# Pharmacological properties of NOACs relevant for bleeding

### Direct thrombin inhibitor dabigatran

Dabigatran etexilate is a prodrug with a bioavailabilty of 6.5% that is rapidly absorbed in the gastrointestinal tract. After oral absorption, dabigatran etexilate is rapidly and completely converted by plasma and liver esterases to dabigatran, a competitive and reversible thrombin inhibitor.1 The maximum anticoagulant effect of dabigatran occurs at the peak of drug plasma concentrations (i.e., after 2h) with the anticoagulant effect decreasing to ~50% of its maximum at 12h after its administration.2-5 Because dabigatran is eliminated mainly by the renal route, its plasma concentration and, in parallel, its anticoagulant effect depend very strongly on changes in renal function. The mean half-life increases to 18h with a creatinine clearance (CrCl) between 30 and 49 ml/min. As recommended by the ESC for all NOAC,6 dabigatran should not be used below a CrCl of <30 mL/min where the half life further increases to 27h. In case of an overdose, administration of oral activated charcoal (30-50g) may be helpful to reduce drug absorption following recent ingestion (<2h). Due to its low plasma protein binding, haemodialysis may be effective in removing dabigatran in cases of severe bleeding or in patients with renal impairment.7 In patients with end-stage renal disease, 68% of active dabigatran was removed after 4h of haemodialysis. While the settings of haemodialysis seems to be less important8, it remains unclear whether these data can be translated to unstable patients undergoing haemodiafiltration.As expected for an anticoagulant, major bleeding increased with the concentration of dabigatran in the RE-LY trial. Median trough concentrations were increased by 55% in patients with major bleeding compared to those without.9 Clinical characteristics associated with high plasma concentrations were age and reduced renal function, reemphasizing the importance of dose reduction in patients over 80 years of age and with moderately reduced renal function (CrCl < 50 ml/min). Close clinical surveillance is recommended in patients with weight < 50kg. Relevant drug interactions increasing dabigatran plasma concentrations have been observed with verapamil, requiring dose reduction, and with systemic ketoconazole, cyclosporine, itraconazole and dronedarone, contraindicating the combination of dabigatran with these agents.10

## Factor Xa inhibitors

Apixaban, edoxaban and rivaroxaban are oral selective, competitive and direct inhibitors of both free FXa and FXa associated with the prothrombinase complex, thus decreasing the conversion of prothrombin to thrombin.11-16 They decrease the generation of thrombin and thereby reduce thrombin-mediated activation of both coagulation and platelets, but allow residual thrombin to carry out its other important functions.17 FXa inhibitors have a much higher bioavailability than the prodrug dabigatran (*Table 1*). Their half-life of 5 to 14h is comparable to that of dabigatran. They are excreted by the kidney to a lesser extent than dabigatran (27-50%).

Rivaroxaban inhibits human FXa (inhibitory constant [Ki*]* 0.4 nM) with >10 000-fold greater selectivity than for related serine proteases.13, 18 In human plasma, it inhibits both clot-bound FXa (half maximal inhibitory concentration [*IC50*] 75 nM) and FXa associated with prothrombinase (IC50 2.1 nM),13, 17 and inhibits the generation of thrombin after activation of the extrinsic tissue factor pathway (IC50 25 nM).19 Rivaroxaban should be taken with food, increasing its bioavailability to almost 100%. It dose-dependently inhibits FXa activity by 20–60% after single- and multiple-dosing11-12, 20 and its plasma concentrations are closely correlated with FXa activity.11-12, 21-22 While having a similar half-life than other NOACs, it has been tested with a once daily (od) regimen for treatment of atrial fibrillation (AF) and venous thromboembolism (in the long-term phase). No dose reduction is required for elderly patients or low or high body weight.

Apixaban concentration-dependently inhibits human FXa (Ki 0.08 nM),23 whereas Ki values ≥ 3 000 nM were determined for other serine proteases.14, 23 It shows a moderate selectivity for clot-bound over free FXa (IC50 1.3 vs. 7.6 nM)24 and inhibits thrombin generation rate (IC50 35 nM). Dose reduction is recommend when at least two of the following characteristics exist: age ≥ 80 years, body weight ≤ 60 kg, or serum creatinine ≥ 1.5 mg/dL (133 µmol/L).

Activated charcoal can be administered if apixaban is taken within 3h or rivaroxaban within 2h. Rivaroxaban and apixaban cannot be removed by haemodialysis due to their high plasma protein binding. Rivaroxaban and apixaban should be used with caution in patients with severe renal impairment (CrCl 15–29 mL/min) and the dose should be reduced (rivaroxaban CrCl < 50 ml/min: 15mg od; apixaban CrCl <30 ml/min: 2.5mg twice daily, bid). Rivaroxaban and apixaban are contraindicated in patients with a CrCl of <15 ml/min or with any hepatic disease associated with coagulopathy, since drug exposure and bleeding risk may be increased (Summary of Product Characteristics of European Medicines Agency). Potent inhibitors of CYP3A4 and P-glycoprotein such as azole-antimycotics (e.g. ketoconazole, itraconazole, voriconazole and posaconazole) or HIV protease inhibitors (e.g. ritonavir) may cause a relevant increase in plasma concentrations of apixaban and rivaroxaban, thus increasing the risk of bleeding, and should therefore be avoided.

Edoxaban, the third FXa inhibitor just approved by the European Medicines Agency, dose-dependently inhibits free FXa (Ki 0.56 nM) and FXa bound to the prothrombinase complex (Ki 2.98 nM) and exhibited >10 000-fold selectivity for FXa relative to thrombin.25-27 The edoxaban dose should be halved (30mg od) if patients have a CrCl 15-50 mL/min, a body weight < 60 kg, or concomitant medication with potent P-glycoprotein interaction such as verapamil, quinidine, or dronedarone.28 These dose reduction criteria overcompensated for a potential increase of edoxaban plasma concentrations.29 Edoxaban is contraindicated in patients with a CrCl below 15 ml/min. In patients with end-stage renal disease, haemodialysis had minimal effects on the clearance of edoxaban.30 In the ENGAGE AF-TIMI 48 phase III trial, exodaban has been tested at doses of 15 to 60mg od. The mean trough plasma concentrations of edoxaban increased from 16.0 ng/ml with 15mg to 48.5 ng/ml with 60mg. The risk of major bleeding moderately increased (20-30%) between the 25th to 75th centiles of edoxaban plasma concentrations for each edoxaban dose.29

# Critical appraisal of surrogate endpoints for measurement of the reversal of NOAC effects with coagulation factor concentrates

As the effect of prohaemostatic agents on NOACs has not been tested in clinical trials so far, our evidence is based on surrogate endpoints such as normalization of coagulation tests, ex-vivo thrombus formation or animal bleeding models only (Supplemental *Table 3*). Global coagulation tests are used in clinical practice to monitor the effect of anticoagulation therapy. Specifically, calibrated laboratory tests measure the effect of NOACs, as recently summarized by Siegal et al..31 Thrombin generation (TG) tests such as the calibrated automated thrombogram and clotting tests may be even more sensitive for testing the reversal of OAC effects.32 The lag-time appears to be the most sensitive TG measurement for the effects of NOACs. However, divergent results have been reported for the effect of reversal agents on routine coagulation tests and TG tests,33 questioning the relevance of the information derived from routine laboratory testing. In the absence of ample clinical experience, animal models are a valuable alternative source of information about the effect on bleeding. Specific settings are developed to reflect the effect of reversal agents.34 These clinical models may be more sensitive in measuring the reversal of the effect of NOACs than routine coagulation assays.35 In particular, measurement of the bleeding time (BT) in these animal models may be more sensitive for the reversal effect than measurement of coagulation tests.36-37 However, these animal models may only partially mimic the complex situation of critically-ill patients with life-threatening bleeding

## Reversal of dabigatran effects

CFCs such as 4-factor PCC did not normalize global coagulation tests including thrombin time (TT) and activated partial thromboplastin time (aPTT) in individuals pretreated with dabigatran.38 However, CFCs improved TG in plasma from healthy individuals pretreated with dabigatran ex-vivo.33, 35, 39-40 The reversal effect was dependent on the concentration of dabigatran and CFCs.35 In animal models CFCs dose-dependently and rapidly decreased bleeding time.35, 37, 41 However, there were inconsistent results with regard to reduction of blood loss.42 In a murine ICH model, a 4-factor PCC but not rFVIIa reduced the expansion of ICH and 24h-mortality.41 Recent data from a lethal porcine polytrauma model suggested that high-dose (50 IU/kg) but not low-dose PCC43 reversed the effects of dabigatran on blood loss and survival to a similar extent than idarucizumab, a specific antibody against dabigatran.44-45 With regard to human data, a few case reports/series addressing very different clinical scenarios reported varying outcomes after reversal of dabigatran-induced bleeding with CFCs.46-50 In case of bleeding in patients with renal impairment, additional haemodialysis may be essential for elimination of dabigatran.50

## Reversal of factor Xa inhibitor effects

CFCs such as 3- and 4-factor PCC with 50 U/kg at least partially reverse prothrombin time (PT) in most studies in healthy individuals pretreated with rivaroxaban.38, 51 rFVIIa may have the most prominent reversal effect on PT.52 Moreover, activated PCC and rFVIIa with the most prominent effects but also high-dose 3- and 4-factor PCCs improved TG tests ex vivo using plasma from healthy donors pretreated with rivaroxaban.39, 51, 53-54 However, contrasting data without an effect of PCCs on PT and TG tests using rivaroxaban-anticoagulated blood samples from healthy volunteers are also published.53 With regard to experimental animal data, 4-factor PCC did not consistently reduce bleeding time and did not reduce blood loss.36, 55 Activated 4-factor PCC and rFVIIa, with an eventually more pronounced effect, reduced BT in animals pretreated with rivaroxaban.55 36 In a mouse model, a 4-factor PCC and rFVIIa prevented excess intracranial haematoma expansion with rivaroxaban.56 Despite correction of clinical bleeding in animal models, the PT remained prolonged, suggesting that procoagulants may attenuate bleeding, but fail to normalise global coagulation tests.36, 56

4-factor PCC, activated 4-factor PCC and rFVIIa improved TG after pretreatment with apixaban.57 In a rabbit model, rFVIIa reduced the BT to some extent but did not reduce blood loss with apixaban. In this setting, a 4-factor PCC did not affect clinical bleeding.58

Activated 4-factor PCC and rFVIIa improved anti-Xa activity in edoxaban-containing blood samples.59 4-factor PCC and, most potently, activated 4-factor PCC and rFVIIa dose-dependently reversed reduced TG by edoxaban.15-16, 59-61 In a recent double-blind, randomized, placebo-controlled, 2-way crossover study with 110 healthy individuals 4-factor PCC dose-dependently reversed the effect of edoxaban on TG and bleeding duration after a punch biopsy, with complete reversal at 50 IU/kg.62 No prothrombotic event was observed. With regard to animal data, high dose 4-factor PCC, activated 4-factor PCC and rFVIIa significantly reduced BT and/or blood loss60, 63 without increasing the risk of thrombosis.60

## Supplemental Table 1. Pharmacologic properties of NOACs

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | **Dabigatran**  **(Pradaxa)** | **Rivaroxaban**  **(Xarelto)** | **Apixaban**  **(Eliquis)** | **Edoxaban**  **(Lixiana)** |
| Company | Boehringer Ingelheim | Bayer | BMS/Pfizer | Daiichi Sankyo |
| Direct inhibition of | FIIa | FXa | FXa | FXa |
| Standard dosage for atrial fibrillation | 150mg twice daily | 20mg once daily | 5mg twice daily | 60mg once daily |
| t max | 1-3h | 2-4h | 1-3h | 1-2h |
| Half life (t ½) | 12-17h | 5-9h young  11-13h elderly | 9-14h | 10-14h |
| Bioavailability | 6.5% | 80% | 60% | 62% |
| Renal elimination | 80% | 35% | 25% | 35% |
| Dialysis? | yes | no | no | no |
| Drug interactions |  |  |  |  |
| P-glycoprotein involved | yes | yes | yes | yes |
| CYP3A4 involved | no | yes | yes | minimal |

## Supplemental Table 2 – Effects of NOACs in *ex vivo/in vitro* and *animal* studies

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | Dabigatran | Rivaroxaban | Apixaban | Edoxaban |
| Global coagulation tests | - Prolonged5, 64 | - Prolonged13, 65 | - Prolonged (modest)23, 66 | - Prolonged25-27 |
| Thrombus formation | - Thrombus formation ↓67 - Thrombin generation ↓5 | - Thrombus formation ↓68  - Thrombin generation ↓19 | - Thrombus formation ↓23 - Thrombin generation ↓23 | - Thrombus formation ↓25, 27, 69 - Thrombin generation ↓26-27 |
| Platelet aggregation | - Thrombin- and TF-induced aggregation↓5- No effect on other platelet-stimulating mechanisms5, 70 | - Platelet-induced thrombin generation ↓71 - Thrombin- and TF-induced aggregation↓70-73  - No effect on other platelet-stimulating mechanisms5, 70, 73 | - TF-induced aggregation ↓ - No effect on other platelet-stimulating mechanisms70 | - Weak inhibition of thrombin-induced aggregation25 - No impact on other mechanisms25 |
| Animal data | - Thrombus formation ↓74-75- No prolonged bleeding time (with normal doses)76- No increased bleeding tendency74 | - Thrombus formation ↓13, 77 - Modest increase in bleeding time76-77 | - Thrombus formation ↓78- Modest increase in bleeding time23, 76, 78 | - Thrombus formation ↓25- No prolonged bleeding time (with normal doses)25 |

## Supplemental Table 3. Experimental data about the reversal of NOAC effects with coagulation factor concentrates

## Table 3A. Reversal of routine laboratory tests

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | DABIGATRAN\* | RIVAROXABAN\*\*\* | APIXABAN\*\* | EDOXABAN\*\* |
| 3-factor PCC | No[35](#_ENREF_1) | - Partial reversal of PT ( no data on dose-dependency)[51](#_ENREF_2" \o "Levi, 2014 #124)  - No effect on anti-FXa activity[51](#_ENREF_2) | ND | ND |
| 4-factor PCC | No35, 38 | - Partial reversal of PT (dose-dependent effect; one negative study)[38, 51-52, 54](#_ENREF_2" \o "Levi, 2014 #124)  - No effect on anti-FXa activity[51](#_ENREF_2) | ND | - Dose-dependent effect on bleeding time and blood loss (human data)62 |
| Activated 4-factor PCC | No[35](#_ENREF_1) | - Partial reversal of PT (dose-dependent effect)[54](#_ENREF_4" \o "Perzborn, 2014 #100)  - No data on anti-FXa activity | ND | - Reversal of PT (not dose-dependent)[59](#_ENREF_6" \o "Halim, 2014 #206)  - Dose-dependent, modest reversal of FX activity59 |

\* aPTT, no data on TT or ECT \*\* Anti-FXa activity \*\*\* PT, anti-FXa activity ND, no data available; PCC prothrombin complex concentrate

## Table 3B. Reversal of thrombin generation

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | DABIGATRAN | RIVAROXABAN | APIXABAN | EDOXABANa |
| 3-factor PCC | Yes (dose-dependent effect)[35](#_ENREF_1) | Yes (dose-dependent effect)[51](#_ENREF_2) | ND\* | ND\* |
| 4-factor PCC | Yes (dose-dependent effect)35, 39 | Inconsistent data (dose-dependent effect)38-39, 51, 53-54 | Yes (no data on dose-dependency) [57](#_ENREF_8) | Yes (dose-dependent effect)62, [60](#_ENREF_9) |
| Activated 4-factor PCC | Yes (dose-dependent effect)35, 39 | Yes (dose-dependent effect)39, 54 | Yes (no data on dose-dependency)[57](#_ENREF_8) | Yes (dose-dependent effect)[60](#_ENREF_9) |
| rFVIIa | Yes35, 39 | Yes39, 54 | Yes (no data on dose-dependency)[57](#_ENREF_8) | Yes[60](#_ENREF_9) |

Reversal was assumed to be “yes” if there was a consistent effect using at least one of the following tests: clotting time, thrombin generation (including lag time, maximum concentration, endogenous thrombin potential), prothrombin time, \*ND, no data available; PCC prothrombin complex concentrate

## Table 3C. Reversal of bleeding in animal models

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | DABIGATRAN | RIVAROXABAN | APIXABAN | EDOXABAN |
| 3-factor PCC | - Bleeding time ↓[35](#_ENREF_1)  - No data on blood loss or ICH | ND\* | ND\* | ND\* |
| 4-factor PCC | - Inconsistent data regarding bleeding time35, 37, 41-42  - Inconsistent data regarding blood loss[37, 41-42](#_ENREF_11)  - ↓ Hematoma size in ICH model and improved survival (with high doses)33, 79 | - Inconsistent data regarding bleeding time36, 55  - No reduced blood loss[55](#_ENREF_15)  - ↓ Hematoma size in ICH model (dose-dependent)[56](#_ENREF_17) | - No effect on bleeding time[58](#_ENREF_14)  - No reduced blood loss[58](#_ENREF_14)  - No data on ICH | - Bleeding time ↓63  - Blood loss ↓63  - No data on ICH |
| Activated 4-factor PCC | - Bleeding time ↓35, 42  - No reduced blood loss[42](#_ENREF_11)  - No data on ICH | - Bleeding time ↓[36](#_ENREF_16)  - No data on blood loss or ICH | ND\* | - Bleeding time ↓[60](#_ENREF_9)  - No data on blood loss or ICH |
| rFVIIa | - Inconsistent data regarding bleeding time35, 42  - No reduced blood loss[42](#_ENREF_11)  - No reduction of hematoma size in ICH model[41](#_ENREF_13) | - Bleeding time ↓36, 55  - No reduced blood loss[55](#_ENREF_15)  -↓ Hematoma size in ICH[56](#_ENREF_17) | - Bleeding time ↓(partially)[58](#_ENREF_14)  - No reduced blood loss[58](#_ENREF_14)  - No data on ICH | - Bleeding time ↓[60](#_ENREF_9)  - No data on blood loss or ICH |

\*ND, no data available; PCC prothrombin complex concentrate; ICH intracranial haemorrhage

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