

# Recombinant Factor VIIa reverses the effects of a high dose of rivaroxaban in baboons

Andras Gruber<sup>1</sup>, Ulla M Marzec<sup>1</sup>, Ulf Buetehorn<sup>2</sup>, Stephen R Hanson<sup>1</sup>, Elisabeth Perzborn<sup>3</sup>

<sup>1</sup>Departments of Biomedical Engineering & Medicine, Oregon Health and Science University School of Medicine, Portland, Oregon;

<sup>2</sup>Bioanalytics and <sup>3</sup>Global Drug Discovery, Bayer HealthCare AG, Wuppertal, Germany

## Introduction

Rivaroxaban is a novel, oral, direct Factor Xa inhibitor in advanced clinical development for the prevention and treatment of thrombosis.<sup>1</sup> Bleeding complications are a potential side-effect of all systemic anticoagulant agents, including rivaroxaban.<sup>2,3</sup> Activated Factor VII (FVIIa) may be able to reverse the antithrombotic effects of rivaroxaban during bleeding emergencies.

## Objective

- To investigate the potential of recombinant human FVIIa (NovoSeven®; rFVIIa) to reverse the antithrombotic effect of high-dose rivaroxaban in baboons

## Methods

- The study used 11 juvenile male baboons (*Papio anubis* or  *cynocephalus*), weight 8.5–11.6 kg

### Dose-finding studies

- In dose-finding studies, animals were given intravenous (i.v.) bolus doses of 0.2, 0.4 or 0.6 mg/kg rivaroxaban to determine a dose that would prolong bleeding time (BT) ≥2-fold, and prolong prothrombin time (PT) 3- to 4-fold

### Rivaroxaban pharmacodynamics

- Five animals were administered rivaroxaban (0.6 mg/kg bolus, followed by 0.6 mg/kg/hour infusion)
- PT, activated partial thromboplastin time (aPTT) and BTs were monitored

### Effect of rFVIIa on haemostasis in animals receiving high-dose rivaroxaban

- Seven animals received 0.6 mg rivaroxaban intravenously, followed by an i.v. infusion of 0.6 mg/kg/hour for 60 minutes
- A bolus of rFVIIa 210 µg/kg was administered 30 minutes after the start of rivaroxaban infusion
- Skin template BT was measured by an incision device (Surgicutt®)
- BT tests were performed before rivaroxaban administration, 15 and 30 minutes after the start of the rivaroxaban infusion, and 5, 15 and 30 minutes after the administration of rFVIIa
- PT, aPTT and plasma thrombin–antithrombin (TAT; measured by ELISA) levels were monitored

## Results

### Dose range

- Ten minutes after single i.v. bolus injections of 0.2, 0.4 and 0.6 mg/kg of rivaroxaban, PT was prolonged 1.7-, 2.4- and 3.7-fold over baseline, respectively. The higher two doses also increased BT
- Based on these results, an i.v. loading dose of rivaroxaban 0.6 mg/kg followed by continuous i.v. infusion of 0.6 mg/kg/hour was selected for the subsequent tests

### Rivaroxaban pharmacodynamics

- In animals given an i.v. loading dose of rivaroxaban 0.6 mg/kg followed by i.v. infusion of 0.6 mg/kg/hour for 60 minutes, a steady state of anticoagulation was obtained over 60 minutes (Table 1)

**Table 1.** Effect of administration of a bolus dose of 0.6 mg/kg rivaroxaban followed by a 0.6 mg/kg/hour continuous infusion on BT, PT and aPTT in baboons (N=5). Values are given as mean±SD

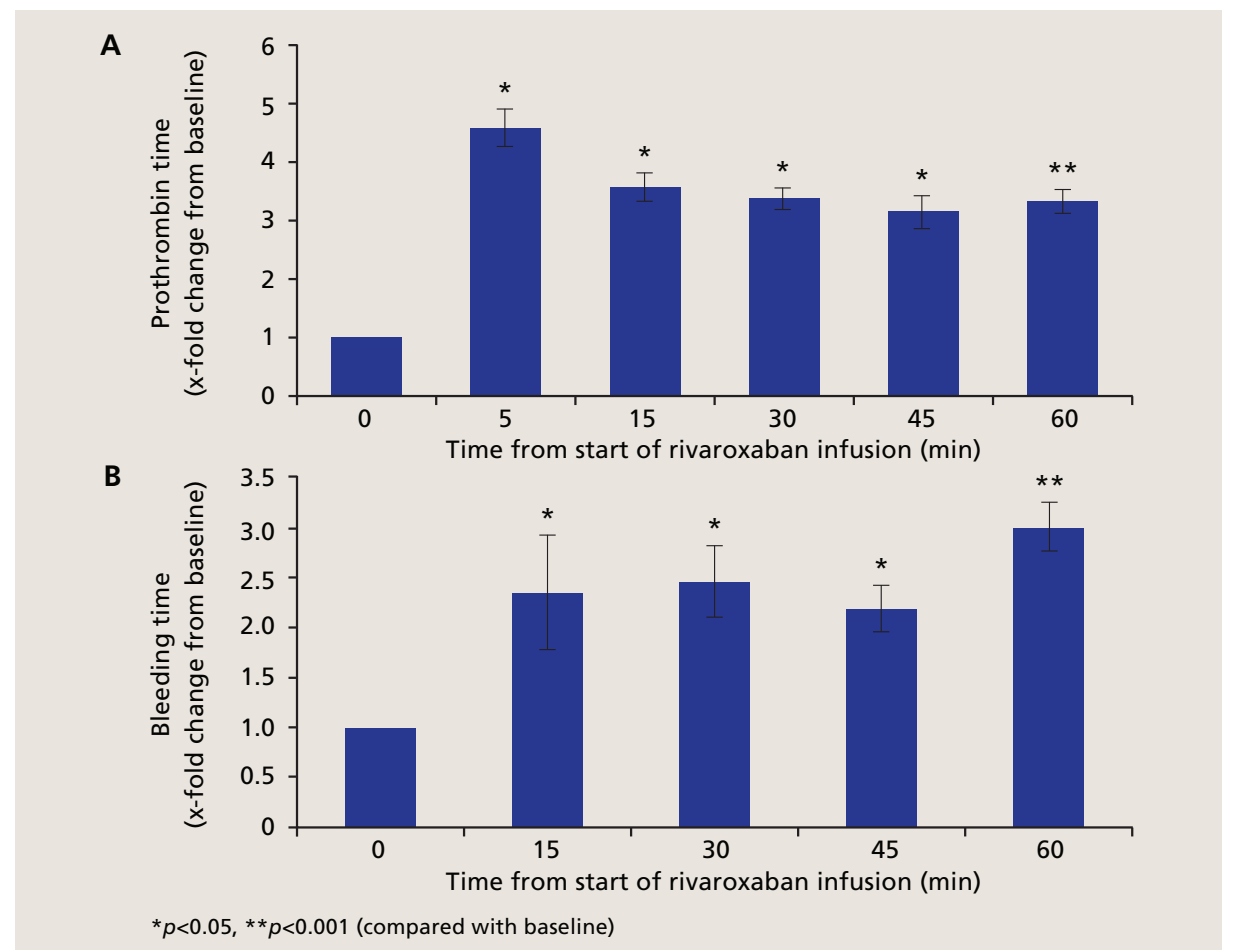
Time (mins)	Mean BT (x-fold change from baseline)	Mean PT (x-fold change from baseline) [Neoplastin <sup>a</sup> ]	Mean PT (x-fold change from baseline) [Innovin <sup>a</sup> ]	Mean aPTT (x-fold change from baseline) [Hemosil SynthASil <sup>b</sup> ]
5	–	4.58±0.66	2.22±0.18	2.04±0.20
15	2.36±1.27	3.58±0.52	1.85±0.13	1.83±0.13
30	2.46±0.81	3.38±0.40	1.76±0.13	1.76±0.13
45	2.19±0.41	3.16±0.47	1.66±0.08	1.74±0.15
60	3.01±0.33	3.34±0.45	1.84±0.14	1.79±0.15

aPTT, activated partial thromboplastin time; BT, bleeding time; PT, prothrombin time; SD, standard deviation.

<sup>a</sup>PT was measured with Innovin™ reagent (Dade Behring, Deerfield, IL, USA) and Neoplastin® CI Plus reagent (Diagnostica Stago, Parsippany, NJ, USA), using an electromechanical fibrometer (FibroSystem™, Becton Dickinson, Franklin Lakes, NJ, USA).

<sup>b</sup>aPTT was measured with Hemosil SynthASil™ reagent (Instrumentation Laboratory, Lexington, MA, USA).

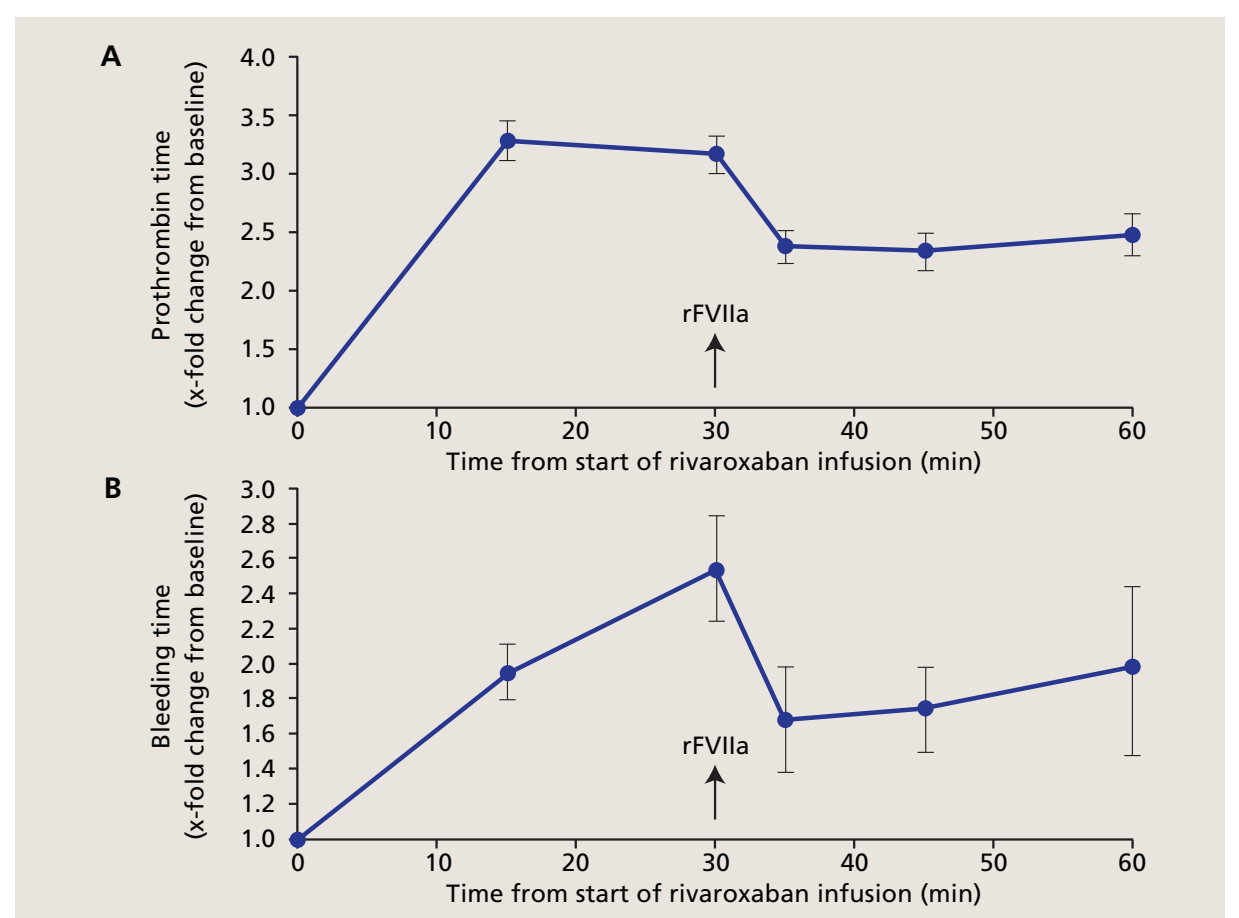
- PT (Neoplastin) was prolonged by 3.58±0.52-fold compared with baseline at 15 minutes ( $p<0.05$ ) and 3.34±0.45-fold compared with baseline at 60 minutes ( $p<0.001$ ) (Figure 1A)
- Mean BT was prolonged by 2.36±1.27-fold compared with baseline at 15 minutes ( $p<0.05$ ) and 2.52±0.56-fold compared with baseline between 45 and 60 minutes ( $p<0.001$ ) (Figure 1B)
- Rivaroxaban prolonged aPTT (Hemosil SynthASil) by 1.79±0.15-fold compared with baseline at 60 minutes



**Figure 1.** Effect of high-dose intravenous rivaroxaban on (A) prothrombin time and (B) bleeding time in baboons (N=5). Values are shown as mean±standard error of the mean.

### Effect of rFVIIa on rivaroxaban-impaired haemostasis

- Administration of rFVIIa 30 minutes after the start of the rivaroxaban infusion reduced PT (Neoplastin) prolongation from 3.17±0.42-fold to 2.38±0.41-fold compared with baseline 5 minutes after rFVIIa injection – a 25% reduction. PT was markedly shortened for at least 30 minutes (Figure 2A; Table 2)
- BT shortened from 2.54±0.79-fold to 1.68±0.80-fold compared with baseline 5 minutes after rFVIIa injection – a 34% reduction (Figure 2B; Table 2)
- TAT levels (a surrogate marker of thrombin activity) decreased during rivaroxaban infusion and this decrease did not change after rFVIIa bolus administration (Table 2)



**Figure 2.** Effect of recombinant human Factor VIIa (rFVIIa; 210 µg/kg at 30 minutes) on (A) prothrombin time and (B) bleeding time in baboons (N=7) receiving high-dose rivaroxaban. Values are shown as mean±standard error of the mean.

**Table 2.** Effect of rFVIIa on BT, PT, aPTT and TAT concentration in baboons (N=7) anticoagulated with high-dose rivaroxaban. Values are given as mean±SD

Time	Mean BT (x-fold change from baseline)	Mean PT (x-fold change from baseline) [Neoplastin <sup>a</sup> ]	Mean aPTT (x-fold change from baseline) [Hemosil SynthASil <sup>b</sup> ]	Mean TAT concentration (µg/l)
Baseline	1.00±0.00	1.00±0.00	1.00±0.00	7.35±4.17
15 minutes	1.95±0.43	3.29±0.44	1.78±0.19	–
30 minutes	2.54±0.79	3.17±0.42	1.76±0.19	2.95±0.79
5 minutes after rFVIIa	1.68±0.80	2.38±0.41	1.45±0.11	2.58±0.52
15 minutes after rFVIIa	1.74±0.65	2.24±0.41	1.41±0.09	–
30 minutes after rFVIIa	1.96±1.26	2.48±0.49	1.45±0.07	4.00±1.12

aPTT, activated partial thromboplastin time; BT, bleeding time; PT, prothrombin time; rFVIIa, recombinant human activated Factor VII; TAT, thrombin–antithrombin; SD, standard deviation.

<sup>a</sup>PT was measured with Neoplastin® CI Plus reagent (Diagnostica Stago, Parsippany, NJ, USA), using an electromechanical fibrometer (FibroSystem™, Becton Dickinson, Franklin Lakes, NJ, USA).

<sup>b</sup>aPTT was measured with Hemosil SynthASil™ reagent (Instrumentation Laboratory, Lexington, MA, USA).

## Conclusion

- Administration of human rFVIIa can rapidly reverse the antithrombotic effect of high-dose rivaroxaban in baboons

### References and disclosures

- Agnelli G *et al.* *Circulation* 2007;116:180–187.
- Levine MN *et al.* *Chest* 2004;126(Suppl 3):2875–3105.
- Perzborn E *et al.* *Haemostasis Thromb* 2005;3:514–521.

This study was supported by Bayer HealthCare AG and Scios, Inc. Rivaroxaban is in clinical development and not yet licensed.

Poster P059 presented at the 20th International Congress on Thrombosis (ICT), Athens, Greece; 25–28 June 2008