Recombinant FXIII (rFXIII-A₂) Prophylaxis Prevents Bleeding and Allows for Surgery in Patients with Congenital FXIII A-Subunit Deficiency

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Recombinant factor XIII-A₂ (rFXIII-A₂) was developed for prophylaxis and treatment of bleeds in patients with congenital FXIII A-subunit deficiency. mentor™2 (NCT00978380), a multinational, open-label, single-arm, multiple-dosing extension to the pivotal mentor™1 trial, assessed long-term safety and efficacy of rFXIII-A₂ prophylaxis in eligible patients (patients with severe [<0.05 IU/mL] congenital FXIII subunit A deficiency) aged ≥6 years. Patients received 35 IU/kg rFXIII-A₂ (exact dosing) every 28 ± 2 days for ≥52 weeks. Primary endpoint was safety (adverse events including immunogenicity); secondary endpoints were rate of bleeds requiring FXIII treatment, haemostatic response after one 35 IU/kg rFXIII-A₂ dose for breakthrough bleeds and withdrawals due to lack of rFXIII-A₂ efficacy. Steady-state pharmacokinetic variables were also summarized. Elective surgery was permitted during the treatment period. Sixty patients were exposed to rFXIII-A₂; their median age was 26.0 years (range: 7.0–77.0). rFXIII-A₂ was well tolerated without any safety concerns. No non-neutralizing or neutralizing antibodies (inhibitors) against FXIII were detected. Mean annualized bleeding rate (ABR) was 0.043/patient-year. Mean spontaneous ABR was 0.011/patient-year. No patients withdrew due to lack of efficacy. Geometric mean FXIII trough level was 0.17 IU/mL. Geometric terminal half-life was 13.7 days. rFXIII-A₂ prophylaxis provided sufficient haemostatic coverage for 12 minor surgeries without the need for additional FXIII therapy; eight procedures were performed within 7 days of the patient’s last scheduled rFXIII-A₂ dose, and four were performed 10 to 21 days after the last dose.

Keywords
► recombinant FXIII-A₂
► safety
► congenital FXIII deficiency
► prophylaxis
► surgery

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Abstract

Recombinant factor XIII-A₂ (rFXIII-A₂) was developed for prophylaxis and treatment of bleeds in patients with congenital FXIII A-subunit deficiency. mentor™2 (NCT00978380), a multinational, open-label, single-arm, multiple-dosing extension to the pivotal mentor™1 trial, assessed long-term safety and efficacy of rFXIII-A₂ prophylaxis in eligible patients (patients with severe [<0.05 IU/mL] congenital FXIII subunit A deficiency) aged ≥6 years. Patients received 35 IU/kg rFXIII-A₂ (exact dosing) every 28 ± 2 days for ≥52 weeks. Primary endpoint was safety (adverse events including immunogenicity); secondary endpoints were rate of bleeds requiring FXIII treatment, haemostatic response after one 35 IU/kg rFXIII-A₂ dose for breakthrough bleeds and withdrawals due to lack of rFXIII-A₂ efficacy. Steady-state pharmacokinetic variables were also summarized. Elective surgery was permitted during the treatment period. Sixty patients were exposed to rFXIII-A₂; their median age was 26.0 years (range: 7.0–77.0). rFXIII-A₂ was well tolerated without any safety concerns. No non-neutralizing or neutralizing antibodies (inhibitors) against FXIII were detected. Mean annualized bleeding rate (ABR) was 0.043/patient-year. Mean spontaneous ABR was 0.011/patient-year. No patients withdrew due to lack of efficacy. Geometric mean FXIII trough level was 0.17 IU/mL. Geometric terminal half-life was 13.7 days. rFXIII-A₂ prophylaxis provided sufficient haemostatic coverage for 12 minor surgeries without the need for additional FXIII therapy; eight procedures were performed within 7 days of the patient’s last scheduled rFXIII-A₂ dose, and four were performed 10 to 21 days after the last dose.
**Introduction**

Factor XIII (FXIII) is the terminal enzyme in the blood coagulation cascade and essential for cross-linking fibrin molecules to form an effective and stable blood clot. In plasma, FXIII circulates as an inactive heterotetramer composed of two catalytic FXIII A-subunits and two carrier FXIII B-subunits (FXIII-A2B2). A deficiency or dysfunction of either the A or B subunit can result in FXIII deficiency. Congenital FXIII deficiency is a rare and serious autosomal recessive coagulation disorder with a high risk of life-threatening bleeding complications. The prevalence of congenital FXIII deficiency is estimated to be approximately 1 in 1 to 2 million people worldwide. Its incidence is much higher in certain parts of the world where consanguinity is common; this is particularly the case in Iran, which has the highest reported rate of FXIII deficiency. Patients with congenital FXIII deficiency experience major spontaneous bleeding episodes (e.g., intracranial, intramuscular and gastrointestinal tract bleeds) more frequently than patients with most other rare bleeding disorders and are at a high risk of intraoperative and delayed postoperative bleeds.

The most common form of congenital FXIII deficiency is A-subunit deficiency, which is more severe and accounts for 95% of all severe deficiency, while B-subunit deficiency is very rare. Patients with congenital FXIII deficiency require FXIII replacement to treat bleeds and, even more importantly, to prevent them (prophylaxis). The long half-life of FXIII, combined with the high risk of life-threatening bleeds associated with FXIII deficiency, makes prophylaxis an attractive and efficacious treatment option. Thus, prophylaxis is strongly recommended for all patients with severe FXIII deficiency. Until recently, only plasma-derived sources of FXIII have been available (fresh frozen plasma [FFP], cryoprecipitate and plasma-derived FXIII [pdFXIII] concentrate). Such products carry a risk of pathogen transmission and do not contain very concentrated forms of FXIII, such that large volumes need to be administered; pdFXIII concentrate contains approximately 62.5 U of FXIII/mL. Recombinant FXIII (rFXIII-A2, Novo Thirteen; Tretten, Novo Nordisk, Bagsvaerd, Denmark) is the only rFXIII product available; it contains a 13-fold higher concentration of FXIII (833 IU/mL) compared with pdFXIII. rFXIII contains only the FXIII-A2 subunit, offering a treatment option to prevent bleeds in patients with the much more common and more severe form of congenital FXIII deficiency—subunit-A deficiency. rFXIII-A2 is a highly purified product manufactured in Saccharomyces cerevisiae; no human or other mammalian blood or tissue products are used during its manufacture; thus, FXIII-A2 does not carry a risk of contamination with infectious agents, including viruses or prions.

The efficacy and safety of rFXIII-A2 in patients with this rare bleeding disorder have been demonstrated in a large-scale clinical trial programme (mentor™). In the pivotal mentor™ trial with patients aged ≥6 years, prophylaxis with 35 IU/kg rFXIII-A2 dosed every 4 weeks provided a good safety profile and resulted in a very low mean annualized bleeding rate (ABR) of only 0.138 bleeds/patient-year. Similarly, the paediatric mentor™5 showed excellent efficacy and safety of prophylaxis with 35 IU/kg rFXIII-A2 dosed every 4 weeks in patients aged <6 years. Previous reports have shown similar pharmacokinetics (PK) in young children to that of older children and adults. In the mentor™ programme to date, no patients treated with rFXIII-A2 have developed FXIII-neutralizing antibodies (i.e., FXIII inhibitors).

The mentor™2 extension trial is the largest clinical trial performed to date in patients with congenital FXIII deficiency. Here, we report the long-term safety and efficacy results for rFXIII-A2 replacement therapy dosed every 4 weeks for bleed prevention and for treatment of breakthrough bleeds in children, adolescents and adults with congenital FXIII A-subunit deficiency. As mentor™2 permitted elective surgeries, we also report data on minor surgical procedures conducted in patients receiving rFXIII-A2 prophylaxis during the trial.

**Methods**

As the mentor™2 trial is an extension of the pivotal mentor™1 trial, the methods have been reported previously, but will be briefly described here.

**Trial Conduct**

Prior to the start of the trial, written informed consent was obtained from each patient. In addition, the protocol, consent form and patient information sheet were reviewed and approved according to local regulations by appropriate health authorities and independent ethics committees/institutional review boards. The trial was conducted in accordance with the U.S. Food and Drug Administration Code of Federal Regulations, the International Conference on Harmonisation Good Clinical Practice Guidelines, applicable regulatory requirements and the Declaration of Helsinki.

**Patients**

Eligible patients were aged ≥6 years, weighed ≥20 kg and had a diagnosis of congenital FXIII A-subunit deficiency (FXIII A subunit level of <0.05 IU/mL with genetic confirmation demonstrating homozygosity or compound heterozygosity for FXIII A subunit deficiency mutation[s]). Key exclusion criteria were any known congenital or acquired coagulation disorder, other than congenital FXIII deficiency; a known history of thromboembolic events; known inhibitors to FXIII; and any medical, social or psychosocial factors expected to impact compliance or safety.

**Trial Design**

The mentor™2 trial (Fig. 1) was a multicentre, multinational, open-label, single-arm, multiple-dosing, non-randomized safety extension to the pivotal mentor™1 trial. Important protocol amendments to mentor™2 included the addition of new patients who did not enrol from mentor™1 and the option to treat breakthrough bleeds with rFXIII-A2 (introduced in 2012). Inclusion of re-enrolled patients was also allowed. The per-protocol prophylaxis dose of rFXIII-A2 was 35 IU/kg (exact dosing given; achieving this required wastage of product when
Patients from mentor™1

New patients
n = 34

Patients exposed to rFXIII
(19) N = 60

Patients undergoing PK study
n = 23 (38.3%)

Withdrawals
Non-compliance 18 (60.0%)
Withdrawing criteria 2 (3.3%)
Other 8 (13.3%)

Completed main phase
n = 42 (70.0%)

Full analysis
N = 60 (100.0%)

Fig. 1 Patient disposition. ID, identification; N, number of patients; PK, pharmacokinetics. Note: A total of 60 patients were enrolled and exposed in the trial, but three were later withdrawn and subsequently re-enrolled with new patient identifications, giving rise to a total of N = 63 patient identifications. *From the mentor™1 trial, there were 34 patients enrolled into mentor™2.

calculated amount was less than full vial of 2,500 U/vial), given intravenously every 28 ± 2 days for a minimum of 52 weeks. Patients who experienced breakthrough bleeds and required additional FXIII treatment were to be treated according to local standard practice, with the option for treatment with rFXIII-A2 (a single 35 IU/kg dose was to be used, if possible).

Elective surgery was permitted during the treatment period. For any surgical procedure, if investigators judged that additional FXIII substitution was required, then pdFXIII was to be administered according to local practice. After any administration of pdFXIII to cover surgery, patients would continue with the original rFXIII-A2 replacement schedule (i.e., dosing every 4 weeks), with the next dose administered 28 ± 2 days after the last scheduled dose.

Objectives and Endpoints
The primary objective was to assess the long-term safety (including any antibody development against rFXIII-A2—if any) of replacement therapy every 4 weeks with rFXIII-A2 when used for the prevention of bleeding episodes and for the treatment of breakthrough bleeds in patients with congenital FXIII A-subunit deficiency. Important endpoints comprised the assessment of all adverse events (AEs; both serious and non-serious, including any AEs of special interest, such as antibody development against rFXIII-A2, medication errors or thromboembolic events). Additional secondary safety endpoints comprised any abnormalities in laboratory variables, vital signs and body measurements. Other secondary objectives were to evaluate the efficacy and summarize the steady-state PK profile of replacement therapy every 4 weeks with rFXIII-A2. Laboratory sampling and medical assessments were performed every 12 weeks throughout the treatment period.

Efficacy assessments, performed by the patient or caregiver in consultation with the investigator, were based on a 4-point scale (“Excellent”, “Good/Effective”, “Moderate/Partly Effective” and “Poor”). Haemostatic outcome was only collected for bleeding episodes treated with rFXIII-A2. The number of patients withdrawn due to lack of treatment efficacy was also recorded.

The Berichrom® FXIII chromogenic activity assay (Siemens Healthcare, Erlangen, Germany) was used to measure FXIII activity. A subgroup of patients participated in steady-state PK assessments in which blood samples were collected at pre-dose, 1 and 2 hours and at 3, 7, 14, 21 and 28 days post-dose. Only patients who had already received a minimum of 10 doses of rFXIII-A2 participated in the PK assessment. PK methods are explained in more detail in Kerlin et al.

For patients undergoing elective surgery, the following assessments were made during the perioperative period: type and severity (major/minor) of surgery, timing of the surgery in relation to the last rFXIII-A2 dose, haemoglobin levels pre- and post-surgery, dose regimen of pdFXIII, additional haemostatic drugs used, the amount of blood and blood products transfused and AEs. Minor surgery was defined as any invasive operative procedure in which only skin, mucous membranes or superficial connective tissue was manipulated. All AEs, concomitant medication and bleeds were monitored up to 4 weeks after the date of surgery.

Antibody Assays
An anti-rFXIII antibody enzyme-linked immunosorbent assay (ELISA) and an in vitro neutralizing antibody assay were used for immune response screening, confirmation and antibody characterization.

Statistical Analysis
The sample size for this study was estimated to cover the patients migrating from mentor™1 and augmented by 10 to 20 additional patients who had not originally participated in mentor™1.

Sixty unique patients enrolled into mentor™2. All patients were included in the full analysis set and all safety endpoints were summarized using descriptive statistics. As in mentor™1, ABR was based on the number of bleeding episodes requiring FXIII treatment and was evaluated by a Poisson regression model with baseline age as a covariate and total observation time as an offset. The number of bleeds requiring treatment with a FXIII-containing product, haemostatic response to FXIII-A2 treatment and number of patients withdrawn due to lack of treatment efficacy were tabulated. PK variables were calculated based on non-compartmental methods without prior baseline adjustments, as described by Kerlin et al.

Results
Patients
Sixty patients (16 children aged 7 to <18 years and 44 adults aged 18–77 years) from 34 centres in 12 countries (Austria, Canada, Finland, France, Germany, Israel, Italy, Japan, Spain,
Switzerland, UK and United States) were enrolled and exposed to rFXIII-A2 between 21 September 2009 and 20 October 2015. Thirty-four patients were enrolled from the mentor™1 trial and an additional 26 newly enrolled patients were included (Fig. 1). The median age of patients at enrolment was 26.0 years (range: 7.0–77.0 years; Table 1); 16 (26.7%) patients were younger than 18 years. The majority of patients were male (n = 38; 63.3%) and most were Caucasians (n = 34; 56.7%).

Eighteen patients (30.0%) withdrew from the trial (Fig. 1). Three withdrew because they became pregnant and five because they failed to comply with the trial treatment or protocol; thus, these eight patients met the protocol-defined withdrawal criteria. Two patients were withdrawn due to non-compliance as they were out of the country for 7 weeks. An additional eight patients withdrew due to non-compliance as they were out of the country for another reason (patient/family withdrew consent without specifying a reason (n = 5); patient elected to stop study treatment and receive commercially available rFXIII-A2 (n = 1); patient undertook a yearlong trip, so could not remain in the study (n = 1); and patient wished to be treated with commercially available pdFXIII (n = 1). Of the 18 patients who withdrew, three later re-enrolled onto the mentor™2 trial and were given new patient IDs (reasons for initial withdrawal of these patients were patient wishes, pregnancy and compliance issues).

### Safety

Patients were exposed to a total of 2,410 rFXIII-A2 doses, corresponding to 186.5 patient-years of cumulative exposure (an average of 3.1 [range: 0.15–5.37] years per patient). rFXIII-A2 prophylaxis once every 4 weeks was well tolerated. There were 920 treatment-emergent AEs reported in 56 (93.3%) patients (Table 2); headache and nasopharyngitis were the most common AEs. The vast majority of AEs (904/920; 90.3%) were mild or moderate in severity, while 16/920 (1.7%) were severe. Of the 920 AEs, 19 (occurring in 12 patients) were considered serious, based on seriousness criteria. All 19 serious AEs (SAE) were evaluated as unlikely to be related to rFXIII-A2 treatment (Table 3); all of these patients with SAEs continued FXIII-A2 treatment at the same dose. Seven AEs in six patients were evaluated as being possibly/probably related to rFXIII-A2 (Table 3); however, all six patients recovered. There were 12 AEs of special interest in eight patients, 10 of which were small deviations from the correct dose, ranging from 28.2 to 37.6 IU/kg (Table 3). One event of overdose (~2.3 times the planned dose) was evaluated by the investigator as mild and did not have any clinical consequences. All AEs of special interest were considered mild and the patients recovered. None of these medication errors were judged to have had an impact on the results of the trial.

No anti-rFXIII antibodies (neutralizing or non-neutralizing) were detected in the mentor™2 trial. There were no thromboembolic events, fatal AEs or AEs leading to withdrawal (Table 2) and no anaphylactic or allergic reactions to rFXIII-A2.

### Efficacy

No patients were withdrawn due to lack of rFXIII-A2 efficacy.

During the trial, the rate of FXIII treatment requiring bleeds was low: eight bleeds in seven patients. Six bleeds were trauma induced and two were spontaneous (Table 4). The spontaneous bleeds were epistaxis (picked nose) in an 8-year-old patient, and a muscular bleed (after considerable physical work) in a 25-year-old patient. There was one intracranial bleeding episode that was the result of a head injury in a severe car accident, occurring 4 days after the patient’s last prophylactic dose of rFXIII-A2. The intracranial bleed was considered unrelated to rFXIII-A2 treatment by the

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### Table 1 Baseline characteristics

| Patients (N)² | Total |
|--------------|-------|
| Adults (≥18 y of age); adolescents/children (6 to <18 y of age) | 44:16 |
| Sex (female: male) | 22:38 |
| Weight, mean (SD), kg | 67.7 (19.9) |
| Body mass index, mean (SD), kg/m² | 24.3 (5.3) |
| Race, N (%) | |
| Caucasian | 34 (56.7) |
| Asian | 9 (15.0) |
| Black or African American | 6 (10.0) |
| American Indian or Alaskan Native | 1 (1.7) |
| Other | 6 (10.0) |
| Unknown | 4 (6.7) |

Abbreviations: N, number of patients; SD, standard deviation.

²Includes three patients who withdrew from the trial and were subsequently re-enrolled with new patient IDs.

### Table 2 Summary of adverse events (AEs)

| Summary | Number of patients²/ events |
|---------|-----------------------------|
| All AEs | 56 (93.3%); 920 |
| AEs by severity | |
| Severe | 12 (20%); 16 |
| Moderate | 26 (43.3%); 119 |
| Mild | 54 (90%); 785 |
| AEs by relationship to rFXIII-A2 treatment | |
| Probably or possibly related | 6 (10%); 7 |
| Unlikely related | 56 (93.3%); 890 |
| Missing | 6 (10%); 23 |
| AEs leading to withdrawal | 0 |

Abbreviations: rFXIII-A2, recombinant factor XIII-A2; %, percentage of patients with AEs.

Note: Non-serious AEs that were symptoms of other AEs were not included.

²When summarizing AEs, the three re-enrolled patients were assigned their initial patient IDs to avoid double counting.
investigator. No internal organ bleeds or severe gastrointestinal bleeds occurred. The majority of patients (four of seven) who had a FXIII treatment requiring bleed were younger than 18 years (►Table 5). Three bleeds occurred within the first 14 days post-rFXIII-A2 infusion when FXIII levels were predicted to be greater than 0.3 IU/mL in most patients, while five occurred between days 17 and 24 post-rFXIII-A2 infusion. As such, the mean ABR for patients on study (treated with 35 IU/kg rFXIII-A2 every 4 weeks) was 0.043 (0.032 and 0.011 for traumatic and spontaneous bleeds/patient-year, respectively; ►Table 4). This mean ABR implies only one bleed per 23 patient-years, while the mean spontaneous bleed ABR of 0.011 bleeds/patient-year implies one spontaneous bleed per 91 patient-years.

### Table 3 Details of serious adverse events (SAEs), possible/probable treatment-related adverse events (AEs) and medical events of special interest

| Type of AE | N  | Description                                                                 | Days since last dose |
|------------|----|------------------------------------------------------------------------------|---------------------|
| SAE        |    |                                                                              |                     |
| Falls or accidents | 10 | • Five SAE related to motor vehicle accidents in two patients (female, aged 23; male, aged 29) | 12 (female, aged 23) and 4 (male, aged 29) |
|            |    | • Two SAE due to fall/chest injury in one patient (female, aged 48)          | 7 and 23, respectively |
|            |    | • Two SAE related to a head injury in one patient (female, aged 8°)          | 8 and 25, respectively |
|            |    | • One SAE related to a laceration to the head in one patient (male, aged 12) | 24                  |
| Left inguinal hernia | 1 | Herina surgical repair under rFXIII-A2 prophylaxis (male, aged 50)           | 33                  |
| Sigmoid diverticulitis | 1 | Female, aged 57                                                             | 24                  |
| Worsening of bilateral chronic otitis media | 1 | Male, aged 55                                                               | 11                  |
| Ectopic pregnancy | 1 | Led to termination of pregnancy at gestation week 7 (female, aged 25)        | 28                  |
| Cerebral ischaemia | 1 | Secondary to accidental, work-related carbon monoxide poisoning (male, aged 60; history of diabetes mellitus) | 8                  |
| Headache    | 1 | Admitted to hospital due to headaches (female, aged 8°)                      | 25                  |
| Suicide attempt | 1 | Overdose with aspirin and ibuprofen (female, aged 14)                       | 20                  |
| Atrial septal defect and persistent ductal arteriosus | 2 | 277 days (8 months) after a woman (aged 24) withdrew from trial due to pregnancy her newborn was born with defects | 277                |

#### Possible/probable rFXIII-A2 treatment-related AE

| Incorrect dose | 1 | Accidentally dosed 3.0 mL instead of 3.2 mL (male, aged 15)                  | 28                  |
| Overdose      | 1 | Approximately 2.3 times overdose (80.03 IU/kg instead of 35 IU/kg), no adverse clinical effect (male, aged 8°) | 29                  |
| Limb injury   | 1 | Bleeding under toenail due to trauma (male, aged 22)                         | 28                  |
| Arthralgia    | 1 | Left knee pain (male, aged 8°)                                               | 22                  |
| Leukopenia    | 1 | Asymptomatic leukopenia (female, aged 15)                                     | 28                  |
| Alanine aminotransferase increased | 1 | Two of 42 tests with elevated values of 52 and 61 U/L (male, aged 26; reference range 6–43 U/L) | 28                  |
| Blood alkaline phosphatase increased | 1 | Male, aged 63, with chronic hepatitis and a liver cyst (elevated values between 126–154 U/L; reference range 35–125 U/L) | 28                  |

#### AEs of special interest

| Incorrect dose | 10 | Small deviations from the correct dose (seven patients)                      | –                   |
| Overdose      | 1  | Approximately 2.3 times overdose (80 IU/kg instead of 35 IU/kg), no adverse clinical effect (male, aged 8°) | 29                  |
| Product preparation error | 1 | Reconstitution with saline (not sterile water; female, aged 14)          | 32                  |

**Abbreviations:** N, number of AEs; rFXIII-A2, recombinant factor XIII-A2.

*Two different AEs in the same patient.

*There were two different AEs in this patient ("arthralgia" and "overdose"); the AE "overdose" was considered a possible/probable rFXIII-A2 treatment-related AE and also an AE of special interest. Patient age refers to age at baseline of the mentor™ trial.
One traumatic bleed was successfully treated with rFXIII-A₂: the patient suffered a trauma-induced muscular bleed 24 days after the last rFXIII-A₂ dose (►Table 5) and a single rFXIII-A₂ dose of 35 IU/kg was administered. The haemostatic response was rated as excellent. No additional FXIII-containing products were necessary to treat the bleed.

**Pharmacokinetic Analysis**

Steady-state PK data with rFXIII-A₂ have been previously reported.²² In the current mentor™2 trial, evaluation of all available pre-dose samples from all 60 patients revealed a geometric mean FXIII trough level of 0.17 IU/mL (coefficient of variation: 0.37). In addition, the geometric mean terminal half-life of rFXIII-A₂ was 13.6 (range: 10.1–24.6) days, in line with previously reported PK results for rFXIII-A₂.²² FXIII trough levels below 0.10 IU/mL were detected sporadically throughout the trial: among a total of 2,245 trough level measurements, only 65 (2.9%) were less than 0.10 IU/mL. In all patients, individual mean FXIII trough levels were greater than 0.10 IU/mL.

**Surgery**

Twelve minor surgical procedures were performed in nine patients: post-traumatic surgical repair in one patient, and 11 other minor (mostly dental) procedures in eight patients (►Table 6). Median (range) patient age at the time of surgery was 31 (19–59) years. Eight of the 12 procedures were performed within 7 days of the patient’s last scheduled rFXIII-A₂ dose, and four were performed 10 to 21 days after the last dose. No additional substitution with pdFXIII or rFXIII-A₂ was given for any procedure. Concomitant antifibrinolytics were used in four procedures (tooth extractions [n = 3] and colonoscopy [n = 1]). In three patients, haemoglobin levels were available before (9.9, 11.8 and 15.8 g/dL, respectively) and immediately after surgery (10.3, 11.6 and 16.5 g/dL, respectively), demonstrating that haemoglobin did not decline in association with surgery. The longest period between a rFXIII-A₂ dose and minor surgery was 3 weeks; based on PK modelling, it was estimated that the patient would have had a FXIII activity level of 0.19 IU/mL (►Table 6). There were no reports of unexpected blood loss, requirements for transfusion with blood or blood products and no surgical complications related to FXIII deficiency or treatment for any of these 12 procedures.

**Discussion**

This paper presents the long-term safety and efficacy results from the mentor™2 extension trial—the largest dataset of any clinical trial in FXIII deficiency involving 186.5 patient-years’ worth of data. This is also the first report on minor surgical procedures conducted in patients receiving rFXIII-A₂ prophylaxis.

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**Table 4** Bleeding episodes requiring treatment with FXIII

|                          | Total |
|--------------------------|-------|
| Number of patients       | 60    |
| Number of patients with bleeds | 7     |
| All bleeds               |       |
| Number of bleeds         | 8     |
| Mean ABR*                | 0.043 |
| Spontaneous bleeds       |       |
| Number of bleeds         | 2     |
| Mean ABR*                | 0.011 |
| Traumatic bleeds         |       |
| Number of bleeds         | 6     |
| Mean ABR*                | 0.032 |

Abbreviations: ABR, annualized bleeding rate; FXIII, factor XIII. *Calculated as the number of bleeds divided by the total patient-years (Poisson estimate).

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**Table 5** Details of bleeding episodes requiring treatment with FXIII, arranged in ascending age of patients at time of bleed

| Age | Gender | Bleed type | Days since last dose | Description                                                                                   | Treatment       |
|-----|--------|------------|----------------------|-----------------------------------------------------------------------------------------------|-----------------|
| 8²  | Female | Traumatic  | 7                    | Bruising on arm/soft tissue                                                                  | pdFXIII         |
| 10² | Male   | Spontaneous| 19                   | Spontaneous epistaxis (possibly after picking her nose)                                      | pdFXIII         |
| 12² | Male   | Traumatic  | 17                   | Injury (bruises) to left knee while playing soccer                                            | pdFXIII         |
| 16² | Male   | Traumatic  | 24                   | Forehead laceration due to fall on a staircase                                               | pdFXIII         |
| 23² | Female | Traumatic  | 12                   | Bruises to hip, lumbar spine, sternum and head after motor vehicle accident                  | rFXIII-A₂       |
| 25² | Male   | Spontaneous| 20                   | Muscular bleed in left shoulder after “massive physical work”                               | pdFXIII         |
| 29² | Male   | Traumatic  | 4                    | Subarachnoid subdural and extradural haemorrhage after motor vehicle accident                | Cryoprecipitate |

Abbreviations: FXIII, factor XIII; pdFXIII, plasma-derived FXIII. ²Age at baseline of the mentor™2 trial. ²²The same patient.
Inhibitor (neutralizing antibody) development is the most serious threat associated with the treatment of FXIII deficiency. This has been reported in several patients treated with pdFXIII-containing products. In the current mentor trial as well as in the entire mentor trial programme, no patients have developed inhibitors and in the entire mentor trial programme only five individuals, all younger than 18 years, have developed transient, non-neutralizing antibodies to rFXIII-A2 [reviewed in Carcao et al]. These antibodies have been shown to have no inhibitory activity and the individuals did not experience any AEs or bleeding as a result of or in association with these antibodies. It should be noted that no additional patients developed non-neutralizing antibodies in this current mentor trial.

At no time would any patient experience a FXIII level in the severe (<0.05 IU/mL) deficiency range.

No patients withdrew due to lack of efficacy of rFXIII-A2 treatment. The spontaneous ABR reported in mentor was comparable to that reported for the marketed pdFXIII product. The treatment of breakthrough bleeds with rFXIII-A2 was first introduced in 2012 as a protocol amendment during the mentor trial. Given the rarity of bleeds in the mentor trial, only one bleed was treated with rFXIII-A2; the haemostatic outcome was excellent. Similarly, a recent case study also reported the rapid and successful treatment of a bleed (major intramuscular bleed) with 35 IU/kg rFXIII-A2 in a patient who was not on the mentor trial. Further case reports would provide valuable information on the effectiveness of rFXIII-A2 for the treatment of breakthrough bleeds.

Efficacy

In the mentor trial, rFXIII-A2 prophylaxis (35 IU/kg every 28 ± 2 days) resulted in high FXIII trough levels in all patients and consequently excellent haemostatic coverage. This was evident in the absence of spontaneous intracranial, internal organ or severe gastrointestinal bleeds during the mentor trial encompassing 186.5 patient-years. Furthermore, there were only two spontaneous bleeds over the 6-year trial duration. It should be noted that on this prophylactic regimen (35 IU/mL every 28 ± 2 days), patients would be expected to have FXIII levels in the range of normal (0.5 IU/mL) for 1 week following an infusion; after this, their FXIII levels would be expected to be in the mild deficiency range (0.3–0.5 IU/mL) for another week before falling into the moderate deficiency range (0.05–0.3 IU/mL).

Surgery

To date, the only published data on surgery in FXIII deficiency derive from a report of six patients receiving pdFXIII to cover major or minor procedures. As elective surgery was permitted during mentor, this trial provided the first data on minor surgical procedures performed in patients receiving rFXIII-A2 prophylaxis. All 12 minor procedures (1 post-traumatic surgical repair and 11 other procedures) were performed within 1 to 21 days of the last scheduled rFXIII-A2 dose and additional FXIII substitution was not administered for any of these procedures. In a few of these, anti-fibrinolytic therapy was also given. These findings suggest that prophylaxis with 35 IU/kg rFXIII-A2 every 4 weeks provides sufficient haemostatic coverage for the perioperative management of minor surgical procedures in patients with congenital FXIII deficiency, without the need for
additional FXIII treatment. While additional FXIII treatment was not deemed necessary by the investigators for surgeries in this study, all FXIII treatment options should still be considered on an individual patient basis.

Of the worldwide number of known patients diagnosed with congenital FXIII deficiency, 4.5% participated in the mentor™2 trial. Despite that, a potential limitation was the low number of children/adolescents (n = 16) participating in the mentor™2 trial; however, the current mentor™2 results support those from the mentor™5 trial, which showed efficacy and safety in six paediatric patients aged 1 to 4 years, in which rFXIII-A2 prophylaxis was well tolerated and provided appropriate haemostatic coverage; the ABR was 0.18.

In conclusion, results from the multicentre, multinational mentor™2 extension trial showed that rFXIII-A2 prophylaxis every 4 weeks is well tolerated without identified safety issues; bleeding rates are very low during prophylaxis among patients of all ages with congenital FXIII A-subunit deficiency. None of the patients receiving rFXIII-A2 as part of the mentor™2 study developed either non-neutralizing or neutralizing antibodies, or experienced spontaneous intracranial haemorrhages, severe bleeds, thromboembolic events, anaphylactic or allergic reactions, exposure to pathogen transmission or any significant changes in haematology or clinical chemistry. In addition, while limited to 12 procedures in nine patients, initial data from mentor™2 for minor surgeries performed in patients receiving rFXIII-A2 prophylaxis are favourable, and add to the very limited body of evidence currently available on surgery in FXIII-deficient patients.21

What is known about this topic?

- Congenital FXIII A-subunit deficiency is a rare, autosomal recessive coagulation disorder with a high risk of life-threatening bleeding.
- Prophylaxis is the preferred treatment option for patients with FXIII A-subunit deficiency.
- rFXIII-A2 is the only recombinant FXIII A2-subunit product available to prevent bleeds in patients with congenital FXIII A-subunit deficiency.

What does this paper add?

- The mentor™2 trial represents the largest clinical trial dataset in congenital FXIII A-subunit deficiency, involving 60 unique patients with the disorder.
- rFXIII-A2 35 IU/kg was given once every 4 weeks (for a total of 186.5 cumulative patient-years) and was well tolerated without hypersensitivity reactions, thromboembolic events or anti-FXIII antibody development.
- rFXIII-A2 provided excellent long-term haemostatic efficacy with a geometric mean FXIII trough of 0.17 IU/mL, with patients demonstrating a spontaneous ABR that corresponded to one bleed per 91 patient-years. rFXIII-A2 prophylaxis allowed for effective haemostasis in 12 minor surgical procedures in nine patients without the need for additional FXIII treatment. This is the first report on the effectiveness of rFXIII-A2 in the surgical setting.

Financial Support

This trial was sponsored by Novo Nordisk A/S (Bagsværd, Denmark).

Author Contributions

M.-L. Garly and A. Rosholm: representing Novo Nordisk A/S study concept and protocol. A. Rosholm: statistical analyses. M. Carcao, C. Altisent, G. Castaman, K. Fukutake, B. Kerlin, C. Kessler, R. Lassila, D. Nugent, J. Oldenburg and A. Inbal: data acquisition. M. Carcao, C. Altisent, G. Castaman, K. Fukutake, B. Kerlin, C. Kessler, R. Lassila, D. Nugent, J. Oldenburg, M.-L. Garly, A. Rosholm and A. Inbal: analysis and interpretation of data. M. Carcao: manuscript drafting. M. Carcao, C. Altisent, G. Castaman, K. Fukutake, B. Kerlin, C. Kessler, R. Lassila, D. Nugent, J. Oldenburg, M.-L. Garly, A. Rosholm and A. Inbal: critical revision and final draft of manuscript.

Conflicts of Interest

M. Carcao has received research funding from Baxalta (now a part of Shire), Bayer HealthCare, Biogen, Novo Nordisk and Pfizer; additionally, he has received honoraria for advisory board participation and for speaking from Baxalta (now a part of Shire), Bayer HealthCare, Biogen, Biotest, CSL Behring, Novo Nordisk, Octapharma and Pfizer. C. Altisent has received honoraria or consultation fees from Baxalta (now a part of Shire), Bayer HealthCare, CSL Behring, Grifols, NovoNordisk, Octapharma, Pfizer and Sobi. G. Castaman has received honoraria for advisory board participation and for speaking from Baxalta (now a part of Shire), Bayer HealthCare, Biogen, CSL Behring, Novo Nordisk, Pfizer and Sobi, and has received research support from CSL Behring and Pfizer. K. Fukutake has received research grants from Baxalta (now a part of Shire), Bayer HealthCare, Biogen, CSL Behring, Japan Blood Products Organization, Kaketsuken, Novo Nordisk, Ortho Clinical Diagnostics and Pfizer, as well as honoraria and personal fees for participating in educational events, advisory board and publications from Abbott, Baxalta (now a part of Shire), Bayer HealthCare, Biogen, CSL Behring, Novo Nordisk, Pfizer and Roche. C. Kessler has received grants from Baxalta (now a part of Shire), Bayer HealthCare, Biogen, CSL Behring, Fujirebio Inc., Kaketsuken, LSI Medience, Novo Nordisk, Pfizer, Roche Diagnostics, Sekisui Medical, Siemens, SRL Inc. and Torii Pharmaceuticals outside the submitted work. B. Kerlin has served as an advisory board member for Bayer Healthcare US and Baxalta (now part of Shire), and has received research support from Novo Nordisk A/S and the CSL Behring foundation. C. Kessler has received honoraria or personal fees for participating in educational events and advisory boards from Baxalta (now a part of Shire), Bayer HealthCare, Biogen, Novo Nordisk, Octapharma, Pfizer and Roche. In addition, he has received honoraria and personal fees for participating in educational events and advisory boards from Baxalta (now a part of Shire), Bayer HealthCare, Biogen, CSL Behring, Grifols, Novo Nordisk, Octapharma, Pfizer and Roche. D. Nugent has received honoraria for advisory board participation from Baxalta (now a part of Shire), CSL Behring, Novo Nordisk, Octapharma and Pfizer. J. Oldenburg has no conflicts of interest to declare.
received reimbursement for attending symposia/congresses, and/or honoraria for speaking, and/or honoraria for consulting, and/or funds for research from Baxalta (now a part of Shire), Bayer HealthCare, Biogen Idec, Biotest, Chugai, CSL Behring, Grifols, Novo Nordisk, Octapharma, Pfizer, Roche and Swedish Orphan Biovitrum. M.-L. Garly is an employee of Novo Nordisk. A. Rosholm is an employee of Novo Nordisk and holds shares in Novo Nordisk. A. Inbal has no conflicts of interest to declare.

Acknowledgements
This trial was sponsored by Novo Nordisk A/S (Bagsværd, Denmark). The authors thank the patients and their families/caregivers and the investigators, pharmacists, nurses and trial staff at each centre for participating in the trial.

The authors directed the data analysis and the development of the manuscript. The sponsor was responsible for trial operations, including data analysis. The Principal Investigator (Dr. Manuel Carcao) assumes full responsibility for the accuracy and completeness of the reported data and for the conformity of the manuscript to the trial protocol and the final trial report.

The authors would also like to thank Frank Dziessler (Novo Nordisk Health Care AG) for his scientific advice. Medical writing support was provided by Jo Fetterman, PhD (PAREXEL UK). This work was funded by Novo Nordisk A/S (Bagsværd, Denmark).

References
1 Chung SI, Lewis MS, Folk JE. Relationships of the catalytic properties of human plasma and platelet transglutaminases (activated blood coagulation factor XIII) to their subunit structures. J Biol Chem 1974;249(03):940–950
2 Schwartz ML, Pizzo SV, Hill RI, McKee PA. Human factor XIII from plasma and platelets. Molecular weights, subunit structures, proteolytic activation, and cross-linking of fibrinogen and fibrin. J Biol Chem 1973;248(04):1395–1407
3 Muszbek L, Katona É. Diagnosis and Management of Congenital and Acquired FXIII Deficiencies. Semin Thromb Hemost 2016;42(04):429–439
4 Peyvandi F, Palla R, Menegatti M, et al; European Network of Rare Bleeding Disorders Group. Coagulation factor activity and clinical bleeding severity in rare bleeding disorders: results from the European Network of Rare Bleeding Disorders. J Thromb Haemost 2012;10(04):615–624
5 Anwar R, Minford A, Gallivan L, Trinh CH, Markham AF. Delayed umbilical bleeding—a presenting feature for factor XIII deficiency: clinical features, genetics, and management. Pediatrics 2002;109(02):E32
6 Dorgalah A, Naderi M, Shamsizadeh M. Morbidity and mortality in a large number of Iranian patients with severe congenital factor XIII deficiency. Ann Hematol 2016;95(03):451–455
7 Janbain M, Nugent DJ, Powell JS, St-Louis J, Frame VB, Leissinger CA. Use of factor XIII (FXIII) concentrate in patients with congenital FXIII deficiency undergoing surgical procedures. Transfusion 2015;55(01):45–50
8 Ivaskevicius V, Seitz R, Kohler HP, et al; Study Group. International registry on factor XIII deficiency: a basis formed mostly on European data. Thromb Haemost 2007;97(06):914–921
9 Hsieh L, Nugent D. Factor XIII deficiency. Haemophilia 2008;14(06):1190–1200
10 European Medicines Agency EMA. 2011. Guideline on plasma-derived medicinal products. Available at: http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2011/07/WC500109627.pdf. Accessed October 11, 2016
11 European Medicines Agency EMA. 2012. European Summary of Product Characteristics. Available at: http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_-_Product_Information/human/002284/WC500132997.pdf. Accessed December 6, 2016
12 Dorey E. First recombinant factor XIII approved. Nat Biotechnol 2014;32(03):210
13 Kohler HP, Ichinose A, Seitz R, Ariens RA, Muszbek L; Factor XIII and Fibrinogen SSC Subcommittee of the ISTH. Diagnosis and classification of factor XIII deficiencies. J Thromb Haemost 2011;9(07):1404–1406
14 Lovejoy AE, Reynolds TC, Visich JE, et al. Safety and pharmacokinetics of recombinant factor XIII-A2 administration in patients with congenital factor XIII deficiency. Blood 2006;108(01):57–62
15 Mumford AD, Ackroyd S, Alkhan R, et al; BCSH Committee. Guideline for the diagnosis and management of the rare coagulation disorders: a United Kingdom Haemophilia Centre Doctors’ Organization guideline on behalf of the British Committee for Standards in Haematology. Br J Haematol 2014;167(03):304–326
16 Keeling D, Tait C, Makris M. Guideline on the selection and use of therapeutic products to treat haemophilia and other hereditary bleeding disorders. A United Kingdom Haemophilia Center Doctors’ Organisation (UKHCDO) guideline approved by the British Committee for Standards in Haematology. Haemophilia 2008;14(04):671–684
17 Inbal A, Oldenburg J, Carcao M, Rosholm A, Tehranchi R, Nugent D. Recombinant factor XIII: a safe and novel treatment for congenital factor XIII deficiency. Blood 2012;119(22):5111–5117
18 Kerlin BA, Inbal A, Will A, et al. Recombinant factor XIII prophylaxis is safe and effective in young children with congenital factor XIII-A deficiency: international phase 3b trial results. J Thromb Haemost 2017;15(08):1601–1606
19 Williams M, Will A, Stenno C, Rosholm A, Tehranchi R. Pharmacokinetics of recombinant factor XIII in young children with congenital FXIIII deficiency and comparison with older patients. Haemophilia 2014;20(01):99–105
20 Brand-Staufer B, Carcao M, Kerlin BA, et al. Pharmacokinetic characterization of recombinant factor XIII (FXIII)-A2 across age groups in patients with FXIII A-subunit congenital deficiency. Haemophilia 2015;21(03):380–385
21 Carcao M, Fukutake K, Inbal A, et al. Developing the first recombinant factor XIII for congenital factor XIII deficiency: clinical challenges and successes. Semin Thromb Hemost 2017;43(01):59–68
22 Kerlin B, Brand B, Inbal A, et al. Pharmacokinetics of recombinant factor XIII at steady state in patients with congenital factor XIII A-subunit deficiency. J Thromb Haemost 2014;12(12):2038–2043
23 Food and Drug Administration FDA. 2016. Electronic Code of Federal Regulations 312.120. Available at: http://www.ecfr.gov/cgi-bin/text-idx?SID¼3ee286332416f26a91d9e6d786a604ab&mc=n=ecfrbrowse/Title21/21tab_02.pl. Accessed February 2, 2016
24 International Conference Harmonisation ICH. 2016. Tripartite Harmonised Guideline: Good Clinical Practice, Consolidated Guideline (E6). Available at: http://www.ich.org/fileadmin/Public_Web_Site/ICh_Products/Guidelines/Efficacy/E6/E6_R1_Guideline.pdf. Accessed February 2, 2016
25 World Medical Association WMA. 2016. Declaration of Helsinki—Ethical Principles for Medical Research Involving Human Subjects. Available at: https://www.wma.net/policies-post/wma-declaration-of-helsinki-ethical-principles-for-medical-research-involving-human-subjects/. Accessed February 2, 2016
26 Solomon C, Korte W, Fries D, et al. Safety of factor XIII concentrate: analysis of more than 20 years of pharmacovigilance data. Transfus Med Hemother 2016;43(05):365–373
27 Lusher J, Pipe SW, Alexander S, Nugent D. Prophylactic therapy with Fibrogammin P is associated with a decreased incidence of bleeding episodes: a retrospective study. Haemophilia 2010;16(02):316–321
28 Ashley C, Chang E, Davis J, Mangione A, Frame V, Nugent DJ. Efficacy and safety of prophylactic treatment with plasma-derived factor XIII concentrate (human) in patients with congenital factor XIII deficiency. Haemophilia 2015;21(01):102–108
29 Árokszállási A, Kerényi A, Katona É, et al. The use of recombinant factor XIII in a major bleeding episode of a patient with congenital factor XIII deficiency—the first experience. Haemophilia 2015;21(01):e118–e121
30 World Federation of Haemophilia. Report on Annual Global Survey 2014. Available at: http://www1.wfh.org/publications/files/pdf-1627.pdf. Accessed February 2, 2016