Time (PT) are normal and cannot be used for the screening. The clot solubility test may also be used (clot is stable for less than 24 hours in case of FXIII deficiency). However, the assay is qualitative, and when performed correctly the test is positive only when the FXIII activity in the sample is close to zero. Differential diagnoses mainly include the other congenital coagulation factor deficiencies: fibrinogen, factors II, V, VII, X, XI, VIII and IX. Antenatal diagnosis is possible if the causal mutations have previously been identified in the family.

Congenital factor XIII deficiency is characterized by hemorrhagic diathesis frequently associated with spontaneous abortions and defective wound healing. Congenital FXIII deficiency can manifest at any age, but diagnosis is often made during infancy. Umbilical stump bleeding manifests in up to 80% of the patients. Other common signs include intracranial hemorrhage (25-30%), soft tissue bleeding, bruising, hemarthroses (20%), and recurrent spontaneous abortions. In most cases, hemorrhages are delayed (12-36hr) after trauma or surgery. Patients may have poor wound healing.

FXIII is a pro-enzyme (pro-transglutaminase) and is the terminal enzyme in the coagulation cascade. In plasma, FXIII circulates as an inactive zymogen heterotetramer [A_2B_2] composed of two FXIII A-subunits [A_2] and two carrier FXIII B-subunits [B_2] held together by strong non-covalent interactions. The FXIII A-subunits possess the catalytic site of the FXIII enzyme. The FXIII B-subunits act as carrier molecules for the FXIII A-subunits in circulation, and prolong the half-life of the FXIII A-subunit when in circulation. The carrier FXIII B-subunits are present in approximately 50% excess in plasma in the form of circulating, uncomplexed proteins. The mechanism of action of FXIII is as follows: following injury to the vessel wall the coagulation cascade is activated. This leads to the conversion of fibrinogen into fibrin monomers, which polymerise and form a fibrin clot at the site of injury. Upon proteolytic activation by thrombin, the FXIII A-subunits are released from the FXIII B-subunits, whereby the active site of the FXIII enzyme is exposed. Activated FXIII binds to fibrin and catalyses the formation of several amide bond cross-links between adjacent fibrin molecules.

This cross-linking increases the mechanical strength of the fibrin clot and prevents premature fibrinolysis. In addition to cross-linking fibrin molecules, FXIII incorporates into the fibrin clot a number of plasma- and extracellular matrix proteins involved in inhibition of fibrinolysis and promotion of wound healing, including the anti-fibrinolytic protein α2 antiplasmin, which is the major inhibitor of

plasmin-induced clot degradation. Thus, FXIII modifies both the mechanical strength and the physiological properties of the blood clot and is vital to normal haemostasis.

In congenital FXIII deficiency, initial clot formation is often sufficient to arrest bleeding. However, if there is insufficient FXIII activity for adequate cross-linking of fibrin and for incorporation of α2 antiplasmin into the clot, clot strength is decreased and premature clot lysis occurs. Bleeding may resume several hours after injury (delayed bleeding). In the absence of FXIII replacement, a single injury may initiate refractory bleeding episodes lasting weeks or months.

Current treatment options in FXIII deficiency include cryoprecipitate, fresh frozen plasma, and – where available – plasma-derived FXIII concentrate. Prophylactic therapy with FXIII concentrate should be indicated to prevent recurrent bleedings such as intracranial hemorrhage. Intracranial hemorrhage can be life threatening, but prognosis is favorable if adequate treatment is provided.

Recombinant FXIII (rFXIII, rFXIII-A_2) is a dimer of two FXIII A-subunits [rA_2] and is identical in structure to the human FXIII A-subunit. Upon injection, the rFXIII A_2 dimers bind to endogenous circulating B-subunits that circulate in excess to form a stable rA_2B_2 heterotetramer.

The Applicant applied for the following therapeutic indication for rFXIII: *prophylactic treatment of bleeding in patients 6 years and above with congenital factor XIII A-subunit deficiency.*

The final indication agreed by the CHMP is *long-term prophylactic treatment of bleeding in patients 6 years and above with congenital factor XIII A-subunit deficiency*

The recommended dose is 35 IU/kg body weight (bw) once monthly (every 28 days +/- 2 days), administered as an intravenous bolus injection. Based on the actual concentration of NovoThirteen, the dose volume (in millilitres) to be administered can be calculated from the formula below:

Dose volume in ml = 0.042 x subject bw (kg)

Dose adjustment can be considered necessary by the physician in certain situations where the prevention of bleeding is not appropriately covered by the recommended 35 IU/kg/month dose. This dose adjustment should be based on FXIII activity levels. Monitoring NovoThirteen activity levels using a standard FXIII activity assay is recommended.

2.2. Quality aspects

2.2.1. Introduction

NovoThirteen contains recombinant human coagulation factor XIII A (rFXIII-A)-subunit produced in yeast cells (*Saccharomyces cerevisiae*) by recombinant DNA technology.

Recombinant FXIII is a dimer of two FXIII A-subunits [rA_2] and is identical in structure to the human FXIII A-subunit. The finished product is presented as powder for solution for injection containing a nominal value of 2500 IU of rFXIII in glass vial. The container closure system for NovoThirteen consists of 12 ml glass vial Type I, chlorobutyl rubber stopper and snap-off. For reconstitution of the finished product, water for injections (WFI) is also supplied in another glass vial containing 3.2 mL.

2.2.2. Active Substance

The structure of recombinant FXIII (rFXIII) is a mature non-glycosylated full-size FXIII A subunit molecule with a native conformation of a non-covalent homodimer consisting of two identical rFXIII A-subunits with a mass of approximately 166 kDa for the dimer.

The FXIII A-subunits are composed of 731 amino acids and possess the catalytic site of the FXIII enzyme. As for the native protein, the N-terminal serine is blocked and there are no post-translational modifications, and no disulfide linkages are present.

The structural characterisation and elucidation of physico-chemical properties using adequate analytical methods presented in the application have confirmed that the structure and properties of rFXIII active substance produced by Novo Nordisk have identical features to the native FXIII-A subunit.

Manufacture

rFXIII-A_2 is produced intracellularly in *Saccharomyces cerevisiae* using fed-batch fermentation after which the cells are homogenized and rFXIII is captured and purified. The cell substrate was derived from *Saccharomyces cerevisiae* cell line which was transformed by a plasmid containing the gene of interest. The rFXIII A-subunit protein is not further processed and collected intracellularly in the cytosol.

The commercial manufacturing process for rFXIII active substance consists of fermentation, recovery and purification.

Fermentation includes propagation of *S. cerevisiae* strain expressing rFXIII in a fed batch process. A two-tiered banking system (MCB/ WCB) is used to assure supply of the production cell line to support manufacture of rFXIII. MCB and WCB were both established without the use of materials of human or animal origin. During the recovery process rFXIII is released from the cells by high pressure homogenisation. The homogenized broth is clarified by centrifugation followed by filtration.

The quantitative information (e.g. volumes/size, mass, load, cell density…) have been described. The description of the fermentation and recovery process and their controls is considered adequate. Detailed information on the in-process tests (including their appropriate limits) has been provided.

The released rFXIII-A_2 is captured using anion exchange (DEAE) chromatography. During the following purification process another three chromatographic steps are performed. The purification process also includes 2 filtrations 0.2 µm. The bulk is then concentrated by ultrafiltration, formulated into the final formulation buffer by diafiltration and filled into LDPE containers.

Genetic stability was investigated on End of production cells (EOP) obtained with commercial scale fermentation up to the "routine limit of cell age", and Late extended culture (LEC), obtained from small scale expansion beyond the routine limit of cell age.

A batch of rFXIII active substance is the final material that is derived from one fed-batch fermentation. No material mixing or splitting is involved as one fermentation batch is recovered into one active substance batch. The range of the batch size initially claimed by the Applicant was not supported by the validation data provided and it was not clear whether the range proposed was due to technical equipment or to active substance yield. Supplementary validation studies were performed and results are within the validation target. The proposed batch size of the active substance is considered acceptable and the batch numbering system appropriately described.

Validation and/or Evaluation

The validation is based on the results of in-process tests and batch analysis of 3 batches produced with the final commercial process. The validation of the fermentation and recovery process was performed on 3 batches. The validation of the purification process consisted of 3 batches. Four batches were

manufactured but one batch did not meet a prerequisite defined in the protocol. However, this was appropriately justified. All the results comply with their acceptance criteria.

Specification

The active substance specification includes test methods for identity, content, specific bioactivity, aggregates, purity, impurities, rFXIIIa° (Non-proteolytically activated rFXIIIa). The Tests for process-related impurities only consists in HCP (Host Cell Protein) since validation removal is claimed for DNA and Nickel. Other general tests (appearance, pH, endotoxins and microbial count) are also included in the specification.

A concern was raised regarding the control of purity and rFXIIIa° as it has not been demonstrated to appropriately monitor oxidized, deamidated and truncated forms. This concern was considered as sufficiently addressed by the Applicant's responses and the purity/impurity profiles can be considered appropriately controlled; specific acceptance criteria have been defined for each individual peak in the finished product specification.

As the HCP acceptance criteria is above the value of the clinical Phase III batches and a trend was observed with the recent Novo Nordisk campaigns with an increase of HCP levels, the CHMP recommended revision of the HCP limit once sufficient data are available. The data provided suggested that the production process is able to reduce HCP below the proposed limit which should be tightened. The Applicant committed to revise this limit when sufficient active substance analysis data are available.

As no international recombinant FXIII-A reference material exists, the biological activity/Total rFXIII bioactivity is performed according to an in-house method based on calibration of the rFXIII primary reference material (PRM) against the WHO 1st International Standard (IS) Factor XIII Plasma. The latter contains FXIII in form of the hetero-tetramer A_2B_2, whereas rFXIII-A exists as homodimer A_2 in NovoThirteen.

A parallel-line analysis has been performed using the Berichrom assay with dilution series of the WHO IS FXIII Plasma and the rFXIII PRM based on the validated range of the method. Statistical evaluation of the data has been provided according to Ph. Eur. chapter 5.3. The evaluation indicated no significance for non-parallelism of the calibration lines in the validated range, therefore it was concluded that rFXIII PRM acts like the WHO 1st IS FXIII plasma (like-versus-like). Thus, traceability to the international standard is considered established and the use of international units (IU) for NovoThirteen rFXIII is regarded justified.

The methods proposed to monitor the substance are generally considered appropriate.

Stability

The primary and supportive studies were performed according to the current ICH guidelines. Based on the data provided a shelf-life of 60 months at -20 ± 5°C for the active substance is considered acceptable.

Comparability exercise for Active Substance

Before the first clinical trials, the manufacture was transferred to the initial active substance manufacturing site. After campaign 1 the process was optimized to increase purity of the active substance before the production for the phase 3a clinical trials. After the phase 3a clinical trial the process has been moved to Novo Nordisk A/S where the commercial production of the rFXIII active substance will take place.

A comparability study was conducted on three initial manufacturing site and two Novo Nordisk representative rFXIII active substance batches. In addition, comparison of batch analysis of seven initial manufacturing site batches and three Novo Nordisk batches was provided. These studies show that the quality profiles of the active substance produced at initial manufacturing site and Novo Nordisk are highly similar.

2.2.3. Finished Medicinal Product

The finished product is formulated with L-histidine, sodium chloride, sucrose, polysorbate 20. The lyophilised powder is dissolved in 3.2 ml water for injections before use.

The rFXIII active substance is formulated with L-Histidine to maintain an optimal pH for stability, sodium chloride to minimise formation of rFXIIIa° in the lyophilised product, sucrose to protect the protein from aggregation, and polysorbate 20 to stabilise the active substance against surface denaturation. All excipients comply with Ph. Eur. monographs.

Pharmaceutical Development

During development different manufacturing processes have been used. The differences between the manufacturing processes relate to the following changes:

- Change in manufacturing process of solution prior to lyophilisation due to change in composition
- Change in filling volume and vial size
- Change in manufacturing site
- Change in lyophilisation process

Development and modifications of the manufacturing process have been made to minimise the formation of rFXIIIa° in the lyophilised finished product and to accommodate to the changes in the formulation, the change in vial size and filling height, and to the actual production site.

The current composition (Formula C) intended for the market corresponds to the composition used in phase 3 clinical trials for treatment of congenital FXIII A-subunit deficiency. The Formula C was used with the active substance manufactured according to the current Novo Nordisk process, and also for some batches manufactured according to the previous manufacturing site campaign 2-3 process.

The lyophilisation process was challenged in a full load extended laboratory scale study. All individual parameters in the lyophilisation program were systematically challenged by worst case combinations of minimum/maximum values. The purity and the content in rFXIII° and aggregates were not modified by the different worst case combinations.

The comparability of the Formula C finished product manufacturing process, according to the active substance initial manufacturing campaign 2-3 process and to the active substance Novo Nordisk process, has been studied.

Release data and stability data have been compared for the different formulations and manufacturing processes used during development and demonstrated that rFXIII finished product manufactured with active substance from the initial manufacturing site and Novo Nordisk is comparable.

Adventitious agents

NovoThirteen is produced without the use of human or animal derived materials.

TSE compliance

The active substance is manufactured and purified in the absence of human or animal derived material. The cell banking system was also established without the use of materials of human or animal origin. Therefore NovoThirteen is free of TSE-risk substances.

Virus safety

The fermentation process of NovoThirteen uses animal and human component free medium. This minimizes a possible contamination for adventitious viruses. The cells used for production of NovoThirteen are fungi (*S. cerevisiae*), therefore no virus safety testing on cell banks and un-processed bulk has been performed and the purification process was not validated for its virus reducing capacity. This approach is in compliance with current guidelines. In summary, the viral safety of NovoThirteen has been sufficiently demonstrated.

Manufacture of the product

The manufacturing process mainly consists in active substance thawing, formulation, double filtrations, filling (by weight) into vials and lyophilisation. The final formulation is sterile filtered and lyophilised. After completion the vials are closed automatically and capped.

The finished product manufacturing process is generally well described. All excipients are controlled according to Ph. Eur.

A comprehensive finished product process validation consisting of process design, process qualification and process confirmation was provided. The lyophilisation program was challenged at laboratory scale by worst case combinations of minimum/maximum values for time, temperature and pressure. Four worst case combinations were tested. The manufacturing process has been transferred to production, with a process challenge made at two worst case conditions identified in laboratory scale (the HighHigh and LowLow combinations) and a process qualification/confirmation at set point.

Process qualification has been performed on 2 batches formulated with active substance from the initial manufacturing site and 3 batches formulated with active substance from Novo Nordisk. These 5 batches were produced with the commercial finished product process. The results of in-process controls (IPCs) are in accordance with the control limits.

Process confirmation has been performed on only 1 batch produced from active substance manufactured through the Novo Nordisk manufacturing process. The CHMP recommended post-approval validation and batch analysis on two additional commercial scale of the finished product.

Batch analysis was provided for 15 finished product batches, including 6 batches produced according to the final process of the active substance and finished product processes. All results meet the acceptance criteria.

Product specification

The finished product (powder) is controlled for appearance, reconstitution time and water content. After reconstitution with WFI, it is controlled for appearance, identity , purity, aggregates, rFXIIIa°, bioactivity assay, protein concentration, sterility, bacterial endotoxins, pH, relative osmotic pressure and particulate matter.

Initially no upper limit has been set for the potency therefore the consistency of the product strength could not be guaranteed. This was raised as a major objection in the LoQ and is considered to be resolved at the time of the opinion. The proposed revised acceptance criterion for total bioactivity at release and shelf life of the finished product is considered acceptable in accordance with the target range of the Ph. Eur. monograph for other coagulation factors. The limits for purity, rFXIIIa° and water content are appropriately defined.

Stability of the product

The studies were performed according to the current ICH guidelines. The container closure system used in all stability studies is identical to the one intended for the market. The batches included in the stability programme were manufactured at the intended commercial manufacturing site.

Based on the data provided, a shelf-life as stated in the SmPC for the finished product is considered acceptable. However, the CHMP recommended the Applicant to perform in-use stability studies at the end of the shelf life for two additional primary batches of the finished product to confirm the proposed in-use time of 24 hours storage at 5°C ± 3°C or 3 hours at maximum 25°C after reconstitution.

Water for Injection (WFI)

Water for injection for rFXIII is used for reconstitution of lyophilised rFXIII 2500 IU before use. The manufacture of WFI for rFXIII has been adequately described. The manufacturing process of the solvent consists in filtration with nitrogen, filling by weight, capping and autoclaving.

(Critical) in-process tests have been established in the production process and are appropriate to control the manufacturing process. The terminal sterilisation process for sterile water for rFXIII has been qualified to provide at least a six log reduction of microorganisms ensuring a safe product.

Stability results of the 2 tested validation batches comply with specification after 12 months storage at +2 to +8°C and 6 months at 25°C. A shelf life of 24 months at +2 to +8°C is supported by stability data.

2.2.4. Discussion on chemical, pharmaceutical and biological aspects

Information about the active substance, catridecacog, was of acceptable quality. In general sufficient evidence regarding the manufacturing process has been provided.

Specification limits and analytical methods are suitable to control the quality of the active substance. However, the CHMP recommended revision of the HCP limit once sufficient data are available.

The finished product was well characterised. In general sufficient evidence regarding the manufacturing process has been provided. The method of manufacture has been satisfactorily described and the validation data shows consistent manufacture. Nevertheless, the CHMP recommended post-approval validation and batch analysis on two additional commercial scale batches of the finished product.

The proposed specifications were justified based on batch and stability results, and are in general adequate for assuring the product quality and therefore were accepted.

The stability program is in general considered satisfactory. The results generated during the stability studies support the proposed shelf life and storage conditions as defined in the SmPC. However, the CHMP recommended performing in-use studies at the end of shelf life for two additional primary batches of the finished product to confirm the proposed in-use time of 24 hours storage at 5°C ± 3°C or 3 hours at maximum 25°C after reconstitution.

2.2.5. Conclusions on the chemical, pharmaceutical and biological aspects

The active substance (catridecacog) and the finished product have been appropriately characterised and in general satisfactory documentation has been provided. The results indicate that catridecacog as well as the finished product can be reproducibly manufactured.

2.2.6. Recommendation(s) for future quality development

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

1. The CHMP recommends post-approval validation and batch analysis on two additional commercial scale batches of the finished product.
2. The CHMP recommends revision of the HCP limit once sufficient data on the active substance are available.
3. The Applicant is recommended to perform in-use studies at the end of shelf life for two additional primary batches of the finished product to confirm the proposed in-use time of 24 hours at 5°C ± 3°C or 3 hours at maximum 25°C.

2.3. Non-clinical aspects

2.3.1. Introduction

The pharmacological activity of rFXIII has been investigated in *in vitro* and *in vivo* studies. The objective was to demonstrate that rFXIII has the same pharmacodynamic properties in plasma as those described for endogenous FXIII.

The pharmacology and PK studies in the development program were conducted in accordance with good scientific principles; however, these studies were not conducted in full compliance with Good Laboratory Practice (GLP) regulations. The studies comprising the toxicology program were conducted in compliance with GLP, with the exception of toxicokinetics assays which were conducted according to good scientific principles, but not in full compliance with GLP regulations.

2.3.2. Pharmacology

Primary pharmacodynamic studies

In vitro studies

Table 3 and 4 shows a summary of the major findings for *in vitro* and *in vivo* primary pharmacodynamic studies.

Table 3: Summary of in vitro primary pharmacodynamic studies

Report No.	Model	Concentration /Dose	Major Findings
RES-FXIII-0025 (2002) RES-FXIII-0025 Addendum (2002)	Binding endogenous FXIII-B subunit to immobilised rFXIII[A2] affinity resin. Specifically bound proteins were analysed by SDS-PAGE gels and conclusively identified by N-terminal sequencing.		Binding to the endogenous FXIII-B subunit in **mouse, rat, dog, monkey and human.** Endogenous FXIII-B subunit from both **human and monkey** serum bound to human rFXIII.
RES-FXIII-0024 (2002)	rFXIII was added to FXIII deficient or depleted human and cynomolgus plasma. The resulting fibrin clot was washed and dissolved and the different fibrin forms were separated on SDS-PAGE gels.	0, 0.313, 0.625, 1.25, 2.5, 5, 10, 20 µg/ml	rFXIII is able to crosslink both **human and monkey fibrin** and this crosslinking readily occurs in a plasma environment. The rFXIII from two different production strains of *S. cerevisiae* was compared and found to have identical activity in this assay system.
RES-FXIII-0063 (2002)	Fibrinogen/fibrin clots were solubilised with urea/DTT and analyzed by SDS-PAGE.	Dilution series of normal human plasma prepared in FXIII-A-deficient plasma (100, 50, 20, 5, 1, 0.5, 0.15, 0 normal FXIII levels) rFXIIIa* or FXIIIA° : 0, 2, 5, 10, 15, 20, 30, 40, 50 µg/ml rFXIII : 0, 20, 100, 200, 400, 800 µg/ml)	rFXIIIa* and rFXIIIa° could **cross fibrinogen** (in absence of thrombin) in a dose-response manner (2-50µg/ml), the crosslinking profiles and dose response were similar between rFXIIIa* and rFXIIIa°. The human and cynomolgus heparinized plasma were shown to have equivalent crosslinking profiles with rFXIIIa* and rFXIIIa°.
LCP051125 (2005)	Clot-lysis analysis in normal and FXIII-A deficient plasma	rFXIII : 0, 30, 60, 120, 240, 480 nM	rFXIII **prolongs clot-lysis time** in a concentration-dependent manner and rFXIII is required for the anti-fibrinolytic activity of TAFI.
EHNO050520 (2005)	Thromboelastography (TEG) was used to evaluate the *in vitro* effects of rFXIII and rFVIIa on clot properties in normal whole blood, a model system that mimic	SCT patients : 0, 60, 120, 240 nM rFXIII combined with 0.25 and 100 nM rFVIIa CS patients : 0, 60, 120, 240 nM rFXIII combined with 0, 12,5 and 100 nM rFVIIa	rFXIII significantly enhanced the mechanical **strength and the resistance against fibrinolysis** in a dose-dependent manner in normal blood and blood from patients with low

	thrombocytopenia based on normal blood as well as an evaluation of rFVIIa and rFXIII spiked in whole blood from cardiac surgery (CS) patients and stem cell-transplantation (SCT) patients		platelet count patient from cardiac surgery and stem cell-transplant. Combining rFVIIa and rFXIII overall enhanced **clot formation, mechanical strength, and resistance against fibrinolysis** and was obtained, as the two individual proteins affected the thromboelastography parameters differently.

In vivo studies

Table 4: Summary of in vivo primary pharmacodynamic studies related to the proposed therapeutic indication

Report No.	Test system	Concentration /Dose	Major Findings
RES-FXIII-0058 (2002)	Cynomolgus monkeys	rFXIII 1-30 mg/kg	rFXIII appear as **crosslinked protein complexes**, including γ-fibrinogen dimers, α-fibrinogen multimers, and/or fibrinogen complexes with α-2-plasmin inhibitor, fibronectin and α-2-macroglobulin. The formation of these crosslinked protein complexes can be expected to have both direct and indirect effects on blood flow and plasma viscosity.
RES-10352 (2003+ statistic 2010)	New Zealand white rabbits	rFXIII : 0.4 mg/kg tPA IV bolus 1 mg/kg, followed by continuous infusion of 1 mg/kg/h	Animals pretreated with rFXIII showed **significantly increased resistance to clot lysis** by tPA indicating increased clot strength.

Secondary pharmacodynamic studies

The ability of rFXIII and the thrombin-activated rFXIII (rFXIIIa*) to interact with human whole blood and bind to a variety of cells was assessed in in vitro tests. An overview of the major findings is presented in Table 5. It was observed that thrombin-activated FXIII may be deposited at the intercellular junction of cultured porcine aortic endothelial cells and FXIII may be linked to gut permeability and inflammatory bowel disease. Hence several cell types were investigated.

Table 5: Summary of secondary pharmacodynamic studies

Report No.	Model	Concentration /Dose	Major Findings
RES-FXIII-0018 (1- 2001 and 2- 2002)	Interaction of rFXIII and rFXIIIa* with human whole blood cells, detected by flow cytometry. Blood cells were isolated by differential centrifugation and rFXIII and rFXIIIa* were incubated with the relevant cell type 1 hour at 4°C. (Human lymphocytes, granulocytes, monocytes and platelets)	rFXIII or rFXIIIa* : 10-400 µg/ml (control cells) 1-20 µg/ml (blood nuclear cells) 5-40 µg/ml (platelets)	Neither activated nor quiescent platelets bound rFXIII. **Activated platelets and quiescent platelets bound to rFXIIIa* at concentration as low at 5µg/ml.** No binding to other blood cell types (rFXIII lot FE92007X). Two batches of rFXIII qualitatively behave in a similar manner with respect to platelet binding.
RES-FXIII-0017 (1- 2000-2001 and 2- 2002)	Detection of rFXIII and rFXIIIa* binding to various human cells lines by immunofluorescence microscopy using a rabbit anti-human rFXIII primary antibody, and an anti-rabbit FITC conjugated secondary antibody. (Human endothelial-, intestinal epithelial-, fibroblast- and smooth muscle cells)	rFXIII or rFXIIIa* : 0, 0.1, 1, 10, 100 µg/ml	**Only the rFXIIIa* bound to the cultured cells** in the first assay (lots FH92009X, FE92007X and F249124). The assay results from rFXIII lot 217-01-001 indicate the same result.

Safety pharmacology programme

The safety pharmacological activity of rFXIII has been investigated in *in vitro* and *in vivo* specific studies (cardiovascular model in rabbit and extra-corporal circulation model in cynomolgus monkey). Moreover, specific endpoints have been incorporated in the design of two toxicology studies (SBI 1394-175 and NN 205255) for assessing safety pharmacology in cynomolgus monkey.

A summary of the major findings in *in vitro* and *in vivo* safety pharmacology studies is shown in Table 6.

Table 6: Summary of *in vitro* and *in vivo* safety pharmacology studies

Report No./Type of study	Test system	Method of administration Concentration/Dose	Major Findings
RES-FXIII-0014 (2001)/ cytoxicity	HUVEC cells	*In vitro* rFXIII : 0.1-100 µg/ml rFXIIIa* : 0.1-100 µg/ml Doxirubicin :0.1 and 10 µg/ml rFXIII batches FH92009X, FE92007X and F249124 (yeast Z M146)	10 µg/ml or less (concentrations compatible with *in vivo* treatment): **neither non-activated nor activated rFXIII[A_2] are toxic to endothelial cells** 100 µg/ml or greater: Activated rFXIII[A_2*] may have **a inhibitory effect on endothelial cell proliferation**
RES-FXIII-0015 (2001)/adhesion molecule up-regulation	Endothelial cells	*In vitro* rFXIII : 0.1, 10 and 100 µg/ml rFXIIIa* : 0.1, 10 and 100 µg/ml TNF :0.1 µg/ml rFXIII batches FH92009X, FE92007X and F249124 (yeast Z M146)	10 µg/ml or less: (concentrations compatible with *in vivo* treatment) rFXIII[A_2] and rFXIII[A_2*] are **not pro-inflammatory**. 100 µg/ml: rFXIII[A_2*] give **a small increase of adhesion** molecule expression; rFXIII[A2*] **falls out of solution in physiological buffer**, this effect may be due to aggregate formation.
RES-FXIII-0016 (2001)/cytokine release	Whole blood	*In vitro (Ex vivo)* rFXIII : 0.1-100 µg/ml rFXIIIa* : 0.1-100 µg/ml LPS: 0.1-100 ng/ml rFXIII batches FH92009X, FE92007X and F249124 (yeast ZM146)	Concentrations up to 100 µg/ml: rFXIII[A_2] and rFXIII[A_2*] **do not elicit an immune/inflammatory response** in whole blood
RES-10351 (2003)/ Clot formation and cardiovascular system in an arteriovenous-shunt model with activated platelets	Rabbits	Intravenous rFXIII: 0.4 mg/kg batch 217-01-001 (yeast BJ2n-5-LA)	**No effect of rFXIII on clot growth, blood flow, blood pressure, and platelet counts**
ZGI 1112-010 (2004)/ Pilot study : Cardiovascular and respiratory system after extracorporal circulation (ECC)	Cynomolgus monkey	Intravenous	ECC alone with particular attention to the coagulation status of the animals may be a preferred model compared to the model involving ECC with sternotomy.
ZGI 1112-011 (2004)/ Cardiovascular and respiratory system after extracorporal circulation (ECC)	Cynomolgus monkey	Intravenous rFXIII: 0, 2.1, 7.1 mg/kg lot 217-04-001 (yeast BJ2n-5-LA)	300 or 1000 U/kg rFXIII after two hours of ECC: **no apparent adverse effects**
ZGI 1112-013 (2004)/ Cardiovascular and respiratory system after extracorporal circulation (ECC)	Cynomolgus monkey	Intravenous rFXIII: 0, 7.1 mg/kg lot 217-04-001 (yeast BJ2n-5-LA)	1000 U/kg rFXIII after two hours of ECC: **no apparent adverse effects**

A summary of the major findings from *in vitro* and *in vivo* pharmacodynamic drug interaction studies is presented in Table 7.

Pharmacodynamic drug interactions

Table 7: Summary of in vitro and in vivo pharmacodynamic drug interactions studies

Report No./Type of study	Test system	Method of administration	Major Findings
RES-10323 (2003)/Interaction with heparin and protamine	Human plasma 1 – heparin (0.75 units/ml) and protamine (2.7µg/ml) 2 – heparin (0.75, 3, 10 units/ml) and protamine (3.75, 16.5, 56.3 µg/ml)	*In vitro* 1 - rFXIII: 0, 1, 3, 10, 30, 100 µg/ml 2 – 0.05 µg/ml lot 217-01-001 (yeast BJ2n-5-LA)	**No cross-reactivity between rFXIII and heparin/protamine**.
NN205070 (2007)/Single dose toxicity study rFXIII and RFVIIa	Cynomolgus monkeys	Intravenous rFVII (NovoSeven) 0.1, 0.33, 1.67, 5) + rFXIII (0.34, 1.12, 5.6, 16.8 mg/kg) batch : ZBC401N (yeast BJ2n-5-LA)	5 mg/kg rFVIIa+16.8 mg/kg rFXIII: early death or termination of both dosed animals (DIC) Up to 1.67 mg/kg rFVIIa + 5.6 mg/kg rFXIII: well tolerated with no microscopic evidence of thrombus formation in the lungs, kidneys, heart, eye or brain. **NOAEL: 1.67 mg/kg rFVIIa + 5.6 mg/kg rFXIII**
NN205148 (2008)/Single dose toxicity study rFXIII and rFVIIa	Cynomolgus monkeys	Intravenous rFVIIa (1, 2, 1, 2, 1, 0.75, 4, -, 4) + rFXIII (3.5, 3.5, 7, 7, 14, 10.5, 3.5, 10.5,-) (batches PR40255, RLDS002, yeast BJ2n-5-LA)	Five different ratio combination rates ranging from (1:14 to 1:0.88): **no deaths or adverse clinical signs** Microscopic evidence of pathological changes were found in animal dosed with the highest levels of either of the two test articles but without any clear evidence of an additive or synergic effect of the two compounds in combination. **NOAEL: 2 mg/kg rFVIIa + 7 mg/kg rFXIII**
NN206100 (2010)/Cardiovascular system rFXIII and RFVIIa	Cynomolgus monkeys	Intravenous rFVIIa+ rFXIII (batch: SLDF010, yeast BJ2n-5-LA) Group 1 : vehicle (0/0) Group 2 : mid dose combination (1/3.5 mg/kg) Group 3 : high dose combination (2/7 mg/kg) Group 4 : low dose combination (0.5/1.75 mg/kg)	Group 2: 1 animal died (early thrombus formation) Group 3: 2 animals had fluctuating blood pressure, left ventricular pressure The cause of death may be a combination of exaggerated pharmacology of combination of rFVIIa and rFXIII in animals undergoing anaesthesia with 6 indwelling catheters which may have caused a pro-coagulant state. **NOAEL: 0.5 mg/kg rFVIIa + 1.75 mg/kg rFXIII**

2.3.3. Pharmacokinetics

The pharmacokinetics of rFXIII were characterized in single-dose administration in cynomolgus monkeys (0.3-20 mg/kg equal to 50-3340 IU/kg), repeated administration in rat (1-15 mg/kg equal to 167-2505 IU/kg) and repeated administration in monkey (1-10 mg/kg equal to 167-1670 IU/kg).

Absorption

Absorption of rFXIII has been studied within dedicated pharmacokinetic studies (SBi-1224-175 and SBi 1241-175) as well as within single and repeat dose toxicology studies (Table 8).

Table 8: Summary of total A2 and A2B2 pharmacokinetic parameters after single dose in cynomolgus monkey study (SBI 1220-175, SBI 1278-175, SBI 1224-175, SBI 1241-175, SBI1266-175)

			Total A_2				A_2B_2		
Study ID	**Male/Female**	**Dose (mg/kg)**	**C_0 (µg/ml)**	**$T_{1/2}$ (h)**	**AUC_{inf} (h*µg/ml)**	**Cl (ml/h/kg)**	**Cmax (µg/ml)**	**$T_{1/2}$ (h)**	**AUC_{inf} (h*µg/ml)**
SBI 1220-175	1F	10	182	161	4722	2.1	46.1	166	14017
	1F	17.5	304	118	4964	3.5	36.2	154	10227
	1F	20	360	113	5462	3.7	49.2	118	11328
SBI 1278-175	1F	20	516	180	7728	2.6	57.4	199	19407
	1F	21.2	536	91	6887	3.1	61.8	180	17651
SBI 1224-175	2M/2F	1	22	122	1620	0.83	28	244	8729
	2M/2F	5	118	167	3853	1.34	38	225	13971
SBI 1241-175	4M/4F	0.5	11	195	656	0.9	17.5	164	3239
	4M/4F	1	19	143	985	1.2	22.8	121	4382
	4M/4F	5	103	131	2677	2.0	38.5	125	10512
SBI1266-175	3M/3F	5	97.4	186	3965	1.29	38.7	72	8660
	3M/3F	8	144	134	3927	2.18	35.9	69	8398
	3M/3F	12.5	232	174	5752	2.2	39.7	65	8365

A comparison of the pharmacokinetic profiles of a single intravenous injection of rFXIII at 1 mg/kg between juvenile and mature monkeys showed significantly lower exposure in juvenile versus mature animals, indicating a higher clearance and/or higher volume of distribution in the juveniles.

Distribution

A subset of the monkeys (2M/2F) in study 7333-101 were allocated for the investigation of the distribution of 125I-labelled rFXIII in a Whole-body Autoradiography (WBA) study following single intravenous administration of 0.5 and 5.0 mg/kg. Radioactivity was mainly distributed to plasma and highly perfused organs. No tissues (except for the thyroid as could be expected due to the Iodine label) were exposed to radioactivity at tissue to plasma ratios above 1 at either 2 or 72 hours post-dose.

Metabolism

No *in vit*ro or *in vivo* metabolism studies were submitted.

Elimination

In the biodistribution study performed in cynomolgus monkeys (study 7333-101_B), 4 monkeys (1 male and 1 female per dose) received a single IV injection of 0.5 or 5 mg/kg 125I-labelled rFXIII. Urine and faeces were collected at 0-4, 4-8, 8-24 and 24-hour intervals through 168 hours postdose. Urinary excretion was the predominant route of elimination. Urine and faeces accounted for average values of 51.5 and 1.93% of the administered radioactive dose, respectively, for the low dose and 41.8 and 2.37%, respectively, for the high dose.

Pharmacokinetic drug interaction

A single-dose study was performed in cynomolgus monkey assessing the toxicity of intravenous (IV) administration of rFVIIa and rFXIII (study NN205148). IV administration of rFXIII in the dose range 0 to 14 mg/kg and rFVIIa in the dose range 0 to 4 mg/kg administered sequentially showed that the exposure (AUC (0-72h)) of the A_2B_2 complex did not change with dosing of rFVIIa or rFXIII.

2.3.4. Toxicology

Single dose toxicity

Three single dose toxicity studies have been performed in cynomolgus monkeys (Table 9).

Table 9: Overview of single dose toxicity studies:

Study ID	**Species/ Number/ Sex/ Group**	**Route/ Dose (mg/kg)/(IU/kg)**		**Note worthy findings**
SBI 1220-175	Cynomolgus monkey 1 F	IV 0, 10, 17.5, 20 (0, 1670, 2923, 3340)	Non-GLP	None, all animals survived the study period (8 days observation period)
SBI 1278-175	Cynomolgus monkey 1 F	IV 0, 20, 21.2, 22.5, 25, 30 (0, 3340, 3540, 3758, 4175, 5010)	Non-GLP	Animals administered 22.5 and 30 died or were sacrificed moribund, DIC
SBI 1249-175	Cynomolgus monkey 1 M (21.9 mg/kg) 1 F and 1 M (22.5 mg/kg)	IV 21.9 and 22.5 (3657, 3758)	GLP	All monkeys died day 1, DIC

F= female, M = male, rFXIII was administered in mg/kg the exact content in IU is not known, the dose level in IU/kg is an approximate dose based on 1 mg= 167 IU

No apparent rFXIII-related effects were observed for clinical signs, food consumption, body weight, clinical pathology, body temperature, heart rate or blood pressure.

Repeat dose toxicity

Repeat dose toxicity studies were performed in the rat (2 studies) for up to 4 weeks and the cynomolgus monkey (4 studies) for up to 27 weeks (Table 10).

Table 10: Overview of repeat-dose toxicity studies

Study ID	Species Number/Sex/group	Route/ Dose (mg/kg)	Dose interval, Duration	NOAEL (mg/kg) [IU/kg]	Noteworthy findings
NN209517 (non-GLP)	Rat 5M/5F	IV 0, 5, 15	daily, 5 days	15 [2505]	none
NN209502 (GLP)	Rat 10M/10F	IV 0, 1, 5, 15	daily 4 weeks	15 [2505] NN DS 5 [835] Initial manufacturing site DS	Reversible lymphoid hyperplasia in the spleen
SBI 1249-175 (GLP)	Monkey 1-2M/0-1F	IV 0, 12.5, 17.5	3 days 3 days	No NOAEL	All monkeys but one died or were sacrificed moribund after 2 repeat doses, DIC
SBI 1394-175 (GLP)	Monkey 3-5M/3-5F	IV 0, 0.3, 3, 6	daily, 2 weeks	6 [1002]	none
SBI 1266-175 (GLP)	Monkey 3-5M/3-5F	IV 0, 5, 8, 12.5	2 weeks, 4 weeks	8 [1336]	Reversible lymphoid hyperplasia in the spleen; Mild glomerulopathy (in one out of six)
NN205255 (GLP)	Monkey 3-15M/3-15F	IV 0, 1, 3, 10	2 weeks, 13 and 27 weeks	3 [501]	One out of 30 in the 10 mg/kg group died due to thrombosis and ischemic necrosis

In study SBI 1249-175, one animal from group 2 dosed on Days 1 and 4 with 12.5 mg/kg rFXIII survived until the terminal sacrifice on Day 15. In this animal, no rFXIII related effects were observed on clinical signs, food consumption, body weight, blood pressure, body temperature, heart rate, clinical pathology, organ weights and gross pathology. Histopathology in this animal revealed minimal vascular thrombosis in one optic nerve. In all seven animals that died or were sacrificed, the underlying pathology was systemic thrombosis in multiple organs, which was attributed to an exaggerated pharmacology of rFXIII/activated rFXIII.

Lymphoid hyperplasia in the spleen was observed in the repeat dose study NN209502 in rat animals and in SBI 1266-175 study in monkeys. A comparison between lymphoid hyperplasia in the spleen and the development of anti-rFXIII antibodies is shown in Table 11 and 12.

Table 11: Incidence of lymphoid hyperplasia in the spleen

		Males						Females					
Dose group		1M	2M	3M	4M	5M	6M	1F	2F	3F	4F	5F	6F
Dose level (IU/kg)		0	167	835	2505	167	835	0	167	835	2505	167	835
No examined - main		10	10	10	10	10	10	10	10	10	10	10	10
Grade	-	7	7	8	4	3	3	8	8	6	6	3	2
Lymphoid	1	3	2	2	6	6	3	2	2	4	4	7	2
hyperplasia	2	0	1	0	0	1	4	0	0	0	0	0	4
	3	0	0	0	0	0	0	0	0	0	0	0	2
No examined – Recovery		5	5	5	5	5	5	5	5	5	5	5	5
Grade	-	4	5	3	3	4	3	5	5	4	4	5	1
Lymphoid hyperplasia	1	1	0	2	2	1	2	0	0	1	1	0	4

Key: "- "= finding not present, 1= minimal, 2= slight, 3= moderate (on a five grade scale)

Group 1= vehicle control, groups 2-4: $rFXIII_{NN}$, groups 5-6: $rFXIII_{Avecia}$

Table 12: Number of animals developing anti-FXIII antibodies out of total number examined after administration of rFXIII

Dose group ($rFXIII_{NN}$ or $rFXIII_{Avecia}$)	2 M/F ($rFXIII_{NN}$)	3 M/F ($rFXIII_{NN}$)	4 M/F ($rFXIII_{NN}$)	5 M/F ($rFXIII_{Avecia}$)	6 M/F ($rFXIII_{Avecia}$)
Dose level (IU/kg)	167	835	2505	167	835
No of animals with anti-FXIII antibodies after end of treatment	8/20	3/20	0/20	13/20	1/20
No of animals with anti-FXIII antibodies after treatment free period	9/10	9/10	3/10	10/10	9/10

Genotoxicity

The Applicant did not submit studies on genotoxicity.

Carcinogenicity

The Applicant did not submit studies on carcinogenicity.

Reproduction Toxicity

The Applicant did not submit studies on reproduction toxicity.

Toxicokinetic data

Toxicokinetic data was assessed as part of the repeat-dose toxicity studies in cynomolgus monkeys. A summary of the data is presented in Table 13.

Table 13: Summary of TK parameters - repeat-dose toxicity study in cynomolgus monkey after IV administration (SBI 1249-175)

			Day 1	
Study ID	**Sex/N**	**Daily Dose Range (mg/kg)**	**C0, total A2 (µg/mL)**	**AUC(0 to 72), total A2 (h*µg/mL)**
SBI 1249-175 (2002)	M/1 F/2	12.5-14.5	292.05 (85.01)	3744.82 (1262.74)
	M/1 F/1	17.0-17.4	396.32 (48.65)	4876.03 (391.10)

NB: Because of acute animal death following dosing on Day 1, TK analyses were not conducted on data collected from animals in Groups 4 and 5. Because of death of animals in Groups 2 and 3 following dosing on Day 4, no TK analyses were conducted on samples collected following dosing on Day 4.

Following a single dose of rFXIII on Day 1, the total A2 data for the animals treated at 12.5-14.5 and 17.0-17.4 mg/kg rFXIII showed a proportional increase in C0(A2) and AUC(0-72), total A2 with respect to dose range.

Table 14: Summary of TK parameters - repeat-dose toxicity study in cynomolgus monkey after IV administration (SBI1394-175, 2003)

		Day 1		Day 14		Accumulation ratio
Study ID	**Daily Dose (mg/kg)**	**C0 (µg/mL)**	**AUC(0.24) (h*µg/mL)**	**C0 (µg/mL)**	**AUC(0.24) (h*µg/mL)**	
SBI 1394-175 (2002)	0.3	6.89	132.94	23.45	428.27	3.60
	3	74.54	774.38	130.98	1320.37	1.71
	6	172.56	1490.57	186.29	2192.14	1.53

Table 15: Summary of TK parameters - repeat-dose toxicity study in rat after IV administration (NN209502)

		Day 1				Week 4			
Study ID	**Daily Dose (mg/kg)**	**$C_{0.25h}$(%)**		**AUC_{0-24h} (%*h)**		**$C_{0.25h}$(%)**		**AUC_{0-24h} (%*h)**	
		F	M	F	M	F	M	F	M
NN209502	1	405	269	405	269	264	153	3450	3800
	5	1340	1180	1340	1180	1470	1350	13400	15800
	15	5600	4020	5600	4020	4660	4380	39100	39500
	1	592	334	592	334	104	130	3840	2410
	5	1700	1310	1700	1310	2070	1080	14500	14600

Local Tolerance

Local tolerance has been assessed within repeated dose toxicity studies in rat and cynomolgus monkey. Generally there were no or minor local reactions noted in the repeat dose toxicity studies.

Furthermore, two separate local tolerance studies have been performed in rabbits (see Table 16).

Table 16: Summary of dedicated local tolerance studies in rabbit (NN205496 and NN209504)

Study number Species/Strain	Method of administration	Doses rFXIII (mg/kg)/ [IU/kg]	Gender and No. per group	Noteworthy Findings
NN205496 (DANAK, 2006)/New Zealand White rabbits	Intravenous intra-arterial perivenous	0.35 /[58] 0.35 /[58] 0.35 /[58]	4 females 4 females 4 females	None
NN209504 (LAB research, 2010)/New Zealand White rabbits	Intravenous intra-arterial perivenous	0.2 /[35] 0.2 /[35] 0.2 /[35]	4 females 4 females 4 females	<u>After intravenous injection:</u> **slightly more severe reaction than the vehicle** at the site of introduction of the needle.

Other toxicity studies

2.3.5. Ecotoxicity/environmental risk assessment

No environmental risk assessment was performed given that the drug substance is a protein. The active substance is a natural substance, the use of which will not alter the concentration or distribution of the substance in the environment. Therefore, rFXIII is not expected to pose a risk to the environment.

2.3.6. Discussion on non-clinical aspects

The pharmacological activity of rFXIII was investigated in *in vitro* and *in vivo* studies by the Applicant. rFXIII was shown to bind to the endogenous FXIII-B subunit in mouse, rat, rabbit, dog, monkey and human forming the heterotetramer [A2B2] complex. The data demonstrated that rFXIII has the same pharmacodynamic properties as has been described for endogenous FXIII. In rat studies, the PK showed a non-linear profile between 1.5 and 15 mg/kg, with no apparent differences between males and females. The accumulation between Day 1 and Day 28 was low (<1.6 fold) and the determined half-life was 6-10 hours. In cynomolgus monkey studies, rFXIII was found to bind to free FXIII- B_2; the complex had a terminal half-life of 50-225 hours, the half-life of free rFXIII was in the order of 4-7 hours. In cynomolgus monkeys, tissue biodistribution appeared to be predominantly in blood and highly perfused organs. Furthermore, the Applicant performed a single dose pharmacokinetic study in juvenile and mature cynomolgus monkeys. The results demonstrate a significantly lower exposure in juvenile animals. Thus clinical pharmacokinetic assessment in children may be necessary to further evaluate the need for any adjustment of the recommended dose.

Urine was identified as the primary route of excretion of 125I-rFXIII-derived radioactivity. The excretion studies are considered sufficient and the results revealed no concerns.

Furthermore, pharmacokinetic drug interaction between rFXIII and rFVIIa has been investigated in a single-dose study in cynomolgus monkey. No signs for pharmacokinetic drug interaction between

rFXIII and rFVII were found in this study. Overall, the pharmacokinetic studies in adult animals submitted in the application are considered sufficient to support marketing authorization of rFXIII.

The safety pharmacological activity of rFXIII was investigated in *in vitro* and *in vivo* specific studies. The safety pharmacological program followed ICH S6 guidance. *In vitro* data showed no adverse effects observed at concentrations compatible with *in vivo* treatment (10µg/ml or less), neither non-activated nor activated rFXIII were toxic to endothelial cells, pro-inflammatory, and did not elicit an immune/inflammatory response. *In vivo* data showed no effects in the clot growth, blood flow, blood pressure, and platelet counts in rabbit. In extra-corporal circulation system in cynomolgus monkey, no treatment-related effects were noted in the safety pharmacological battery of tests, i.e. respiration, ECG, CVS parameters, body temperature, clinical chemistry, haematology, coagulation endpoints and gross necropsy. A single dose of 2.1 and 7.1 mg/kg after two hours of extra-corporal circulation was well tolerated and with no apparent adverse effects through six hours of observation. *In vivo* data in toxicological studies showed no treatment-related effects in vital organs, CNS or CVS with rFXIII in cynomolgus monkey. The rat and monkey animal studies, NN209502 and SBI 1266-175, described the finding of lymphoid hyperplasia in the spleen. A comparison between lymphoid hyperplasia in the spleen and the development of anti-rFXIII antibodies demonstrated that there was no direct correlation between the two occurrences. Mild glomerulopathy was observed in the monkey study SBI 1266-175. However, this pathological finding was reviewed and considered as normal background finding and no further concerns were expected.

Results from a cardiovascular model study in cynomolgus monkeys where rFVIIa was administered concomitantly with rFXIII suggest that this combination could have a synergic effect which could lead to thrombus and death. The results are highlighted in section 5.3 of the SmPC.

The Applicant did not submit *in vitro* or *in vivo* metabolism studies, genotoxicity studies, carcinogenicity studies and reproduction studies. The metabolism of rFXIII was expected to follow the same catabolic routes as its endogenous counterpart and the CHMP considered the lack of studies acceptable. No effects on reproductive organs were noted in the repeated dose toxicity studies. Given the biological nature of the product, the lack of genotoxicity and carcinogenicity studies was also considered acceptable and consistent with the ICH S6 guideline.

2.3.7. Conclusion on the non-clinical aspects

The non-clinical studies submitted for the marketing authorisation application for rFXIII-A_2 (rFXIII) were considered adequate and acceptable for the assessment of non-clinical aspects for the product catridecacog. The safety pharmacology aspects in the repeat dose toxicity studies revealed no further concerns. The lack of metabolism, genotoxicity, carcinogenicity and environmental risk assessment studies was justified and considered acceptable.

2.4. Clinical aspects

2.4.1. Introduction

GCP

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

The clinical studies were conducted in the EU, Switzerland, US, Canada, Japan and Israel. 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

	Trial ID & Status	Description / Doses / Type and Number of Dosed Subjects
Phase I		
	NN1841-3788 (Complete)	Investigation of bioequivalence and pharmacokinetics of $rFXIII_{IASMS}$ and $rFXIII_{NN}$ after single doses in healthy male subjects. 35 IU/kg 50 Healthy Subjects
	F13-1662 (112C01.0) (Complete)	Evaluation of safety and pharmacokinetics of five consecutive daily doses of rFXIII in healthy subjects. 12 IU/kg, 30 IU/kg or placebo daily for 5 days 24 Healthy Subjects
	F13-1661 (UKHV-1) (Complete)	Evaluation of safety and pharmacokinetics after single dose exposure to rFXIII in healthy subjects. 2, 6, 12, 30 or 60 IU/kg, or placebo 50 Healthy Subjects
	NN1810-3733 (Complete)	Evaluation of safety and pharmacokinetics of rFXIII in healthy Japanese subjects. 12 IU/kg, 35 IU/kg or placebo 24 Healthy Subjects
	F13-1663 (CD1.3) (Complete)	Evaluation of safety after single dose exposure to rFXIII in patients with congenital FXIII A-subunit deficiency. 2, 7, 24, 60 and 89 IU/kg 9 Patients
Phase III		
	F13CD-1725 (Complete)	Evaluation of efficacy and safety of monthly replacement therapy with rFXIII for prevention of bleeding episodes in subjects with congenital FXIII A-subunit deficiency. 35 IU/kg 41 Patients
	F13CD-3720 (Ongoing) *	Extension trial to F13CD-1725. Evaluation of safety of monthly replacement therapy with rFXIII in subjects with congenital FXIII A-subunit deficiency. 35 IU/kg 33 Patients
	F13CD-3760 (Ongoing)*	Evaluation of pharmacokinetics and safety of a single dose of rFXIII in paediatric patients (1 to less than 6 years old) with congenital FXIII A-subunit deficiency. 35 IU/kg 1 Patient
	F13CD-3835 (Ongoing)*	Extension trial to F13CD-3760. Evaluation of safety of monthly replacement therapy with rFXIII in paediatric patients (1 to less than 6 years old) with congenital FXIII A-subunit deficiency. 35 IU/kg 1 Patient
Phase IV		
	NN1841-3868 (Planned)	A prospective, observational study of the safety of rFXIII during commercial use. Doses according to label.
Other studies		
	F13CARD-1660 (Complete)	Evaluation of safety of single-dose exposure to rFXIII following cardiac surgery requiring cardiopulmonary bypass 11.9, 25, 35 or 50 IU/kg rFXIII, or placebo 43 Cardiac surgery Patients
	NN1810-3450 (Ongoing) *	Evaluation of efficacy and safety of single-dose exposure to rFXIII following cardiac surgery requiring cardiopulmonary bypass 17.5 or 35 IU/kg rFXIII, or placebo

		409 cardiac surgery patients

* Cut-off date: 11 Feb 2011

2.4.2. Pharmacokinetics

The pharmacokinetics of rFXIII was investigated in five clinical pharmacology trials; four trials in healthy subjects and one trial in patients with congenital FXIII A-subunit deficiency. In addition, a phase 3 trial (F13CD-1725) contribute to the assessment, as blood sampling for pharmacokinetics and clot solubility was performed immediately before and one hour after each monthly dose of rFXIII as well as 14 days after the initial dose. Furthermore, preliminary results from an ongoing phase 3 safety extension trial (F13CD-3720) on FXIII activity and clot solubility were also analysed (cut-off date of 11 February 2011). Samples were drawn at pre-dose only.

The main features of the clinical pharmacology studies are shown in Table 17.

Table 17: Overview of Trials providing Evidence of Pharmacokinetics

Trial ID	Phase	Trial design	Dose level	Exposure	PK sampling
Healthy subjects					
NN1841-3788	1	Double-blind, cross-over trial investigating bioequivalence and pharmacokinetics of single doses of $rFXIII_{NN}$ and $rFXIII_{Avecia}$.	35 IU/kg	49 exposures to each drug substance.	At baseline, at hours 0.5, 1, 8, 24 and days 3, 7, 14, 21 and 28 post-dose
F13-1661	1	Single-dose, double-blind trial investigating PK and safety	0, 2, 6, 12, 30 and 60 IU/kg[a]	8 rFXIII exposures at each dose tier; 10 placebo doses	At baseline, at hours 0.5, 1, 4, 8, 24, 48, 72 and days 7, 14 and 28 post-dose
F13-1662	1	Single-dose, double-blind trial investigating PK and safety	0, 12 and 30 IU/kg daily for 5 days	8 exposures at each dose level	At baseline, at hours 0.5, 1, 4, 8 and days 5, 6, 7, 19 and 33
NN1810-3733	1	Single-dose, double-blind trial investigating PK and safety in healthy Japanese subjects	0, 12 and 35 IU/kg rFXIII	8 exposures at each dose level	At baseline, at hours 0.5, 1, 4, 8, 24, 48, 72 and days 7, 14, 21 and 28 post-dose
Patients with congenital FXIII deficiency					
F13-1663	1	Single-dose, open-label trial on safety and pharmacokinetics	2, 7, 24, 60 and 89 IU/kg[b]	11 exposures in 9 patients[c]	At baseline, at hours 0.5, 1, 4, 8, 24, 48, 72 and days 7, 14, 21 and 28 post-dose
F13CD-1725	3	Open-label trial of monthly replacement therapy (treatment period of 12 months)	35 IU/kg	471 exposures in 41 patients	Before and 1 hour after each monthly dose of rFXIII as well as 14 days after the initial dose
F13CD-3720	3b	Open-label; monthly replacement therapy (safety extension trial to F13CD-1725).	35 IU/kg	439 exposures in 33 patients[d] (cut-off date: 11 February 2011)	Before each monthly dose

[a] PK assessment performed at the two highest dose levels for Berichrom® assay. For enzyme-linked immunosorbent assays (ELISAs) more dose levels were investigated.
[b] PK assessment performed at the three highest dose levels
[c] Two patients receiving 2 IU/kg rFXIII were re-enrolled at higher dose levels.
[d] To expand the safety data base, the F13CD-3720 protocol was amended to allow for inclusion of additional patients into the trial. As of the cut-off date of 11 February 2011, this had resulted in exposure to a single additional dose in one such patient (not included in the 33 patients).

Pharmacokinetics in Healthy Subjects

An overview of the study results for pharmacokinetics in healthy volunteers is presented in Table 18.

Table 18: Pharmacokinetic Parameters in Healthy Subjects across Studies

	Trial ID	Dose (IU/kg)	N[a] M; F	$AUC_{0-\infty}$ (h*IU/mL) mean (SD)	C_{max} (IU/mL) mean (SD)	V_{ss} (mL/kg) mean (SD)	CL (mL/h/kg) mean (SD)	$t_{½}$ (h) mean (SD)	MRT (h) mean (SD)
Single dose	NN1841-3788[b]	35	50M	*278 (47)*[c] 301 (142)	*0.85 (24)*[c] 0.87 (0.21)[d]	*47 (25)*[c] 48 (12)	*0.13 (37)*[c] 0.14 (0.05)	*266 (64)*[c] 303 (195)	*372 (67)*[c] 423 (282)
	F13-1661	30	4M; 4F[e]	207 (61)	0.80 (0.14)	45 (18)	0.15 (0.05)	219 (80)	313 (139)
	F13-1661	60	5M; 3F[f]	342 (204)	1.02 (0.14)	69 (18)	0.24 (0.16)	273 (161)	402 (232)
Multiple dose	F13-1662	12	6M; 2F	AUC_{0-24h} 16.9 (2.1)	0.87 (0.19)	-	-	346 (215)	-
	F13-1662	30	2M; 6F	AUC_{0-24h} 37.4 (5.5)	1.80 (0.25)	-	-	167 (50)	-

PK endpoints derived from Berichrom® activity assay

[a] N = number of subjects exposed. M = male; F = female.

[b] Pooled data presented ($rFXIII_{Avecia}$ and $rFXIII_{NN}$) giving a total of 98 exposures (NN1841-3788).

[c] Geometric mean (CV) in *italics* (NN1841-3788).

[d] C_{30min} (NN1841-3788).

[e+f] Of these, 4[e] and 7[f] subjects contributed to the PK assessment (F13-1661).

Pharmacokinetics in Patients with FXIII deficiency

Study F13-1663 was a phase 1 escalating dose (2, 7, 24, 60 and 89 IU/kg IV single dose) study of the safety and pharmacokinetics of recombinant factor XIII in patients with congenital factor XIII deficiency (n=9, 5 male/4 female). The terminal half-life ranged from 6 to 12 days, when excluding a very short half-life (9 hours) of a FXIII B-subunit deficient subject. PK characteristics from the F13-1663 trial supported the chosen dose (35 IU/kg) in the F13CD-1725 trial. The reported values for $AUC_{0\text{-}inf}$ and initial concentration (C_0) are shown in Table 19 . There is an approximated linear relation between response and the 3 highest tested doses from 20 U/kg to 75 U/kg (corresponding to approximately 24 IU/kg and 89 IU/kg, respectively). Figure 1 represents a graphical presentation of the values in Table 19. The figure indicates that AUC and C_0 at the dose level of 35 IU/kg (29 U/kg) are predicted by linear interpolation from the observed values at the surrounding dose range from 20 U/kg to 75 U/kg.

Table 19: Pharmacokinetic parameters for berichrom, FXIII total A2, and A2B2 assays (individual data for subjects treated with 20, 50, and 75 U/kg rFXIII – Study F13-1663

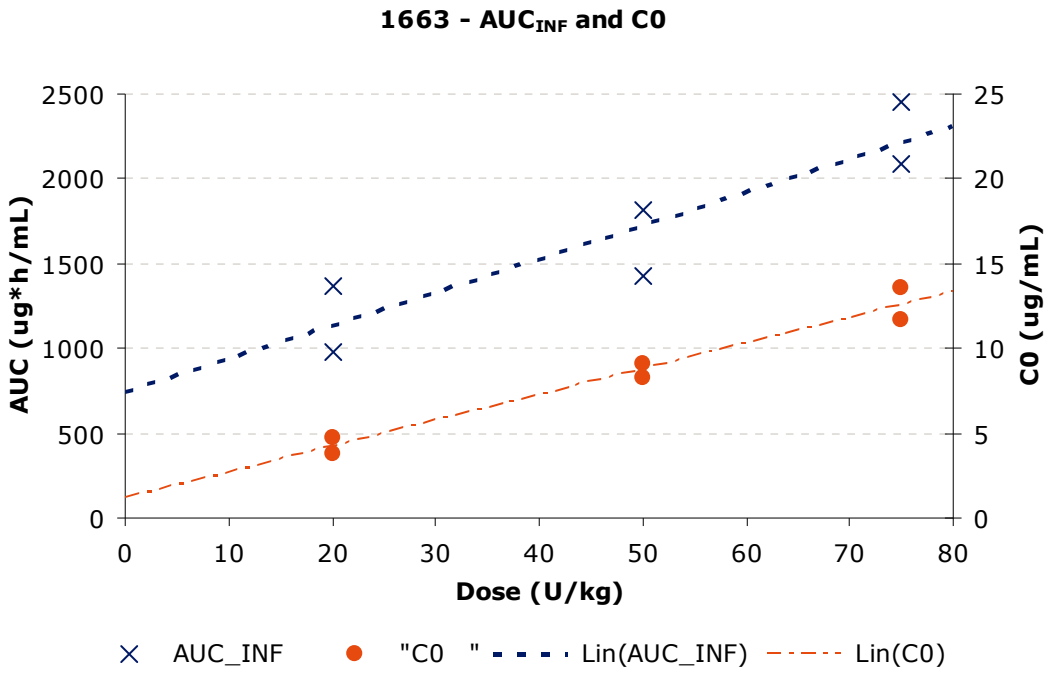
Assay	rFXIII Dose (U/kg)	Subject ID#	$t_{1/2}$ (h)	Conc[a] (µg/mL)	AUC_{0-t} (h•µg/mL)	AUC_{INF} (h•µg/mL)	CL (ml/h/kg)	MRT_{INF} (h)	V_{ss} (mL/kg)
Berichrom®	20	5	278	3.80	1080	1370	0.10	399	41.0
		6[b]	213	4.66	842	984	0.14	320	45.6
	50	7[c]	8.90	8.58	68.5	81.7	4.28	12.0	51.5
		8	149	8.24	1100	1430	0.24	208	50.8
		9	156	9.08	1410	1820	0.19	219	42.2
	75	10	202	13.6	2230	2450	0.22	270	58.4
		11	220	11.7	1820	2090	0.25	300	76.1
Total A_2	20	5	153	3.08	300	625	0.22	220	49.2
		6[b]	208	3.56	644	711	0.20	304	59.8
	50	7[c]	10.3	13.6	98.2	121	2.90	13.2	38.4
		8	156	5.39	484	902	0.39	220	85.2
		9	148	6.60	925	1160	0.30	205	62.1
	75	10	190	12.6	1120	1570	0.34	262	88.7
		11	217	9.21	959	1430	0.37	298	111
A_2B_2	20	5	219	8.17	1650	2540	0.06	312	17.2
		6[b]	264	7.90	1960	2480	0.06	419	23.6
	50	7[c]	NA	NA	NA	NA	NA	NA	NA
		8	151	13.1	2270	2980	0.12	212	24.6
		9	155	17.3	3060	3930	0.09	219	19.5
	75	10	214	18.5	4980	5560	0.10	287	27.3
		11	217	13.4	4110	4690	0.11	303	34.2

a For the Berichrom® and Total A_2 assays, initial concentration was estimated. For the A_2B_2 assay, maximum concentration was estimated.

b Subject 6 had normal levels of FXIII pre-dose, data was baseline corrected before PK analyses.

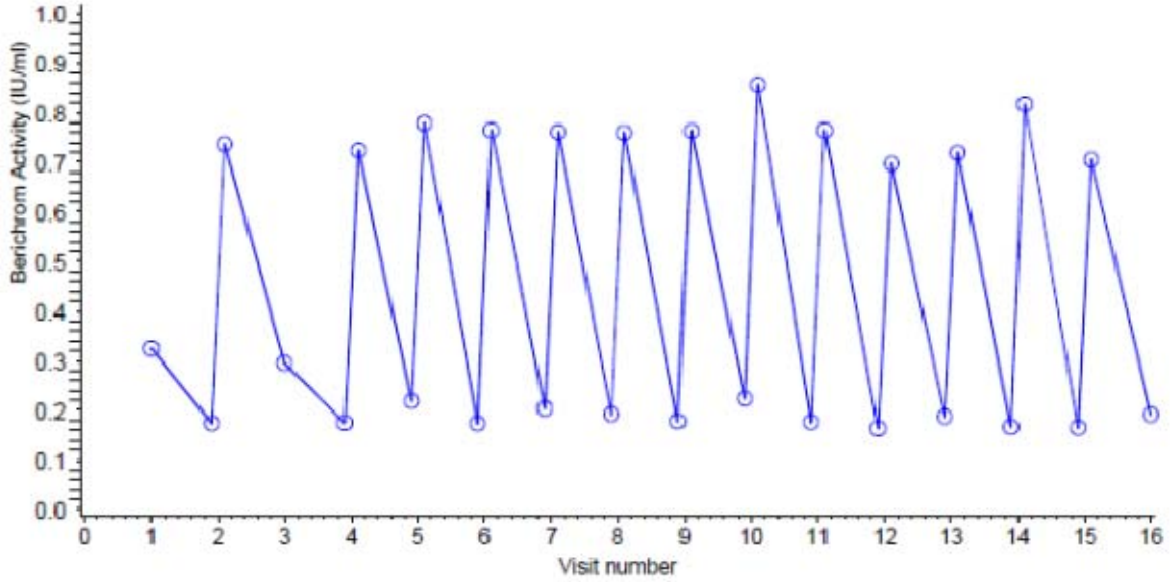
c Subject 7 was FXIII B subunit deficient as well as FXIII A subunit deficient.

Figure 1: Linear relation between response and the doses 20, 50, and 75 U/kg rFXIII – Study F13-1663



Study F13CD-1725 was a prospective, open-label, single arm trial evaluating the efficacy and safety of monthly replacement therapy with 35 IU/kg rFXIII in subjects with congenital factor XIII A-subunit deficiency (n=41). Blood sampling for pharmacokinetics was performed immediately before and one hour after each monthly dose as well as 14 days after the initial dose of rFXIII. Based on FXIII activity as measured for 471 monthly doses of rFXIII in the trial, the estimate of the half-life of rFXIII was 11.5 days.

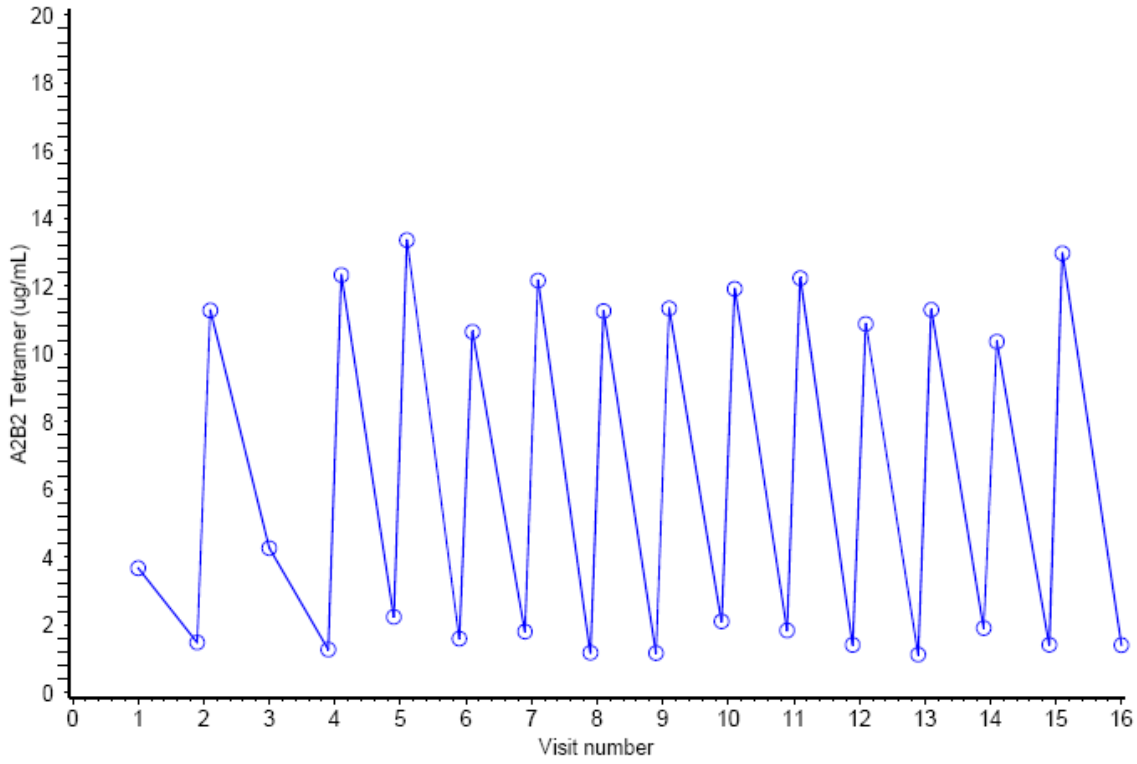
Figure 2: Mean Profile of FXIII Activity (IU/mL) in Patients with Congenital FXIII Deficiency Following Monthly Dosing of rFXIII for 52 Weeks



Visit 1: Screening visit. *Visit 2* and *Visits 4–15*: monthly (28 ± 2 days) dosing visits, during which FXIII activity (Berichrom®) was assessed immediately before rFXIII dosing (trough values) and at one hour after dosing (peak values). *Visit 3*: An additional follow-up visit conducted two weeks after the first dose of trial product. *Visit 16*: end-of-trial visit.

The shape of the mean profiles for A2B2 tetramer (Figure 3) and total FXIII A2 (data not shown) corresponded to the FXIII activity profile, reflecting that concentrations increased sharply after each rFXIII administration followed by a gradual decline during the subsequent month.

Figure 3: Mean Profile of A2B2 Activity (µg/mL) in Patients with Congenital FXIII Deficiency Following Monthly Dosing of rFXIII for 52 Weeks –Full Analysis Set



Distribution

There were no distribution studies submitted.

Elimination

There were no elimination studies submitted.

Dose proportionality and time dependencies

Information regarding the dose proportionality was obtained from Study F13-1661. This study was conducted in healthy volunteers after administration of a single escalating dose. The following doses were tested in parallel cohorts: 0, 2, 5, 10, 25 and 50 U/mL.

The main results of the study are shown in Table 20.

Table 20: Summary of NCA PK parameters determined from the Berichrom assay (µg/mL)

Assay	rFXIII Dose		$t_{1/2}$ (h)	C_{max} (µg/mL)	AUC_{0-t} (h•µg/mL)	AUC_{INF} (h•µg/mL)	CL (ml/h/kg)	MRT_{INF} (h)	V_{ss} (mL/kg)
Berichrom ®	2 U/kg (0.014 mg/kg)	N	0	0	0	0	0	0	0
		Mean	NA	NA	NA	NA	NA	NA	NA
		StDev	NA	NA	NA	NA	NA	NA	NA
		Min	NA	NA	NA	NA	NA	NA	NA
		Median	NA	NA	NA	NA	NA	NA	NA
		Max	NA	NA	NA	NA	NA	NA	NA
	5 U/kg (0.035 mg/kg)	N	0	0	0	0	0	0	0
		Mean	NA	NA	NA	NA	NA	NA	NA
		StDev	NA	NA	NA	NA	NA	NA	NA
		Min	NA	NA	NA	NA	NA	NA	NA
		Median	NA	NA	NA	NA	NA	NA	NA
		Max	NA	NA	NA	NA	NA	NA	NA
	10 U/kg (0.070 mg/kg)	N	0	1	1	0	0	0	0
		Mean	NE	4.070	1416.96	NE	NE	NE	NE
		StDev	NE	NE	NE	NE	NE	NE	NE
		Min	NE	4.07	1416.96	NE	NE	NE	NE
		Median	NE	4.07	1416.96	NE	NE	NE	NE
		Max	NE	4.07	1416.96	NE	NE	NE	NE
	25 U/kg (0.17 mg/kg)	N	4	4	4	4	4	4	4
		Mean	218.84	4.79	920.31	1236.55	0.15	313.464	44.909
		StDev	79.69	0.81	321.14	365.41	0.05	138.677	17.969
		Min	99.91	4.08	667.41	755.39	0.11	144.99	28.98
		Median	252.79	4.65	811.42	1290.29	0.14	322.68	40.81
		Max	269.85	5.77	1390.98	1610.24	0.23	463.50	69.04
	50 U/kg (0.35 mg/kg)	N	7	7	7	7	7	7	7
		Mean	272.77	6.14	1195.59	2047.18	0.24	402.183	69.383
		StDev	161.24	0.83	683.70	1222.28	0.16	231.536	17.982
		Min	79.57	4.51	491.64	631.69	0.08	109.30	56.77
		Median	221.95	6.57	993.22	1771.57	0.20	347.16	60.90
		Max	506.46	6.99	2651.33	4468.67	0.56	742.94	107.11

NA Not Analyzed due to exclusion criteria
NE Not Estimated due to inability to estimate λz or n=1

2.4.3. Pharmacodynamics

The pharmacodynamic properties of rFXIII were evaluated in phase 3 by testing for clot lysis.

Mechanism of action

The Applicant did not submit studies on the mechanism of action.

Primary and Secondary pharmacology

At present there are no markers that can quantitatively assess the *in vivo* pharmacodynamics of FXIII. As it is the quality of the clot that is affected in FXIII deficiency, the results of standard coagulation screening tests such as prothrombin time, activated partial thromboplastin time, fibrinogen level, and platelet count and bleeding time are all normal in FXIII deficiency. Therefore, clot lysis results from inadequate cross-linking of fibrin molecules is used as an indicator of FXIII deficiency.

Five *in vitro* studies were performed with normal and FXIII-deficient human blood. Clot lysis results were used in the assay. The clot solubility assay was included in pivotal F13CD-1725 phase 3 trial in patients with congenital FXIII deficiency. At unscheduled visits, samples were drawn before and one hour after administration of any FXIII containing product. Results are summarised in the table below:

Table 21: Clot Lysis Observations in Patients with Congenital FXIII Deficiency Following Monthly Dosing of FXIII for 52 Weeks - Trial F13CD-1725

	rFXIII 35 IU/kg	
	Pre-Dose N (%)	Post-Dose N (%)
Screening (Visit 1)	2 (4.9)	
Week 0 (Visit 2 - Baseline)	8 (19.5)	1 (2.4)
Week 2 (Visit 3)	0 (0.0)	
Week 4 (Visit 4)	4 (9.8)	0 (0.0)
Week 8 (Visit 5)	1 (2.4)	0 (0.0)
Week 12 (Visit 6)	5 (12.2)	1 (2.4)
Week 16 (Visit 7)	0 (0.0)	0 (0.0)
Week 20 (Visit 8)	3 (7.3)	0 (0.0)
Week 24 (Visit 9)	1 (2.4)	0 (0.0)
Week 28 (Visit 10)	7 (17.1)	1 (2.4)
Week 32 (Visit 11)	2 (4.9)	2 (4.9)
Week 36 (Visit 12)	2 (4.9)	1 (2.4)
Week 40 (Visit 13)	3 (7.3)	1 (2.4)
Week 44 (Visit 14)	1 (2.4)	1 (2.4)
Week 48 (Visit 15)	3 (7.3)	0 (0.0)
Week 52 (Visit 16)	3 (7.3)	
Unscheduled Visit	1 (2.4)	0 (0.0)
All Dosing Visits[b]		
All Visit Patients count	533	533
Normal	419 (78.6)	444 (83.3)
Lysis	40 (7.5)	8 (1.5)
Not recorded	74 (13.9)	81 (15.2)

ssay used: clot solubility assay
For all dosing visits, only observations from planned dosing visits were considered, i.e. screening visit, additional follow-up visit two weeks after first dose (Visit 3) and end-of-trial visit (Visit 16) were excluded.

2.4.4. Discussion on clinical pharmacology

The rFXIII drug substance used for nonclinical studies and the larger part of the clinical development program was produced at the initial AS manufacturing site ($rFXIII_{IASMS}$). One clinical trial, NN1841-3788, was conducted in 50 healthy male subjects to demonstrate PK characterization of both $rFXIII_{IASMS}$ and $rFXIII_{NN}$. The outcome of the study suggests that both active substances may behave similarly.

The findings of Study NN-1841-3788 (based on Berichrom technique) suggest that NovoThirteen is not distributed to extra-vascular compartment (Vss is approximately 47 mL/kg) with a clearance of approximately 0.13 mL/h/kg and a half-life of approximately 10-12 days. As the product under consideration is an endogenous substance obtained by DNA technique, no formal investigations of the elimination route are required.

Available data from studies in healthy subjects as well as in patients suggest strongly that plasma levels of rFXIII increase less than proportionally to the dose in a pronounced manner.

The findings of Study F13CD-1725 where patients were treated for up to 16 months do not suggest a shift in the PK of NovoThirteen over time.

The inter-subject variability of AUC as estimated by the coefficient of variation (CV) is moderate (approximately 25).

Because the recombinant protein is similar to the endogenous protein, no PK interaction studies were submitted.

Out of the five phase I trials, the proposed treatment of 35 IU/kg body weight once monthly has been investigated in two studies (NN1841-3788 and NN1810-3733) including healthy subjects. There is no Pk study performed using the chosen dose of 35 IU/kg $rFXIII_{NN}$ in FXIII deficient patients in the phase III studies. The claimed 35 IU/kg dose has been predicted from an extrapolation of absolute FXIII activity results obtained in the F13-1663 phase I safety study where several doses (2, 7, 24, 60 and 89 IU/kg), were tested. A well approximated linear relation between the administered dose and increase in FXIII plasma activity was observed in the dose interval 24 IU/kg to 89 IU/kg, see Figure 1. Administration of 24 IU/kg, 60 IU/kg or 89 IU/kg resulted in absolute FXIII activity increases of 57-59%, 105-129% and 160-181%, respectively, up to one hour after dosing. The 35 IU/Kg dose is predicted to have FXIII activity well within the normal range in patients with congenital FXIII deficiency, but below 100%, at the time of injection and maintain the activity level >10% at four weeks. A mean 10% trough level was targeted to ensure haemostatic coverage in all patients when considering the variability in bleeding tendency at relatively low FXIII activity levels. The CHMP requested the Applicant to evaluate full PK data using the intended dose of 35 IU/kg.

No PK investigations have been performed in children over 6 years. So far only 9 (6-11 years) and 6 (12-17 years) paediatric patients have been included into the pivotal and extension trial. The CHMP requested further clinical data in children in the extension study applying the regimen of 35 IU/kg every 4 weeks. The results would be an increase of patient number (doubled) and also longer time for the investigation (see also Discussion on clinical efficacy).

The pharmacodynamic properties of rFXIII were evaluated in phase 3 by testing for clot lysis. Clotting dynamics were also exploratively assessed by thromboelastography in the F13-1663 phase 1 trial in patients with congenital FXIII deficiency. Coagulation methods such as rotational thromboelastography (ROTEM) or clot solubility test have not been validated as surrogate efficacy endpoints in this setting. Therefore, surrogate PK and PD data from clot lysis are considered as only qualitative data and do not directly support the pharmacodynamic effect of FXIII.

2.4.5. Conclusions on clinical pharmacology

The CHMP was of the opinion that as no validated pharmacodynamic markers to measure the activity of rFXIII were available, the investigations submitted by the Applicant were regarded as sufficient. The clinical pharmacology studies submitted by the Applicant were adequate but questions remained on the full PK assessment in patients dosed with the chosen dose 35 IU/kg administered in phase III studies. In addition, the available data from study F13CD-3720 showed that the target level of rFXIII (10% of plasma level in non deficient patients at day 28 post administration) was not achieved in approximately 5 % of patients. Thus, the CHMP considers the following measures as stated in the RMP necessary to address the issues related to pharmacology:

- To amend the currently ongoing extension study (F13CD-3720) to include PK sampling of 5 patients at steady state that are dosed with the recommended posology of 35 IU/kg. The PK sampling will generate full PK profiles, with blood samples being taken pre-dose, 1 hour after dosing and 7, 14, 21, and 28 days after dosing. The details are outlined in a draft protocol amendment and the results of the additional PK assessments will be provided post marketing authorisation.

2.5. Clinical efficacy

The following table provides an overview of the trials that were submitted in support of the sought indication.

Table 22: Overview of Trials Providing Evidence of Efficacy

Study ID	No. of study centres / locations	Design	Study Posology	Study Objective	Subjs by arm entered/ compl.	Duration	Gender M/F Median Age	Primary Endpoint
F13CD-1725	23 sites AT, CA, FI, FR, DE, IL, IT, ES, CH, GB, US	Multi-centre, multi-national Open-label; Single-arm	35 IU/kg rFXIII Monthly doses (28± 2 days) (i.v)	Confirmatory phase 3 efficacy and safety trial	CD patients 41 /33	Multi-dose 52 weeks (Complete)	23M/18F 23 years	Rate of bleeding episodes requiring treatment with a FXIII-containing product
F13CD-3720	19 sites AT, CA, FI, FR, DE, IL, IT, ES, CH, GB, US	Similar to above	Similar to above	Safety extension trial to the phase 3 F13CD-1725 trial	CD patients 33 completers from F13CD-1725 *	Multi-dose Minimum 52 weeks. (Ongoing)	20M,13F	Adverse events

2.5.1. Dose response study

No dose response study was submitted.

An overview of the different doses investigated in Phase I trials is presented below.

Table 23: Overview of dose levels used in clinical trials

Trial ID	Trial design	Dose level : N (M/F)
Healthy subjects		
NN1841-3788	Double-blind, cross-over trial investigating bioequivalence and pharmacokinetics of single doses of $rFXIII_{NN}$ and $rFXIII_{IASMS}$.	35 IU/kg : 50 (50/0)
F13-1661	Single-dose, double-blind trial investigating PK and safety	Placebo: 10 (7/3) 2 IU/kg: 8 (6/2) 6 IU/kg: 8 (6/2) 12 IU/kg: 8 (1/7) 30 IU/kg: 8 (4/4) 60 IU/kg: 8 (5/3)
F13-1662	Multiple-dose (once-daily for 5 days), double-blind trial investigating PK and safety	Placebo: 8 (5/3) 12 IU/kg: 8 (6/2) 30 IU/kg: 8 (2/6)
NN1810-3733	Single-dose, double-blind trial investigating PK and safety in healthy Japanese subjects	Placebo: 8 (8/0) 12 IU/kg: 8 (8/0)

		35 IU/kg: 8 (8/0)
Patients with congenital FXIII deficiency		
F13-1663	Single-dose, open-label trial on safety and pharmacokinetics	2 IU/kg: 2 (1/1) 7 IU/kg: 2 (1/1) 24 IU/kg: 2 (2/0) 60 IU/kg: 3 (1/2) 89 IU/kg: 2 (1/1)

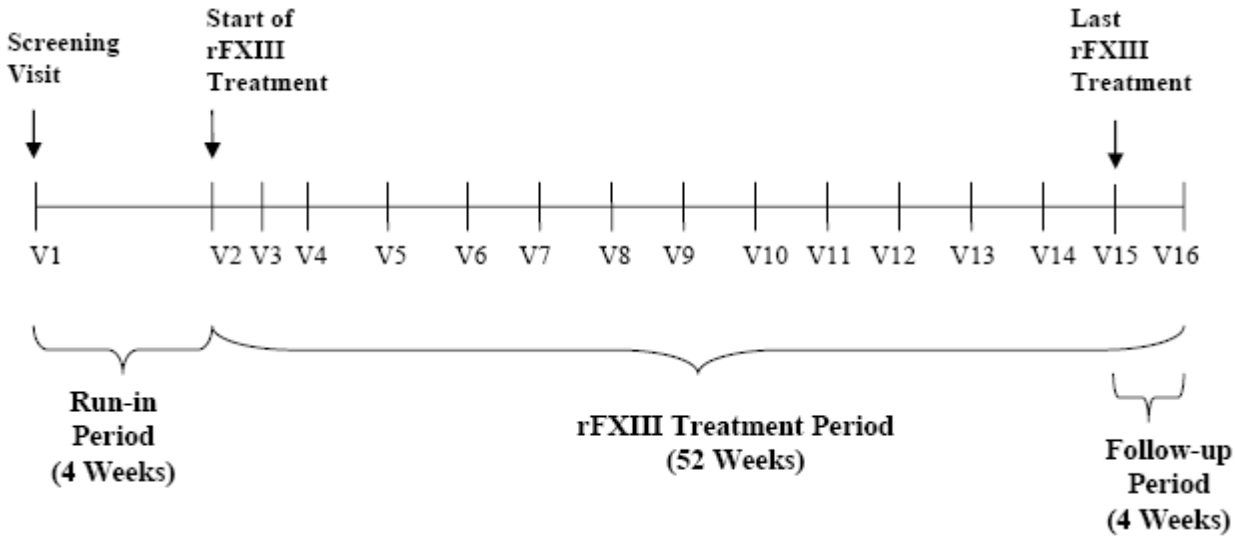
The estimations from the PK characteristics from the F13-1663 trial supported the chosen dose (35 IU/kg) in the F13CD-1725 trial.

2.5.2. Main studies

F13CD-1725: A Multi-Centre, Open-Label, Single-Arm and Multiple Dosing Trial on Efficacy and Safety of Monthly Replacement Therapy with Recombinant Factor XIII (rFXIII) in Subjects with Congenital Factor XIII Deficiency

Methods

An overview of the study design is shown below.



Patients who completed the F13CD-1725 trial could enter into the F13CD-3720 extension period.

Study Participants

Main Inclusion Criteria

Diagnosis of congenital FXIII A-subunit deficiency (confirmed by genotyping at screening visit).

Prior to screening, for patients

- on regular replacement therapy: Treatment with regular FXIII replacement therapy initiated at least 6 months prior to screening and one of the following: a documented history of ≥1 treatment-requiring bleeding episode prior to initiation of regular replacement therapy or a documented family history of FXIII congenital deficiency.

- receiving on-demand treatment: Documented history of ≥ 2 bleeding episodes requiring treatment with FXIII containing blood products within the last 12 months prior to screening

Patients with age ≥ 6 years and a weight ≥20 kg. Before enrolling patients ≥ 6 to < 12 years of age in the EU countries, 7 patients had to be exposed for 12 weeks (3 exposures) to trial product with a safe safety profile.

- German sites were only to include patients above or equal to 12 year of age.
- In Israel: Patients with age > 6 years and a weight > 20 kg. Before enrolling patients of > 6 to < 18 years of age, 7 patients had to be exposed for 12 weeks (3 exposures) to trial product with a safe safety profile.

Main Exclusion Criteria

- Known neutralizing antibodies (inhibitors) towards FXIII.
- Documented history of ≥ 2 treatment-requiring bleeding episodes per year during previous regular replacement therapy with FXIII containing blood products (FFP, pd FXIII and cryoprecipitate).
- Known or suspected allergy to trial product(s) or related products.
- Planned major surgery during the trial period. Catheter, ports and dental extractions and do not count as surgeries and will not exclude the patient.
- Platelet count (thrombocytes) < 75 × 10^9/L; Renal insufficiency defined as current dialysis therapy.
- Any known congenital or acquired coagulation disorder other than congenital FXIII deficiency; Any history of confirmed venous or arterial thromboembolic events.
- Patient that received any anti-thrombotic or anti-platelet drugs within 7 days of trial enrolment.
- Females of childbearing potential who are pregnant, breastfeeding or intend to become pregnant or are not using adequate contraceptive methods.
-

Treatments

- Treatments administered

During the treatment period of 52 weeks (visit 2 through to visit 15), patients received 35 IU/kg rFXIII every 4th week (28±2 days). At each visit, the dose was adjusted according to the actual weight of the patient. The reconstituted trial product was administered as a slow intravenous injection (at a rate that was not to exceed 1-2 mL per minute).

The trial comprised 16 scheduled visits (Visit 1: "Screening visit" at Week -4; Visits 2-15: "Treatment period" at Week 0, 2, 4, 8 and every 4 weeks up to Week 48; Visit 16 "End-of-trial visit" at Week 52) and additional visits in case of treatment-requiring bleeding episodes.

- Prior and concomitant therapy

It was a requirement for inclusion into the trial that patients receiving regular replacement therapy had received treatment with FXIII-containing blood products for at least 6 months prior to screening, and that patients receiving only on-demand treatment had received FXIII-containing blood products on at least two occasions within 12 months of the screening visit.

Non-emergency use of FXIII-containing products other than rFXIII was not allowed during the trial period. In case of acute bleeding episodes, any additional treatment as per investigator judgment was to be according to local standard practice, and additional doses of rFXIII could therefore not be used to treat such breakthrough bleedings. Treatment of bleeding episodes was to be under direct supervision of the investigator or delegated medically qualified staff.

Objectives

Primary Objective: To evaluate the efficacy of monthly replacement therapy with rFXIII in prevention of bleeding episodes in patients with congenital FXIII deficiency.

Secondary Objective: To evaluate the safety of monthly replacement therapy with rFXIII.

Outcomes/endpoints

Primary endpoint:

- Rate (number per patient year) of "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period

Secondary endpoints:

- Percentage of patients without "bleeding episodes requiring treatment" with a FXIII-containing product during the treatment period
- Rate (number per patient year) of spontaneous "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period
- Rate (number per patient year) of traumatic "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period
- Rate (number per patient year) of intracranial "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period
  - *Bleeding episodes:* Throughout the 52-week rFXIII treatment period, details of all bleeding episodes were recorded at scheduled visits as well as at unscheduled visits, the latter in case of treatment-requiring bleedings. The following details were recorded for bleeding events that required treatment with a FXIII-containing product: Date and time of onset of bleeding / Cause of bleed (spontaneous or traumatic) / Site of bleeding (central nervous system bleeding, haemarthrosis, gastrointestinal bleeding, subcutaneous bleeding, muscular bleeding or other) / Haemostatic drug used for treatment of bleeding episodes (drug name, dose and time of administration) / Other therapy used (e.g., compression, ice) / Date and time of bleeding resolution
- Percentage of patients having a normal clot solubility one hour after rFXIII administration and 28 days after rFXIII administration
  - *Clot solubility:* At screening, at visits 1-16 and at any unscheduled visit, blood samples were drawn for analysis of clot solubility. At dosing visits (visits 2-15 except visit 3), samples were to be drawn before and one hour after administration of trial product. At unscheduled visits, samples were to be drawn before administration of any FXIII-containing product. The applied

assay is a qualitative assay typically used to screen for FXIII deficiency. The assay is based on the ability to dissolve fibrin clots that have not undergone FXIII-induced stabilization. Normal blood clots generally remain stable for 24 hours or more, while clots in which fibrin molecules have not been cross-linked are soluble within minutes. The assay was performed at the designated central laboratory.

- Number of patients withdrawn from the trial due to lack of efficacy of rFXIII treatment
- Level of FXIII activity one hour after rFXIII administration and 28 days after rFXIII administration

Safety Endpoints

- Adverse events
- Number of patients with rFXIII antibody development
- Number of patients with rFXIII inhibitor development

In addition, overall safety was evaluated by investigating laboratory parameters, vital signs, physical examination and body measurements. In addition to haematology, biochemistry and urinalysis parameters, clinical laboratory tests comprised a FXIII Berichrom activity assay, FXIII antigen ELISAs, FXIII genotyping, immunogenicity assays and tests for coagulation parameters.

Sample size

Based on retrospectively collected data, the bleeding rate for patients treated only on-demand was estimated by a Poisson model. The estimate was 2.91 bleedings per year with a 95% confidence interval (CI) of [2.18; 3.87]. The same was done for patients receiving regular replacement therapy, and the yearly bleeding rate was estimated at 0.33 with a 95% CI of [0.22; 0.52].

Assuming a yearly bleeding rate for patients receiving monthly replacement therapy with rFXIII of 0.52 and comparing with a fixed rate of 2.91 bleedings yearly by means of a Poisson model with a type I error rate of 5%, a total of 40 patients was calculated to yield 99% power.

Randomisation

The study was a single arm study.

Blinding (masking)

The study was an open label study.

Statistical methods

All patients who receive at least one dose of trial product were to be included in the Full Analysis Set (FAS). Data from patients that have discontinued the trial for any reason would be included in the analysis up to the point of discontinuation.

The per-protocol analysis set (PP) comprised all patients completing the 52-week rFXIII treatment period, and furthermore excluded any patients with protocol violations that are judged to affect the primary efficacy evaluation.

The safety analyses were performed for the FAS population, and the efficacy analyses were performed for both the FAS and the PP Populations. The FAS was the primary analyses, with PP analyses providing supporting evidence.

- Statistical Methodology

All analyses were pre-specified in the statistical analysis plan (SAP).

Unclear or missing data were queried.

- Primary Endpoint

The primary endpoint was evaluated by a poisson model (log-link), comparing the data to a fixed placebo rate of 2.91 bleedings yearly. Age was included as a continuous covariate and the total observation time during the treatment period was used as an offset in the model. The model also took into account those patients withdrawing before end of trial, by adjusting the length of time under observation. The estimated rate was calculated, adjusting for overdispersion. The overdispersion was estimated by Pearson's chi-square statistic divided by its degrees of freedom.

Monthly replacement therapy with rFXIII was concluded as superior to on-demand treatment with FXIII containing products (defined as placebo) if the yearly bleeding rate for the rFXIII group, λ, was lower than 2.91. The null hypothesis of no difference between rFXIII and placebo was rejected if the upper limit of the 95% confidence interval for λ was less than 2.91.

The number of bleeding episodes was tabulated along with point estimate and 95% confidence interval.

Additional Analyses: Bleeding rate was summarised by age group (<18 vs >=18) as well as by time since last dose (<14 days vs >=14 days).

- Secondary Efficacy Endpoints

The percentage of patients without "bleeding episodes requiring treatment" with FXIII containing products was evaluated by a binomial model including age as a covariate, comparing the data to a fixed placebo probability of whether or not patients were experiencing any treatment-requiring bleedings.

As for the primary endpoint the fixed placebo probability was based on the historical data collected.

The probability of not having any bleeding episodes requiring treatment for patients treated on-demand was estimated by a binomial model. Estimate was a probability of 0.25 with a 95% confidence interval of [0.10; 0.51].

Monthly replacement therapy with rFXIII was concluded as superior to on-demand treatment with FXIII containing products (defined as placebo) if the probability of not having any treatment-requiring

bleeding episodes for the rFXIII group, p, was greater than 0.25. The null hypothesis of no difference between rFXIII and placebo was rejected if the lower limit of the 95% confidence interval for p was greater or equal to 0.25.

The percentage of patients without any treatment-requiring bleeding episodes was tabulated along with point estimate and 95% confidence interval.

The rate (number per patient year) of spontaneous, traumatic and intracranial "bleeding episodes requiring treatment" with FXIII containing products during the rFXIII treatment period was tabulated.

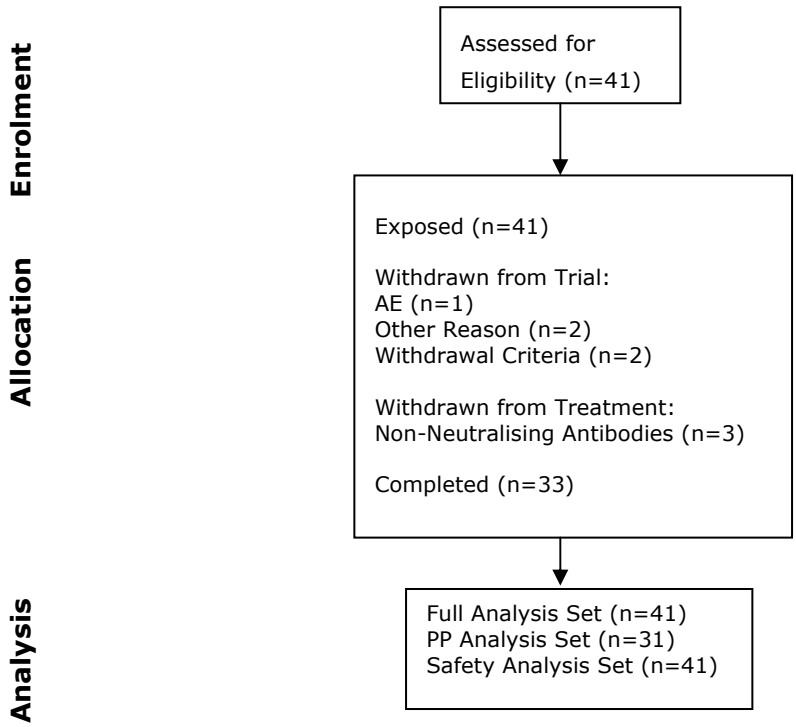
The percentage of patients having normal clot solubility one hour after rFXIII administration and 28 days after rFXIII administration was tabulated and listed.

The level of FXIII activity one hour after rFXIII administration and 28 days after rFXIII administration was tabulated and listed.

The number of patients withdrawn from the trial due to lack of efficacy of rFXIII treatment was tabulated and listed.

Results

Participant flow



Recruitment

Of a total of 29 initiated trial sites, 23 sites enrolled and dosed at least one patient. The country distribution was as follows (number of actively recruiting sites per country in parenthesis): Austria (1), Canada (1), Finland (1), France (1), Germany (3), Israel (2), Italy (1), Spain (1), Switzerland (1), UK (3) and USA (8). The trial was initiated on 18 August 2008 and completed on 15 April 2010.

Conduct of the study

A total of 41 patients were enrolled and exposed to trial drug following screening procedures.

Of the 41 enrolled and dosed patients, 5 patients were withdrawn from the trial:

- One patient was withdrawn by the parents at visit 5 because parents and patient felt that there were too many blood samples.
- One patient was withdrawn by the investigator after visit 6 due to worsening of neutropenia and leucopenia.
- Two patients became pregnant and were withdrawn after visits 11 and 14, respectively.
- One patient withdrew after visit 9 for personal reasons.

In addition, 3 patients were withdrawn from treatment due to development of antibodies but remained in the trial for safety monitoring purposes.

The remaining 33 patients completed the pre-defined trial treatment period. All 33 patients who completed trial F13CD-1725 were enrolled and exposed to trial drug in trial F13CD3720. Of the 33 enrolled and dosed patients, 3 patients were withdrawn from the trial. One patient was withdrawn by the investigator after 3 doses of rFXIII due to pregnancy (withdrawal criteria) and another patient withdrew her consent due to a wish to become pregnant after 4 doses of rFXIII. A third patient wished to withdraw after 13 doses due to relocation of the clinical trial site.

Protocol Deviations

All trial participants met inclusion criteria and none fulfilled any of the exclusion criteria. In addition to withdrawal from treatment or trial, the following major protocol deviations led to exclusion from PP analysis:

- One patient received preventive plasma-derived FXIII during hospitalisation even though no bleeding was present.
- One patient did not receive any trial product at visit 4.

A total of 6 substantial amendments were implemented after initiation of patient enrolment.

Baseline data

The demographics of the trial population at baseline are presented in Table 24 below.

Table 24: Baseline Demographics - Full Analysis Set

	rFXIII 35 IU/Kg
Number of Subjects	41
Age (years)	
N	41
Mean (SD)	26.4 (15.9)
Median	23.0
Min ; Max	7.0 ; 60.0
Sex, N(%)	
N	41 (100)
Female	18 (44)
Male	23 (56)
Race, N(%)	
N	41 (100)
Black or African American	2 (5)
White	28 (68)
Asian	5 (12)
Other	5 (12)
Unknown	1 (2)

The French patients are marked as Unknown as per the French Authorities Guidelines

FXIII A-subunit deficiency was confirmed for all patients. One patient furthermore had a heterozygous missense mutation in the F13B gene. The effect of the F13B gene mutation is unknown, but ELISA results for FXIII B-subunits showed that this patient did not appear to have abnormally low levels of B-units and that the B-subunits were functionally capable of binding to A-subunits as reflected in reduced levels of B-subunits and increased levels of A2B2 following injection of rFXIII.

All patients except two had received regular replacement therapy with FXIII-containing products prior to enrolment into the trial, which further attests to the severity of FXIII deficiency in the investigated patient population.

A total of 8 of the 41 patients included in the trial exhibited abnormal clot solubility (i.e., clot lysis) at baseline. The vast majority of physical examinations were rated as normal.

Historical controls

Historical data from the Congenital FXIII-deficiency Questionnaire obtained between June and September 2005 were available for a total of 92 patients. Patients were included from 13 countries with 1 to 16 centers per country. The majority of the patients were collected at 16 sites in Germany. A total of 45 female and 47 male patients were included and the majority of the patients were White. The reported mean and median age of the patients were 26.7 and 25.0 years, respectively. A total of 87 of the 92 patients included were reported to be alive at the time of data collection. The reported mean and median body weight of the patients were 59 and 61 kg, respectively. For the majority of the patients, the subtype of the FXIII-deficiency (deficiency of the FXIII subunit A and/or B) was unknown. A total of 39 patients were classified as FXIII-subunit A deficient, 2 patients as FXIII-subunit B-deficient, and 4 patients as both FXIII-subunit A- and B deficient. The measured mean and median FXIII activity were 7.1 and 3.5 percent of normal, respectively, and ranged from 0.0 to 55.0% (n=78).

A total of 69 of the 92 (75%) patients had a history of regular FXIII replacement therapy for bleeding prophylaxis and the majority of these patients initiated prophylactic treatment in childhood. Fibrogammin P (plasma-derived FXIII) was the most frequently used FXIII-containing product. The mean and median doses of Fibrogammin P administered for prophylactic therapy were 20.1 and 14.9 units per kg, ranging from 5.7 to 64.1 units per kg, administered with a frequency ranging from twice a week to once every six weeks. Overall, a total of 17 of 64 patients (27%) with a history of prophylactic treatment and available bleeding information had experienced breakthrough bleedings.

The number of breakthrough bleeds per year ranged from 0 to 7 per patient, with a mean and median number of bleeds of 0.3 and 0.0 per patient, respectively.

A total of 3 patients had never received any type of treatment and a total of 20 patients were treated on-demand for the management of acute bleeding episodes without having any current prophylactic treatment. Only 1 patient with a history of CNS bleedings had been treated on-demand only. The number of bleeding episodes requiring on-demand treatment ranged from 0 to 12 per patient, with a mean and median number of bleeds of 2.9 and 2.0 per patient year, respectively (see Table 25).

Table 25: Summary of Bleeding Frequency by Treatment Modality in Historical Controls

		Prophylaxis	On-demand
Total Number of Patients		69	23
Number of Patients	With Bleeds	17*	12
	Without Bleeds	47	4**
	With Unknown Bleeding History	5	7
Number of Patients use for frequency calculation***		60	16
Number of Bleeds per Year****	Total	20.0*****	46.5******
	Range	0- 7	0-12
	Average	0.3 (20.0/60)	2.9 (46.5/16)

* Only 13 patients have data available on number of bleedings. ** Only 4 patients have at least one year exposure to FXIII. *** Excluded patients showed incomplete data for calculation. **** Counted as total for those patients used for the frequency calculation. ***** All types of bleeds (both treatment requiring and non-treated. ****** Only treatment requiring bleeds.

Numbers analysed

All 41 patients who received at least one dose of trial product were included in the full analysis set and the safety analysis set in trial F13CD-1725.

The per-protocol (PP) analysis set comprised patients completing the 52-week rFXIII treatment period and furthermore excluded any patients with protocol violations that were judged to affect the primary efficacy evaluation. A total of 33 patients completed the pre-planned treatment period, two of whom were excluded from PP analysis due to significant protocol violations (one patient received preventive plasma-derived FXIII treatment, and another patient did not receive trial drug at visit 4 (week 8)). The remaining 31 patients comprised the per protocol (PP) analysis set.

Outcomes and estimation

- **Primary endpoint**

Rate (number per subject year) of "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period

The results for the rate of bleeding episodes that required treatment with a FXIII-containing product (in the following labelled treatment-requiring bleeds) are summarised in Table 26.

Table 26: Treatment-requiring Bleeding Episodes - Full Analysis Set

		Historical controls

	rFXIII 35 IU/kg	Prophylaxis	On-demand
Number of patients*	41	60	16
Number of patients with bleed	4	na	na
Total number of bleeds**	5	20	46.5
Range of bleedings	0 - 2	0 - 7	0 - 12
Mean bleedings per patients	0.122	na	na
Mean observation period (days)	322	na	na
Mean yearly bleeding rate	0.138	0.3	2.9

* for historical controls: number of patients used for frequency calculation
** for historical controls: total number of bleeds per year
Cross-reference: Trial F13CD-1725 (M5.3.5.1), EOT Table 14.2.3, Trial F13CD-QUEST, EOT Table 27

In the primary endpoint analysis, the age-adjusted rate (number per subject year) of treatment-requiring bleeding episodes during the rFXIII treatment period was 0.048/year (95% CI: 0.0094 - 0.2501), which is less than 2.91 yearly bleeding rate. Thus, endpoint met the criteria for superiority.

There was a 4-week run-in period before attending the screening visit for subjects who were to receive their usual replacement therapy dose to ensure that the previous treatment did not influence the bleeding frequency in the first month of the trial. As the 4-week run-in period may be considered too short considering the maximum half-life of rFXIII of around 12 days, the primary analysis of the rate of bleeding episodes requiring treatment was repeated, taking into account the data collected following a second administration. The outcome of the analysis is shown in Table 27.

Table 27: Analysis of bleeding rates excluding data in the first month compared with the original analysis

Analysis	N	Number of bleeds	Mean period (days)	Crude bleeding rate (yearly)	Poisson-based bleeding rate (yearly)	95% CI
Modified*	41	5	294	0.151	0.053	[0.010; 0.272]
Original	41	5	322	0.138	0.048	[0.009; 0.250]

*Excluding data in the first month (from first to second dosing with rFXIII).

During the rFXIII treatment period (mean observation period was 322 days), there were five treatment-requiring bleeding episodes observed in four patients. All five events were traumatic bleeding episodes.

- **Secondary endpoints**

Percentage of patients without "bleeding episodes requiring treatment" with a FXIII-containing product during the treatment period

Thirty seven of the 41 patients comprising the full analysis set did not experience any treatment requiring bleeding episodes during the trial. Age-adjusted statistical analysis determined the probability of not having any treatment-requiring bleedings during the trial period to 0.9581/year for the trial period of approximately one year (Table 28).

Table 28: Probability of not having any bleeding episodes the require treatment in patients treated with rFXIII - Full Analysis Set

Treatment	N	Mean(p)	SE	Confidence Interval
rFXIII 35 IU/kg	41	0.9581	1.1045	[0.7242 ; 0.9950]

The probability of not having any bleeding episodes requiring treatment for subjects treated on-demand is estimated by a binomial model including age as a covariate, comparing the data to a fixed placebo probability of whether or not subjects are experiencing any treatment-requiring bleedings.

Rate (number per patient year) of spontaneous "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period

There were no spontaneous treatment-requiring bleeds occurred during the rFXIII treatment period.

Rate (number per patient year) of traumatic "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period

A summary of the results for the rate of traumatic bleeding episodes requiring treatment with a FXIII-containing product during the rFXIII treatment period is shown in Table 29.

Table 29: Summary of rate of Traumatic treatment requiring bleeding episodes - Full Analysis Set

	rFXIII 35 IU/kg
Number of subjects	41
Number of subjects with bleed	4
Total number of bleeds	5
Range of bleedings	0 ; 2
Mean bleedings per subject	0.122
Mean observation period (days)	343
Mean yearly bleeding rate	0.140

Rate (number per subject year) of intracranial "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period

There were no intracranial treatment-requiring bleeds occurring during the rFXIII treatment period.

Percentage of subjects having a normal clot solubility one hour after rFXIII administration and 28 days after rFXIII administration

The number of patients with an abnormal finding (i.e., clot lysis) prior to dosing ranged from 0 to 7 across visits when considering time points corresponding to 4 weeks after the preceding administration of rFXIII (i.e., pre-dose from week 4 to week 52). The proportion of patients exhibiting clot lysis at 1 hour after rFXIII dosing was lower as compared to pre-dose (1.5% versus 7.5% of patients).

Number of subjects withdrawn from the trial due to lack of efficacy of rFXIII treatment

There were no patients withdrawn due to lack of efficacy of rFXIII treatment.

Ancillary analyses

Analyses were performed on bleeding rate by age group (<18 vs ≥18) (Table 30) and by time since last dose (<14 days vs ≥14 days) (Table 31).

Table 30: Summary of rate of treatment requiring bleeding episodes by age group - Full Analysis Set

	rFXIII 35 IU/kg
Number of subjects	41
Age <18	
Number of subjects	15
Number of subjects with bleed	3
Total number of bleeds	4
Range of bleedings	0 ; 2
Mean bleedings per subject	0.267
Mean observation period (days)	269
Mean yearly bleeding rate	0.362
Age >=18	
Number of subjects	26
Number of subjects with bleed	1
Total number of bleeds	1
Range of bleedings	0 ; 1
Mean bleedings per subject	0.038
Mean observation period (days)	353
Mean yearly bleeding rate	0.040

Table 31: Summary of rate of treatment requiring bleeding episodes by Time since last dose – Full Analysis Set

	rFXIII 35 IU/kg
Number of subjects	41
Time since last dose <14 days	
Number of subjects	41
Number of subjects with bleed	1
Total number of bleeds	1
Range of bleedings	1 ; 1
Mean bleedings per subject	0.024
Mean observation period (days)	175
Mean yearly bleeding rate	0.051
Time since last dose >=14 days	
Number of subjects	41
Number of subjects with bleed	3
Total number of bleeds	4
Range of bleedings	1 ; 2
Mean bleedings per subject	0.098
Mean observation period (days)	147
Mean yearly bleeding rate	0.242

Title: F13CD-3720: A Multi-Centre, Open-Label, Single-Arm and Multiple Dosing Trial on Safety of Monthly Replacement Therapy with Recombinant Factor XIII (rFXIII) in Subjects with Congenital Factor XIII Deficiency (cut-off date 11 February 2011).

Methods

The trial F13CD-3720 is a safety extension to Trial F13CD-1725 intended to document long-term safety of monthly replacement therapy with recombinant coagulation factor XIII (rFXIII) in patients previously exposed to rFXIII as part of Trial F13CD-1725. Overall, the design is similar to trial F13CD-1725.

Study Participants

Main Inclusion Criteria

- Previous participation in F13CD-1725 (up to and including Visit 16 [end-of-trial]).

Main Exclusion Criteria

- Known neutralizing antibodies (inhibitors) towards FXIII.
- Any known congenital or acquired coagulation disorder other than congenital FXIII deficiency; Any history of confirmed venous or arterial thrombo-embolic events, including myocardial infarction or stroke
- Documented history of ≥ 3 spontaneous and haemostatic treatment-requiring bleeding episodes per year during previous regular replacement therapy with rFXIII;
- Previous participation in this trial. (Defined as screened at visit 1): these criteria were removed by Substantial Amendment No. 8.
- Platelet count (thrombocytes) < 75 × 10^9/L then modified by Substantial Amendment Nos. 4 and 8 to read: Platelet count (thrombocytes) < 50 × 10^9/L. For subjects who participated in F13CD-1725, platelet count from visit 15 in F13CD-1725 must be used for evaluation.
- Known or suspected allergy to trial product(s) or related products.
- Renal insufficiency defined as currently requiring dialysis therapy.

Treatments

All patients received 35 IU/kg rFXIII every 4th week (28±2 days). At each visit, the dose was adjusted according to the actual weight of the patient. The reconstituted trial product was administered as a slow intravenous injection (at a rate that was not to exceed 1-2 mL per minute).

- Prior and Concomitant Therapy

All patients included in Trial F13CD-3720 as of the cut-off date of 11 February 2011 completed Trial F13CD-1725 and therefore had received regular replacement therapy with rFXIII for at least 52 weeks.

Non-emergency use of FXIII-containing products (other than rFXIII) was not allowed during the trial period. In case of acute bleeding episodes, any additional treatment as per investigator judgment was to be according to local standard practice, and additional doses of rFXIII could therefore not be used to treat such breakthrough bleedings.

Patients requiring elective surgical procedures during the course of the trial were to receive plasma-derived FXIII according to local standard care during and after the surgical procedures. As in Trial F13CD-1725, patients were to be withdrawn from the trial if they received any anti-thrombotic drug during the trial period.

Objectives

Primary Objective: To assess the long term safety of monthly replacement therapy with rFXIII when used for prevention of bleeding episodes in subjects with congenital FXIII deficiency.

Secondary Objective: To evaluate the efficacy of monthly replacement therapy with rFXIII when used for prevention of bleeding episodes in subjects with congenital FXIII deficiency.

Outcomes/endpoints

Primary endpoint:

- Adverse events (serious and non-serious) occurring from first trial-related activity after signing the informed consent to the end of patient's participation in the trial

Secondary safety endpoints:

- Antibody and inhibitor development
- Laboratory parameters
- Vital signs
- Body measurements

Secondary efficacy endpoints:

- Rate (number per patient year) of bleeding episodes requiring treatment with a FXIII containing product during the rFXIII treatment period (spontaneous bleeding episodes / traumatic bleeding episodes / intracranial haemorrhages)
- Number of patients withdrawn from the trial due to lack of efficacy of rFXIII treatment (as judged by the investigator)

Sample size

Sample size was partly determined by the number of patients completing Trial F13CD-1725 (N=33). Substantial Amendment No. 8 allowed inclusion of patients outside the context of the F13CD-1725 trial. Approximately 10-20 additional patients were expected to be enrolled, and to contribute to the documentation of the long-term safety of monthly replacement therapy with rFXIII.

Randomisation

The study was a single arm trial.

Blinding (masking)

The study was an open label study.

Statistical methods

No formal testing of statistical hypotheses was performed. Safety endpoints were summarised descriptively. Adverse events recorded by the investigator to be symptoms related to an adverse event diagnosis were not included in the summary tables. All adverse events were presented in the listings and events related to a diagnosis were flagged.

Number of bleeding episodes requiring treatment were evaluated by a Poisson model (log-link) similar to the model used in the evaluation of efficacy in Trial F13CD-1725. The remaining efficacy endpoints were summarised descriptively.

Results

Participant flow

Subject disposition is presented in the Table 32. All 33 patients who completed Trial F13CD-1725 were enrolled and exposed to trial drug. Of the 33 enrolled and dosed patients, 3 patients were withdrawn from the trial.

Table 32: Patient disposition – Study F13CD-3720

	Avecia	Novo Nordisk	Total
	rFXIII 35 IU/Kg N (%)	rFXIII 35 IU/Kg N (%)	rFXIII 35 IU/Kg N (%)
Randomised	26	31	33
Exposed	26 (100.0)	31 (100.0)	33 (100.0)
Withdrawn from Trial			
Other Reason	1 (3.8)	1 (3.2)	2 (6.1)
Withdrawal Criteria	1 (3.8)	0 (0.0)	1 (3.0)
Full Analysis Set	26 (100.0)	31 (100.0)	33 (100.0)
Safety Analysis Set	26 (100.0)	31 (100.0)	33 (100.0)

N: Number of subjects
%: Proportion of exposed subjects

Recruitment

Of the 23 actively recruiting sites in Trial F13CD-1725, 4 sites did not include patients into Trial F13CD-3720. A total of 19 sites enrolled and dosed at least one patient. The country distribution was as follows (number of actively recruiting sites per country in parenthesis): Austria (1), Canada (1), Finland (1), France (1), Germany (3), Israel (1), Italy (1), Spain (1), Switzerland (1), UK (2) and USA (6).

Conduct of the study

A total of six substantial amendments were implemented after initiation of patient enrolment. The protocol was amended (Substantial Amendment No. 8) in order to offer additional patients (patients who were not included in Trial F13CD-1725) with FXIII subunit A-deficiency participation in Trial

F13CD-3720. The trial was initiated on 21 September 2009 and is not completed. Interim analyses were planned to take place when all patients had been exposed to rFXIII for at least 3 and 6 months. An additional interim analysis covering all endpoints was planned to take place when all patients had completed 52 weeks in the trial.

There were no important protocol deviations in the trial as of the cut-off date 11 February 2011 .

Baseline data

The demographics of the trial population at baseline are presented in the Table 33.

Table 33: Baseline Demographics - Full Analysis Set

	Avecia	Novo Nordisk	Total
	rFXIII 35 IU/Kg	rFXIII 35 IU/Kg	rFXIII 35 IU/Kg
Number of Subjects	26	31	33
Age (years)			
N	26	31	33
Mean (SD)	29.7 (15.6)	29.0 (16.9)	28.8 (16.4)
Median	25.0	25.0	25.0
Min ; Max	8.0 ; 57.0	7.0 ; 60.0	7.0 ; 60.0
Sex, N(%)			
N	26 (100)	31 (100)	33 (100)
Female	11 (42)	11 (35)	13 (39)
Male	15 (58)	20 (65)	20 (61)
Race, N(%)			
N	26 (100)	31 (100)	33 (100)
Black or African American	0 (0)	2 (6)	2 (6)
White	18 (69)	22 (71)	23 (70)
Unknown	1 (4)	1 (3)	1 (3)
Asian	3 (12)	2 (6)	3 (9)
Other	4 (15)	4 (13)	4 (12)

The French patients are marked as Unknown as per the French Authorities Guidelines
SD: standard deviation

Numbers analysed

All analyses were performed on the full analysis set, which was identical to the safety analysis set.

All patients who received at least one dose of trial product were included in the full analysis set. Data from patients who discontinued the trial for any reason was included in the analysis up to the point of discontinuation.

All withdrawn patients were excluded from the per protocol (PP) analysis set.

Outcomes and estimation

Primary endpoint

Of a total of 98 treatment-emergent adverse events reported, two (2) events were evaluated by the investigator to be possibly or probably related to trial product

The overall rates of adverse events were similar for the two drug substances ($rFXIII_{IASMS}$: 31.1 adverse events per 100 exposures; $rFXIII_{NN}$: 25.5 adverse events per 100 exposures) and no noteworthy differences in rate of any adverse event was apparent between the two drug substances.

Secondary efficacy endpoints

Rate (number per patient year) of bleeding episodes requiring treatment with a FXIII containing product during the rFXIII treatment period (spontaneous bleeding episodes / traumatic bleeding episodes / intracranial haemorrhages)

The occurrence of treatment-requiring bleeding episodes as of the cut-off date of 11 February 2011 is summarised for the total exposure period as well as separately for the two drug substances ($rFXIII_{IASMS}$ and $rFXIII_{NN}$) in the table 34.

Table 34: Summary of Rate of Treatment-Requiring Bleeding Episodes - Full Analysis Set

	Avecia	Novo Nordisk	Total
	rFXII 35 IU/Kg	rFXIII 35 IU/Kg	rFXIII 35 IU/Kg
Number of subjects	26	31	33
Number of subjects with bleed	2	2	3
Total number of bleeds	2	3	5
Range of bleedings	0 ; 1	0 ; 2	0 ; 3
Mean bleedings per subject	0.077	0.097	0.152
Mean observation period (days)	131	201	293
Mean yearly bleeding rate	0.214	0.176	0.189

Details of the five treatment-requiring bleeding episodes are listed below.

Table 35: Details of Treatment-requiring Bleeding Episodes - Full Analysis Set

Subject ID	Drug substance origin	Age at baseline	Cause of Bleed	Location of bleeding	Time since last dose of rFXIII(Days)
12001	Avecia		Trauma	Wrist	23
1	Novo Nordisk	8	Spontaneous	Mucosal/nostril	19
	Novo Nordisk		Spontaneous	Bruising on arm/ Soft tissue	13
2	Novo Nordisk	12	Trauma	Forehead laceration	24
3	Avecia	25	Spontaneous	Muscular	17

Table 36: Analysis of rate of treatment requiring bleeding episodes - Full Analysis set

Evaulation	N	Mean¤ (Lambda)	Confidence Interval	Conclusion*	Covariate coefficient	P-value
rFXIII 35 IU/kg	33	0.046	[0.0046; 0.4706]	Superior		
Covariates						
AGE					-0.1347	0.043

The estimate is from a Poisson model with Age as a covariate and the total observation time during the treatment period as an offset in the model. The estimated rate is adjusted for overdispersion.
¤ Mean (Lamba) refer to the estimate of the annualised bleeding rate.
* The null hypothesis of no difference between rFXIII and placebo is rejected if the upper limit of the 95% Confidence interval for the yearly bleeding rate (Lambda) is less than 2.91

Thirty of the 33 patients comprising the full analysis set did not experience any treatment-requiring bleeding episodes during the trial.

Number of patients withdrawn from the trial due to lack of efficacy of rFXIII treatment (as judged by the investigator)

No patients were withdrawn due to lack of efficacy of rFXIII treatment.

Ancillary analyses

No ancillary analyses were submitted.

Summary of main study

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

Table 37: Summary of Efficacy for trial F13CD-1725 and F13CD-3720

Study F13CD-1725: A Multi-Centre, Open-Label, Single-Arm and Multiple Dosing Trial on Efficacy and Safety of Monthly Replacement Therapy with Recombinant Factor XIII (rFXIII) in Subjects with Congenital Factor XIII Deficiency			
Study identifier	F13CD-1725		
Design	Open-Label, Single-Arm		
	Duration of main phase: Duration of Run-in phase: Duration of Extension phase:		52 weeks 4 weeks not applicable
Hypothesis	Superiority: The null hypothesis of no difference between rFXIII and placebo was to be rejected if the upper limit of the 95% confidence interval for the yearly bleeding rate for rFXIII was less than a fixed rate of 2.91 based on retrospectively collected historical data.		
Treatments groups	rFXIII		35 IU/kg every 4th week (28±2 days), 41 patients. At each visit, the dose was adjusted according to the actual weight of the patient. The reconstituted trial product was administered as a slow intravenous injection (at a rate that was not to exceed 1-2 mL per minute).
Endpoints and definitions	Primary efficacy endpoint	λ	Rate (number per patient year) of bleeding episodes that required treatment with a FXIII-containing product during the rFXIII treatment period
	Secondary efficacy endpoint	p	Percentage of patients without "bleeding episodes requiring treatment" with a FXIII-containing product during the treatment period
	Secondary efficacy endpoint	Spontaneous λ	Rate (number per patient year) of spontaneous "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period
	Secondary efficacy endpoint	Traumatic λ	Rate (number per patient year) of traumatic "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period
	Secondary efficacy endpoint	Intracranial λ	Rate (number per patient year) of intracranial "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period
	Secondary efficacy endpoint		Percentage of patients having a normal clot solubility one hour after rFXIII administration and 28 days after rFXIII administration
	Secondary efficacy endpoint		Number of patients withdrawn from the trial due to lack of efficacy of rFXIII treatment

	Secondary efficacy endpoint		Level of FXIII activity one hour after rFXIII administration and 28 days after rFXIII administration
Database lock	03 June 2010		

Results and Analysis

Analysis description	**Primary Analysis**			
Analysis population and time point description	Intent to treat (Full Analysis Set) Week 52			
Descriptive statistics and estimate variability	Treatment group	rFXIII	Regular replacement therapy (historical control)	On demand treatment (historical control)
	Number of patient	41	60	16
	Λ (mean, age-adjusted) 95% CI	0.048 0.094;0.2501	0.3 --	2.91 --
	P (mean, age-adjusted) 95% CI	0.9581 0.7242;0.9950	-- --	0.25 --
	Spontaneous λ (mean)	0	--	--
	Traumatic λ (mean)	0.140	--	--
	Intracranial λ (mean)	0	--	--
	Patients having normal clot solubility at 1 hour and 28 days post-dose (percentage)	83.3	--	--
	Number of patients withdrawn from the trial due to lack of efficacy of rFXIII treatment	0	--	--
	Level of FXIII activity one hour after rFXIII administration (mean)	0.782	--	--
	SD	0.342	--	--
	Level of FXIII activity 28 days after rFXIII administration (mean)	0.189	--	--

	SD	0.102	--	--

Effect estimate per comparison	Mean (λ)	Comparison groups	rFXIII vs On demand treatment (historical control)
		Poisson model (log link) P-value	 0.022
	Patients having normal clot solubility (percentage)	Comparison groups	Pre-dose vs 1 hour and 28 days post-dose
		Absolute difference	4.7
	Level of FXIII activity 1 hour after rFXIII administration (mean)	Comparison groups	Pre-dose vs. 1 hour post-dose
		Absolute difference	0.599
		SD	0.368
	Level of FXIII activity 28 days after rFXIII administration (mean)	Comparison groups	Pre-dose vs. 28 hour post-dose
		Absolute difference	0.005
		SD	0.187

Study F13CD-3720: A Multi-Centre, Open-Label, Single-Arm and Multiple Dosing Trial on Safety of Monthly Replacement Therapy with Recombinant Factor XIII (rFXIII) in Subjects with Congenital Factor XIII Deficiency			
Study identifier	F13CD-3720		
Design	Open-Label, single-Arm, extension study following F13CD-1725		
	Duration of main phase: Duration of Run-in phase: Duration of Extension phase:		52 weeks 4 weeks not applicable
Hypothesis	Exploratory		
Treatments groups	rFXIII		35 IU/kg every 4th week (28±2 days), 33 patients. At each visit, the dose was adjusted according to the actual weight of the patient. The reconstituted trial product was administered as a slow intravenous injection (at a rate that was not to exceed 1-2 mL per minute) During the trial, Novo Nordisk-produced rFXIII was introduced at all sites
Endpoints and definitions	Secondary efficacy endpoint	λ	Rate (number per patient year) of bleeding episodes that required treatment with a FXIII-containing product during the rFXIII treatment period
	Secondary efficacy endpoint	Spontaneous λ	Rate (number per patient year) of spontaneous "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period
	Secondary efficacy endpoint	Traumatic λ	Rate (number per patient year) of traumatic "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period
	Secondary efficacy endpoint	Intracranial λ	Rate (number per patient year) of intracranial "bleeding episodes requiring treatment" with a FXIII-containing product during the rFXIII treatment period
	Secondary efficacy endpoint		Number of patients withdrawn from the trial due to lack of efficacy of rFXIII treatment

Database lock	11 February 2011	
Results and Analysis		
Analysis description	**Interim Analysis (trial ongoing)**	
Analysis population and time point description	Intent to treat (Full Analysis Set) 3 months	
Descriptive statistics and estimate variability	Treatment group	rFXIII
	Number of patient	33
	Λ (mean, age-adjusted) 95% CI	0.046 0.0046;0.4706
	Spontaneous λ (mean)	0.113
	Traumatic λ (mean)	0.076
	Intracranial λ (mean)	0
	Number of patients withdrawn from the trial due to lack of efficacy of rFXIII treatment	0
Notes	Trial ongoing, the final analysis will be performed at week 52 as planned.	

2.5.3. Discussion on clinical efficacy

Design and conduct of clinical studies

The pivotal trial F13CD-1725 was a single arm trial in patients that were FXIII deficient. The trial was designed as a superiority trial, comparing the efficacy of monthly rFXIII replacement therapy against a historical control group that required treatment with a FXIII-containing product per patient per year. The historical data was derived from analysis of retrospective data from 16 patients with congenital FXIII A-subunit deficiency that were treated on-demand with FXIII-containing products. The trial was extended to evaluate the long term safety of monthly replacement therapy with rFXIII in patients with congenital FXIII A-subunit deficiency (Study F13CD-3720) in patients previously exposed to rFXIII as part of Trial F13CD-1725 and other additional patients with FXIII subunit A-deficiency participation.

Given the rarity of the condition, the trial design (single-arm), the low number of patients (41 and 33, respectively) and duration of both trials were considered acceptable. The CHMP noted that such trial could not be recognised as a true superiority trial, considering that the on-demand population was different than the historical data and thus could not be compared. However, the listing of history bleeding (type and number) did not reveal any associated clinical issue, and thus the proposed comparison (prophylaxis vs on demand) was considered acceptable. However, the need for statistical significance should be weighed against the need for clinically relevant/interpretable results (the guideline on clinical trial in small populations CHMP/EWP/83561/2005). The statistical analysis methods applied were considered acceptable.

Efficacy data and additional analyses

The single pivotal phase 3 trial (F13CD-1725) and the extension trial (F13CD-3720) investigated monthly replacement therapy for prevention of bleeding episodes (prophylaxis). The trial F13CD-1725 met its primary endpoint which was superiority over the historical group where the age-adjusted rate (number per subject year) of treatment-requiring bleeding episodes during the rFXIII treatment period was 0.048/year (95% CI: 0.0094 - 0.2501), which is less than 2.91 yearly bleeding rate from historical controls. The baseline age for patients who suffered treatment-requiring bleeds was 8, 10, 16 and 19 years. When considering that the mean age of the trial population was 26 years, there was a tendency for treatment-requiring bleeds to occur primarily at a younger age, consistent with the effect of age being statistically significant in the primary analysis (p=0.022). Efficacy of a long-term prophylactic therapy with rFXIII will be evaluated in the extension trial, which is still ongoing.

In the extension trial, surgeries were not part of the exclusion criteria. The definition of severe bleeders (documented history of ≥ 3 spontaneous and haemostatic treatment-requiring bleeding episodes per year during previous regular replacement therapy with rFXIII) had also been modified. In both trials the bleeding occurred at a younger age. There appeared to be a higher likelihood of traumatic bleeds in children. However, there were also two spontaneous treatment-requiring bleeds in the extension period, in an 8 years old child. The available data suggests that no difference in FXIII activity profiles after dosing of 35 IU/kg rFXIII in patients with congenital FXIII deficiency is observed. However, the need for dose adjustment in paediatric patients should be discussed further when the full reports on the already initiated investigation in young children (under 6 years) and those planned in adult patients become available.

No dose response study has been performed.

The main objective of the trial F13CD-1725 was to assess the efficacy of rFXIII in the prevention of bleeding episodes. In case of acute bleeding episodes during the pivotal trial, additional doses of rFXIII could not be used to treat such breakthrough bleedings. No investigations of rFXIII in on-demand treatment setting are available. Thus, the CHMP had a concern over the off label use of rFXIII in the routine management of breakthrough bleedings. Current practice for patients on long-term prophylaxis involves administration of the FXIII-containing product treatment at home and in advance of the next scheduled dose to treat any breakthrough bleedings. As the treatment of breakthrough bleeding was not part of the investigation in the phase III clinical trial, the treatment of breakthrough bleedings with NovoThirteen was not recommended. A warning was included in section 4.4 to inform the prescriber that the on-demand treatment of acute bleeds or breakthrough bleeds with NovoThirteen has not been studied in clinical trials and that an alternative treatment should be considered in such situations. In addition, monitoring of off-label use of NovoThirteen has been included in the RMP.

There were no efficacy data on the peri-operative management of bleeding in patients with congenital FXIII deficiency with the rFXIII product provided by the Applicant. Thus, the CHMP was of the opinion that the indication required restriction from the general sense of prophylaxis to specifically long-term prophylaxis. Thus, the indication was modified to "Long-term prophylactic treatment of bleeding in patients 6 years and above with congenital factor XIII A-subunit deficiency".

As previously mentioned, clot solubility test has not been validated as surrogate efficacy endpoint in this setting and was not be taken into account.

The pivotal phase 3 trial included 15 patients with congenital FXIII A-subunit deficiency below 18 years (9 between 6 and 11 years, 6 between 12 and 17 years) out of 41 patients. In the currently ongoing extension trial, 11 patients out of 33 were below 18 years (7 between 6 and 11 years, 4 between 12 and 17 years). In accordance with an EMA Paediatric Investigation Plan commitment, a single-dose

trial specifically designed to investigate the pharmacokinetics and safety profile of rFXIII in children less than 6 years old with congenital FXIII deficiency (F13CD-3760) was initiated on 07 November 2010. Inclusion into a long-term safety follow-on trial (F13CD-3835) was offered to those children who complete the F13CD-3760 trial. So far, only 1 patient has been included as of the cut-off date of 11 February 2011.

2.5.4. Conclusions on the clinical efficacy

The pivotal trial F13CD-1725 provided satisfactory evidence that NovoThirteen reduced the rate (number per subject year) of bleeding episodes requiring treatment with a FXIII-containing product during the rFXIII treatment period. Efficacy has been demonstrated. However, the CHMP highlighted the fact that the efficacy and safety of rFXIII in the treatment of breakthrough bleedings had not been demonstrated. Moreover, as no dose response study has been performed in patients with the 35IU/kg/month dose, the CHMP requested further data from the ongoing extension study (F13CD-3720) in order to collect additional information on the 35 IU/kg dose of rFXIII. The results of the additional PK assessments will be provided post marketing authorisation.

The CHMP considers the following measures as stated in the RMP necessary to address issues related to efficacy:

- A post approval commitment to amend the ongoing trial, F13CD-3720, to investigate the clinical efficacy and safety of rFXIII in treatment of breakthrough bleedings during prophylactic treatment with rFXIII for one year after implementation of the protocol amendment. Any breakthrough bleeding during the trial period which requires additional FXIII treatment (by investigator's judgment) should be treated with rFXIII 35 IU/kg in addition to the routine prophylaxis.

In addition, the CHMP recommends the following point for further investigation:

- To date only 9 (6-11 years) and 6 (12-17 years) children have been included into the pivotal and extension trial. Thus, in children further clinical data need to be collected in the extension study applying the dose regimen of 35 IU/kg every 4 weeks. This refers to an increase of patient number (doubled) and also to long-time investigation.

2.6. Clinical safety

The safety data in patients with congenital FXIII A-subunit deficiency included one phase III study F13CD-1725, one phase III extension study F13CD-3720 and one phase I study F13CD-1663.

The safety data in healthy subjects comprised of four phase I studies, F13CD-1662, F13CD-1661, NN1810-3733 and NN1841-3788.

The safety database also included, one phase I study F13CARD-1660 in patients undergoing cardiac surgery requiring cardiopulmonary bypass and one randomised, double-blind, phase II and placebo controlled trial (NN1810-3540) of rFXIII replenishment with two different doses of recombinant factor XIII following cardiopulmonary bypass surgery which is ongoing.

Patient exposure

Patient exposure and the dose levels received are present in Table 38 and 39. Concerning patients with congenital FXIII deficiency, a total of 50 patients were exposed to rFXIII in three studies. The extent of exposure varied from a single dose administration up to 33 infusions. Otherwise, 122 healthy patients were exposed to rFXIII and 26 healthy subjects received placebo. Concerning patients

undergoing cardiac surgery requiring cardiopulmonary bypass, 444 patients were exposed to rFXIII in two studies and 8 patients received placebo.

Table 38: Number of patients exposed to rFXIII or placebo

Study	Number of patients exposed				Dose levels
	rFXIII $_{IASMS}$	**rFXIII NN**	**Placebo**	**Total**	
Congenital FXIII deficiency					
F13CD-1725	41	-	-	41	35 IU/kg (multiple dose, 52 weeks)
F13CD-3720	26	45	-	33*	35 IU/kg (multiple dose, 52 weeks)
F13CD-1663	9	-	-	9	2, 7, 24, 60, 89 IU/kg (single dose)
Healthy subjects					
NN1841-3788	49	49	-	50	35 IU/kg (dual dose**)
F13-1662	16	-	8	24	Placebo, 12, 30 IU/kg (five consecutive daily doses)
F13-1661	40	-	10	50	Placebo, 2, 6, 12, 30, 60 IU/kg (single dose)
NN1810-3733	16	-	8	24	Placebo, 12, 35 IU/kg (single dose)
Cardiac surgery					
NN1810-3540	409			409	Placebo, 17.5 , 35 IU/kg (single dose)
F13CARD-1660	35	-	8	43	Placebo, 11.9, 25, 35, 50 IU/kg (single dose)

* These patients are included in the 41 patients in trial F13CD-1725

** one dose each of $rFXIII_{IASMS}$ and $rFXIII_{NN}$

Table 39: Extent of rFXIII Exposure in all studies (data cut-off 31 July 2011)

	$rFXIII_{IASMS}$	$rFXIII_{NN}$	rFXIII Total
Congenital FXIII Deficiency			
F13CD-1725	471		471
F13CD-3720 [a]	122	559	681
F13-1663	11		11
F13CD-3760 [a]		2	2
F13CD-3835 [a]		13	13
Healthy Subjects			
NN1841-3788	49	49	98
F13-1662	16		16
F13-1661	40		40
NN1810-3733	16		16
Cardiac Surgery			
F13CARD-1660	35		35
NN1810-3540	281		281
Total	**1041**	**623**	**1664**

Adverse events

A total of 352 adverse events were reported in 32 of 41 (78%) of the exposed patients until the cut-off date (studies F13CD-1725 and F13CD-3720 pooled). 270 events were reported after exposure to $rFXIII_{IASMS}$ and 82 events were reported after exposure to $rFXIII_{NN}$. The overall rates of adverse events were 45.5 adverse events per 100 exposures for $rFXIII_{IASMS}$ and 25.9 adverse events per 100 exposures for $rFXIII_{NN}$. A summary of the adverse event recorded is presented in Table 40.

Table 40: Summary of adverse events: F13CD-1725 and F13CD-3720 Pooled (data cut-off July 31, 2011

MedDRA SOC **Adverse events**	**IASMS (N=41)**	**Novo Nordisk (N=45)**	**Total (N=54)**
Number of exposures	**593**	**559**	**1152**
All adverse events	**273 (46%)**	**162 (29%)**	**435 (37.8%)**
Blood and lymphatic disorders	**7 (1.2%)**	**3 (0.5%)**	**10 (0.9%)**
Neutropenia	3 (0.5%)	2 (0.4%)	5 (0.4%)
Cardiac disorders	**1 (0.2%)**	**1 (0.2%)**	**2 (0.2%)**
Ear and labyrinth disorders	**2 (0.3%)**	**2 (0.4%)**	**4 (0.3%)**
Ear pain	1 (0.2%)	2 (0.4%)	3 (0.3%)
Eye disorders	**7 (1.2%)**	**1 (0.2%)**	**8 (0.7%)**
Eye swelling	2 (0.3%)	0 (0%)	2 (0.2%)
Gastrointestinal disorders	**23 (3.9%)**	**10 (1.8%)**	**33 (2.9%)**
Abdominal pain	4 (0.7%)	0 (0%)	4 (0.3%)
Dental caries	1 (0.2%)	3 (0.5%)	4 (0.3%)
Diarrhoea	3 (0.5%)	0 (0%)	3 (0.3%)
Dyspepsia	0 (0%)	2 (0.4%)	2 (0.2%)
Gastritis	1 (0.2%)	1 (0.2%)	2 (0.2%)
Toothache	5 (0.8%)	1 (0.2%)	6 (0.5%)
Vomiting	3 (0.5%)	0 (0%)	3 (0.3%)
General disorders and administration site conditions	**18 (3.0%)**	**9 (1.6%)**	**27 (2.3%)**
Chest pain	2 (0.3%)	0 (0%)	2 (0.2%)
Fatigue	2 (0.3%)	2 (0.4%)	4 (0.3%)
Injection site pain	3 (0.5%)	0 (0%)	3 (0.3%)
Pyrexia	7 (1.2%)	6 (1.1%)	13 (1.1%)
Infections and infestations	**55 (9.3%)**	**28 (5.0%)**	**83 (7.2%)**
Bronchitis	3 (0.5%)	1 (0.2%)	4 (0.3%)
Cystitis	2 (0.3%)	0 (0%)	2 (0.2%)
Ear infection	1 (0.2%)	1 (0.2%)	2 (0.2%)
Gastroenteritis	0 (0%)	2 (0.4%)	2 (0.2%)
Gastroenteritis viral	2 (0.3%)	0 (0%)	2 (0.2%)
H1N1 influenza	2 (0.3%)	0 (0%)	2 (0.2%)
Influenza	4 (0.7%)	1 (0.2%)	5 (0.4%)
Nasopharyngitis	12 (2.0%)	10 (1.8%)	22 (1.9%)
Oral herpes	3 (0.5%)	1 (0.2%)	4 (0.3%)
Otitis media	1 (0.2%)	1 (0.2%)	2 (0.2%)
Pharyngitis streptococcal	1 (0.2%)	1 (0.2%)	2 (0.2%)
Sinusitis	3 (0.5%)	4 (0.7%)	7 (0.6%)
Upper respiratory tract infection	2 (0.3%)	1 (0.2%)	3 (0.3%)
Urinary tract infection	3 (0.5%)	0 (0%)	3 (0.3%)
Viral infection	2 (0.3%)	0 (0%)	2 (0.2%)
Vulvovaginal mycotic infection	2 (0.3%)	1 (0.2%)	3 (0.3%)
Injury, poisoning and procedural complications	**49 (8.3%)**	**32 (5.7%)**	**81 (7.0%)**
Contusion	3 (0.5%)	2 (0.4%)	5 (0.4%)
Excoriation	4 (0.7%)	2 (0.4%)	6 (0.5%)
Fall	1 (0.2%)	4 (0.7%)	5 (0.4%)
Incorrect dose administered	12 (2.0%)	2 (0.4%)	14 (1.2%)
Injury	1 (0.2%)	1 (0.2%)	2 (0.2%)
Joint injury	1 (0.2%)	1 (0.2%)	2 (0.2%)
Joint sprain	4 (0.7%)	3 (0.5%)	7 (0.6%)
Laceration	2 (0.3%)	2 (0.4%)	4 (0.3%)
Limb injury	3 (0.5%)	1 (0.2%)	4 (0.3%)
Post traumatic pain	0 (0%)	2 (0.4%)	2 (0.2%)
Road traffic accident	1 (0.2%)	1 (0.2%)	2 (0.2%)

Splinter	0 (0%)	4 (0.7%)	4 (0.3%)
Thermal burn	2 (0.3%)	0 (0%)	2 (0.2%)
Tooth fracture	2 (0.3%)	0 (0%)	2 (0.2%)
Underdose	2 (0.3%)	0 (0%)	2 (0.2%)
Wound			
Investigations	**6 (1.0%)**	**1 (0.2%)**	**7 (0.6%)**
Antibody test positive	4 (0.7%)	0 (0%)	4 (0.3%)
Metabolism and nutrition disorders	**2 (0.3%)**	**1 (0.2%)**	**3 (0.3%)**
Musculoskeletal and connective tissue disorders	**34 (5.7%)**	**18 (3.2%)**	**52 (4.5%)**
	6 (1.0%)	2 (0.4%)	8 (0.7%)
Arthralgia	7 (1.2%)	4 (0.7%)	11 (1.0%)
Back pain	2 (0.3%)	0 (0%)	2 (0.2%)
Bone cyst	1 (0.2%)	1 (0.2%)	2 (0.2%)
Musculoskeletal pain	2 (0.3%)	1 (0.2%)	3 (0.3%)
Musculoskeletal stiffness	2 (0.3%)	0 (0%)	2 (0.2%)
Myalgia	1 (0.2%)	1 (0.2%)	2 (0.2%)
Myosclerosis	3 (0.5%)	1 (0.2%)	4 (0.3%)
Neck pain	5 (0.8%)	4 (0.7%)	9 (0.8%)
Pain in extremity			
Neoplasms benign, malignant and unspecified	**1 (0.2%)**	**0 (0%)**	**1 (0.1%)**
Nervous system disorders	**30 (5.1%)**	**18 (3.2%)**	**48 (4.2%)**
Headache	25 (4.2%)	16 (2.9%)	41 (3.6%)
Sciatica	2 (0.3%)	1 (0.2%)	3 (0.3%)
Psychiatric disorders	**1 (0.2%)**	**0 (0%)**	**1 (0.1%)**
Renal and urinary disorders	**6 (1.0%)**	**1 (0.2%)**	**7 (0.6%)**
Dysuria	2 (0.3%)	0 (0%)	2 (0.2%)
haematuria	1 (0.2%)	1 (0.2%)	2 (0.2%)
Pollakiuria	2 (0.3%)	0 (0%)	2 (0.2%)
Reproductive system and breast disorders	**1 (0.2%)**	**1 (0.2%)**	**2 (0.2%)**
Respiratory, thoracic and mediastinal disorders	**21 (3.5%)**	**25 (4.5%)**	**46 (4.0%)**
	4 (0.7%)	7 (1.3%)	11 (1.0%)
Cough	2 (0.3%)	0 (0%)	2 (0.2%)
Epistaxis	6 (1.0%)	6 (1.1%)	12 (1.0%)
Nasal congestion	6 (1.0%)	9 (1.6%)	15 (1.3%)
Oropharyngeal pain			
Skin and subcutaneous tissue disorders	**7 (1.2%)**	**7 (1.3%)**	**14 (1.2%)**
Eczema	0 (0%)	3 (0.5%)	3 (0.3%)
Vascular disorders	**1 (0.2%)**	**0 (0%)**	**1 (0.1%)**

*only adverse events reported at least twice in one of the groups are listed in the Table

In phase I study F13CD-1163, 4 patients (36%) experienced at least one adverse event: one subject in the 6 U/kg dose cohort experienced headache, pharyngolaryngeal pain, throat tightness, arthralgia (knee and ankle) and contusion (ankle), one subject in the 20 U/kg cohort experienced headache the day of the study treatment and recovered the same day, one subject in the 50 U/kg cohort experienced headache the day of the study treatment, pain in limb on Day 2 and pain on Day 8 and one subject in the 50 U/kg cohort experienced arthralgia on Day 2 and recovered the same day.

In study F13CD-1662, adverse events occurring in two or more patients are presented below:

Table 41: Adverse events occurring in two or more patients – Study F13CD-1662

Adverse event	Placebo (N=8)	10 U/kg (N=8)	25 U/kg (N=8)	Pooled rFXIII (N=16)
Headache	5 (62.5%)	4 (50.0%)	4 (50.0%)	8 (50.0%)
Somnolence	1 (12.5%)	2 (25.0%)	1 (12.5%)	3 (18.8%)
Upper respiratory tract infection NOS	1 (12.5%)	3 (37.5%)	0	3 (18.8%)
Paraesthesia	0	1 (12.5%)	1 (12.5%)	2 (12.5%)
Fatigue	2 (25.0%)	2 (25.0%)	0	2 (12.5%)
Dysmenorrhea	0	1 (12.5%)	1 (12.5%)	2 (12.5%)

In study NN1841-3788, a total of 46 treatment-emergent adverse events were reported (21 events in 15 patients after administration of $rFXIII_{IASMS}$ and 25 events in 17 patients after administration of $rFXIII_{NN}$). Of these, 3 events (myalgia, pain in extremity and headache) were evaluated by the investigator to be possibly or probably related to trial product after administration of $rFXIII_{NN}$, and 2 events (rFXIII antibody test positive and muscle tightness) were evaluated to be possibly or probably related to trial product after administration of $rFXIII_{IASMS}$. The most frequently reported adverse events were headache, somnolence, upper respiratory tract infection, paraesthesia, fatigue and dysmenorrhoea.

In study F13CD-1661, adverse events occurring in two or more patients are presented in the table below:

Table 42: Adverse events occurring in two or more patients – Study F13CD-1661

Adverse events	Placebo (N=10)	2 U/kg (N=8)	5 U/kg (N=8)	10 U/kg (N=8)	25 U/kg (N=8)	50 U/kg (N=8)
Headache	4 (40%)	4 (50%)	3 (37.5%)	4 (50%)	3 (37.5%)	3 (37.5%)
Fibrin D-dimer increased	2 (20%)	2 (25%)	0 (0%)	0 (0%)	2 (25%)	0 (0%)
Muscle cramp	1 (10%)	0 (0%)	0 (0%)	2 (25%)	1 (12.5%)	1 (12.5%)
Pain in limb	1 (10%)	1 (12.5%)	0 (0%)	2 (25%)	0 (0%)	1 (12.5%)
Nasopharyngitis	0 (0%)	2 (25%)	0 (0%)	1 (12.5%)	0 (0%)	0 (0%)
Thrombin time prolonged	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	**3 (37.5%)**
Upper respiratory tract infection NOS	1 (10%)	1 (12.5%)	0 (0%)	1 (12.5%)	1 (12.5%)	0 (0%)
Abdominal pain upper	0 (0%)	1 (12.5%)	0 (0%)	1 (12.5%)	0 (0%)	0 (0%)
Abrasion NOS	0 (0%)	1 (12.5%)	1 (12.5%)	0 (0%)	0 (0%)	0 (0%)
Contusion	0 (0%)	1 (12.5%)	0 (0%)	0 (0%)	1 (12.5%)	0 (0%)
cough	0 (0%)	0 (0%)	1 (12.5%)	0 (0%)	1 (12.5%)	0 (0%)

NOS: no otherwise specified

One AE of increased AST was reported at 8 hours, 1, 2 and 3 days following administration of 5 U/kg rFXIII. This event was considered possibly related to study treatment. The Day 0 pre-dose value for AST was not available for this subject although the screening laboratory value was within normal range (23 U/L). The maximum value of AST in this subject was 82 U/L (normal range: 4-40 U/L). The AST value had returned to normal range by study day 7.

In summary, the most frequently reported adverse events were headache, increased fibrin D-dimer, muscle cramp and pain in limb.

Adverse drug reactions

A total of 14 adverse events in 9 patients were evaluated by the investigator to be possibly or probably related to trial product in Trials F13CD-1725 and F13CD-3720 (Table 21). All these events occurred after administration of $rFXIII_{IASMS}$. All 14 events were characterised by full recovery except for an event of pain in extremity (both legs) from which the patient recovered from the event approximately 10 months after onset of the event.

A total of 15 adverse events were evaluated by the investigator to be possibly or probably related to trial product in Trials F13CD-1725 and F13CD-3720. These comprised events of positive antibody test (4 events in 4 patients), incorrectly administered dose (3 events in 2 patients), injection site pain (1 event), increase in fibrin D-dimer (1 event), pain in extremity (1 event), headache (1 event), limb injury (1 event), overdose (1 event) and coinciding events of neutropenia and leukopenia in one patient (worsening of mild neutropenia initially diagnosed before first trial drug administration).Of these events, 3 of the 4 cases of positive antibody test were classified as serious adverse events.

Coinciding and possibly related adverse events of worsening of neutropenia and leucopenia were reported for one patient. The patient had mild neutropenia (neutrophil count: 1200/µL; normal range: 2500-7500/µL) before the initial trial drug administration. The neutrophil count dropped to 940/µL at week 12, at which point the patient was withdrawn from the trial. The neutrophil count at the end-of-trial visit at week 16 remained suppressed at 1350/µL.

Table 43: Listing of Possibly or Probably Related Adverse Events in Trials F13CD-1725 and F13CD-3720 – Patients with Congenital FXIII Deficiency

Treatment group	Patient age[1] (years)	Preferred term	Days from dosing to onset[2]	Relation to trial drug[3]	Serious	Outcome
rFXIII 35 IU/kg	7	Antibody test positive	14	PROBABLE	Y	RECOVERED
	25	Pain in extremity	22	POSSIBLE	N	RECOVERED[5]
	26	Headache	0	POSSIBLE	N	RECOVERED
	8	Leucopenia[4]	32	POSSIBLE	N	RECOVERED
		Neutropenia[4]	32	POSSIBLE	N	RECOVERED
	7	Incorrect dose administered	0	PROBABLE	N	RECOVERED
		Incorrect dose administered	0	PROBABLE	N	RECOVERED
		Antibody test positive	28	PROBABLE	N	RECOVERED
	60	Incorrect dose administered	0	PROBABLE	N	RECOVERED
	16	Antibody test positive	16	POSSIBLE	Y	RECOVERED
	14	Antibody test positive	16	POSSIBLE	Y	RECOVERED
	8	Injection site pain	2	POSSIBLE	N	RECOVERED
		Fibrin D dimer increased	14	PROBABLE	N	RECOVERED
		Overdose	0	PROBABLE	N	RECOVERED

[1]Age at baseline, [2]Days since the preceding dose of rFXIII, [3]As judged by the investigator, [4]Reported (lower level MedDRA term) as worsening of neutropenia and leucopenia, [5]Outcome was recorded as 'not recovered' at the end-of-trial visit. Follow-up inquiries confirmed that this patient had recovered from the event approximately 10 months after onset of the event.

Serious adverse event/deaths/other significant events

For the study F13CD-1725, 6 patients experienced eight serious adverse events (SAEs):

- One 57 year-old patient with a medical history of sigmoid colon diverticulosis experienced "diverticulitis" 3 days after treatment with rFXIII. A treatment with antibiotic and Fibrogammin for prevention of an intestinal bleeding was initiated. This patient recovered without any further complications;
- One 55 year-old patient experienced "non-cardiac chest pain" and "headache" 23 and 24 days after treatment with rFXIII. The patient recovered few days later;
- One 19 year-old patient experienced a road traffic accident which was unrelated to study treatment;
- Three patients experienced "antibody test positive" associated with a small intestinal obstruction in one case. Please see the next section for a full assessment of these cases.

In the extension study F13CD-3720, two serious adverse events have been recorded as of the cut-off date (11 February 2011):

- One 12 year-old patient experienced gastroenteritis and headache (treated with ibuprofen) on 18 Aug 2010. On 19 Aug 2010, this patient still felt unwell and fell on the staircase. He experienced a temporal laceration of his forehead and a wrist sprain. On 20 Aug 2010, the patient was treated with Fibrogammin for prevention of bleeding and for wound healing. On 06 Sep 2010, the patient recovered;
- One 55 year-old patient began experiencing carpal tunnel syndrome of the right hand and wrist on 16 Jun 2010. No relevant tests were performed. On 22 Jun 2010, this patient was treated with $rFXIII_{NN}$. On 23 Aug 2010, the patient underwent planned surgery. No bleeding was recorded. On 24 Aug 2010, the patient was discharged and was evaluated as recovered.

In the phase I study F13CARD-1660; two SAEs possibly related to trial product were of interest:

- One 55 year-old patient receiving 11.9 IU/kg experienced an anaphylactic reaction 57 minutes after the treatment. This patient recovered within 6.5 hours. It can be noted that protamine had been given shortly before the trial product was administered. This patient received also Promiten/Macrodex which can be associated with allergic reactions. This patient had a baseline titer of yeast antibodies of 0.23 kU/L and a post-dose titer of < 0.11 kU/L.

- One 73 year old patient receiving 25 IU/kg experienced soft tissue necrosis at the site of the vein harvesting, 10 days after trial drug administration. The patient fully recovered.

No serious adverse events were reported in Trials F13-1663, F13CD-3760 and F13CD-3835. No serious adverse events were reported in healthy patients (Trials NN1841-3788, F13-1661, F13- 1662 and NN1810-3733).

No deaths were reported in the two main phase III studies.

No adverse events of anaphylactic or allergic reaction, bleeding episodes or changes in pharmacokinetics have been observed in any of the patients at any time during the presence of the non-neutralising antibodies or during the follow-up period.

Table 44: Listing of Serious Adverse Events in Trials F13CD-1725 and F13CD-3720 – Patients with Congenital Deficiency

Treatment group	Patient age (years)	Preferred term	Days from dosing to onset	Relation to trial drug*	Outcome
rFXIII 35IU/kg	7	Antibody test positive	15	PROBABLE	RECOVERED
	57	Diverticulitis	3	UNLIKELY	RECOVERED
	55	Non-cardiac chest pain	23	UNLIKELY	RECOVERED
		Headache	24	UNLIKELY	RECOVERED
		Carpal tunnel syndrome	17	UNLIKELY	RECOVERED
	19	Road traffic accident	28	UNLIKELY	RECOVERED
	16	Antibody test positive	17	POSSIBLE	RECOVERED
	14	Small intestinal obstruction	4	UNLIKELY	RECOVERED
		Antibody test positive	17	POSSIBLE	RECOVERED
	12	Skin laceration	24	UNLIKELY	RECOVERED

* As judged by the investigator.

Apart from the antibody development, the serious adverse event were all judged as unrelated by the investigator.

- Medication error

Medication errors were observed in phase III study F13CD-1725 and in the extension phase III study F13CD-3720. A total of 14 medication errors (7 patients) were recorded in study F13CD-1725 and 3 medication errors were recorded in study F13CD-3720.

Table 45: Overview of medication errors

Type of error	**Number of events in number of patients/ subject IDs** **Whole vial**
Whole vial diluent used (4.3 instead of 3.2)	8 events in 2 patients
Dose chart rounding error	3 events 3 patients
Patient weight not taken between doses	2 events 1 patient
Saline instead of sterile water	1 event 1 patient
Administration of solvent only	1 event 1 patient
Error of dose calculation (Home treatment)	1 event 1 patient
Error of reconstitution procedure	1 event 1 patient

Of the 17 medication errors, 14 are deviations from the protocol-described dose and are related to the trial product and trial conduct. The 14 events include 8 events that were reported in 2 patients at one trial site.

Laboratory findings

No clinically relevant dose-related changes were observed for parameters of haematology, biochemistry and urinalysis.

- Anti-rFXIII antibody

There were four patients in study F13CD-1725, and one patient in the phase I study NN1841-3788 that developed anti-rFXIII antibodies. The anti-rFXIII antibodies were transient, low-titre anti-rFXIII antibodies that had no neutralising activity.

- Thromboembolic events

One thromboembolic event was reported. One 60 year-old patient in the study F13CD-1725 experienced a superficial phlebitis.

The incidence of post-treatment coagulation abnormalities for study F13CD-1661 is summarized in the Table 46.

Table 46: Number of patients with coagulation abnormalities by treatment group

Analyte	**Placebo (N=10)**	**2 U/kg (N=8)**	**5 U/kg (N=8)**	**10 U/kg (N=8)**	**25 U/kg (N=8)**	**50 U/kg (N=8)**	**Pooled rFXIII (N=40)**
PT	3 (30%)	2	3	2	2	1	10 (25%)
aPTT	4 (40%)	3	2	4	1	4	14 (35%)
INR	0 (0%)	0	0	0	0	0	0 (0%)

Thrombin time	4 **(40%)**	3	0	6	5	6	20 **(50%)**
fibrinogen	2 **(20%)**	2	3	4	0	4	13 **(32.5%)**
D-Dimer	8 (80%)	5	6	4	6	6	27 (67.5%)

At each level of classification, patients are counted only once

Thrombin time showed a trend toward longer values with increasing doses of rFXIII. Otherwise, a trend toward increase in thrombin time in subjects receiving rFXIII relative to placebo was observed.

In addition, a trend toward increase in fibrinogen levels in subjects receiving rFXIII relative to placebo was observed.

Three subjects had a post treatment DVT score of 1 indicating a moderate probability of DVT.

Safety in special populations

Paediatric population

Factor XIII A-subunit deficient patients aged from 6 to less than 18 years old have been included in the study F13CD-1725 and in the extension study F13CD-3720. No assessment of this specific population was performed.

There was no data submitted in paediatric patients aged 1 to less than 6 years old with congenital FXIII A-subunit deficiency.

Elderly (> 65 years)

One elderly patient with congenital deficiency has been treated with rFXIII. No safety concerns were identified.

Fertility, pregnancy and lactation:

There was no data on fertility, pregnancy and lactation submitted.

Pregnancy

There are no clinical data on the use of NovoThirteen in pregnant women. Animal studies are insufficient with respect to reproductive toxicity (see section 5.3) as NovoThirteen has not been studied in pregnant animals. The risk to humans is not known. However, based on the therapeutic need the use of NovoThirteen as a replacement therapy may be considered during pregnancy.

Breast-feeding

It is unknown whether rFXIII is excreted in human breast milk. The excretion of rFXIII drug substance in milk has not been studied in animals. A decision on whether to continue/discontinue breast-feeding or to continue/discontinue therapy with NovoThirteen should be made taking into account the benefit of breast-feeding to the child and the benefit of NovoThirteen therapy to the mother.

Fertility

No effects on reproductive organs have been seen in non-clinical studies. There are no human data on potential effects on fertility.

Safety related to drug-drug interactions and other interactions

There was no clinical data submitted on interaction between rFXIII and other medicinal products.

Discontinuation due to adverse events

In study F13CD-1725, of the 41 patients, 8 patients were withdrawn from the trial. 3 patients were withdrawn due to development of antibodies but remained in the trial for safety monitoring purposes. One patient was withdrawn by the investigator after visit 6 due to worsening of neutropenia and leucopenia. Other patients were withdrawn due to pregnancy (2), personal reasons (1) or because there were too many blood samples (1).

In study F13CD-3720, of the 33 enrolled and dosed patients, 3 patients were withdrawn from the trial. One patient was withdrawn by the investigator after 3 doses of rFXIII due to pregnancy (withdrawal criteria) and another patient withdrew her consent due to a wish to become pregnant after 4 doses of rFXIII. A third patient wished to withdraw after 13 doses due to relocation of the clinical trial site.

Post marketing experience

There has been no post-marketing experience submitted for the product.

2.6.1. Discussion on clinical safety

In the two phase III studies (congenital FXIII deficiency), a total of 352 adverse events were reported in 32 of 41 (78%) of the exposed patients until the cut-off date of February 2011 (studies F13CD-1725 and F13CD-3720 pooled). Of these, 270 events were reported after exposure to $rFXIII_{IASMS}$ and 82 events were reported after exposure to $rFXIII_{NN}$. The overall rates of adverse events were 45.5% for $rFXIII_{IASMS}$ and 25.9 % for $rFXIII_{NN}$.

The main adverse event reported was "headache" (32 AEs – 4.2% $rFXIII_{IASMS}$ vs 2.2% $rFXIII_{NN}$ of adverse events) reported in 31.7% of patients. "Nasopharyngitis" (19 AEs – 2.0% vs 2.2%) was also frequently reported in 26.8% of patients. Other AEs frequently observed included "pyrexia" (10 AEs – 1.2% vs 0.9%), "incorrect dose administered" (12 AEs – 2% vs 0%, all with $rFXIII_{IASMS}$) and "oropharyngeal pain" (10 AEs – 1.0% vs 1.3%).

No anaphylactic reaction has been observed until the cut-off date of February 2011 in studies F13CD-1725 and F13CD-3720 but some adverse events as cough (6), conjunctivitis (1), rash papular (1), asthma (1) or eye swelling (2) have been observed and could have an allergic origin. One allergic reaction-related event has been observed in the clinical development program. As rFXIII is produced in yeast and due to the two cases of eczema which could have an allergic origin, a wording concerning the risk of allergic reaction has been included in section 4.4 of the SmPC. This risk was also included as a potential risk in the risk management plan. Finally, NovoThirteen is contraindicated in cases of known hypersensitivity to catridecacog ($rFXIII\text{-}A_2$).

A total of 15 adverse events were evaluated by the investigator to be possibly or probably related to NovoThirteen in Trials F13CD-1725 and F13CD-3720. These comprised events of positive antibody test (4 events in 4 patients), incorrectly administered dose (3 events in 2 patients), injection site pain (1 event), increase in fibrin D-dimer (1 event), pain in extremity (1 event), headache (1 event), limb injury (1 event), overdose (1 event) and coinciding events of neutropenia and leukopenia in one patient (worsening of mild neutropenia initially diagnosed before first trial drug administration).

No death was reported. Three serious reports of "antibody formation" were probably related to the study treatment and one serious case of anaphylactic reaction in a patient undergoing cardiac surgery is of interest. All patients recovered.

As there is a risk of clotting and fibrinolysis during sampling due to an incorrect storage, the risk of thromboembolic events remains of concern. This has been included in the SmPC in section 4.4 and the RMP. Additional risk minimisation will be implemented via educational tools.

No events of thromboembolism were observed during replacement therapy with rFXIII. However, cases of increase in fibrin D-Dimer, a trend toward increase in fibrinogen levels in patients receiving rFXIII relative to placebo, cases of prolonged thrombin time and a trend toward increase in thrombin time in patients receiving rFXIII relative to placebo have been observed. The risk of increased levels of non-proteolytically activated NovoThirteen in case of incorrect storage is of concern as it may increase the risk of thrombosis. In addition, there is an increased risk of embolic and thrombotic events including the risk of vessel occlusion in patients with thrombotic risk. The SmPC included wording in section 4.4 of the SmPC, for both confirmed venous or arterial thromboembolic events. "In case of predisposition to conditions of thrombosis, caution should be exercised due to the fibrin-stabilizing effect of NovoThirteen. A stabilization of the thrombus might occur, resulting in increased risk of vessel occlusions."

Laboratory and other safety variables revealed no clinically relevant findings and did not indicate any safety issues.

Assessment of safety in Factor XIII A-subunit deficient patients aged from 6 to less than 18 years old was performed. No safety issues other than the ones identified in the adult population have emerged in this population.

From the safety database all the adverse reactions reported in clinical trials have been included in the Summary of Product Characteristics. These were headache, leucopenia and aggravated neutropenia, pain in extremity, injection site pain, non-neutralising antibodies and fibrin D-dimer increase.

Four patients with congenital FXIII A-subunit deficiency and one healthy subject developed low-titre, transient, non neutralising antibodies rapidly following exposure to rFXIII. No inhibitor formation to rFXIII has been observed in clinical trials. However, neutralising antibodies have been observed in pivotal repeat-dose toxicity studies and this remains a potential risk of rFXIII. Long-term safety data are not sufficient to conclude on this safety issue. Inhibitors may be suspected in the event of lack of therapeutic response which is observed as bleeding or demonstrated by laboratory findings including FXIII activity that fails to reach expected levels. In the event that inhibitors are suspected analysis for antibodies should be performed. Moreover, patients known to have neutralising antibodies to FXIII should not be treated with NovoThirteen without close monitoring.

17 medication errors were recorded without any adverse events. Most of them were due to errors during the reconstitution or errors concerning the weight of the patient.

There are no clinical data on the use of NovoThirteen in pregnant women. Animal studies are insufficient with respect to reproductive toxicity (see section 5.3) as NovoThirteen has not been studied in pregnant animals. The risk to humans is not known. However, based on the therapeutic need the use of NovoThirteen as a replacement therapy may be considered during pregnancy.

It is unknown whether NovoThirteen is excreted in human breast milk. The excretion of NovoThirteen in milk has not been studied in animals. A decision on whether to continue/discontinue breast-feeding or to continue/discontinue therapy with NovoThirteen should be made taking into account the benefit of breast-feeding to the child and the benefit of NovoThirteen therapy to the mother.

Exposure during pregnancy will be recorded during routine pharmacovigilance, as part of the observational PASS study and in patients included in the PRO-RBDD registry with which cooperation has been agreed.

The safety profile of rFXIII in elderly has been described as important missing information, as there has been limited exposure to rFXIII in this population. The elderly population will be monitored via routine pharmacovigilance in the post marketing period. The SmPC included the wording "There is very limited clinical experience administering rFXIII to elderly patients with congenital FXIII deficiency" in Section 4.4 special warnings and precautions of the SmPC.

In patients with FXIII deficiency, NovoThirteen is not effective if used for monthly prophylactic treatment of bleeding in patients with congenital FXIII B-subunit deficiency. FXIII B-subunit deficiency is associated with a much reduced half-life of the administered pharmacologically active A-subunit. The subunit deficiency of patients should be determined prior to treatment by appropriate diagnostic procedures including factor XIII activity and immunoassay and if applicable genotyping.

Patients with hepatic impairment have not been studied. NovoThirteen may not be effective in patients with hepatic impairment if the hepatic impairment is severe enough to result in decreased levels of FXIII B-subunits. FXIII activity levels should be monitored in patients with severe hepatic impairment.

2.6.2. Conclusions on the clinical safety

The AES reported for patients being treated with rFXIII appear to be mostly of low grade and manageable. From the safety database, there were no safety signals that were raised by the safety data. However, it is of note that there was very limited safety data in patients with congenital FXIII deficiency who have received only $rFXIII_{NN}$ (10 patients – 45 doses). The long term safety will be assessed and monitored in congenital FXIII deficient patients who have received only rFXIII at the recommended dose, including the elderly and paediatric patients, as part of the cumulative safety reviews.

The CHMP was concerned over the off-label use of the product in breakthrough bleedings. No investigations of rFXIII in on-demand treatment setting and for breakthrough bleedings were performed. The monitoring of off-label use is covered by the RMP. However, to investigate the potential efficacy of NovoThirteen in breakthrough bleeding, the CHMP requested a post approval commitment to amend the ongoing trial, F13CD-3720, to investigate the clinical efficacy and safety of rFXIII in treatment of breakthrough bleedings during prophylactic treatment with rFXIII.

2.7. Pharmacovigilance

Detailed description of the pharmacovigilance system

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

Risk Management Plan

The Applicant submitted a risk management plan.

Table 47: Summary of the risk management plan

Safety concern	Proposed pharmacovigilance activities	Proposed risk minimisation activities
Identified risks		
Non-neutralising antibodies	• Continued analysis of safety data from clinical trials and safety data collected during post-marketing period including PASS NN1841-3868 • Structured follow-up of reports where clinical findings or laboratory findings may indicate a lack of expected effect in order to determine the cause • Analysis of blood samples for formation of antibodies when there is lack of effect indicated by clinical signs or laboratory findings reported • Case reporting on an expedited basis regardless of case type (seriousness and expectedness)	The SmPC Sections 4.2 and 4.8 state: • "Treatment should be initiated under the supervision of a doctor experienced in the treatment of rare bleeding disorders." • That frequency of non-neutralising antibodies is common (≥1/100 to <1/10), based on exposure in 51 patients.
Important potential risks		
Neutralising antibodies	• Continued analysis of safety data from clinical trials and safety data collected during post-marketing period including PASS NN1841-3868 and PRO-RBDD registry • Structured follow-up of reports of suspected neutralising antibodies and also reports where clinical findings or laboratory findings may indicate a lack of expected effect • Analysis of blood samples for formation of antibodies when there is lack of effect indicated by clinical signs or laboratory findings or there is suspicion of neutralising antibodies reported • Case reporting on an expedited basis regardless of case type (seriousness and expectedness)	The SmPC Sections 4.2 and 4.4 state: • "Patients known to have neutralising antibodies to FXIII should not be treated with NovoThirteen without close monitoring." • "Treatment should be initiated under the supervision of a doctor experienced in the treatment of rare bleeding disorders." • "Inhibitor formation to NovoThirteen therapy has not been detected in clinical trials. Inhibitors may be suspected in the event of lack of therapeutic response which is observed as bleeding or demonstrated by laboratory findings including FXIII activity that fails to reach expected levels. In the event that inhibitors are suspected analysis for antibodies should be performed. Patients known to have neutralising antibodies to FXIII should not be treated with NovoThirteen without close monitoring."
Allergic reactions	• Continued analysis of safety data from clinical trials and safety data collected during post-marketing period including PASS NN1841-3868 and PRO-RBDD registry • Structured follow-up of reports indicating a possible allergic reaction • Case reporting on an expedited basis regardless of seriousness and expectedness	The SmPC Sections 4.2 and 4.4 state: • "Treatment should be initiated under the supervision of a doctor experienced in the treatment of rare bleeding disorders." • "As NovoThirteen contains a recombinant protein it may cause allergic reactions including anaphylactic reaction. Patients should be informed of the early signs of hypersensitivity reactions (including hives, generalised urticaria, tightness of the chest, wheezing, hypotension) and anaphylaxis. If allergic or anaphylactic-type reactions occur, the administration should be immediately discontinued and further treatment with NovoThirteen should not be given."
Embolic and thrombotic events	• Continued analysis of safety data from clinical trials and safety data collected during post-marketing period including PASS NN1841-3868 and PRO-RBDD registry • Structured follow-up of reports indicating embolic and thrombotic events	The SmPC Sections 4.2 and 4.4 state: • "Treatment should be initiated under the supervision of a doctor experienced in the treatment of rare bleeding disorders." • "In case of predisposition to conditions of thrombosis, caution should be exercised due to the fibrin-stabilising effect of rFXIII. A stabilisation of the thrombus might occur, resulting in increased risk of vessel occlusions." • "Incorrect storage of the product after reconstitution must be avoided as it may result in loss of sterility and in increased levels of activated NovoThirteen. Increased levels of activated NovoThirteen may increase the risk of

		thrombosis." Patient educational material and physician information brochure are being prepared to decrease the risk of toxicity in connection with incorrect storage (draft in Annex 10 and Annex 11).
Lack of efficacy	• Continued analysis of safety data from clinical trials and safety data collected during post-marketing period including PASS NN1841-3868 and PRO-RBDD registry • Structured follow-up of reports indicating lack of efficacy	The SmPC Sections 4.1, 4.2 and 4.4 state: • "rFXIII is indicated for long-term prophylactic treatment of bleeding in patients 6 years and above with congenital factor XIII A-subunit deficiency." • "In patients with FXIII deficiency, NovoThirteen is not effective if used for monthly prophylactic treatment of bleeding in patients with congenital FXIII B-subunit deficiency. FXIII B-subunit deficiency is associated with a much reduced half life of the administered pharmacologically active A-subunit. The subunit deficiency of patients should be determined by appropriate diagnostic procedures including factor XIII activity and immunoassay and if applicable genotyping." • "Treatment should be initiated under the supervision of a doctor experienced in the treatment of rare bleeding disorders. The congenital factor XIII A-subunit deficiency should be confirmed by appropriate diagnostic procedures including factor XIII activity and immunoassay and if applicable genotyping." • "Patients with hepatic impairment have not been studied. NovoThirteen may not be effective in patients with hepatic impairment if the hepatic impairment is severe enough to result in decreased levels of FXIII B-subunits. FXIII activity levels should be monitored in patients with severe hepatic impairment."
Drug interaction of rFXIII with rFVIIa when used outside of the approved indication	• Collect exposure and follow-up on adverse reaction reports associated with off-label use during post-marketing period including PASS NN1841-3868 and PRO-RBDD registry.	Mandatory training for all relevant Novo Nordisk A/S employees in SOPs concerning off-label information is anticipated to improve communication and decrease potential off-label use. The SmPC Sections 4.1, 4.2 and 4.5 state: • "rFXIII is indicated for long-term prophylactic treatment of bleeding in patients 6 years and above with congenital factor XIII A-subunit deficiency." • "Treatment should be initiated under the supervision of a doctor experienced in the treatment of rare bleeding disorders. The congenital factor XIII A-subunit deficiency should be confirmed by appropriate diagnostic procedures including factor XIII activity and immunoassay and if applicable genotyping." • "There are no clinical data available on interaction between NovoThirteen and other medicinal products. Based on the non-clinical study (see section 5.3 in SmPC) it is not recommended to combine NovoThirteen and rFVIIa."
Medication error related to reconstitution and administration	• Continued analysis of safety data from clinical trials and safety data collected during post-marketing period including PASS NN1841-3868 and PRO-RBDD registry • Structure follow-up forms in cases of embolic and thrombotic events	The SmPC Section 4.4 states: "Incorrect storage of the product after reconstitution must be avoided as it may result in loss of sterility and in increased levels of activated NovoThirteen. Increased levels of activated NovoThirteen may increase the risk of

		thrombosis." Patient educational material and a physician information brochure are being prepared to avoid medication error related to reconstitution and administration (draft in Annex 10 and Annex 11).
Off-label use for management of bleedings	• An amendment to F13CD-3720 will be made to allow the collection of efficacy and safety data for the management of breakthrough bleeds with rFXIII. • Continued analysis of safety data from clinical trials (F13CD-3720) and safety data during the post-marketing period including PASS NN1841-3868 and PRO-RBDD registry. • Collection of drug utilisation data during the post-marketing period including PASS NN1841-3868 and PRO-RBDD registry.	As a routine risk minimisation, the SmPC Sections 4.1 and 4.4 state: • "rFXIII is indicated for long-term prophylactic treatment of bleeding in patients 6 years and above with congenital factor XIII A-subunit deficiency." • "The on-demand treatment of acute bleeds or breakthrough bleeds with NovoThirteen has not been studied in clinical trials. An alternative treatment could be considered in such situations." A physician information brochure is being prepared to create awareness among physicians about the indications of the product (draft in Annex 11).
Important missing information		
Children (<6 years of age)	• Continued analysis of safety data (F13CD-3760, F13CD-3835, NN1841-3868) • Continued analysis of safety data collected during post-marketing period including PASS NN1841-3868 and PRO-RBDD registry	The SmPC Sections 4.2 and 4.8 states: • "The safety and efficacy of NovoThirteen in children less than 6 years have not yet been established. No data are available." • "In clinical studies, adverse reactions were more frequently reported in patients aged from 6 to less than 18 years old than in adults. 4 patients (25%) under 18 years experienced serious adverse reactions in comparison to 3 patients over 18 years (8.5%) that experienced serious adverse reactions. Four cases of non-neutralising antibodies were reported at the start of the treatment in patients under 18 years of age. 3 of these patients discontinued the study due to the adverse reaction."
Elderly	• Continued analysis of safety data from clinical trials and safety data collected during post-marketing period including PASS NN1841-3868 and PRO-RBDD registry	The SmPC Section 4.4 states: "There is limited clinical experience in administering rFXIII to elderly patients with congenital FXIII deficiency."
Pregnant and lactating women	• Continued analysis of safety data collected during post-marketing period including PASS NN1841-3868 and PRO-RBDD registry	The SmPC Section 4.6 states: • "There are no clinical data on the use of NovoThirteen in pregnant women. Animal studies are insufficient with respect to reproductive toxicity as NovoThirteen has not been studied in pregnant animals. The risk to humans is not known. However, based on the therapeutic need the use of NovoThirteen as a replacement therapy may be considered during pregnancy. It is unknown whether rFXIII is excreted in human breast milk. The excretion of rFXIII drug substance in milk has not been studied in animals. A decision on whether to continue/discontinue breast-feeding or to continue/discontinue therapy with NovoThirteen should be made taking into account the benefit of breast-feeding to the child and the benefit of NovoThirteen therapy to the mother." • "No effects on reproductive organs have been seen in nonclinical studies. There are no human data on potential effects on fertility."
Patients with renal insufficiency	• Continued analysis of safety data collected during post-marketing period including PASS NN1841-3868 and PRO-RBDD registry	The SmPC Section 4.4 states: "Patients with renal insufficiency requiring dialysis have not been studied in clinical trials."

The CHMP, having considered the data submitted, was of the opinion that the below pharmacovigilance activities in addition to the use of routine pharmacovigilance are needed to investigate further some of the safety concerns:

Description	Due date
F13CD-3720 A multi-centre, open-label, single-arm and multiple dosing trial on safety of monthly replacement therapy with recombinant factor XIII (rFXIII) in subjects with congenital factor XIII deficiency. The Applicant commits to collect full PK-data of the 35IU/kg/month claimed dose in 5 patients and to investigate the clinical efficacy and safety of rFXIII in treatment of breakthrough bleedings during prophylactic treatment with rFXIII for one year.	2016
F13CD-3760 A phase 3b trial investigating the pharmacokinetics and safety profile of a single IV dose of rFXIII in paediatric (1 to less than 6 years old) subjects with congenital FXIII A-subunit deficiency	2012
F13CD-3835 A multi-centre, multinational, open-label, single-arm and multiple dosing trial on safety and efficacy of monthly replacement therapy with recombinant factor XIII (rFXIII) in paediatric subjects with congenital factor XIII A-subunit deficiency (safety extension trial to F13CD-3760)	2014
NN1841-3868 Treatment of congenital FXIII deficiency, a prospective multi-centre observational study	2018 Submission of the draft protocol to the CHMP by 30 June 20012
Reports from PRO-RBDD registry	in the PSURs until 2015

The following additional risk minimisation activities were required:

The Marketing Authorisation Holder (MAH) shall ensure that, at launch, a letter is sent to all expected and actual prescribers of NovoThirteen with an Educational Pack containing the following:

1. Physician brochure
2. Patient brochure

Both documents are to be used as part of an educational plan aiming to minimise risks of medication errors, risk of thromboembolic events due to increased levels of non-proteolytically activated rFXIII in connection with incorrect storage, and risk of off-label use for treatment of breakthrough bleeding. The MAH should ensure harmonisation between terminology used in the brochures and the product information.

The physician brochure should contain the following key elements and item:

- indication of the product
- the risks of off-label use within FXIII congenital deficiency
- appropriate diagnostic procedures to confirm FXIII A-subunit deficiency
- warning of the difference of both posology and concentration between NovoThirteen and other FXIII containing products (The recommended dose of NovoThirteen is 35 IU/kg body weight (bw) once monthly, administered as an intravenous bolus injection. The dose volume in millilitres should

be calculated based on body weight for each patient using the following formula: Dose volume in ml = 0.042 x subject bw (kg).)

- correct handling and the risks associated with mishandling
- embolic and thrombotic events including the increased risk of vessel occlusion in patients at risk of thrombosis
- what to do in the event of incorrect storage, thrombosis or embolism
- contraindication of hypersensitivity
- warning and precautions regarding anaphylaxis
- the importance of collecting safety data and how to enrol patients in the PASS and other registries
- distribution and use of the patient brochure and the need to ensure that the patient has read and understood the brochure
- Summary of Product Characteristics

The patient brochure, to be distributed to patients by the prescribers, should contain the following key elements and item:

- indication of the product
- the risks of off-label use within FXIII congenital deficiency
- how to safely store, handle, reconstitute and administer the product
- the risks associated with incorrect storage and mishandling
- how to recognise the potential side effects (thrombosis and embolism)
- what to do in the event of incorrect storage, thrombosis or embolism
- Package Leaflet

The Marketing Authorisation Holder must implement this educational plan nationally, prior to marketing. The final content, format and distribution modalities of both documents should be agreed with the national competent authority in each Member State.

In addition, the CHMP considered that the Applicant should take the following minor points into consideration when an update of the Risk management Plan is submitted:

1. Table of contents: The hyperlinks should be corrected, e.g. for section 4.
2. Table 37, last row and table 31 should be aligned with the middle column of the respective risk in table 48, which the Applicant changed in the latest RMP.
3. Each reference should be hyperlinked to eCTD section 5.4.

2.8. 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.*

3. Benefit-Risk Balance

Benefits

Beneficial effects

The pivotal trial met its primary endpoint and the efficacy of NovoThirteen in the indication "Long term prophylactic treatment of bleeding in patients 6 years and above with congenital factor XIII A-subunit deficiency" is considered established. During the rFXIII prophylactic treatment period, five traumatic bleeding episodes that required treatment with a FXIII-containing product were observed in 4 patients out of 41 subjects enrolled. No intracranial or life-threatening bleedings occurred.

Current treatment options for patients with a congenital FXIII deficiency include cryoprecipitate, fresh frozen plasma, and plasma-derived FXIII concentrate. NovoThirteen, a recombinant derived FXIII A_2 subunit protein, represents an additional choice with an advantage over the other products of no potential virus safety risk.

Uncertainty in the knowledge about the beneficial effects

The final dosing proposal of 35 IU/kg was derived from phase III results and correlation between dose and response is missing. No PK data with the recommended posology was formally assessed. In addition, most bleedings occurred in the middle of the prophylaxis interval where FXIII levels should be higher than 10%. As this result may indicate the need for dose adjustment, the CHMP requested to amend the current ongoing extension study (F13CD-3720) in order to collect full PK-data from congenital FXIII deficient patients administered the recommended posology.

Risks

Unfavourable effects

The safety database showed no major concerns over adverse reactions. 14 adverse events were considered possibly or probably related to rFXIII in the two phase III studies. These comprised 4 events of positive antibody test (non-neutralising), 3 events of incorrectly dose administered, 1 event of increase D-Dimer, 1 event of pain in extremity, 1 headache, 1 injection site pain, 1 overdose and coinciding events of neutropenia and leukopenia. All these adverse events have been included in the SmPC of rFXIII. As rFXIII is produced in yeast and it is probable that the cases of eczema could have an allergic origin, the risk of allergic reaction remains a potential risk.

No neutralising antibodies have been reported. However, neutralising antibodies were included in the RMP as an identified risk.

Adverse events were more frequently reported in patients aged from 6 to less than 18 years old than in older patients: 4 patients (25%) under 18 years experienced serious adverse reactions in comparison to 3 patients over 18 years (8.5%) that experienced serious adverse reactions. Out of the 4 cases of non-neutralising antibodies that were reported, 3 of these patients discontinued the study due to the adverse reaction. This is adequately reflected in the SmPC of NovoThirteen (section 4.8 "paediatric population").

The data from paediatric patients from 1 to less than 6 years old was very limited. Thus, this population was not included in the indication and information in the SmPC.

The risk of increased levels of non-proteolytically activated rFXIII in case of incorrect storage was of concern as this may increase the risk of embolic and thrombotic events including the risk of vessel occlusion in patients with thrombotic risk. The CHMP considered that the addition of risk minimisation measures was necessary and educational materials were proposed for physicians and patients on the appropriate storage, handling, reconstitution and administration of rFXIII.

Uncertainty in the knowledge about the unfavourable effects

The magnitude of the risk of thrombotic and embolic events derived from the increased levels of non-proteolytically activated NovoThirteen when the medication is not stored properly is unknown. In addition, the risk of medication errors when patients receive their treatment at home was highlighted in the list of ADRs. These risks were included in the RMP as important potential risk, to be monitored via routine PV and PASS. However, as the risk minimisation activities was considered insufficient to adequately address some risks, an educational program, including physician and patient brochures, has been developed.

Benefit-risk balance

Importance of favourable and unfavourable effects

Overall, the pivotal study provided satisfactory results with respect to efficacy (no occurrence of intracranial or life-threatening bleedings) and safety (no emergence of a major safety signal). The availability of a recombinant form of FXIII as a therapeutic option for the treatment of congenital FXIII deficiency was of interest given the viral safety. Thus, the beneficial effect of treatment with NovoThirteen in the prevention of bleeding episodes in patients with severe congenital FXIII deficiency was regarded as an important aspect for patients with this rare disease.

Benefit-risk balance

Based on the results of the pivotal trial F13CD-1725 and the extension study F13CD-3720, the benefits of NovoThirteen treatment in the prevention of bleeding episodes in patients with severe congenital FXIII deficiency outweighed the adverse events (leucopenia and aggravated neutropenia, headache, pain in extremity, injection site pain, non-neutralising antibodies, fibrin D-dimer). Therefore, the CHMP considers that the benefit-risk balance for rFXIII in the long term prophylactic treatment of bleeding in patients 6 years and above with congenital factor XIII A-subunit deficiency is positive.

4. Recommendations

Outcome

Based on the CHMP review of data on quality, safety and efficacy, the CHMP considers by consensus that the risk-benefit balance of NovoThirteen in the long-term prophylactic treatment of bleeding in patients 6 years and above with congenital factor XIII A-subunit deficiency is favourable and therefore recommends the granting of the 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).

Conditions and requirements of the Marketing Authorisation

Risk Management System

The MAH must ensure that the system of pharmacovigilance, presented in Module 1.8.1 of the marketing authorisation, is in place and functioning before and whilst the product is on the market.

The MAH shall perform the pharmacovigilance activities detailed in the Pharmacovigilance Plan, as agreed in edition 7 of the Risk Management Plan (RMP) presented in Module 1.8.2 of the marketing authorisation and any subsequent updates of the RMP agreed by the CHMP.

As per the CHMP Guideline on Risk Management Systems for medicinal products for human use, the updated RMP should be submitted at the same time as the next Periodic Safety Update Report (PSUR).

In addition, an updated RMP should be submitted:

- When new information is received that may impact on the current Safety Specification, Pharmacovigilance Plan or risk minimisation activities
- Within 60 days of an important (pharmacovigilance or risk minimisation) milestone being reached
- at the request of the EMA

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

The Marketing Authorisation Holder (MAH) shall ensure that, at launch, a letter is sent to all expected and actual prescribers of NovoThirteen with an Educational Pack containing the following:

1. Physician brochure
2. Patient brochure

Both documents are to be used as part of an educational plan aiming to minimise risks of medication errors, risk of thromboembolic events due to increased levels of non-proteolytically activated rFXIII in connection with incorrect storage, and risk of off-label use for treatment of breakthrough bleeding. The MAH should ensure harmonisation between terminology used in the brochures and the product information.

The physician brochure should contain the following key elements and item:

- indication of the product
- the risks of off-label use within FXIII congenital deficiency
- appropriate diagnostic procedures to confirm FXIII A-subunit deficiency
- warning of the difference of both posology and concentration between NovoThirteen and other FXIII containing products (The recommended dose of NovoThirteen is 35 IU/kg body weight (bw) once monthly, administered as an intravenous bolus injection. The dose volume in millilitres should be calculated based on body weight for each patient using the following formula: Dose volume in ml = 0.042 x subject bw (kg).)
- correct handling and the risks associated with mishandling
- embolic and thrombotic events including the increased risk of vessel occlusion in patients at risk of thrombosis
- what to do in the event of incorrect storage, thrombosis or embolism

- contraindication of hypersensitivity
- warning and precautions regarding anaphylaxis
- the importance of collecting safety data and how to enrol patients in the PASS and other registries
- distribution and use of the patient brochure and the need to ensure that the patient has read and understood the brochure
- Summary of Product Characteristics

The patient brochure, to be distributed to patients by the prescribers, should contain the following key elements and item:

- indication of the product
- the risks of off-label use within FXIII congenital deficiency
- how to safely store, handle, reconstitute and administer the product
- the risks associated with incorrect storage and mishandling
- how to recognise the potential side effects (thrombosis and embolism)
- what to do in the event of incorrect storage, thrombosis or embolism
- Package Leaflet

The Marketing Authorisation Holder must implement this educational plan nationally, prior to marketing. The final content, format and distribution modalities of both documents should be agreed with the national competent authority in each Member State.

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

The Member States should ensure that all conditions or restrictions with regard to the safe and effective use of the medicinal product described below are implemented:

The Marketing Authorisation Holder (MAH) shall ensure that, at launch, a letter is sent to all expected and actual prescribers of NovoThirteen with an Educational Pack containing the following:

1. Physician brochure
2. Patient brochure

Both documents are to be used as part of an educational plan aiming to minimise risks of medication errors, risk of thromboembolic events due to increased levels of non-proteolytically activated rFXIII in connection with incorrect storage, and risk of off-label use for treatment of breakthrough bleeding. The MAH should ensure harmonisation between terminology used in the brochures and the product information.

The physician brochure should contain the following key elements and item:

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once monthly, administered as an intravenous bolus injection. The dose volume in millilitres should be calculated based on body weight for each patient using the following formula: Dose volume in ml = 0.042 x subject bw (kg).)

- correct handling and the risks associated with mishandling
- embolic and thrombotic events including the increased risk of vessel occlusion in patients at risk of thrombosis
- what to do in the event of incorrect storage, thrombosis or embolism
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- indication of the product
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- what to do in the event of incorrect storage, thrombosis or embolism
- Package Leaflet

The Marketing Authorisation Holder must implement this educational plan nationally, prior to marketing. The final content, format and distribution modalities of both documents should be agreed with the national competent authority in each Member State.

New Active Substance Status

Based on the CHMP review of data on the quality properties of the active substance, the CHMP considers that catridecacog (rFXIII-A_2) is qualified as a new active substance.

Paediatric Data

Furthermore, the CHMP reviewed the available paediatric data of studies subject to the agreed Paediatric Investigation Plan P/273/2010 and the results of these studies are reflected in the Summary of Product Characteristics (SmPC) and, as appropriate, the Package Leaflet.