

Recombinant Activated Factor VII (rFVIIa) Acutely Normalizes Prothrombin Time in Patients with Cirrhosis During Bleeding from Oesophageal Varices

E. Ejlsen, T. Melsen, J. Ingerslev, R. Borum Andreasen & H. Vilstrup

Dept. of Medicine V and Centre for Haemophilia and Thrombosis, Aarhus University Hospital, Aarhus, Denmark; Clinical Drug Development, Novo Nordisk A/S, Bagsvaerd, Denmark

Ejlsen E, Melsen T, Ingerslev J, Borum Andreasen R, Vilstrup H. Recombinant activated factor VII (rFVIIa) acutely normalizes prothrombin time in patients with cirrhosis during bleeding from oesophageal varices. *Scand J Gastroenterol* 2001;36:1081–1085.

Background: Patients with cirrhosis have low levels of coagulation factors, the most pronounced deficiency being that of FVII. This may compromise haemostasis during bleeding from ruptured oesophageal varices. The objective of this trial was to evaluate the effect of rFVIIa on prothrombin time in cirrhosis patients with ongoing variceal bleeding. Safety, including signs of DIC, was monitored. **Methods:** The study is a single centre, open-label trial. Ten consecutive patients with known alcoholic cirrhosis and oesophageal variceal bleeding were included. The patients received routine treatment, including Terlipressin. Each patient received one i.v. injection of rFVIIa (80 µg/kg bw). The study observation time was 12 h per patient. **Results:** The mean age of the patients was 48 years (8 men and 2 women). The cirrhosis was classified as Child B in 5 patients and Child C in 5. At baseline, all patients had prothrombin time levels above the normal range, and all but one had FVII coagulation activity (FVII:C) levels below the normal range. rFVIIa normalized the prothrombin time in all patients within 30 min. The effect lasted for more than 4 h in 7 patients, and for about 2 h in the remaining 3 patients. Immediate bleeding control was obtained in all patients, and no patient died within the study time. There was no sign of DIC. **Conclusions:** rFVIIa is effective in transiently reversing the prolonged prothrombin time in cirrhosis patients with haematemesis from varices. This indicates a potential of improving haemostasis and survival in patients with compromised coagulation due to liver disease.

Key words: Activated recombinant factor VII; cirrhosis; haematemesis; Novo Seven; oesophagogastric varices; prothrombin time

Hendrik Vilstrup, M.D., D.Sc., Dept. of Medicine V, Aarhus University Hospital, 44 Noerrebrogade, DK-8000 Aarhus C, Denmark (fax. +45 89 49 28 60, e-mail. vilstrup@aaa.dk)

The liver is the principal site of synthesis and clearance of coagulation factors, the components of the fibrinolytic system, and the naturally occurring anti-coagulants. The most frequently encountered abnormalities in haemostasis in cirrhosis patients include thrombocytopenia, prolonged prothrombin time and hyperfibrinolysis. The progressive loss of liver parenchymal cells associated with cirrhosis results in a decreased synthesis of the vitamin K-dependent coagulation factors: FII, FVII, FIX, FX and protein C (and S) (1).

Cirrhosis patients with prolonged prothrombin time have been shown to have low levels of coagulation factors, the most pronounced deficiency being that of FVII (1). The coagulation abnormalities are similar, irrespective of the type of liver damage, and the extent of the abnormalities depends upon the degree of liver damage and loss of function (1). Cirrhosis patients with low platelet counts and/or prolonged prothrombin time have an increased risk of bleeding from oesophagogastric varices or gastric mucosa. The majority of

patients with cirrhosis develop oesophageal varices, one-third bleed within the first year from the time of diagnosis, and bleeding accounts for one-third of all deaths. The mortality of each bleeding episode is 30%–50% depending on the clinical status of the patient (2). Early control of the bleeding episode is associated with better survival (3), possibly because secondary coagulopathy is prevented. Correspondingly, it may be important to correct coagulopathy during bleeding.

The current treatment of coagulopathy caused by liver cirrhosis in an actively bleeding patient is based on replacement of coagulation factors by administration of fresh frozen plasma. However, it is not documented that plasma improves survival, and the volume necessary to correct the coagulation status may be prohibitively large. Correction of single specific key deficiencies, therefore, is an attractive alternative. FVII may be such an alternative, since lack of this factor is the most pronounced deficiency (cf. above). FVII acts via its activated form, FVIIa, which is not haemostatically active itself. It requires binding to tissue factor present in the deeper layers of

the vessel wall. FVIIa forms complexes with the tissue factor at the site of injury, that is, where the endothelial barrier is injured and the deeper layers are exposed, and induces a local haemostasis by activating FX. This selective mechanism of action minimizes the risk of inducing a generalized activation of the coagulation cascade, and thus of disseminated intravascular coagulation (4). FVIIa, produced by a recombinant technique (rFVIIa) is available, and has been tested with promising results in non-bleeding cirrhosis patients with disturbed coagulation (5).

The objective of the present trial was to evaluate the short-term effect of rFVIIa on prothrombin time in cirrhosis patients actively bleeding from oesophageal varices. Additionally, safety, including signs of DIC, was monitored.

Materials and Methods

Patients

Ten consecutive patients with known alcoholic liver cirrhosis acutely admitted to the Dept. of Medicine V, Aarhus University Hospital, Denmark, were included. The department is a primary and secondary referral centre for patients with liver disease. Patients were eligible for inclusion if they (a) had haematemesis; (b) had a prolonged prothrombin time; (c) had known or presumed liver cirrhosis; (d) had oesophageal variceal bleedings, either known from previous encounters or endoscopically proven at the present episode; (e) were aged 18 years or older; and (f) were able to provide informed consent (either from the patient or from relatives). No patient fulfilled the exclusion criteria, that is, none: (a) had known hypersensitivity to rFVIIa or any of its components; (b) had received treatment with prothrombin-complex concentrates within the last 7 days; (c) had received treatment with 1-desamino-8-D-arginine vasopressin or antifibrinolytics within the last 3 days; (d) had taken other investigational drug(s) within 3 months before screening; (e) were women of child-bearing potential.

Study design

The study was a single-centre, open-label pilot trial—duration 12 h per patient. The primary effect variable was the prothrombin time after administration of the drug. Clinical outcomes during and beyond the study period were recorded (6). Each patient received one intravenous (i.v.) injection of rFVIIa (80 µg/kg bw). Informed consent was obtained from the patient or relatives if the patient was incapable of doing so him/herself. At baseline, the following procedures were performed: physical examination including concomitant medication and illnesses; recording of haemostatic therapy for the last 7 days; laboratory tests (haemoglobin, leukocytes, alanine aminotransferase, alkaline phosphatase, bilirubin, albumin, s-creatinine, potassium and sodium); coagulation status (prothrombin time, aPTT, FVII coagulation activity, FVII:C); safety parameters (platelets, D-dimer, AT-III and F₁₊₂). Other safety data included vital signs and monitoring

of adverse events. Blood samples (coagulation status and safety parameters) were drawn 0.5, 2, 4, 6, 8 and 12 h after dosing of rFVIIa. Laboratory tests were measured 12 h post-dosing. The tourniquet time for drawing of all blood samples was <1 min per draw. The protocol was approved by the Danish Medicines Agency and the Biomedical Ethics Committee for Aarhus County.

Materials

Recombinant FVIIa is identical in structure and activity to human FVII. The risk of contamination by human viruses is eliminated and the preparation contains no foreign proteins. Recombinant FVIIa (NovoSeven[®], Novo Nordisk A/S, Copenhagen, Denmark) was supplied as a freeze-dried powder in 2 ml vials. Each vial was reconstituted immediately before use in 2.2 ml sterile water. Reconstituted rFVIIa contains 0.6 mg/ml rFVIIa (1.2 mg/2 ml). Recombinant FVIIa was administered as a slow i.v. bolus injection (80 µg/kg bw) over 2 min.

Coagulation assays

Coagulometric global tests of coagulation, activated partial thromboplastin time (aPTT) and prothrombin time, were recorded on an ACL-3000 Coagulation Analyser (Instrumentation Laboratory, Milan, Italy), using Platelin LS reagent (Organon Teknika, Turnhout, Belgium) for aPTT measurements and IL-test prothrombin-fibrinogen reagent (Instrumentation Laboratory) for prothrombin time determinations. The normal ranges were 10–12 sec for prothrombin and 25–35 sec for aPTT.

Antithrombin (AT-III) was analysed using the Coamatic Antithrombin kit-method (Chromogenix, Mölndal, Sweden). The normal range for AT-III was 0.89–1.25 U/ml. FVII coagulation activity (FVII:C) was recorded on a Thrombolyzer (Behnk Elektronik, Norderstedt, Germany) utilizing Simplastin S tissue factor preparation (Organon Teknika) and true FVII deficiency plasma as a test base. The reference range for FVII:C was 0.6–1.4 U/ml. Prothrombin fragment₁₊₂ (F₁₊₂) was measured using an ELISA kit method (Behring Diagnostika, Marburg, Germany). The reference range for F₁₊₂ was below 2.36 mmol/l.

Results

Patients

The mean age was 48 years (range 34–67 years; 8 men and 2 women) (Table I). The aetiology of cirrhosis was alcohol in all cases. The stage of the cirrhosis was classified as Child-Pugh class B in 5 patients and class C in 5 patients. All patients had known varices that had bled before. Acute endoscopy was not performed before entry into the study. All patients had a prothrombin time above the normal range and all but one had FVII:C levels below the normal range (Table I). One patient (no. 7) had a sudden life-threatening rupture of a varix and rFVIIa was administered before blood was drawn

Table I. Patient characteristics

Patient	Sex	Age (years)	Child-Pugh	Initial PT (sec) (NL, 10–12)	Initial FVIIa:C (U/ml) (NL, 0.6–1.4)
1	M	45	B	15	0.37
2	F	39	B	18	0.39
3	M	46	B	13	0.73
4	M	41	C	21	0.28
5	F	43	C	22	0.12
6	M	46	C	23	0.09
7	M	57	B	18	—
8	M	67	C	16	0.28
9	M	64	C	21	0.15
10	M	34	B	14	0.38

NL = normal limits; PT = Prothrombin time.

for baseline values. The immediately preceding prothrombin time was used as entry value.

Varices were accepted as bleeding source based on endoscopy after study entry in six patients, by contrast filling of varices and control of bleeding at portography and TIPS in three patients that had earlier received several banding treatments, and by immediate bleeding control by balloon tamponate in one patient who could not undergo endoscopy.

All patients were treated throughout the study period with Terlipressin (Glypressin[®]), initially 2 mg as an iv bolus dose, followed by 1 mg every 4 h. During the study, the patients received a median of 2 units of packed red cells (range 0–5). In addition, 1 patient (no. 5) received 4 units of plasma terminally in the study period.

Immediate bleeding control was obtained in all patients. However, within the study period of 12 h, 2 patients (Child-Pugh class C) had haematemesis. Beyond the study time period, 1 further patient (class C) had haematemesis and 3 patients (class B) received a TIPS within 24 h. Thus, 5 (non-TIPS) patients did not rebleed within 24 h. No patient died within the study observation period of 12 h. Six patients (5 in class C, 1 in class B, and those with the longest initial prothrombin times) died after 2–10 days, that is in all cases related to the bleeding episode (6).

Prothrombin time

The initial prothrombin time values were variable (mean at baseline 18 sec, range 13–23 sec). There was a trend towards parallelism among the patients' individual time curves (Table I, Fig. 1). Prothrombin time normalized in all patients 30 min after injection of rFVIIa. In 7 patients, the prothrombin time remained within the reference range 4 h after treatment, while it returned to above normal limits after 2 h in the remaining 3 patients, who all had prothrombin time above 20 sec at baseline (Table I) (patients nos. 4, 5, 9).

FVII coagulation activity

Mean initial factor VII:C activity was 0.19 mU/ml. The mean peak FVII:C value was 17.7 U/ml (range 8.3–

25.1 U/ml). Two hours after dosing, about half of the measured peak activity had disappeared (Fig. 2).

Safety

No adverse event was reported during the study. There was no clinical or biochemical sign of DIC. At baseline, the patients displayed fibrin D-dimers ranging from <500 mg/l to 15.300 mg/l, suspectedly related to intestinal absorption from large amounts of clotted blood. Likewise, the prothrombin fragment F1 + 2 was dissimilar among patients. Antithrombin, albeit abnormally low in all patients at all times during the observation period of 12 h, was reviewed, and data did not demonstrate a tendency to change in either direction. Hence, consumption of antithrombin was not detected.

Discussion

This study shows that it is possible to correct the coagulopathy of patients with decompensated cirrhosis and active bleeding from oesophageal varices by means of specific replacement therapy with rFVIIa. The effect, quantified by the prothrombin time, reached a maximum within 30 min and lasted for 4–6 h in most patients, and for only 2 h in a minority of patients. The therapy increased the factor VII:C activity by a factor of 100.

It has earlier been shown that rFVIIa can normalize prothrombin time in haemophiliacs (7) and in subjects on anticoagulation regimens (8). Furthermore, the use of rFVIIa is under implementation in clinical hepatology. It is reported to correct haemostasis during alcohol injection therapy of hepatocellular cancer (9), and reports on its use in complicated liver transplantation are under way (e.g. 10). It is able to correct the prothrombin time in non-bleeding cirrhosis patients (5). The present results extend these findings by demonstrating that rFVIIa exerts its corrective effects on haemostasis even in the case of active bleeding and ongoing loss of coagulation factors in liver patients already depleted of coagulation factors. This suggests a clinical utility of rFVIIa in cirrhosis patients, besides its effects on laboratory tests.

The patients with the shortest effect of the treatment (nos.

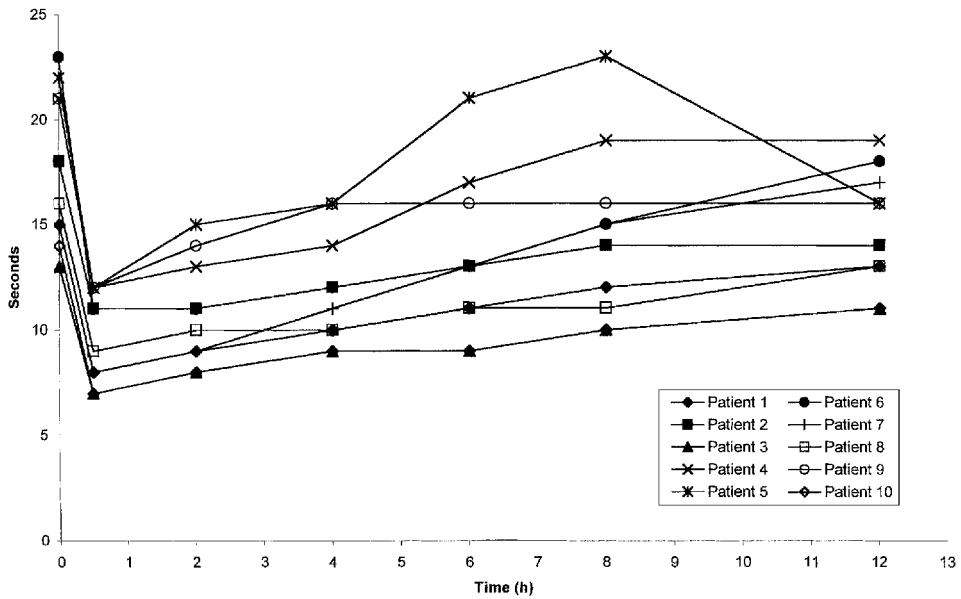


Fig. 1. Individual prothrombin time levels. The figure displays all 10 time courses. Some data points coincide, since prothrombin time is given in seconds.

4, 5, 9) all had baseline prothrombin times above 20 sec, had class C cirrhosis, actively drank up till admission, had alcoholic hepatitis, and were in a generally deteriorated nutritional and social state. This may suggest that the activation of the coagulation cascade by rFVIIa is less efficient in patients that are universally and severely depleted of plasma proteins.

We administered the same dose per kg weight of rFVIIa to all patients, namely the dose that has been shown to be large enough to normalize artificial coagulopathy (8). It is not

known whether a dose adjusted in accordance with initial factor level may have a larger or more protracted effect.

The safety laboratory parameters, although a difficult issue in patients with advanced liver disease, bleeding and intestinal blood absorption, showed no sign of general activation of the coagulation cascade. This is in accordance with the described local effect of the drug, which depends on activation at the site of the lesion. The effect of the drug was not due to other replacement therapy. No fresh plasma was given within the first 12 h after administration of rFVIIa

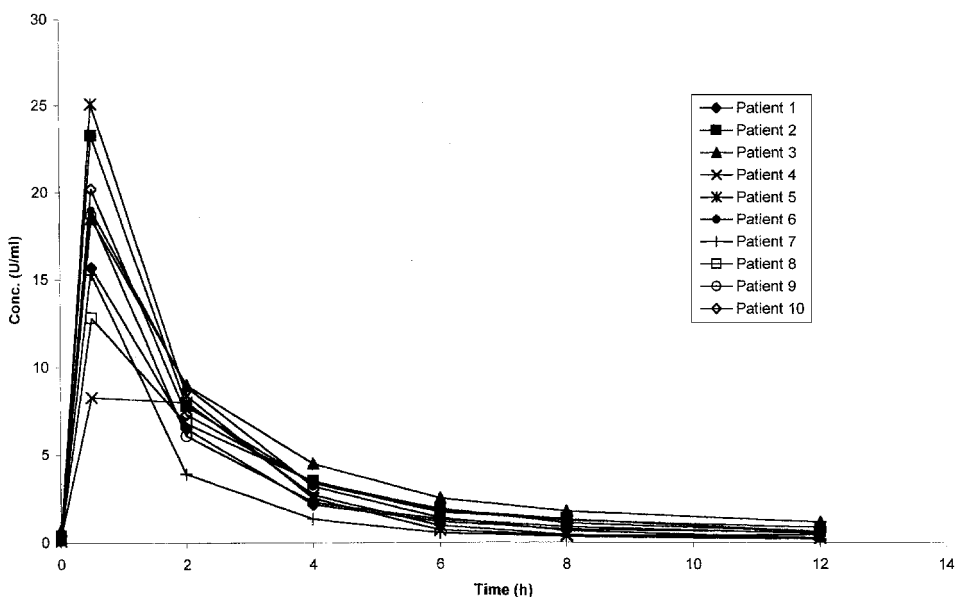


Fig. 2. Individual factor VII:C activities.

except for patient no. 5, who received 4 units a short time before the final study blood sample (Fig. 1). With the exception of the three patients with the most marked pretreatment coagulopathy, the decrease in FVII level was similar to that reported earlier in non-bleeding cirrhosis patients in the same Child-Pugh class (5). This may indicate that the FVII level is not immediately further decreased by bleeding.

Haematemesis from varices is the major cause of death in patients with cirrhosis. Several risk factors carrying a poor prognosis during a bleeding episode have been identified, e.g. large varices, thin mucosal cover ('cherry red spots'), and high portal pressure. It is biologically reasonable to expect that also the degree of coagulopathy may be an independent risk factor. This has not, however, been definitely settled, but is suggested by primary (11) or secondary analysis of data from prospective trials (12). Whether the higher mortality is due to impaired liver function reflected by the decreased prothrombin level or is due to coagulopathy in itself is not established. The present data indicate that it will be possible and safe to elucidate this important issue by way of rFVIIa administered in a randomized, prospective trial. However, the clinical courses and outcomes reported here in themselves give no indication of the prognostic importance of correcting the coagulopathy, and the effect of sustained correction of the coagulopathy by means of repeated administration of rFVIIa is not known.

In conclusion, one dose of rFVIIa to cirrhosis patients with bleeding from oesophagogastric varices can correct coagulopathy. This may purport a potential in the treatment of this life-threatening complication to cirrhosis, where all new possible therapeutical modalities are welcomed. A prospective randomised trial is warranted.

Acknowledgements

We thank Joan Dideriksen for dedicated day and night assistance during the trial. This trial was supported by a grant from Novo Nordisk A/S and was conducted at the Dept. of Medicine V, Aarhus University Hospital, Denmark.

Received 9 August 2000

Accepted 28 February 2001

References

1. Mammen EF. Coagulation abnormalities in liver disease. *Hematol Oncol Clin North Am* 1992;6:1247-57.
2. Graham DY, Smith JL. The course of patients after variceal hemorrhage. *Gastroenterology* 1981;80:800-9.
3. Levacher S, Letoumelin P, Pateron D, Blaise M, Lapandry C, Pourriat J-L. Early administration of terlipressin plus glyceryl trinitrate to control active upper gastrointestinal bleeding in cirrhotic patients. *Lancet* 1995;346:865-8.
4. Rapaport SI, Rao LVM. Initiation and regulation of tissue factor-dependent blood coagulation. *Arterioscler Thromb* 1992;12:1111-21.
5. Bernstein DE, Jeffers L, Erhardtson E, Reddy KR, Glazer S, Squiban P, et al. Recombinant factor VIIa corrects prothrombin time in cirrhotic patients: a preliminary study. *Gastroenterology* 1997;113:1930-7.
6. de Franchis R, Pascal JP, Ancona E, Burroughs AK, Henderson M, Fleig W, et al. Definitions, methodology and therapeutic strategies in portal hypertension: a Consensus Development Workshop, Baveno, Lake Maggiore, Italy, 5 and 6 April 1990. *J Hepatol* 1992;15:256-61.
7. Shapiro AD, Gilchrist GS, Hoots WK, Cooper HA, Gastineau DA. Prospective, randomised trial of two doses of rFVIIa (NovoSeven) in haemophilia patients with inhibitors undergoing surgery. *Thromb Haemost* 1998;80:773-8.
8. Erhardtson E, Nony P, Dechavanne M, Ffrench P, Boissel JP, Hedner U. The effect of recombinant factor VIIa (NovoSeven) in healthy volunteers receiving acenocoumarol to an International Normalized Ratio above 2.0. *Blood Coagul Fibrinolysis* 1998;9:741-8.
9. Papatheodoridis GV, Chung S, Keshav S, Pasi J, Burroughs AK. Correction of both prothrombin time and primary haemostasis with therapeutic alcohol injection of hepatocellular cancer in liver cirrhosis. *J Hepatol* 1999;31:747-50.
10. Kalicinski P, Kaminski A, Drewniak T, Ismail H, Szymczak M, Markiewicz M, et al. Quick correction of hemostasis in two patients with fulminant liver failure undergoing liver transplantation by recombinant activated factor VII. *Transplant Proc* 1999;31:378-9.
11. The North Italian Endoscopic Club for the Study and Treatment of Esophageal Varices. Prediction of the first variceal hemorrhage in patients with cirrhosis of the liver and esophageal varices. *N Engl J Med* 1988;319:983-9.
12. The PROVA study group. Prophylaxis of first hemorrhage from esophageal varices by sclerotherapy, propranolol or both in cirrhotic patients: a randomized multicenter trial. *Hepatol* 1991;14:1016-24.