The Combined Use of Bypassing Agents with Antithrombin Reduction in Plasma of Hemophilia A and B Patients with Inhibitors

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Introduction

Treatment Options for Patients with Inhibitors

Patients with hemophilia with high responding inhibitors are treated with bypassing agents

Bypassing agents aim to enable the burst of thrombin generation as the final common mediator for clot formation, despite the absence of FVIII or FIX

The most commonly used bypassing agents are recombinant activated FVII (rFVIIa; i.e. NovoSeven) and activated prothrombin complex concentrate (aPCC; i.e. FEIBA)

Recently, new non-factor replacement therapies have emerged for patients with hemophilia, including patients with inhibitors

Fitusiran

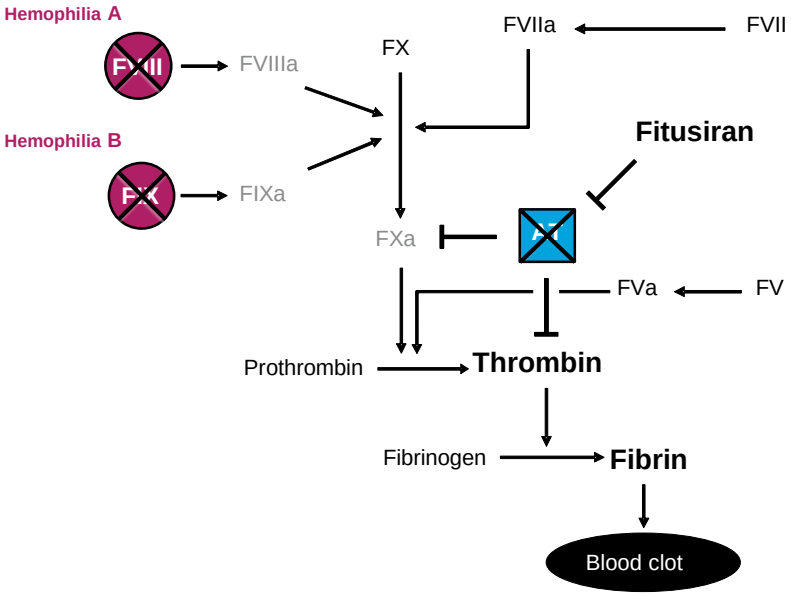
Investigational RNAi Therapeutic for Treatment of Hemophilia

Fitusiran (ALN-AT3)

- SC-administered small interfering RNA (siRNA) therapeutic targeting antithrombin (AT)
  - Non-biologic, chemically-synthesized, with targeting ligand to specifically deliver to liver—site of AT synthesis
  - Harnesses natural RNA interference (RNAi) mechanism for regulation of plasma AT levels

Therapeutic hypothesis

- Hemophilia A and B are bleeding disorders characterized by ineffective clot formation due to insufficient thrombin generation
- Fitusiran is designed to lower AT, with goal of promoting sufficient thrombin generation to restore hemostasis and prevent bleeding
  - Observation of ameliorated bleeding phenotype in patients with co-inheritance of thrombophilic traits in hemophilia[1-4]
  - Supported by pre-clinical data[5] and emerging Phase 1 clinical results[6-7]



[1]Kurnik K, et al. *Haematologica*. 92:982-985 (2007); [2]Ettingshausen E, et al. *Thromb Haemost*. 85:218-220 (2001); [3]Negrier C, et al. *Blood*. 81:690-695 (1993); [4]Shetty S, et al. *Br J Haematol*. 138:541-544 (2007); [5]Seghal A, et al. *Nat Med*. 21:492-497 (2015); [6]Pasi KJ et al. *Blood*. 2016, 128: 1397; [7]Pasi KJ, et al. *N Engl J Med. 2017;* epub ahead of print.

Aims

Patients with hemophilia with inhibitors treated with fitusiran may experience breakthrough bleeding episodes

As bleeding episodes in inhibitor patients are treated with bypassing agents, we aimed to predict the effect of bypassing agents in the background of fitusiran therapy

Thrombin generation was evaluated in plasma of patients with hemophilia A and B with inhibitors in the presence of bypassing agent and reduced antithrombin activity

Methods

Plasma samples from patients with severe hemophilia A (HA) and hemophilia B (HB) with high responding inhibitors were spiked with anti-AT antibody to target reduction of AT activity by approximately 50% or 90%, to model fitusiran treatment

Patient plasma was spiked with rFVIIa (1.25 and 2.5 µg/ml corresponding to doses of 45 and 90 mcg/kg, respectively[1,2]) or aPCC (0.5 and 1 U/ml corresponding to doses of 37.5 and 75 U/kg, respectively[1,2]) either alone or in combination with AT activity reduction

TG was measured by calibrated automated TG assay using 1 pM tissue factor and 4 µM phospholipid

aPCC, activated prothrombin complex concentrate; rFVIIa, recombinant factor VIIa; TG, thrombin generation

1. Turecek PL, et al.. *Pathophysiol Haemost Thromb.* 2003;33(1):16-22.
2. Livnat T, & Kenet G. Blood Cells, Molecules and Diseases. 2017 [In Press]

Results

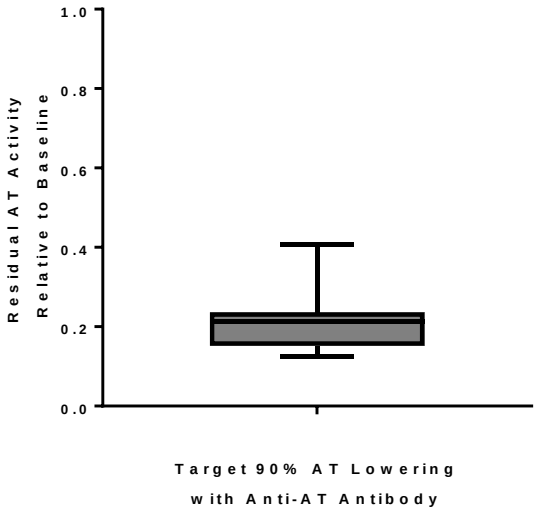
Demographics and AT Lowering

15 patients with high responding inhibitors provided plasma samples

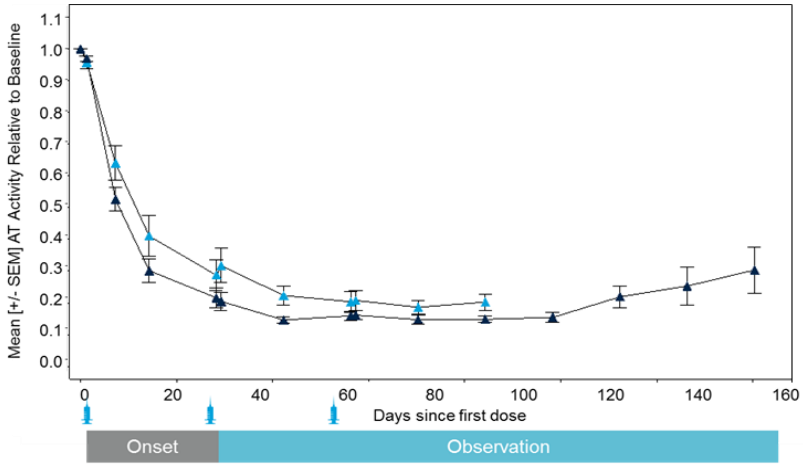
- 12 HA patients
- 3 HB patients
- Median age = 6 (range 1-50)

AT lowering, in samples from 14 patients, with anti-AT antibody targeting 50 or 90% lowering AT lowering was similar to fitusiran (~ 80% lowering)

Antithrombin lowering with anti-AT antibody in samples from patients with hemophilia A or B with inhibitors



Antithrombin lowering with fitusiran in patients with hemophilia A or B with inhibitors

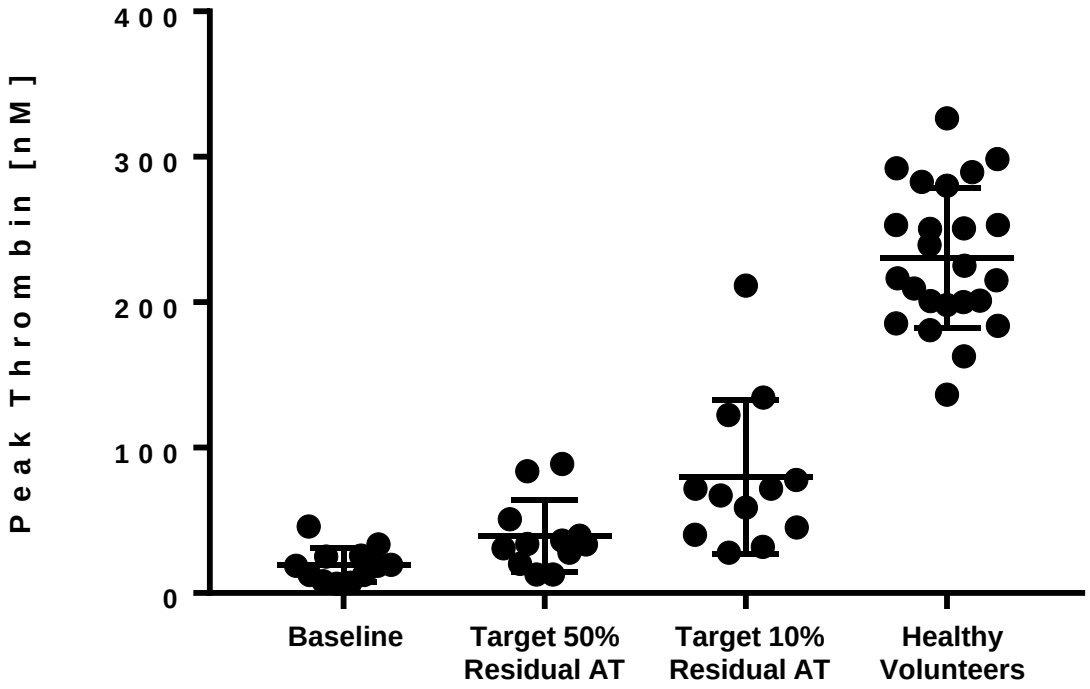


AT, antithrombin; HA, hemophilia A; HB, hemophilia B

Results

Antithrombin Reduction Improved Thrombin Generation in Hemophilia Plasma

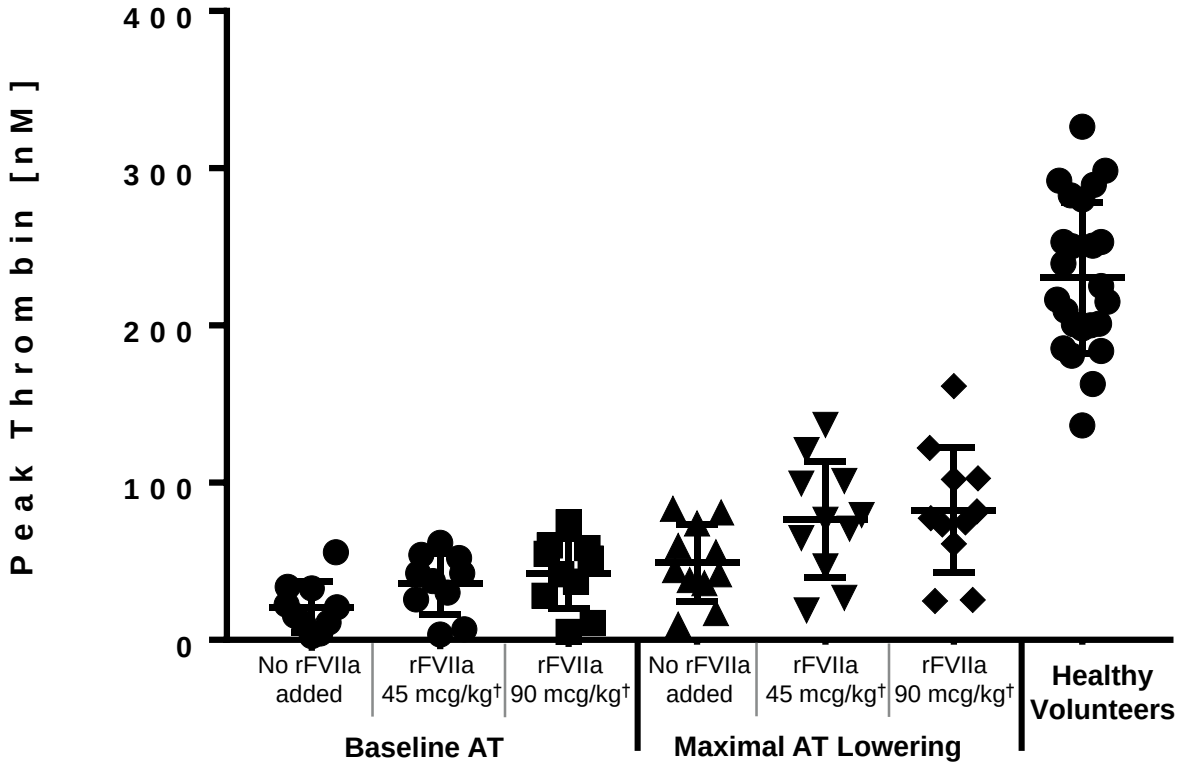
Median baseline peak thrombin generation was substantially lower in patients with hemophilia compared to healthy volunteers. Decrease in antithrombin activity resulted in a corresponding improvement in thrombin generation



Results

Thrombin Generation Further Improved with Addition of rFVIIa to AT Reduction and Does not Exceed Healthy Volunteer Range

Addition of 45 and 90 mcg/kg rFVIIa to the plasma samples induced an additive increase in peak thrombin generation

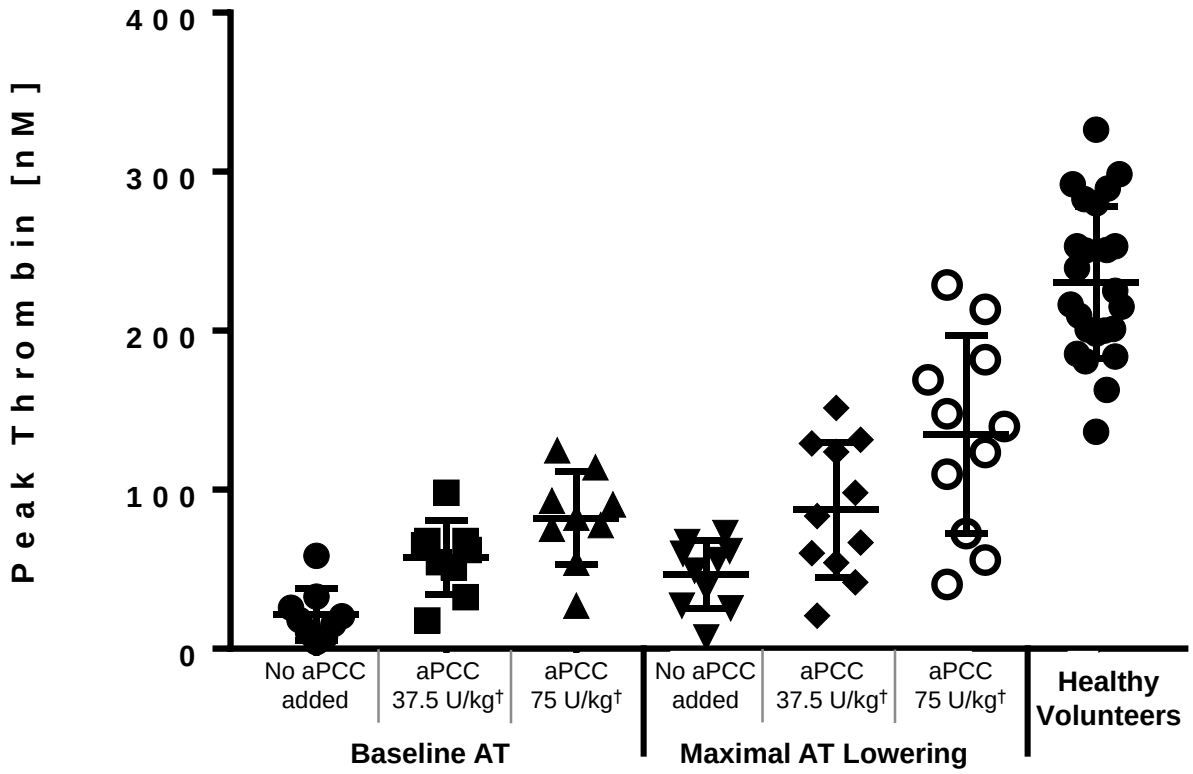


AT, antithrombin; rFVIIa, recombinant factor VIIa
†Doses of 45 and 90 mcg/kg of rFVIIa correlate to approximately 1.25 and 2.5 mcg/ml, respectively

Results

Thrombin Generation Further Improved with Addition of aPCC to AT Reduction and Does Not Exceed Healthy Volunteer Range

Addition of 38 and 75 U/kg aPCC to the plasma samples induced higher peak thrombin generation



aPCC, activated prothrombin complex concentrate; AT, antithrombin
†Doses of 37.5 and 75 U/kg of aPCC correlate to approximately 0.5 and 1 U/ml, respectively

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Limitations

Small, heterogeneous sample size makes this challenging to generalize broadly to hemophilia subpopulations

TG was assayed in platelet poor plasma samples, which may underestimate the level of TG for rFVIIa, since platelets play an important role in its pharmacodynamics effects

TG, thrombin generation; rFVIIa, recombinant factor VIIa

Summary

Enhanced thrombin generation was achieved with bypassing agents in the context of reduced plasma AT activity in this ex vivo study

Peak thrombin levels did not exceed the normal range, either with AT lowering alone or with the addition of bypassing agents

These data suggests that bypassing agents may potentially be used in conjunction with AT reduction

Further clinical investigations are required to better understand the implications, relevance, and translatability of these ex vivo findings to clinical practice

AT, antithrombin