EXPERT REVIEW OF HEMATOLOGY, 2016
VOL. 9, NO. 2, 115–122
http://dx.doi.org/10.1586/17474086.2016.1135046

Taylor & Francis
Taylor & Francis Group

DRUG PROFILE

# Andexanet alfa for the reversal of Factor Xa inhibitor related anticoagulation

Kamrouz Ghadimi[a], Keith E. Dombrowski[b], Jerrold H. Levy[a] and Ian J. Welsby[a]

[a]Divisions of Cardiothoracic Anesthesiology & Critical Care Medicine, Duke University Medical Center, Durham, NC, USA; [b]Department of Neurology, Duke University Medical Center, Durham, NC, USA

**ABSTRACT**

Andexanet alfa is a specific reversal agent for Factor Xa inhibitors. The molecule is a recombinant protein analog of factor Xa that binds to Factor Xa inhibitors and antithrombin:LMWH complex but does not trigger prothrombotic activity. In ex vivo, animal, and volunteer human studies, andexanet alfa (AnXa) was able to dose-dependently reverse Factor Xa inhibition and restore thrombin generation for the duration of drug administration. Further trials are underway to examine its safety and efficacy in the population of patients experiencing FXa inhibitor-related bleeding.

**ARTICLE HISTORY**
Received 12 August 2015
Accepted 18 December 2015
Published online
1 February 2016

**KEYWORDS**
Morpholines; Thiophenes; Andexanet alfa; Factor Xa inhibitors; Prothrombinase; Hemostasis; Thrombosis; Thrombin Generation; Apixaban; Rivaroxaban

## Introduction

Andexanet alfa (AnXa) is a modified, recombinant derivative of factor Xa (FXa) that was specifically engineered to act as a binding and reversal agent for the direct FXa inhibitor (FXai) class of direct oral anticoagulants (DOACs). The potential for this drug to meet this increasingly important clinical need is the topic of this review.

Oral (rivaroxaban [Xarelto, Janssen Pharmaceuticals, Beerse, Belgium], apixaban [Eliquis, Bristol-Meyers Squibb, New York City, NY, USA], edoxaban [Savaysa®, Lixiana®, Daiichi Sankyo, Japan]) and a direct thrombin inhibitor (Dabigatran [Pradaxa®, Boehringer Ingelheim, Ridgefield, CT, USA]) are rapidly gaining market share from the established oral vitamin K antagonists (VKAs) (e.g. warfarin) for the prevention of stroke in patients with nonvalvular atrial fibrillation [1,2]. Dabigatran [3] has a recent US FDA-approved reversal agent, idarucizumab, while AnXa, a FXai reversal agent, currently shows great promise in clinical trials for both oral FXa and low molecular weight heparin (LMWH) reversal. LMWHs are the current mainstay for the prevention of venous thromboembolism (VTE) in hip or knee replacement surgery, and the treatment and secondary prevention of pulmonary embolism and/or deep vein thrombosis [4]. Anticoagulant mechanism of action also involves FXa inhibition. Oral FXais have a similar onset time, as well as efficacy and safety profile, compared with parenteral LMWHs, with the added advantage of oral administration. The increasing use of DOACs worldwide has afforded several advantages compared with the administration of VKAs. These include fewer drug interactions, absence of the necessity for routine monitoring and titration, and the rapid onset of these medications obviating the need for bridging strategies. The lack of an effective reversal strategy for rare events of major or life-threatening bleeding, or need for urgent surgery, however, is an increasing safety concern.

In VKA-related bleeding, efficient reversal of anticoagulation is associated with improved hemostatic efficacy [5] and reduced mortality [6]. Efficient reversal of anticoagulation results in decreased hematoma growth with prothrombin complex concentrates (PCCs) during intracranial hemorrhage [7]. International guidelines support reversal of severe VKA-related bleeding with PCCs and vitamin K as soon as bleeding has been identified [8–17]. Specifically, PCCs and vitamin K administration within the first 8 h is associated with a 50% decrease in 7-day mortality [18]. Understanding the bleeding risk in patients receiving FXais is clinically important as there are no currently approved agents available for reversing FXai-related anticoagulation. Most treatments are based on limited clinical data or preclinical studies especially with respect to intracranial bleeding. Prospective evaluation of 1775 chronic rivaroxaban users, treated for atrial fibrillation-related stroke prevention or VTE, identified a bleeding rate of 59.4 events per 100 patient-years. Of these, 36.3 events per 100 patient-years were International Society on Thrombosis and Haemostasis (ISTH)-defined 'minor' bleeding events, while 19.7 events per 100 patient-

CONTACT Ian J. Welsby ian.welsby@duke.edu DUMC, Box 3094, Durham, NC, USA

years were ISTH-defined 'nonmajor, clinically relevant' bleeding events and 3.4 events per 100 patient-years were ISTH-defined 'major' bleeding events [6]. Increased usage of FXais accelerates the urgency for determining an effective reversal strategy or antidote, which has driven the development of AnXa (Portola Pharmaceuticals, South San Francisco, CA).

## Mechanism of AnXa

AnXa has been structurally designed to neutralize the anticoagulant effect of direct and indirect FXais (Figure 1). This molecule is a modified FXa protein that retains similar affinity for FXais to that of native FXa [19]. In doing so, AnXa competitively inhibits binding of the native FXa molecule to the FXai. This mechanism reverses the anticoagulant effect of the FXai, provided a sufficient number of AnXa molecules are available to bind free FXais. Importantly, within the AnXa molecule, the serine on the FXa-active site has been mutated to alanine, removing the ability to cleave prothrombin to thrombin [19]. Therefore, AnXa has been inactivated so that it lacks intrinsic, physiological blood coagulation factor activity. In addition, the membrane-binding domain of plasma-derived factor X has been deleted, rendering AnXa unable to assemble into the prothrombinase complex (Figure 1). AnXa is effectively a decoy molecule with high affinity binding for FXais. Sequestering the FXai causes rapid reduction of the free plasma concentration and neutralization of the anticoagulant effect. This releases the inhibition of FXa in the prothrombinase complex, which then results in the generation of thrombin. AnXa additionally binds to antithrombin complexed with LMWH (e.g. enoxaparin [Lovenox®]), reversing the anticoagulant effect of these subcutaneously administered FXais. The administration of AnXa should, therefore, be effective in the reversal of oral and parenteral FXais.

## Preclinical experience with AnXa

In vivo animal models have been used to demonstrate the correlation between reversal of laboratory measures of anticoagulation (International Normalized Ratio (INR), prothrombin time (PT), thrombin generation and anti-FXa activity), plasma unbound fractions of the FXai

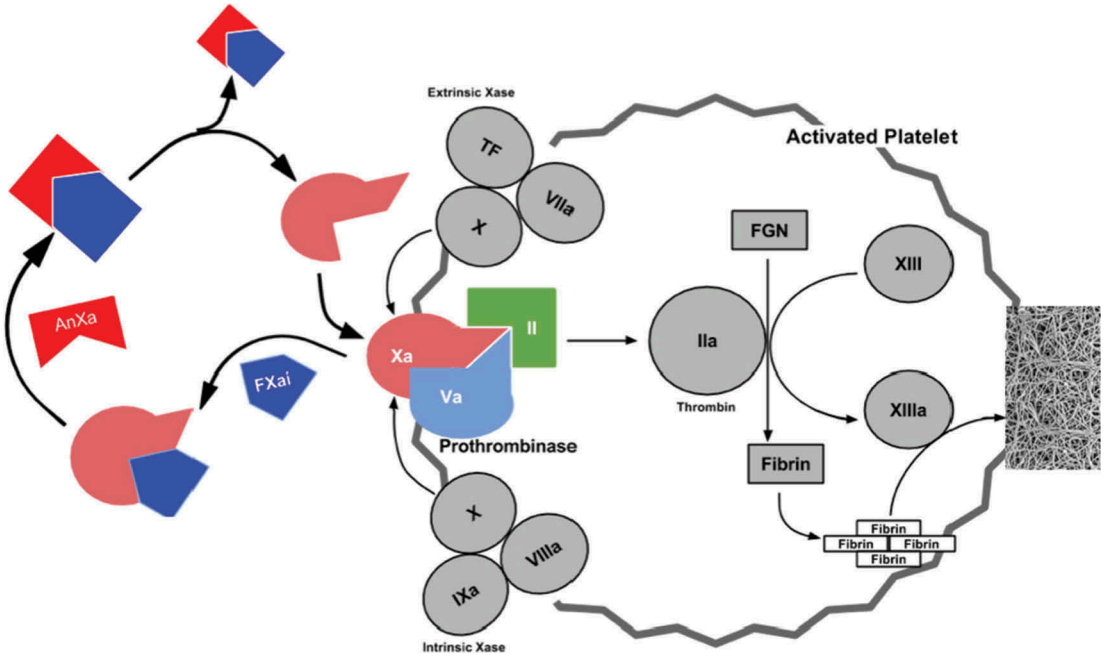


Figure 1. Pharmacologic mechanism of Andexanet alfa within the coagulation pathway involves the promotion of the Prothrombinase Complex. The figure is depicted as two pathways, which intersect at the Prothrombinase complex, on the surface of the activated platelet. The classic coagulation pathways, illustrated in terms of coagulation complexes, depict the generation of prothrombinase from both extrinsic and intrinsic tenase (Xase) complex molecules. Prothrombinase then cleaves prothrombin (Factor II, green) into thrombin (Factor IIa). Factor IIa promotes the conversion of fibrinogen to fibrin and fibrin monomers, which cross-link to polymerize into a fibrin clot (scanning electron micrographic image insert). In the setting of a FXa inhibitor, the FXa molecule binds to the FXa inhibitor molecule and the prothrombinase complex function is inhibited. The FXa inhibitor can bind both free FXa and FXa in the prothrombinase complex and inhibit thrombin generation. With the addition of andexanet alfa (AnXa), the AnXa and FXa inhibitor bind together and facilitate the reversal of inhibition of FXa, leading to the restoration of thrombin generation and downstream fibrin cross-linkage. Abbreviations: AnXa = Andexanet alfa; FXai = Factor Xa inhibitor; FGN = Fibrinogen; TF = Tissue factor; Xase = Tenase complex.

anticoagulant and reduction of actual blood loss [20,21]. In rodents anticoagulated with supratherapeutic doses of enoxaparin, AnXa elicited a dose-dependent decrease in ex vivo plasma anti-FXa activity when given either prophylactically or as treatment. When administered prophylactically prior to tail transection, blood loss was reduced to levels seen in non-anticoagulated animals [22]. Administration of AnXa (75 mg/rabbit) reduced blood loss due to rivaroxaban anticoagulation by >85% and decreased peak anti-FXa activity by 98%, PT by 74%, aPTT by 66% and the percent free fraction of rivaroxaban in plasma from 26% to <0.5% [20]. The decrease in plasma anti-FXa activity following AnXa administration correlated with a reduction in blood loss and restored hemostasis for long enough to stop or significantly reduce blood loss in this study design. The short half-life of AnXa, however, requires testing in a more robust model.

Prophylactic andexanet was compared to recombinant FVIIa (rFVIIa, Novoseven®, NovoNordisk) or three-factor PCCs (Bebulin®, Baxter) for reversal of anticoagulation and reduction in blood loss in an in vivo model of liver laceration in rivaroxaban-anticoagulated rabbits [20,21]. Intravenous bolus AnXa administration reversed rivaroxaban-induced anticoagulation as measured by plasma anti-FXa activity (almost 100% decrease), PT (77% decrease) and unbound fraction of the anticoagulant (>90% decrease), which correlated with a 75% reduction of blood loss. In contrast, rFVIIa or three-factor PCC had no effect on blood loss or anti-fXa activity. Of note, plasma-free fraction of FXais is not a relevant measure for rFVIIa or PCC as these agents aim to overdrive FXa production in order to overcome FXa inhibition. In addition, PCCs function to promote downstream thrombin generation by FXa that escapes FXa inhibition by increasing prothrombin levels. This is in contrast to AnXa, which serves to bind FXai molecules and therefore reduce circulating levels of this drug. Interestingly, rFVIIa reduced PT in non-anticoagulated (36% decrease) and in rivaroxaban-anticoagulated (89% decrease) animals, but this was not associated with reduced bleeding. PT is very dependent on FVII levels and sensitive to rFVIIa, an effect not predictive of efficacy [23].

Andexanet administered as postinjury treatment in the same rabbit liver laceration model [21] was given over a wide dose range (5–75 mg/rabbit) to explore the stoichiometric ratio of andexanet to total rivaroxaban concentration capable of decreasing blood loss. In this study, the lowest effective dose of andexanet (35 mg) to decrease blood loss reduced the anti-FXa activity, the plasma-free fraction of rivaroxaban and PTs by >90%.

There was no evidence of neurological, pulmonary or cardiovascular toxicity identified in nonhuman studies. No histopathological evidence of thrombus formation was seen despite the transient increase in thrombin–antithrombin (TAT) and D-dimer (fibrin degradation) values with 60 mg/kg doses of AnXa in cynomolgus monkeys. Activation of these coagulation markers is possibly due to AnXa-binding of tissue factor pathway inhibitor (TFPI), a phenomenon later illustrated in phase 2 studies of young healthy volunteers [24,25].

In summary, FXai-anticoagulated, in vivo animal injury models identified that rFVIIa and PCCs may affect PT or aPTT but not blood loss [23]. Furthermore, the hemostatic efficacy of these factor concentrates cannot be quantified by measuring signature laboratory values. In contrast, strong correlation between decreased free anticoagulant levels and reversal of anti-FXa activity characterize the restoration of hemostasis seen with AnXa.

## Clinical phase 1 and 2 studies

AnXa has been evaluated in one completed phase 1 study of healthy subjects, one ongoing phase 2 study in healthy subjects and two recently completed phase 3 studies in older healthy subjects ($n$ = 152, age 50–75 years, ANNEXA-A and ANNEXA-R), as described below [26]. Preliminary data have been presented at the ISTH and American Heart Association. Data surrounding the reversal of FXais (particularly rivaroxaban, apixaban and enoxaparin) are summarized below.

AnXa has been shown to be well tolerated in phase 1 and 2 clinical studies, which have included >100 healthy volunteers, without adverse or thromboembolic events. In addition, antibodies to factor X or FXa were not observed [24–26]. Long-term safety outcomes of using such a biologic at high concentrations, however, remain to be elucidated.

AnXa was associated with dose-dependent increases in $F_{1+2}$, TAT complex, D-dimer and with concomitant decreases in TFPI activity, all of which reversed quickly after discontinuation of the drug [25]. These changes returned to baseline on average by day 10 (4 days after discontinuation). These findings were not associated with a clinical thromboembolic event in any subject. Compared with administration of andexanet alone, the effects on $F_{1+2}$, TAT, D-dime, and TFPI were attenuated (all to a similar extent) in the presence of apixaban, rivaroxaban, enoxaparin and edoxaban [24].

Phase 2, randomized, double-blind, placebo-controlled studies have evaluated the safety, pharmacokinetics and pharmacodynamics of AnXa in healthy subjects who had received one of four direct or indirect FXais: apixaban, rivaroxaban, edoxaban or enoxaparin.

Multiple dosing regimens of AnXa were given in cohorts of these healthy subjects ($n = 9$) after steady-state anticoagulation was achieved over 6 days. The anticoagulation regimens include the administration of (1) apixaban 5 mg PO given twice daily, (2) rivaroxaban 20 mg PO administered once daily, (3) enoxaparin 40 mg injected subcutaneously once daily or (4) edoxaban 60 mg given orally once daily. AnXa was administered 3 h after the last anticoagulant dose on day 6 to coincide with maximum estimated plasma levels.

Subjects were followed through study day 13 in an inpatient facility then up to day 48 as outpatients. The dosing regimens evaluated in the apixaban module were single boluses ranging between 90 and 420 mg; a double bolus regimen (420 mg bolus + 180 mg bolus, 45 min apart) and a combination of a 420 mg bolus followed by either a 180 mg infusion over 45 min or by a 480 mg infusion over 120 min [27]. The regimens evaluated in the rivaroxaban module were single boluses of 210 , 420 , and 600 mg; a 720 mg bolus followed by a 240 mg infusion administered over 60 min, and an 800 mg bolus followed by a 960 mg infusion administered over 120 min [25]. The regimens evaluated in the enoxaparin module were single bolus doses of 210 and 420 mg [28].

AnXa exhibited dose-dependent pharmacokinetics, with a mean terminal half-life of approximately 5 h and an effective half-life of approximately 1 h. Administration of AnXa was associated with a rapid, almost immediate, decrease in unbound apixaban, rivaroxaban and edoxaban that was dose-dependent, with the greatest effect observed at the highest bolus doses tested and clinically significant when compared with placebo ($p < 0.001$). The decrease in unbound FXai was sustained when AnXa was administered as a bolus followed by an infusion. Again, the degree of reduced unbound FXai was sustained with higher initial bolus doses followed by higher infusion doses. The reduction in anti-FXa activity paralleled the decrease in unbound drug concentration and restoration of thrombin generation [26,28]. A similar pattern is seen among other tested FXais.

These data demonstrate the ability to reverse the laboratory-measured effects of anticoagulation after in vivo administration to humans. Note, however, that restoration of thrombin generation is entirely dependent on sufficient AnXa molecules being available to bind free FXai molecules. AnXa has a short half-life, whereas, for example, rivaroxaban may have a prolonged half-life in the elderly or those with renal dysfunction. Half-life mismatch between AnXa and the FXai could result in transient or incomplete reversal, if AnXa clears but the FXai remains. Brief reversal of anticoagulant effects may halt bleeding, but a rebound of anti-FXa activity may result in resumption of bleeding. Careful evaluation of pharmacokinetics and careful dosing with bolus followed by infusion of AnXa will likely be needed to match the duration of anticoagulant effect.

## Clinical phase 3 studies

Two randomized, double-blind, placebo-controlled studies to evaluate reversal of anticoagulation in older subjects (ages 50–75 years) anticoagulated with apixaban or rivaroxaban (ANNEXA-A and ANNEXA-R, respectively) have been recently completed [26]. The administration of AnXa or placebo occurs on the fourth day, following the last dose of the anticoagulant (at steady state). The subjects are then followed for 4 days in an inpatient study unit and, subsequently, for >30 days as outpatients. Reversal of anticoagulation is being measured using anti-FXa activity, anticoagulant-free fraction, thrombin generation and other coagulation markers. Thus far, results from current phase 3 studies show promise with similar time course of plasma concentrations of unbound FXai, correction of anti-FXa activity (Figure 2) and thrombin generation (Figure 3), as was seen in phase 2 studies [26,29]. The cumulative nature of this data will be similar to those in phase II studies, with the exception that they are derived from elderly rather than younger healthy subjects [26].

## Conclusion

AnXa is a direct-acting reversal agent specific to both oral (e.g. rivaroxaban and apixaban) and parenteral (LMWHs and fondaparinux) FXais. This agent directly binds to the FXai (or antithrombin III:inhibitor complex), blocking the anticoagulant action and releasing the inhibition of native FXa for promotion of thrombin generation and downstream hemostasis (Figure 1). The clearance of AnXa as well as its behavior as a depot for sequestering (and later release) of the anticoagulant has yet to be elucidated in the clinical setting, but the ongoing Annexa-4, phase 3b trial in patients experiencing bleeding on FXais (NCT02329327) may provide some answers. Promising results, however, in preclinical animal trials suggest that blood loss significantly decreases once the anticoagulant activity has been sufficiently (>80%) reversed.

Thus far, it has a favorable safety efficacy profile and, while theoretically having no direct procoagulant effect, it does bind to TFPI, leading to biochemical activation of coagulation without clinical thrombosis in healthy,

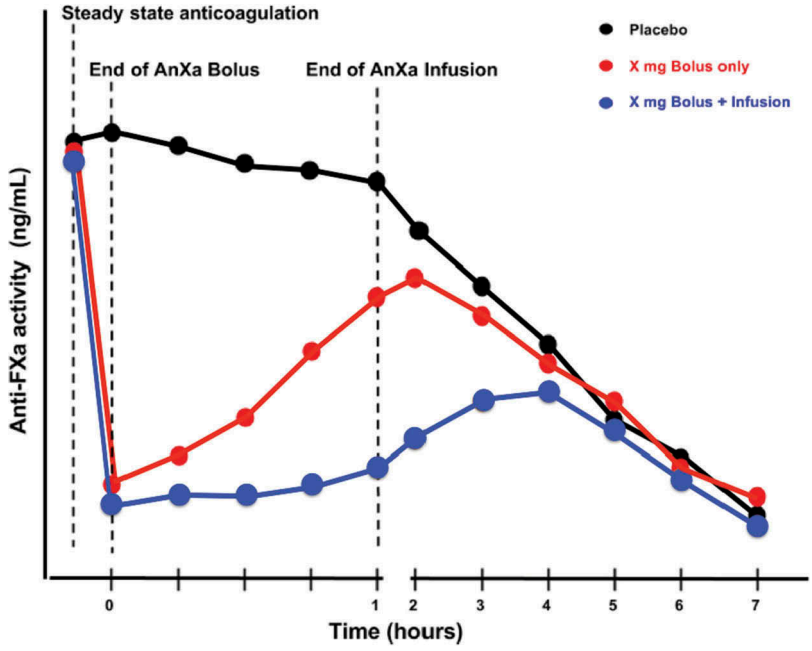


Figure 2. Reversal of Anti-Xa activity is sustained for longer duration when a bolus is followed by an infusion in older, healthy volunteers. Anti-Xa activity reflects the unbound drug levels of either oral (e.g., rivaroxaban or apixaban) or parenteral FXa inhibitors (e.g., LMWH or fondaparinux). Steady state anticoagulation is denoted prior to the end of Andexanet alfa bolus (time = 0 hrs). Placebo is noted in black. "X mg" denotes a certain dosage of Andexanet alfa administered. Subjects receiving X mg bolus only of Andexanet alfa is depicted in red and the subject receiving X mg bolus and an infusion is depicted in blue. Compared to the placebo treatment, reversal of anti-FXa activity can last for 1-2 hours after the end of andexanet bolus administration before it returns to the placebo level. Subjects receiving bolus and infusion sustain reduced activity until end of andexanet alfa infusion and display a slower rise in Anti-FXa activitiy until the FXa inhibitor undergoes metabolic clearance (~2 to 4 hrs). Note that Anti-FXa activity with respect to parenteral FXa inhibitors is reflected as IU/ml, but this unit measurement is not depicted on the abscissa in this illustration. Abbreviations: AnXa = Andexanet alfa; FXa = Factor Xa. Figure modified from Siegal, et al.[26]**

ambulatory volunteers. Currently, off-label FXai reversal strategies may use PCCs to promote thrombin generation to a level that can overwhelm the FXai effect, with the theoretical disadvantage that excessive thrombin generation may persist beyond the duration of FXai anticoagulant action, thus promoting a clinical thrombosis risk (Figure 3) [30]. Thrombotic risk is, however, dependent upon circulating levels of vitamin K-dependent factors II, VII, IX and X present at the time of PCC administration. The administration of AnXa may obviate the need for PCCs in the clinical setting of an early FXai-associated bleed. The challenge, however, may prove to be matching AnXa levels to FXai concentrations in order to sustain the reversal effect for a long enough duration to achieve hemostasis. These AnXa levels must last long enough to persist beyond any subsequent rebound of FXai anticoagulant effect while awaiting final metabolic clearance of the FXai from circulation. Similar considerations may apply to other investigational reversal agents that are in a less advanced phase of development than AnXa [31]. For now, the results of the phase 3b clinical trial for AnXa are heavily anticipated and subjects are being actively enrolled who meet study criteria while experiencing bleeding on FXais (NCT02329327).

## Expert commentary

AnXa is a recombinant, specific FXai-reversal agent with a 'FXa decoy' mechanism of action (Figure 1). It has an encouraging and favorable preliminary safety and efficacy profile, but regulatory approval for clinical use is dependent on an ongoing, pivotal, registration-enabling study in bleeding patients. Despite the potential for future clinical applications, in patients who present following severe FXai-related bleeding and consumptive coagulopathy, a multimodal approach to hemostatic resuscitation should occur and may include allogeneic blood products and factor concentrates to restore global hemostatic mechanisms. Further studies are needed to evaluate the safety and efficacy of using AnXa within a coagulation management algorithm.

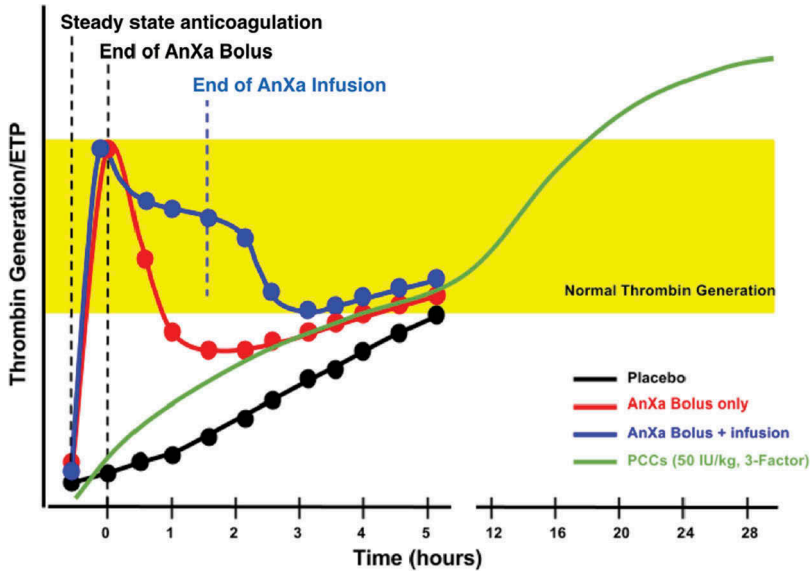


Figure 3. Thrombin generation is preserved for longer durations when andexanet alfa bolus is followed by an infusion, compared with bolus alone, in young, healthy volunteers. Thrombin generation is prolonged with PCC usage and outlasts the duration of circulating Factor Xa inhibitors, highlighting the thrombotic potential of PCCs as a treatment modality for Factor Xa inhibitor anticoagulation. Steady state anticoagulation is denoted prior to the start of Andexanet alfa bolus (time = 0 hrs). Placebo is noted in black. The normal range of thrombin generation as denoted by baseline measurement of the healthy study cohorts is depicted in yellow. Both groups received bolus administrations of andexanet alfa and almost immediately returned to normal thrombin generation range. Subjects receiving an infusion following the bolus remain in the normal range of thrombin generation longer than those receiving bolus only. The anticipated trajectory of thrombin generation after 3-Factor PCC (Profilnine®, Grifols Pharmalogicals, Los Angeles, California, USA) administration is illustrated and adopted from a study of healthy volunteers anticoagulated with rivaroxaban and then administered 50 IU/kg of Profilnine or 4-Factor PCCs (Beriplex®, CSL Behring, Germany).[32] 3F-PCCs have been illustrated in this figure to emphasize prolonged thrombin generation due to higher concentrations of prothrombin (half-life 48-72 hours) in Profilnine (150 IU of Factor II per 100 IU of Factor IX) compared with Beriplex (130 IU of Factor II per 100 IU of Factor IX). Thrombin generation reaches baseline values approximately 4 hours after PCC administration and remains elevated well beyond the duration of circulating Factor Xa inhibitor.[32] Abbreviations: AnXa = Andexanet alfa; PCCs = Prothrombin complex concentrates; ETP = Endogenous thrombin potential. Figure modified from Siegal, et al.,[26] Thrombin generation after 3-Factor PCCs has been adopted from Levi, et al.[32]

## Five-year view

AnXa shows great promise to becoming a reversal agent for FXai-related anticoagulation, which also includes LMWH where a reversal agent is also not currently available. Comprehensively, the field of anticoagulation with respect to stroke prevention in the setting of nonvalvular atrial fibrillation and VTE continues to evolve. Newer, designer medications have been manufactured with fewer side effect profiles as compared with previous drugs. They have shorter half-lives than the traditional VKAs, but in the setting of massive hemorrhage and coagulopathy, a cascade of factor consumption may outlast this short half-life. Hemodynamic and hemostatic resuscitation while source control is sought should remain the cornerstone of management in the actively bleeding patient. Nonetheless, growing trends toward prescribing these medications continue to be more commonplace; the introduction of approved reversal agents makes this a safer prospect.

AnXa is poised to be the next DOAC-reversal antidote and may encounter a similar important reception as idarucizumab. With that said, caution must be exercised, however, given the fact that the apparent short half-life of AnXa, relative to FXais, may be insufficient to correct coagulopathy, particularly in the setting of renal dysfunction or older age. With that said, the reversal of anticoagulation was maintained during the bolus plus infusion dosing strategies, supporting this method of AnXa administration [26]. Furthermore, insufficient dosage administration of the reversal agent may provide inadequate neutralization of FXai molecules with resulting persistent anticoagulant effects. The phase 3 trials were designed and performed by administering the maximum approved dose of each of the direct FXais, and AnXa was administered at the timing of the maximum concentration of the inhibitor. Under these conditions, the dose of AnXa was administered to reverse anticoagulation by >90% and this extent of reversal could be maintained

Key issues

- Andexanet alfa (AnXa) is a modified, recombinant derivative of factor Xa (FXa) specifically engineered to act as a binding and reversal agent for the direct FXa inhibitor class of direct oral anticoagulants.
- Serine on the FXa-active site of the AnXa molecule has been mutated to alanine, removing the ability to cleave prothrombin to thrombin, and theoretically eliminate thrombotic potential.
- Current phase 3 studies in healthy, elderly volunteers illustrate that administration of AnXa is able to reduce plasma concentrations of unbound FXa inhibitor, reduce anti-FXa activity and restore thrombin generation to baseline values.
- Patients receiving boluses followed by infusions experienced stronger reversal effects when compared with patients receiving bolus alone, without increased thromboembolic events.
- Long-term safety and efficacy remains to be elucidated, but enrollment is ongoing in a phase 3b trial evaluating patients with FXa inhibitor-associated bleeding (NCT02329327).

throughout the additional infusion period. These conditions represent a 'high hurdle' for reversal, and the majority of patients presenting to the hospital with a severe bleed in need of urgent reversal will be presenting at a time that is beyond the maximum concentration of the inhibitor. This assumes that the concentration of the inhibitor will likely be lower in the clinical setting than what was tested in these phase 3 studies. The clinical setting is dynamic and concentration of the inhibitor may be higher than what has been tested in trials, which may warrant continuation of an AnXa infusion. The next five years shows great potential for defining the role of AnXa and other agents in development (e.g. PER977) as a means to safely administer FXais in order to provide alternative anticoagulation for the reversible, yet problematic, VKAs. With that said, a balanced, laboratory-guided (e.g. platelet count, fibrinogen levels), multimodal approach to hemostasis management will remain the safest and most efficacious strategy in order to promote the best clinical outcomes in patient care.

## Financial & competing interests disclosure

*K Ghadimi and KE Dombrowski are Co-Investigators in a prospective, open label study of Andexanet-Alfa in patients receiving Factor Xa inhibitors with acute major bleeding, sponsored by Portola Pharmaceuticals. JH Levy serves on steering committees for Boehringer-Ingelheim, CSL Behring, Grifols, and Janssen. JH Levy is a consultant to Instrumentation Laboratories. IJ Welsby is the Principal Investigator in a prospective, open-label study of Andexanet Alfa in patients receiving Factor Xa inhibitors with acute major bleeding, sponsored by Portola Pharmaceuticals and has recently received grant support from CSL Behring and Terumo BCT. The authors and their work were supported by Duke Anesthesiology And Duke Neurology. The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.*

## References

Papers of special note have been highlighted as:
•• of considerable interest

1. Crowther M, Crowther MA. Antidotes for novel oral anticoagulants: current status and future potential. Arterioscler Thromb Vasc Biol. 2015;35(8):1736–1745.
   •• **Review of future antidotes for DOACs**
2. Levy JH, Spyropoulos AC, Samama CM, et al. Direct oral anticoagulants: new drugs and new concepts. JACC Cardiovasc Interv. 2014;7(12):1333–1351.
   •• **State-of-the-art-Review outlining DOACs, mechanisms, tables, evidence, as well as snap shot of future reversal agents**
3. Pollack CV Jr, Reilly PA, Eikelboom J, et al. Idarucizumab for dabigatran reversal. N Engl J Med. 2015;373(6):511–520.
4. Enriquez A, Lip GY, Baranchuk A. Anticoagulation reversal in the era of the non-vitamin K oral anticoagulants. Europace. 2015.
   •• **Review of upcoming reversal agents with figures, tables, and evidence**
5. Sarode R, Milling TJ Jr, Refaai MA, et al. Efficacy and safety of a 4-factor prothrombin complex concentrate in patients on vitamin K antagonists presenting with major bleeding: a randomized, plasma-controlled, phase IIIb study. Circulation. 2013;128(11):1234–1243.
6. Beyer-Westendorf J, Forster K, Pannach S, et al. Rates, management, and outcome of rivaroxaban bleeding in daily care: results from the Dresden NOAC registry. Blood. 2014;124(6):955–962.
7. Huttner HB, Schellinger PD, Hartmann M, et al. Hematoma growth and outcome in treated neurocritical care patients with intracerebral hemorrhage related to oral anticoagulant therapy: comparison of acute treatment strategies using vitamin K, fresh frozen plasma, and prothrombin complex concentrates. Stroke. 2006;37(6):1465–1470.
8. American Society of Anesthesiologists Task Force on Perioperative Blood Management. Practice guidelines for perioperative blood management: an updated report by the American Society of Anesthesiologists Task Force on Perioperative Blood Management*. Anesthesiology. 2015;122(2):241–275.
9. Baglin TP, Keeling DM, Watson HG. Guidelines on oral anticoagulation (warfarin): third edition–2005 update. Br J Haematol. 2006;132(3):277–285.
10. Baker RI, Coughlin PB, Gallus AS, et al. Warfarin reversal: consensus guidelines, on behalf of the Australasian

Society of Thrombosis and Haemostasis. Med J Aust. 2004;181(9):492–497.
11. Ferraris VA, Brown JR, Despotis GJ, et al. 2011 update to the Society of Thoracic Surgeons and the Society of Cardiovascular Anesthesiologists blood conservation clinical practice guidelines. Ann Thorac Surg. 2011;91(3):944–982.
12. Holbrook A, Schulman S, Witt DM, et al. Evidence-based management of anticoagulant therapy: antithrombotic Therapy and Prevention of Thrombosis, 9th ed: American college of chest physicians evidence-based clinical practice guidelines. Chest. 2012;141(2_suppl):e152S–e184S.
13. Morgenstern LB, Hemphill JC 3rd, Anderson C, et al. Guidelines for the management of spontaneous intracerebral hemorrhage: a guideline for healthcare professionals from the American Heart Association/American Stroke Association. Stroke. 2010;41(9):2108–2129.
14. National Advisory Committee on Blood and Blood Products. Recommendations for use of prothrombin complex concentrates in Canada. 2011.
15. Pernod G, Godier A, Gozalo C, et al. French clinical practice guidelines on the management of patients on vitamin K antagonists in at-risk situations (overdose, risk of bleeding, and active bleeding). Thromb Res. 2010;126(3):e167–e174.
16. Spahn DR, Bouillon B, Cerny V, et al. Management of bleeding and coagulopathy following major trauma: an updated European guideline. Crit Care. 2013;17(2):R76.
17. Steiner T, Kaste M, Forsting M, et al. Recommendations for the management of intracranial haemorrhage - part I: spontaneous intracerebral haemorrhage. The European Stroke Initiative Writing Committee and the Writing Committee for the EUSI Executive Committee. Cerebrovasc Dis. 2006;22(4):294–316.
18. Tazarourte K, Riou B, Tremey B, et al. Guideline-concordant administration of prothrombin complex concentrate and vitamin K is associated with decreased mortality in patients with severe bleeding under vitamin K antagonist treatment (EPAHK study). Crit Care. 2014;18(2):R81.
19. Lu G, DeGuzman FR, Hollenbach SJ, et al. A specific antidote for reversal of anticoagulation by direct and indirect inhibitors of coagulation factor Xa. Nat Med. 2013;19(4):446–451.
20. Hollenbach S, Lu G, Tan S, et al. PRT064445 but not recombinant FVIIa reverses rivaroxaban induced anticoagulation as measured by reduction in blood loss in a rabbit liver laceration model. Blood. 2012;120(Abstract 3414, 21).
21. Hollenbach S, Tan S, Deguzman F, et al. PRT064445 reverses rivaroxaban induced anticoagulation in a rabbit liver laceration "treatment" model. Eur Heart J. 2013;34:24.
22. Lu G, DeGuzman FR, Hollenbach SJ, et al. Reversal of low molecular weight heparin and fondaparinux anticoagulation by a recombinant antidote (r-antidote). Circulation. 2010;122(21).
23. Dzik WS. Reversal of drug-induced anticoagulation: old solutions and new problems. Transfusion. 2012;52(Suppl 1):45S–55S.
24. Crowther M, Kitt M, Lorenz T, et al. A phase 2 randomized, double-blind, placebo-controlled trial of PRT4445, a novel, universal antidote for direct and indirect Factor Xa inhibitors. Presented at: The XXIV Congress of the International Society on Thrombosis and Haemostasis Congress; 2013 July 2; Amsterdam, The Netherlands. 2013.
25. Crowther M, Vandana M, Kitt M, et al. A phase 2 randomized, double-blind, placebo-controlled trial demonstrating reversal of rivaroxaban-induced anticoagulation in healthy subjects by andexanet alfa (PRT064445), an antidote for FXa inhibitors. Presented at: The 55th American Society of Hematology Annual Meeting and Exposition; 2013 Dec 9; New Orleans, LA. 2013.
26. Siegal DM, Curnutte JT, Connolly SJ, et al. Andexanet alfa for the reversal of factor Xa inhibitor activity. N Engl J Med. 2015;373:2413–2424.

**•• Phase 3 publication of Andexanet alfa, outlining safety and efficacy of this reversal agent in healthy, elderly volunteers**

27. Crowther M, Lu G, Conley P, et al. Sustained reversal of apixaban anticoagulation with andexanet alfa using a bolus plus infusion regimen in a phase 2 placebo controlled trial [abstract]. Eur Heart J. 2014;35:137.
28. Crowther M, Levy GG, Lu G, et al. Reversal of enoxaparin-induced anticoagulation in healthy subjects by andexanet alfa (PRT064445), an antidote for direct and indirect FXa inhibitors. Presented at: The XXIV Congress of the International Society on Thrombosis and Haemostasis Congress; 2014 Jun 23; Milwaukee, WI. 2014.
29. Crowther MA, Levy GG, Lu G, et al. ANNEXATM-A: a phase 3 randomized, double-blind, placebo-controlled trial, demonstrating reversal of apixaban-induced anticoagulation in older subjects by andexanet alfa (PRT064445), a universal antidote for factor Xa (fXa) inhibitors [abstract]. Circulation. 2014;130:2105–2136.
30. Babilonia K, Trujillo T. The role of prothrombin complex concentrates in reversal of target specific anticoagulants. Thromb J. 2014;12:8.
31. Ansell JE, Bakhru SH, Laulicht BE, et al. Use of PER977 to reverse the anticoagulant effect of edoxaban. N Engl J Med. 2014;371(22):2141–2142.
32. Levi M, Moore KT, Castillejos CF, et al. Comparison of three-factor and four-factor prothrombin complex concentrates regarding reversal of the anticoagulant effects of rivaroxaban in healthy volunteers. J Thromb Haemost. 2014;12(9):1428–1436.