Safety and dose-dependency of eptacog beta (activated) in a dose escalation study of non-bleeding congenital haemophilia A or B patients, with or without inhibitors

Ducore, J; Lawrence, J B; Simpson, M; Boggio, L; Bellon, A; Burggraaf, J; Stevens, Jasper; Moerland, M; Frielong, J; Reijers, J

Published in:
Haemophilia

DOI:
10.1111/hae.13357

IMPORTANT NOTE: You are advised to consult the publisher's version (publisher's PDF) if you wish to cite from it. Please check the document version below.

Document Version
Publisher's PDF, also known as Version of record

Publication date:
2017

Link to publication in University of Groningen/UMCG research database

Citation for published version (APA):

Copyright
Other than for strictly personal use, it is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), unless the work is under an open content license (like Creative Commons).

Take-down policy
If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

Downloaded from the University of Groningen/UMCG research database (Pure): http://www.rug.nl/research/portal. For technical reasons the number of authors shown on this cover page is limited to 10 maximum.

Download date: 31-12-2018
Safety and dose-dependency of eptacog beta (activated) in a dose escalation study of non-bleeding congenital haemophilia A or B patients, with or without inhibitors


1University of California, Davis Health System, Sacramento, CA, USA
2LFB USA, Framingham, MA, USA
3Rush University Medical Center, Chicago, IL, USA
4LFB SA, Paris, France
5Centre for Human Drug Research, Leiden, The Netherlands
6Hemophilia & Thrombosis Center, University of Colorado, Aurora, CO, USA

Correspondence
Jeffry B. Lawrence, LFB USA Inc., Framingham, MA, USA.
Email: Jeffry.Lawrence@lfb-usa.com

Introduction: Varying initial doses of activated eptacog beta (recombinant human FVIIa, rhFVIIa) may provide therapeutic options when treating bleeding in patients with congenital haemophilia who have developed inhibitory antibodies to factor VIII (FVIII) or factor IX (FIX). This study evaluated escalated doses of a new rhFVIIa product as a prelude to selecting the doses for clinical efficacy evaluation in haemophilia patients.

Aim: To assess the safety, pharmacokinetics, and laboratory pharmacodynamics of 3 doses of rhFVIIa in non-bleeding patients with congenital haemophilia A or B with or without inhibitors.

Methods: Adult male patients (18-75 years old) with congenital haemophilia A or B (with or without inhibitors) received infusions of rhFVIIa at doses of 25, 75 or 225 μg/kg body weight. Ten patients were treated at each dose level, and each patient received 2 different dose levels. Descriptive methods were used to analyse the data.

Results: Administration of rhFVIIa at all doses was well tolerated. Pharmacokinetic analyses showed that peak FVIIa plasma levels (C_{max}) were approximately proportional to dose and correlated well with peak thrombin generation. Total AUC_{0-inf} also was approximately dose proportional. Clot formation and duration correlated with FVIIa activity. Repeat doses did not produce an immunological response.

Conclusion: In the first dose-escalation study of rhFVIIa to support product registration, eptacog beta at doses of 25, 75, and 225 μg/kg was pharmacodynamically active and well tolerated in non-bleeding patients with congenital haemophilia A or B.

KEYWORDS
bypassing agents, eptacog beta, inhibitors, Phase Ib, recombinant activated factor VII, rhFVIIa

INTRODUCTION

Haemophilia A and B are blood clotting disorders caused by congenital deficiency of factors VIII (FVIII) or IX (FIX) respectively. Bleeding episodes (BEs) in these patients are treated with factor replacement therapy; however, during their lifetime, 20%-30% of patients with haemophilia A, and 5% with haemophilia B, will develop inhibitory antibodies (inhibitors) to FVIII or FIX. In such patients, haemostasis may not be achievable with replacement of the deficient factor (depending upon the inhibitor titre) and thus may require administration of bypassing
agents, such as plasma-derived activated prothrombin complex concentrates (aPCC) or activated recombinant factor VII (rFVIIa).\textsuperscript{2-5}

Primarily out of concern for thrombosis, most clinicians use a titrated, "lowest effective" dose of bypassing agent in acute bleeding episodes.\textsuperscript{6,7} In an early review of rFVIIa dosing, Abshire and Kenet\textsuperscript{8} proposed that optimization of the initial thrombin burst in patients at low thrombotic risk may lead to earlier and possibly more durable haemostasis. Optimal dose finding is further complicated by the absence of validated pharmacodynamic markers to predict clinical efficacy. Retrospective rFVIIa utilization data in the congenital haemophilia inhibitor population indicate the risk of thrombin generation at sites where pathologic thrombosis is triggered, rather than physiologic haemostasis, is related to the presence of pre-existing risk factors for thrombosis.\textsuperscript{9,10} To date, clinical evidence suggestive of dose-dependent efficacy has been generated, but this has been difficult to confirm or reproduce for reasons that are not well understood.\textsuperscript{11-15}

Historically, various dosing regimens have been employed attempting improvements in overall rFVIIa haemostatic efficacy and dosing efficiency. In on-demand settings, there is weak evidence that the AUC of a bypassing agent is the primary correlate for the achievement of haemostasis, though the effect of sustained thrombin generation is thought to improve healing and may delay clot dissolution.\textsuperscript{16,17} Not surprisingly, continuous infusion in the setting of surgical interventions (with ongoing wound manipulation) has been shown to be efficacious, thus confirming that a steady concentration of agent may have benefit in certain clinical circumstances. In contrast, animal experiments and some prospective human studies make clear that the peak plasma concentration ($C_{\text{max}}$) of administered FVIIa is correlated with thrombin generation, clot strength and faster onset of haemostasis.\textsuperscript{18-22} Whether incremental dosing could delay otherwise achievable early haemostasis is unclear.

Within this clinical context, activated eptacog beta (rhFVIIa), a new recombinant human coagulation FVIIa molecule produced by LFB USA using Pro\textsuperscript{\textregistered} Technology,\textsuperscript{2} is being developed as a haemostatic bypassing agent for treating bleeding episodes in patients with haemophilia A or B who develop inhibitors. A dose-escalation study was performed to assess the safety, pharmacokinetic profile (PK), and laboratory pharmacodynamic markers (PD) of rhFVIIa activity in non-bleeding subjects with haemophilia A or B, with or without inhibitors. Computationally modelled pharmacokinetic behaviour supported the selection of doses for Phase 3 clinical trials of rhFVIIa for the treatment of bleeding episodes in patients with inhibitors.\textsuperscript{23,24}

The primary sequence and the degree of gamma carboxylation of eptacog beta and eptacog alfa are identical. The ex vivo molar potency of eptacog beta is greater, and differences in glycosylation and binding to cellular sites such as Endothelial Protein C Receptor and platelets have been observed.

2 | MATERIALS AND METHODS

This study received approval by institutional review boards, and was conducted in compliance with established good clinical practice as stated in the current Declaration of Helsinki.\textsuperscript{25} Written informed consent was obtained from all subjects at the time of their enrollment. The study is registered at clinicaltrials.gov (NCT01708564).

2.1 | Study design

Three doses of rhFVIIa (25, 75 and 225 µg/kg) were examined using a Phase Ib dose-escalation study design. Doses were preselected based on preclinical in vitro and in vivo studies of activity [LFB SA Data on File]. The first 10 subjects received the lowest dose of rhFVIIa. Following review of the 30 ± 6 hour safety and PD coagulation data by an independent, external Data Monitoring Committee (DMC), 5 of these subjects were randomly selected and 5 new subjects received the second treatment dose. Following a similar safety review by the DMC, the 5 remaining subjects treated at dose level 1, and the 5 newly treated subjects from dose level 2 were treated with the highest dose. Open label study drug was administered intravenously over 2-3 minutes; a single lot of drug product was used throughout the study. Two centres in the US and 1 in The Netherlands enrolled subjects Figure 1.

2.2 | Patients

Male subjects, aged 18-75 years, with moderate or severe congenital haemophilia A or B (FVIII or FIX levels <5% of normal) were eligible to enrol. Subjects with prothrombotic risk factors were excluded from the study. Key inclusion and exclusion criteria are listed in Table 1.

2.3 | Pharmacokinetic, pharmacodynamic and safety evaluations

PK, PD and safety assessments were obtained not more than 30 minutes prior to dosing (baseline); at 5, 15 and 30 minutes; and at 1, 2, 5, and 15 minutes post-dose. A composite endpoint was defined as the mean peak concentration ($C_{\text{max}}$) of administered FVIIa was correlated with thrombin generation, clot strength and faster onset of haemostasis. Whether incremental dosing could delay otherwise achievable early haemostasis is unclear.

Within this clinical context, activated eptacog beta (rhFVIIa), a new recombinant human coagulation FVIIa molecule produced by LFB USA using Pro\textsuperscript{\textregistered} Technology,\textsuperscript{2} is being developed as a haemostatic bypassing agent for treating bleeding episodes in patients with haemophilia A or B who develop inhibitors. A dose-escalation study was performed to assess the safety, pharmacokinetic profile (PK), and laboratory pharmacodynamic markers (PD) of rhFVIIa activity in non-bleeding subjects with haemophilia A or B, with or without inhibitors. Computationally modelled pharmacokinetic behaviour supported the selection of doses for Phase 3 clinical trials of rhFVIIa for the treatment of bleeding episodes in patients with inhibitors.\textsuperscript{23,24}

The primary sequence and the degree of gamma carboxylation of eptacog beta and eptacog alfa are identical. The ex vivo molar potency of eptacog beta is greater, and differences in glycosylation and binding to cellular sites such as Endothelial Protein C Receptor and platelets have been observed.

2 | MATERIALS AND METHODS

This study received approval by institutional review boards, and was conducted in compliance with established good clinical practice as stated in the current Declaration of Helsinki.\textsuperscript{25} Written informed consent was obtained from all subjects at the time of their enrollment. The study is registered at clinicaltrials.gov (NCT01708564).

2.1 | Study design

Three doses of rhFVIIa (25, 75 and 225 µg/kg) were examined using a Phase Ib dose-escalation study design. Doses were preselected based on preclinical in vitro and in vivo studies of activity [LFB SA Data on File]. The first 10 subjects received the lowest dose of rhFVIIa. Following review of the 30 ± 6 hour safety and PD coagulation data by an independent, external Data Monitoring Committee (DMC), 5 of these subjects were randomly selected and 5 new subjects received the second treatment dose. Following a similar safety review by the DMC, the 5 remaining subjects treated at dose level 1, and the 5 newly treated subjects from dose level 2 were treated with the highest dose. Open label study drug was administered intravenously over 2-3 minutes; a single lot of drug product was used throughout the study. Two centres in the US and 1 in The Netherlands enrolled subjects Figure 1.

2.2 | Patients

Male subjects, aged 18-75 years, with moderate or severe congenital haemophilia A or B (FVIII or FIX levels <5% of normal) were eligible to enrol. Subjects with prothrombotic risk factors were excluded from the study. Key inclusion and exclusion criteria are listed in Table 1.

2.3 | Pharmacokinetic, pharmacodynamic and safety evaluations

PK, PD and safety assessments were obtained not more than 30 minutes prior to dosing (baseline); at 5, 15 and 30 minutes; and at 1, 2,
pharmacodynamic parameters were determined using a non-compartmental PK analysis. For each PD variable, descriptive statistics were analyzed by time point and dose group for actual values and change from baseline. Baseline-corrected levels were reported as indicated. The 24-hour safety evaluation time point was not used in the PK analysis. Due to the study design, significance testing was not performed.

3 | RESULTS

3.1 | Subject population

Eighteen subjects were screened: 15 of these satisfied the inclusion and exclusion criteria and received both assigned doses of rhFVIIa. 14 of these subjects completed all follow-up evaluations, with 1 subject being lost to both the 14 and 28-day safety assessments following the second dose. Demographics are shown in Table 2.

3.2 | Pharmacokinetics

Table 3 summarizes the baseline-corrected PK parameters for all 3 doses. Both the C_max and the AUC were dose dependent across all
TABLE 2  Demographics of subjects receiving study medication

<table>
<thead>
<tr>
<th>Parameter, N = 15</th>
<th>Value (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age, y</strong></td>
<td></td>
</tr>
<tr>
<td>Mean [SD]</td>
<td>33.0 [10.8]</td>
</tr>
<tr>
<td>Range</td>
<td>20/61</td>
</tr>
<tr>
<td><strong>Race/ethnicity, n</strong></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>10 (66.7)</td>
</tr>
<tr>
<td>Asian</td>
<td>1 (6.7)</td>
</tr>
<tr>
<td>Hispanic/Latino</td>
<td>2 (13.3)</td>
</tr>
<tr>
<td>Indian</td>
<td>1 (6.7)</td>
</tr>
<tr>
<td>Moroccan</td>
<td>1 (6.7)</td>
</tr>
<tr>
<td><strong>Haemophilia type</strong></td>
<td></td>
</tr>
<tr>
<td>Haemophilia A</td>
<td>11 (73.3)</td>
</tr>
<tr>
<td>Haemophilia B</td>
<td>4 (26.7)</td>
</tr>
<tr>
<td><strong>Haemophilia severity</strong></td>
<td></td>
</tr>
<tr>
<td>Severe</td>
<td>12 (80.0)</td>
</tr>
<tr>
<td>Moderate</td>
<td>3 (20.0)</td>
</tr>
<tr>
<td>None</td>
<td>11 (73.3)</td>
</tr>
<tr>
<td>≤0.8 BU</td>
<td>4 (26.7)</td>
</tr>
<tr>
<td>HIV infection</td>
<td>1 (6.7)</td>
</tr>
</tbody>
</table>

TABLE 3  Geometric mean (CV) of non-compartmental PK parameter analyses

<table>
<thead>
<tr>
<th>Parameter, N = 15</th>
<th>25 μg/kg</th>
<th>75 μg/kg</th>
<th>225 μg/kg</th>
</tr>
</thead>
<tbody>
<tr>
<td>CL, L/h</td>
<td>9.0 (42.9)</td>
<td>10.0 (23.5)</td>
<td>7.9 (34.2)</td>
</tr>
<tr>
<td>Vss, L</td>
<td>29.8 (50.3)</td>
<td>30.4 (32.2)</td>
<td>20.0 (40.2)</td>
</tr>
<tr>
<td>Cmax, ng/mL</td>
<td>230 (42.8)</td>
<td>717 (32.2)</td>
<td>1870 (36.6)</td>
</tr>
<tr>
<td>AUC0-inf, ng·h/mL</td>
<td>212 (41.1)</td>
<td>617 (28.8)</td>
<td>2240 (26.2)</td>
</tr>
<tr>
<td>t1/2, h</td>
<td>2.3 (29.6)</td>
<td>2.1 (23.4)</td>
<td>1.8 (14.1)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Parameter, N = 15</th>
<th>25 μg/kg</th>
<th>75 μg/kg</th>
<th>225 μg/kg</th>
</tr>
</thead>
<tbody>
<tr>
<td>CL, L/h</td>
<td>9.0 (42.9)</td>
<td>10.0 (23.5)</td>
<td>7.9 (34.2)</td>
</tr>
<tr>
<td>Vss, L</td>
<td>29.8 (50.3)</td>
<td>30.4 (32.2)</td>
<td>20.0 (40.2)</td>
</tr>
<tr>
<td>Cmax, ng/mL</td>
<td>230 (42.8)</td>
<td>717 (32.2)</td>
<td>1870 (36.6)</td>
</tr>
<tr>
<td>AUC0-inf, ng·h/mL</td>
<td>212 (41.1)</td>
<td>617 (28.8)</td>
<td>2240 (26.2)</td>
</tr>
<tr>
<td>t1/2, h</td>
<td>2.3 (29.6)</td>
<td>2.1 (23.4)</td>
<td>1.8 (14.1)</td>
</tr>
</tbody>
</table>

CL, apparent clearance; Vss, apparent volume of distribution at steady state; Cmax, maximum observed concentration; AUC0-inf, area under the plasma-concentration time curve, time of dosing to infinity; t1/2, terminal half-life. For all PK parameters, baseline FVIIa levels were subtracted from the measured FVIIa values.

dosing levels; with linear dose proportionality being observed between the 25 and 75 μg/kg arms. Clearance, volume of distribution and terminal half-life remained consistent irrespective of dose. With all 3 doses, the measured FVIIa activity decreased to baseline levels by 24 hours (Figure 2).

While there appears to be a trend toward a reduction in half-life with increasing dose, the large coefficients of variation make this statistically unlikely. The kinetics of rhFVIIa are linear in haemophilia patients [LFB SA Data on File].

### 3.3  Pharmacodynamics

The effects of binding of rhFVIIa to platelets is more prominent than those of tissue factor binding. Thus, a platelet-spiked TGA assay (TGAp) is more representative of its clinical mode of action than the platelet-free assay. Plasma samples were obtained at prespecified times after rhFVIIa administration. Thrombin generation was measured and mean thrombin generation peaks were calculated. Dose-dependent effects were seen in the peak thrombin generation when measured from 5 minutes to 24 hours (Figure 3 and Table 4). The platelet-free TGA showed significant variability, and the results are not reported here.

Baseline aPTT uniformly exceeded the upper limit of the normal range (23 to 34 seconds) (Table 4). The aPTT following treatment with rhFVIIa was dose-dependent, with the 225 μg/kg dose producing the greatest effect: 5 minutes postinfusion, the mean aPTT was reduced by 6.9, 11.5 and 22.2 seconds for the 25, 75, and 225 μg/kg dose groups respectively. The aPTT returned to baseline values after 1 hour following the 25 μg/kg dose and by 4-12 hours following the higher doses.

Reduction in prothrombin time was observed at all doses, but greatest in the 75 and 225 μg/kg dose groups (Table 4). At baseline, the mean PTs were comparable for all groups and were within the normal range (13.1-15.7 seconds). Full evidence of dose dependency was obscured, as the lower limit of the assay (approximately 8 seconds) was reached with both the 75 and 225 μg/kg doses. The PT returned to baseline by 12 hours following the 25 μg/kg dose, and by 24 hours following the higher doses.

### 3.4  ROTEM (modified)

Clot firmness is a measure of the absolute strength of the fibrin and platelet clot and in this study, the change from baseline also exhibited a dose-dependent relationship (Figure 4). Maximum clot firmness (MCF) was observed 5 minutes postinfusion, with the change from baseline being 5.9 ± 3.7, 7.0 ± 2.8, and 9.7 ± 3.3 mm for the 25, 75, and 225 μg/kg dose groups, respectively. The aPTT returned to baseline values after 1 hour following the 25 μg/kg dose and by 4-12 hours following the higher doses.

### 3.5  Measures of coagulation activation

Baseline D-dimer levels were variable among subjects; however, individual subject levels remained largely unchanged throughout the evaluation period, as did the mean levels. One subject had elevated levels at the 24 hours time point on the 225 μg/kg dose, being 2-fold higher than baseline, and occurring 24 hours postinfusion.

Prothrombin fragment F1+2 showed a dose-dependent increase with a peak 1 to 2 hours postinfusion, indicating the formation of thrombin following rhFVIIa administration. Levels returned to baseline between 6 and 12 hours postinfusion. When the change from baseline value was calculated, a low level dose-response was observed: the median change from baseline at 2 hours was 33, 94 and 164 pmol/L for the 25, 75, and 225 μg/kg dose groups, respectively. (Table 4)

TAT, a complex between thrombin and antithrombin, is a marker of coagulation activation, and is highly variable among normal individuals. Overall, the 25 μg/kg dose appeared to have no effect on thrombin-antithrombin complex behaviour. TAT levels for the 75 and 225 μg/kg dose groups were variable and inconsistent between patients, with

Overall, the 25 μg/kg dose appeared to have no effect on thrombin-antithrombin complex behaviour. TAT levels for the 75 and 225 μg/kg dose groups were variable and inconsistent between patients, with
some patients showing evidence of minimal thrombin generation immediately following rhFVIIa administration.

3.6 | Immunogenicity

No hypersensitivity or humoral immunogenicity was detected in any subject during primary or re-exposure assessments.

3.7 | Safety

In all 3 cohorts, both doses of rhFVIIa (25/75, 25/225 or 75/225 µg/kg) were well tolerated. Three adverse events that occurred within 36 hours of infusion were possibly related to the drug based upon the temporal relationship between the events and drug infusion. One patient experienced mild dizziness of short duration after receiving doses of 25 and 75 µg/kg rhFVIIa (11 and 3 minutes’ duration respectively). A second patient experienced a mild headache that lasted for 2 hours following administration of 25 µg/kg rhFVIIa. All events resolved without intervention. There were no clinically meaningful excursions in hematologic or serum chemistry measures following administration. There was no evidence of administration-related allergic reaction.

### Table 4

<table>
<thead>
<tr>
<th>Assay</th>
<th>25 µg/kg, (N = 10)</th>
<th>75 µg/kg, (N = 10)</th>
<th>225 µg/kg, (N = 10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>TGAp, nmol/L</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Preinfusion (~30 min)</td>
<td>0.86 (0.39)</td>
<td>0.82 (0.49)</td>
<td>0.68 (0.55)</td>
</tr>
<tr>
<td>Peak thrombin generation postinfusion</td>
<td>11.8 (3.7)</td>
<td>16.7 (4.1)</td>
<td>20.5 (3.5)</td>
</tr>
<tr>
<td>aPTT, s</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Preinfusion (~30 min)</td>
<td>55.4 (9.0)</td>
<td>53.2 (13.3)</td>
<td>57.7 (13.7)</td>
</tr>
<tr>
<td>5 min postinfusion</td>
<td>48.5 (6.3)</td>
<td>41.7 (7.8)</td>
<td>35.5 (3.6)</td>
</tr>
<tr>
<td>Prothrombin time, s</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Preinfusion (~30 min)</td>
<td>14.0 (1.0)</td>
<td>13.6 (0.5)</td>
<td>13.9 (0.8)</td>
</tr>
<tr>
<td>5 min postinfusion</td>
<td>9.0 (0.5)</td>
<td>8.3 (0.5)</td>
<td>8.0 (0.6)</td>
</tr>
<tr>
<td>Maximum clot firmness, mm</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Preinfusion (~30 min)</td>
<td>5.5 (3.5)</td>
<td>5.2 (2.9)</td>
<td>4.4 (2.1)</td>
</tr>
<tr>
<td>5 min postinfusion</td>
<td>11.5 (1.9)</td>
<td>12.2 (2.8)</td>
<td>14.1 (2.3)</td>
</tr>
</tbody>
</table>

Normal range 13.1-15.7 s.

aNormal range 23–34 s.
bThe shortest measurable time in the PT assay was 8 s.
Bypassing agent therapy is a core treatment modality for haemophilia patients who develop inhibitors to FVIII or FIX. This Phase Ib study evaluated a second-generation bypassing agent, recombinant human FVIIa (rhFVIIa; activated eptacog beta),2,4 to guide dosing levels and provide initial safety data for Phase 3 clinical studies. Multiple surrogate pharmacodynamic markers for haemostasis demonstrated dose dependent changes upon treatment with rhFVIIa, a result that was consistent with the dose-dependent PK observed in the same patients. The findings indicate that the highest 2 doses (75 and 225 μg/kg) produced the greatest effect in these assays and were therefore considered suitable for further clinical investigation.

The interaction between the endothelial protein C receptor (EPCR) and FVIIa is known to enhance FVIIa-driven haemostasis in vivo.29 In vitro studies, Grandoni et al5 demonstrated that rhFVIIa binds to EPCR 25%-30% more effectively than the predecessor rFVIIa product, eptacog alfa. In addition, it has been suggested that a second minor binding site may exist for rhFVIIa, but not eptacog alfa.5 Based upon these observations, preclinical models, and the reported clinical experience of eptacog alfa,30 a dose of 25 μg/kg was selected as the minimum dose at which a nominal clinical response might be expected. The higher doses of 75 and 225 μg/kg were based upon the 3-fold dosing range reported for eptacog alfa (all doses were within the 7-fold safety margin established in the determination of MTD in New World primates (LFB SA Data on File)).

The baseline-corrected FVIIa peak plasma activities and AUCs were non-linearly dose proportional, a result comparable to that previously reported for in vitro studies of FVIIa thrombin generation. As rhFVIIa exerts its pharmacological effect in plasma, the PK results were analyzed using a non-compartmental methodology; however, as in vivo endothelial and platelet receptor binding does affect the plasma concentration, the non-compartmental analysis may not accurately reflect levels of bioavailable rhFVIIa.5

When correcting haemophilic coagulopathy with replacement factor, classic laboratory assessment of intrinsic and extrinsic coagulation, along with factor level assays, represent the standard of care for guiding therapy. For bypassing agents, however, no laboratory assay has been validated for monitoring or anticipating clinical coagulation response. Several pharmacodynamic markers of coagulation activation, although not validated as surrogate markers of in vivo haemostasis, did indicate a dose-dependent response after infusion of rhFVIIa. Specifically, for the TGA with platelets, MCF, PT and aPTT, a dose-dependent relationship was observed. Measures of clot formation (TAT, F1+2) and dissolution (D-dimer) were highly variable and subject to confounding secondary to the use of citrate for coagulation inactivation.31 There were substantial inter-patient variations in F1+2, with some patients demonstrating a proportional relationship with study dose. The PT assay demonstrated persistent shortening; however, the initial dose-response relationship was obscured as the lower limits of the assay were reached.

Only 2 subjects in this study had inhibitors to FVIII or FIX. Deficient thrombin generation in patients with haemophilia A or B is common to patients with or without inhibitors30 though the severity of the defect may be greater in the inhibitor population. Variations in FVIIa pharmacologic half-life have only been demonstrated in patients with acquired FVIIa inhibitors; nevertheless, the trends observed in these data provide a meaningful basis for the selection of doses to be evaluated in Phase 3 clinical studies examining on-demand treatment of mild/moderate bleeding in patients with inhibitors.

All doses of rhFVIIa were well tolerated. No consistent elevations of D-dimer or TAT were noted. D-dimer measurement in samples collected in citrate have been shown to vary widely, a result of time to analysis due to Ca++-independent plasmin activity in vitro.31,32 Nevertheless, while D-dimer is non-specific and does not exclude pathologic clot formation, there were no consistent elevations following rhFVIIa.

No hypersensitivity or other clinical events occurred that would indicate an immunological response following primary or repeat

**FIGURE 4** Rotational thromboelastometry (ROTEM) to assess clot firmness level. A dose-dependent effect on clot firmness was observed, with the maximum clot firmness being observed at 5 min.
5 | CONCLUSION

Data from this dose escalation study provide guidance for clinical dose selection for future rhFVIIa studies. The peak plasma concentration of rhFVIIa has relevant effects on platelet augmented thrombin generation, suggesting dose-dependent increases in local thrombin concentration at the site of bleeding. Further, time to clot formation and clot structure, strength and retractive force development are directly related to the peak thrombin generation within the forming clot.\(^\text{18,33-35}\) Thus, \(C_{\text{max}}\) considerations should guide the choice of the initial dose for on-demand treatment of bleeding episodes. In addition, the total AUC determined at different doses confirms the viability of a 3-hour dosing interval for the 75 µg/kg and suggests that a 9-hour dose-dependent interval between the initial 225 µg/kg and subsequent doses are pharmacokinetically supported. This key consideration permits design of an on-demand dosing regimen. Without clinically validated PD markers to predict bypassing agent efficacy in bleeding haemophilia subjects with inhibitors, a subsequent Phase 3 study was initiated (using 75 and 225 µg/kg as initial doses) to validate these results.\(^\text{22,24}\)

Activated eptacog beta (rhFVIIa), a new second-generation rhFVIIa, was well tolerated in haemophilia patients with or without inhibitors, and had dose-dependent PK and PD profiles that support further evaluation of its potential efficacy in studies of bleeding episodes in haemophilia patients with inhibitors.

ACKNOWLEDGEMENTS

The authors wish to thank the study investigators at each study center and the expert steering committee members who guided the study design. N.S. Rudolph helped edit the manuscript and was paid by LFB USA. Mel Carlson, rEVO Biologics, and Ian Mitchell, HEMA Biologics, helped edit the manuscript.

DISCLOSURES

JD, MLS, LB and MW were study investigators and have received compensation from Hema Biologics/LFB/rEVO Biologics. JD has provided consultation or been an investigator for Bayer, Shire, Octapharma, Biogen, CSL, Sparks, OPKO. MLS has been a consultant for Biogen, CSL Behring, Grifols, NovoNordisk, Octapharma, and Shire. LB has served as a consultant or investigator for Biogen, Bayer, Shire, OPKO, NovoNordisk, Grifols and Octapharma. MW has provided consultation for Shire, NovoNordisk, Biogen, CSL Behring. JL (USA), AB (Paris) and JF are/were employees of LFB. JB, MM, JS and JR are employees of CHDR, Leiden, The Netherlands, and were compensated for analytical work supporting this study. JF is no longer employed by LFB Paris. He currently has no competing interests which may be perceived as posing a conflict or bias.

REFERENCES


24. Wang MD. The Global, Multi-Center, Phase III, Randomized, Efficacy, Pharmacokinetic and Safety Cross-Over Study (PERSEPT 1) of Two Dose Regimens of Eptacog Beta (rhFVIIa) in Congenital Haemophilia A and B Patients with Inhibitors. WFH 2016 Presentation; 2016.


How to cite this article: Ducore J, Lawrence JB, Simpson M, et al. Safety and dose-dependency of eptacog beta (activated) in a dose escalation study of non-bleeding congenital haemophilia A or B patients, with or without inhibitors. Haemophilia. 2017;23:848–851. https://doi.org/10.1111/hae.13357