Translational Success Stories highlight how basic discoveries have led to clinical advances, such as the use of new drugs or diagnostic modalities in patients. This initiative reflects the renewed emphasis of our Journal on translational research. It is hoped that these articles will stimulate efforts to translate basic insights into clinical practice.

Oral Direct Factor Xa Inhibitors

Calvin H. Yeh, James C. Fredenburgh, Jeffrey I. Weitz

Abstract: Vitamin K antagonists, such as warfarin, have been the mainstay of oral anticoagulation for many decades. Although effective, warfarin has numerous limitations, including a variable dose requirement from patient to patient because of differences in dietary vitamin K intake, common genetic polymorphisms, and multiple drug interactions that affect its pharmacodynamics and metabolism. Consequently, warfarin requires frequent monitoring to ensure that a therapeutic anticoagulant effect has been achieved because excessive anticoagulation can lead to bleeding, and because insufficient anticoagulation can result in thrombosis. Such monitoring is burdensome for patients and physicians and is costly for the health care system. These limitations have prompted the development of new oral anticoagulants that target either factor Xa or thrombin. Although the path to the development of these drugs has been long, the new drugs are at least as effective and safe as warfarin, but they streamline clinical care because they can be administered in fixed doses without routine coagulation monitoring. This article focuses on rivaroxaban, apixaban, and edoxaban, the oral factor Xa inhibitors in the most advanced stages of development. After 20 years of discovery research, these agents are already licensed for several indications. Thus, the long path to finding replacements for warfarin has finally reached fruition. Therefore, development of the oral factor Xa inhibitors represents a translational science success story. (Circ Res. 2012;111:1069-1078.)

Key Words: apixaban ■ edoxaban ■ factor Xa inhibitors ■ heparin ■ rivaroxaban ■ thrombosis ■ warfarin

Anticoagulants are widely used for the prevention and treatment of venous and arterial thrombosis. Both parenteral and oral anticoagulants are available. Parenteral anticoagulants, which are administered by intravenous or subcutaneous injection, are mainly used for short-term prevention or for initial treatment of thrombosis, whereas oral anticoagulants are preferred for long-term management.1 The prototypical parenteral and oral anticoagulants are heparin and warfarin, respectively; these agents have been on the market for decades. Although effective, heparin and warfarin have limitations that complicate their use.2

Heparin acts as an anticoagulant by activating antithrombin and accelerating the rate at which it inhibits thrombin, factor (f) Xa and multiple other upstream coagulation enzymes.3 In addition to its requirement for parenteral administration, the limitations of heparin include its poor bioavailability after subcutaneous injection and short half-life when administered intravenously, which explains why heparin usually is administered via a continuous intravenous infusion. Not only is such treatment cumbersome but also the anticoagulant response to heparin is variable from patient to patient, which necessitates frequent coagulation monitoring to ensure that a therapeutic anticoagulant effect has been achieved. Finally, there is a risk of heparin-induced thrombocytopenia, a life-and limb-threatening immunological reaction in up to 5% of the heparin-treated patients.3

Low-molecular-weight heparins (LMWHs), which were introduced in the 1980s, overcome many of the limitations of heparin. With greater bioavailability after subcutaneous injection, longer half-lives, and more predictable anticoagulant responses, LMWHs can be administered by subcutaneous injection in fixed or weight-adjusted doses on a once or twice daily basis and coagulation monitoring is rarely required. In addition, the risk of heparin-induced thrombocytopenia is lower with LMWHs than with heparin. Consequently, LMWHs are safer and more convenient to administer than heparin, and with self-injection, LMWHs enable out-of-hospital
prophylaxis or treatment. For these reasons, LMWHs have replaced heparin for most indications.2

Whereas heparin inhibits FXa and thrombin to a similar extent, LMWHs have greater inhibitory activity against FXa, an observation that first brought attention to FXa as a potential target for new anticoagulants.4 The interest in FXa as a target was solidified with the introduction of fondaparinux, a synthetic analog of the unique pentasaccharide sequence in heparin and LMWHs that mediates their interaction with antithrombin.5,6 In contrast to heparin and LMWHs, fondaparinux has no inhibitory activity against thrombin and mainly targets FXa, although it also inhibits FIXa.7,8 Despite the absence of activity against thrombin, fondaparinux was at least as effective as heparin or LMWHs for prevention or treatment of venous thromboembolism (VTE),9 or prevention of recurrent ischemia in patients with acute coronary syndrome (ACS).10 These observations highlighted the suitability of FXa as a target for new anticoagulants and prompted the development of direct FXa inhibitors, small molecules that bind to FXa and inhibit its activity without the involvement of plasma factors. With proof-of-principle initially provided by FXa inhibitors isolated from hematophagous organisms,11,12 attention then focused on the development of synthetic agents that could be administered orally, with the ultimate goal being the replacement of warfarin with drugs that would streamline the extended prevention and treatment of thrombosis. The first new oral anticoagulant to be licensed, albeit briefly, was ximelagatran, a thrombin inhibitor. Although ximelagatran was withdrawn from the market because of potential hepatic toxicity, the results with this agent, which was administered in fixed doses without coagulation monitoring, illustrated the potential of targeted anticoagulant therapy. With the subsequent licensing of oral FXa inhibitors, we now have agents that are at least as effective as warfarin but are easier to administer.13–15 Consequently, the development of oral FXa inhibitors is a shining example of a translational science success story.

Why has replacement of warfarin been the "holy grail" of anticoagulant development? Although effective, warfarin and other vitamin K antagonists have numerous limitations.16,17 Warfarin has a slow onset of action because its anticoagulant activity depends on clearance of the vitamin K–dependent clotting proteins and replacement with factors with impaired function, a process that takes several days to occur. Because of its slow onset of action, warfarin must be overlapped with a rapidly acting parenteral anticoagulant when treatment is initiated in patients with established thrombosis or in patients at high risk for thromboembolic events. In addition, warfarin dosing is problematic because dietary vitamin K intake, common genetic polymorphisms, and multiple drug–drug interactions affect the pharmacodynamics and metabolism of warfarin. As a result, the warfarin dose requirement is highly variable both within and between patients, which necessitates frequent coagulation monitoring to ensure that a therapeutic anticoagulant effect is achieved. This is important because excessive anticoagulation increases the risk of serious bleeding, including intracranial hemorrhage, and thrombotic events can occur if the level of anticoagulation is subtherapeutic. Consequently, optimal warfarin management requires the institution of expensive systems that include specialized anticoagulation clinics, which are often staffed by well-trained pharmacists or nurses, point-of-care testing facilities, computerized warfarin-dosing algorithms, and patient self-testing and self-management programs.18 Despite the benefits of these systems, many patients using long-term warfarin therapy do not have access to specialized care, and community-based studies indicate that the level of anticoagulation is frequently outside the therapeutic range, thereby placing patients at risk for thrombosis or bleeding.19 The complexities of warfarin therapy contribute to its underuse, particularly for stroke prevention in patients with atrial fibrillation (AF), or for prevention of mechanical valve thrombosis in developing countries where facilities for coagulation monitoring are lacking.20 These limitations highlight the need for new oral anticoagulants that are unaffected by dietary vitamin K intake, have few drug–drug interactions, and have a sufficiently wide therapeutic index that they can be administered in fixed doses without the need for routine coagulation monitoring.21 The new oral FXa inhibitors possess these characteristics, which is why their development is such a success story. Consequently, they will streamline long-term anticoagulation and the oral FXa inhibitors also will have the potential to increase the uptake of anticoagulant prophylaxis in eligible patients with AF, thereby reducing the risk of a disabling or fatal stroke.20

In this article, we highlight the role of FXa as the gatekeeper of coagulation, review the evidence supporting FXa as a suitable target for new anticoagulants, describe the pathway of development of synthetic direct FXa inhibitors from parenteral agents to orally active drugs, outline the pharmacological properties of the oral FXa inhibitors and compare and contrast them with those of warfarin, indicate how the results of recent clinical trials support the use of oral FXa inhibitors, and provide insight into the advantages and potential drawbacks of the oral FXa inhibitors.

**FXa: The Gatekeeper of Coagulation**

The generation of thrombin from its precursor prothrombin is the pivotal event in blood coagulation because thrombin is essential for hemostasis.22 Thus, insufficient thrombin generation can result in bleeding, whereas excess thrombin generation leads to thrombosis.23 Thrombin is generated via a complex series of proteolytic reactions that are initiated when cell-bound tissue factor, which is normally cryptic, becomes exposed at sites of vascular injury (Figure 1).24 Tissue factor binds circulating FVIIa to form extrinsic tenase, an enzyme complex that activates FIX and FX, thereby generating FXa and FIIa, respectively. Intrinsic tenase, the complex of FXa and FVIIa that assembles
Validation of FXa as a Target: Studies With Antistasin, TAP, and Fondaparinux

The first direct FXa inhibitors, antistasin and TAP, are naturally occurring proteins derived from hematophagous organisms. Antistasin, a 119 amino acid polypeptide, was isolated from the salivary glands of the Mexican leech Haementeria officinalis in 1987, whereas TAP, which is composed of 60 amino acids, was extracted from the soft tick Ornithodoros moubata in 1990. Both proteins are slow, tight-binding, Kunitz-type inhibitors that are specific for FXa and have inhibition constant ($K$) values in the range of 0.3 to 0.6 nM.11

The availability of natural and recombinant forms of antistasin and TAP enabled in vitro and in vivo studies aimed at better defining the role of FXa in thrombosis. Studies in vitro revealed that like fibrin-bound thrombin, thrombus-associated FXa also contributes to the procoagulant activity of thrombi.29 Heparin had limited activity against thrombus-associated FXa, which when incorporated into the prothrombinase complex is protected from inhibition by the heparin–antithrombin complex.30,31 In contrast, direct FXa inhibitors, such as TAP, inhibit FXa in prothrombinase and free FXa equally well, which is a finding that suggested that direct FXa inhibitors would be superior to indirect inhibitors, such as heparin or LMWH. Furthermore, the observation that TAP inhibited clot-induced fibrinopeptide A generation to a similar extent as hirudin, a potent inhibitor of thrombin, raised the possibility that the procoagulant activity of thrombi was driven by FXa-induced thrombin generation rather than preformed thrombin, a concept that would enable the use of FXa inhibitors for treatment of thrombosis.32 This concept was supported by the results of studies in vivo.33–37 Thus, when compared with heparin or hirudin in a variety of animal models of venous and arterial thrombosis, recombinant TAP was more effective than heparin and was at least as effective as hirudin but produced less bleeding. These and other observations prompted speculation that anticoagulants that target FXa would be safer than those that inhibit thrombin because FXa inhibitors do not preclude the generation of sufficient amounts of thrombin to effect hemostasis.

Despite the promising preclinical findings with antistasin and TAP, these agents were never assessed in humans. However, the suitability of FXa as a target was confirmed by large clinical trials with fondaparinux. When compared with enoxaparin, a LMWH, for thromboprophylaxis in patients undergoing hip or knee replacement or surgery for hip fracture, fondaparinux exhibited superior efficacy but was associated with producing less bleeding. These and other observations prompted speculation that anticoagulants that target FXa would be safer than those that inhibit thrombin because FXa inhibitors do not preclude the generation of sufficient amounts of thrombin to effect hemostasis.

With the unique position of FX in the coagulation pathway and the critical role of FXa as the effector of thrombin generation, FXa has emerged as an attractive target for new anticoagulants. Although inhibition of thrombin generation is an obvious strategy for prevention of thrombosis, confirmation was needed that drugs that target FXa would be adequate for treatment of established thrombosis when thrombin generation already has occurred. Such confirmation was provided by the results of preclinical studies with antistasin and tick anticoagulant peptide (TAP), naturally occurring FXa inhibitors isolated from hematophagous organisms, and the results of clinical trials with fondaparinux, an antithrombin-dependent inhibitor of FXa.

on the platelet surface, also activates FX, thereby amplifying FXa generation beyond that produced by extrinsic FX.25 Thus, FX is uniquely positioned at the convergence of both extrinsic and intrinsic tenase, and its activation is essential for the propagation of coagulation. FXa generated on the platelet surface binds FXa to form prothrombinase, the enzyme complex that converts prothrombin to thrombin.26 The rate of prothrombin activation by FXa generated in the prothrombinase complex is approximately 106-fold faster than that of FXa alone.27 Consequently, each molecule of FXa generates approximately 1000 molecules of thrombin; this is a critical amplification step in coagulation that results in a burst of thrombin generation at sites of injury, thereby facilitating rapid hemostatic plug formation.28

With the unique position of FX in the coagulation pathway and the critical role of FXa as the effector of thrombin generation, FXa has emerged as an attractive target for new anticoagulants. Although inhibition of thrombin generation is an obvious strategy for prevention of thrombosis, confirmation was needed that drugs that target FXa would be adequate for treatment of established thrombosis when thrombin generation already has occurred. Such confirmation was provided by the results of preclinical studies with antistasin and tick anticoagulant peptide (TAP), naturally occurring FXa inhibitors isolated from hematophagous organisms, and the results of clinical trials with fondaparinux, an antithrombin-dependent inhibitor of FXa.
studies with fondaparinux validated fXa as a suitable target for both prevention and treatment of venous and arterial thrombosis. With this information, the race to develop small-molecule, orally available, direct fXa inhibitors was on.

Development of Synthetic Oral Direct fXa Inhibitors
Initial identification of fXa inhibitors relied on high-throughput screening assays based on inhibition of fXa-induced chromogenic or clotting activity. The first leads contained isoxazoline or isoxazole derivatives, such as benzamidine, guanidine, or naphthylamide, which are thought to mimic Glu-Gly-Arg, the sequence in prothrombin that is recognized by fXa.42 The first such compound was DX-9065a, which contained highly basic amidine groups as nonpeptidic mimetics of the Arg residue in the prothrombin recognition sequence.43 Because it exploited interactions with the S4 and S1 subsites of the active site (Figure 2), DX-9065a was specific for fXa and inhibited the enzyme with a $K_i$ value of 46 nmol/L.43 After establishing its antithrombotic activity in a variety of animal models of venous and arterial thrombosis,44–46 DX-9065a was compared with heparin in a small, phase II, dose-finding study in ACS patients.47 Although the results of the study were promising, development of DX-9065a was halted because the oral bioavailability of the drug in humans was only 2% to 3% because of its highly basic amidine content.42 Nonetheless, DX-9065a provided the groundwork for future orally active fXa inhibitors.

Once lead compounds were identified through high-throughput screening, a structure-based approach was used to characterize their interaction with fXa. Using molecular modeling based on the crystallographic structure of fXa in complex with the first generation of fXa inhibitors, such as DX-9065a, quantitative structure–activity relationship analyses were used to determine how modifications affected the potency of the leads.48–51 These techniques revealed that the highly basic amidine moiety in the P1 position of isoxazoline derivatives participates in a two-component interaction with the carboxylate group of the Asp residue at position 189 in the floor of the S1 pocket that flanks the catalytic triad of fXa. The first component involved a Coulombic interaction between positive and negative charges (so-called ion pairing), whereas the second reflected a bidentate hydrogen bond.52 Elimination of either of these interactions increased the $K_i$ by one to two orders of magnitude.52 Building on this information, the amidine group in the P1 position was replaced with nonbasic moieties in an attempt to increase oral bioavailability.53 Various linker or P4 elements were then added to overcome the subsequent loss in potency by maximizing interactions at the S4 site. A decade of development led to the synthesis of several orally active fXa inhibitors.54 Of these, rivaroxaban (Bayer Healthcare),13 apixaban (Bristol-Myers Squibb),14 and edoxaban (Daiichi Sankyo)15 are the drugs in the most advanced stages of development. All of these agents contain nonbasic moieties in the P1 position: chlorothiophene in rivaroxaban, methoxyaryl in apixaban, and chloro-substituted pyridine rings in edoxaban54 (Figure 3). With the drugs developed and their antithrombotic activities established in animal models, testing in humans was performed to determine their pharmacological properties, to identify appropriate doses, and to compare their efficacy and safety with those of conventional anticoagulants.

Pharmacological Properties of Oral fXa Inhibitors
In contrast to warfarin, which targets multiple coagulation factors, the oral fXa inhibitors are specific for fXa. The pharmacological properties of the oral fXa inhibitors differ from those of warfarin in several ways. Because the fXa inhibitors are active compounds with good oral bioavailability, they have a rapid onset of action, thereby eliminating the need for bridging therapy with a parenteral anticoagulant in most situations.55,56 Unlike warfarin, dietary vitamin K intake has
no effect on the activity of the oral fXa inhibitors, and the potential for drug–drug interactions is much lower with the new agents than with warfarin. These features together with the predominantly dose-dependent pharmacokinetic and pharmacodynamic profiles of the oral fXa inhibitors result in a predictable anticoagulant response after fixed-dose administration. Because of this feature, routine coagulation monitoring is unnecessary. Finally, all the oral fXa inhibitors exhibit a dual mechanism of excretion with both a renal and a fecal component, thereby attenuating the risk of drug accumulation in patients with renal impairment. The pharmacological profiles of rivaroxaban, apixaban, and edoxaban are compared in Table 1. With these properties defined, the next step was to evaluate the oral fXa inhibitors in clinical trials.

### Clinical Trials With Oral fXa Inhibitors

All the oral fXa inhibitors followed similar development pathways. Initially, the drugs were compared with enoxaparin for thromboprophylaxis after elective hip or knee arthroplasty; studies that provided proof-of-principle of the antithrombotic efficacy of the new agents and identified doses that had favorable benefit-to-risk profiles in the postoperative setting. Based on the results of phase II studies in VTE treatment (which were performed with rivaroxaban and apixaban or stroke prevention in AF (the pathway followed with edoxaban), appropriate drug doses were chosen for phase III evaluation for VTE treatment and for stroke prevention in AF. Rivaroxaban and apixaban also were evaluated for thromboprophylaxis in medically ill patients and for prevention of recurrent ischemia in stabilized ACS patients. Table 2 lists the phase III, randomized, clinical trials performed for the various indications with the oral fXa inhibitors and highlights the large number of patients included in the development programs with these agents. An overview of the results of these trials is provided.

### Rivaroxaban

For thromboprophylaxis, rivaroxaban (10 mg once daily) was compared with enoxaparin in two trials in patients undergoing elective hip arthroplasty (Regulation of Coagulation in Orthopedic Surgery to Prevent Deep Vein Thrombosis and Pulmonary Embolism, RECORD-1 and –2) and in 2 trials in those undergoing elective knee arthroplasty (RECORD-3 and –4). The same dose of rivaroxaban also was compared with enoxaparin for thromboprophylaxis in medically ill patients (Venous Thromboembolic Event Prophylaxis in Medically Ill Patients, MAGELLAN). In the orthopedic setting, the rate of VTE, which included asymptomatic DVT detected by routine venography, as well as symptomatic DVT or pulmonary embolism, was significantly lower with rivaroxaban than with enoxaparin, and the two agents had similar safety profiles. These findings formed the basis for the licensing of rivaroxaban in the United States for VTE prophylaxis after elective hip or knee arthroplasty. Although extended prophylaxis with rivaroxaban was more effective than a shorter period of prophylaxis with enoxaparin at reducing the rate of symptomatic VTE or asymptomatic DVT detected by routine lower extremity ultrasonography in medically ill patients, rivaroxaban produced more bleeding, a finding that limits its use in this setting.

### Table 1. Pharmacological Properties of Warfarin, Rivaroxaban, Apixaban, and Edoxaban

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Warfarin</th>
<th>Rivaroxaban</th>
<th>Apixaban</th>
<th>Edoxaban</th>
</tr>
</thead>
<tbody>
<tr>
<td>Target</td>
<td>VKORC1</td>
<td>fXa</td>
<td>fXa</td>
<td>fXa</td>
</tr>
<tr>
<td>Molecular weight, Da</td>
<td>308</td>
<td>436</td>
<td>460</td>
<td>548</td>
</tr>
<tr>
<td>Bioavailability, %</td>
<td>100</td>
<td>80</td>
<td>≈50</td>
<td>≈50</td>
</tr>
<tr>
<td>Dosing</td>
<td>Once daily</td>
<td>Once or twice daily</td>
<td>Twice daily</td>
<td>Once daily</td>
</tr>
<tr>
<td>Time-to-peak effect</td>
<td>4–5 d</td>
<td>2–3 h</td>
<td>1–2 h</td>
<td>1–2 h</td>
</tr>
<tr>
<td>Half-life, h</td>
<td>40</td>
<td>7–11</td>
<td>15</td>
<td>5–11</td>
</tr>
<tr>
<td>Renal clearance, %</td>
<td>None</td>
<td>33 (66)*</td>
<td>25</td>
<td>35</td>
</tr>
<tr>
<td>Monitoring</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>Interactions</td>
<td>Multiple</td>
<td>3A4/P-gp</td>
<td>3A4/P-gp</td>
<td>P-gp</td>
</tr>
<tr>
<td>Antidote</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
</tr>
</tbody>
</table>

3A4 indicates cytochrome P450 3A4 enzyme; P-gp, P-glycoprotein; VKORC1, C1 subunit of the vitamin K epoxide reductase enzyme.

*33% cleared as unchanged drug and 33% as inactive metabolites.

Rivaroxaban was compared with conventional anticoagulant therapy for treatment of DVT or pulmonary embolism in the Oral Direct Factor Xa Inhibitor Rivaroxaban in Patients With Acute Symptomatic Deep-Vein Thrombosis or Pulmonary Embolism (EINSTEIN-DVT) and pulmonary embolism (EINSTEIN-PE) trials, respectively. For these indications, rivaroxaban was administered at an initial dose of 15 mg twice daily for 3 weeks, and the dose was reduced to 20 mg once daily thereafter. Rivaroxaban was noninferior to conventional anticoagulation therapy for prevention of recurrent symptomatic VTE in both patient groups and was associated with similar or lower rates of major bleeding. Based on these findings, rivaroxaban is currently undergoing evaluation for this indication by the Food and Drug Administration in the United States and is already licensed for treatment of DVT in Europe and Canada.

In patients with AF, rivaroxaban (20 mg once daily and 15 mg once daily for those with impaired renal function) was compared with warfarin for prevention of stroke or systemic embolism in AF patients (Rivaroxaban Once Daily Oral Direct Factor Xa Inhibition Compared with Vitamin K Antagonism for Prevention of Stroke and Embolism Trial in Atrial Fibrillation, ROCKET-AF), more than half of whom had experienced a previous stroke or other cardioembolic event. Rivaroxaban was noninferior to warfarin, with annual rates of stroke or systemic embolism of 1.7% and 2.2%, respectively, and was associated with a similar rate of major bleeding (5.55% and 5.42%, respectively). However, the annual rate of hemorrhagic stroke was significantly lower with rivaroxaban than with warfarin (0.26% and 0.44%, respectively; $P=0.024$), as was the rate of fatal bleeding (0.2% and 0.5%, respectively; $P=0.003$), mainly reflecting the reduction in the rate of intracranial bleeding (0.5% and 0.7%, respectively; $P=0.02$). Based on these data, rivaroxaban has been licensed in the United States as an alternative to warfarin for stroke prevention in AF.

Low-dose rivaroxaban (2.5 or 5 mg twice daily) was compared with placebo for prevention of recurrent ischemic events in stabilized ACS patients (Anti-Xa Therapy to Lower Cardiovascular Events in Addition to Aspirin with/without Thienopyridine Therapy in Subjects with Acute Coronary
Syndrome, ATLAS-2),\(^{73}\) most of whom were also receiving dual antiplatelet therapy with aspirin and clopidogrel. Both doses of rivaroxaban significantly reduced the rate of the primary efficacy end point, a composite of cardiovascular death, myocardial infarction, or stroke. Although both doses of rivaroxaban increased the rate of major bleeding (including intracranial bleeding) compared with placebo, the lower-dose rivaroxaban regimen (2.5 mg twice daily) was associated with less bleeding than the higher-dose regimen. Despite the increased risk of bleeding, the lower-dose rivaroxaban regimen produced a significant reduction in cardiovascular death and death from any cause compared with placebo. The Food and Drug Administration is currently evaluating rivaroxaban for ACS indication.

**Apixaban**

For thromboprophylaxis, apixaban (2.5 mg twice daily) was compared with enoxaparin in two trials in patients undergoing elective knee arthroplasty (The Apixaban Dosed Orally Versus Anticoagulation with Injectable Enoxaparin to Prevent Venous Thromboembolism, ADVANCE-1 and -2)\(^{74,75}\) and in one trial of those having elective hip arthroplasty (ADVANCE-3).\(^{76}\) In patients undergoing knee or hip replacement surgery, apixaban was superior for prevention of VTE when enoxaparin was used at a dose of 40 mg once daily, but it was associated with a similar rate of VTE in patients undergoing knee arthroplasty when the enoxaparin dose was 30 mg twice daily. VTE in these studies included asymptomatic DVT detected by routine venography, as well as symptomatic events. Rates of bleeding were numerically lower with apixaban than with enoxaparin, but the differences were not statistically significant except when apixaban was compared with the higher-dose enoxaparin regimen. Based on these data, apixaban is licensed for thromboprophylaxis after elective hip or knee arthroplasty in Europe and Canada, but not in the United States.

An extended course of apixaban (2.5 mg twice daily) also was compared with shorter-term thromboprophylaxis in medically ill patients (Apixaban Dosing to Optimize Protection from Thrombosis, ADOPT).\(^{77}\) The rates of the primary efficacy end point, a composite of VTE-related death, symptomatic VTE, or asymptomatic DVT detected by routine lower extremity ultrasonography, were similar with apixaban and enoxaparin. Although rates of major bleeding were low, there was significantly more bleeding with apixaban. Therefore, the results of this trial do not support the use of apixaban for this indication.

Two trials assessing the use of apixaban for treatment of VTE are underway. The first trial compares apixaban with conventional anticoagulant therapy for treatment of acute VTE (Apixaban after the Initial Management of Pulmonary Embolism and Deep Vein Thrombosis with First-Line Therapy, AMPLIFY). In this trial, apixaban is administered at a dose of 10 mg twice daily; after 1 week, the dose is reduced to 5 mg twice daily. The second trial compares two doses of apixaban (2.5 or 5 mg twice daily) with placebo for prevention of recurrent VTE in patients who have received at least a 6-month course of anticoagulant therapy for an initial VTE event (AMPLIFY-Extension). The hypothesis underlying this trial is that the lower-intensity apixaban regimen may retain most of the benefit of the high-dose regimen but may be associated with less bleeding.

Apixaban has been evaluated for stroke prevention in AF in two trials. The first compared apixaban with aspirin in patients with AF who were unwilling or unable to use warfarin (Apixaban Versus Acetylsalicylic Acid (ASA) to Prevent Strokes, AVERROES),\(^{83}\) whereas the second compared apixaban with warfarin (Apixaban for Reduction in Stroke and Other Thromboembolic Events in Atrial Fibrillation, ARISTOTLE).\(^{79}\) The AVERROES trial was stopped early when it was evident that apixaban was superior to aspirin for prevention of stroke or systemic embolism (annual rates of 1.6% and 3.7%, respectively; \(P<0.001\)) and was associated with a similar rate of major bleeding (1.4% and 1.2%, respectively; \(P=0.57\)). Apixaban

### Table 2. Landmark Phase III Clinical Trials and the Number of Patients Enrolled for Rivaroxaban, Apixaban, and Edoxaban for Various Indications

<table>
<thead>
<tr>
<th>Indication</th>
<th>Condition</th>
<th>Rivaroxaban</th>
<th>Apixaban</th>
<th>Edoxaban</th>
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<tr>
<td>VTE prevention</td>
<td>THA</td>
<td>RECORD-1</td>
<td>4542</td>
<td>ADVANCE-3</td>
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<td></td>
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<td>TKA</td>
<td>RECORD-3</td>
<td>2531</td>
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<td></td>
<td>RECORD-4</td>
<td>3148</td>
<td>ADVANCE-2</td>
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<td></td>
<td>Medically ill</td>
<td>MAGELLAN</td>
<td>8101</td>
<td>ADOPT</td>
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<td>VTE treatment</td>
<td>DVT</td>
<td>EINSTEIN-DVT</td>
<td>3449</td>
<td>AMPLIFY</td>
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<td>PE</td>
<td>EINSTEIN-PE</td>
<td>4833</td>
<td>AMPLIFY-EXT</td>
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<td></td>
<td>VTE Extended</td>
<td>EINSTEIN-EXT</td>
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<tr>
<td>AF</td>
<td>Versus aspirin</td>
<td>ROCKET-AF</td>
<td>14264</td>
<td>AVERROES</td>
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<td></td>
<td>Versus warfarin</td>
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<td></td>
<td>ARISTOTLE</td>
</tr>
<tr>
<td>ACS</td>
<td>Versus Placebo</td>
<td>ATLAS-A CS2-TIMI-51</td>
<td>15526</td>
<td>APPRAISE-2</td>
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<tr>
<td>Total patients</td>
<td></td>
<td>60100</td>
<td>60077</td>
<td>29933</td>
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</table>

ACS indicates acute coronary syndrome; AF, atrial fibrillation; DVT, deep vein thrombosis; EXT, extension; PE, pulmonary embolism; STARS, Studying Thrombosis After Replacement Surgery; THA, total hip arthroplasty; TIMI, thrombolysis in myocardial infarction; TKA, total knee arthroplasty; VTE, venous thromboembolism.
also was superior to warfarin for prevention of stroke or systemic embolism (annual rates of 1.27% and 1.60%, respectively; \( P=0.01 \)) and produced significantly less major bleeding (0.96% and 1.69%, respectively; \( P<0.001 \)) and intracranial bleeding than warfarin (0.33% and 0.80%, respectively; \( P<0.001 \)), and it was associated with a significant reduction in all-cause mortality (3.52% and 3.94%, respectively; \( P=0.047 \)). Based on these studies, apixaban is undergoing consideration for licensing by the Food and Drug Administration for the AF indication and by other regulatory agencies worldwide.

Like rivaroxaban, apixaban also was compared with placebo in stabilized ACS patients (Apixaban for Prevention of Acute Ischemic and Safety Events, APPRAISE-2).\(^80\) The dose of apixaban used in this trial was 5 mg twice daily, the same dose that was evaluated for stroke prevention in patients with AF. The study was stopped early because there was no evidence of efficacy and apixaban was associated with a significant increase in major hemorrhage, including intracranial bleeding, compared with warfarin. Therefore, this trial does not support the use of apixaban in the ACS setting.

**Edoxaban**

Edoxaban is licensed in Japan for VTE prophylaxis after elective hip or knee arthroplasty based on the results of two trials comparing it with enoxaparin. Because the prophylactic dose of enoxaparin used in Japan is lower than that used elsewhere, these trials do not support the widespread use of edoxaban for this indication. However, two large, international, phase III, clinical trials with edoxaban are underway. Edoxaban is being compared with warfarin for treatment of VTE (Safety and Efficacy of Edoxaban in the Treatment and Prevention of Recurrent Thromboembolic Events in Patients with Deep-Vein Thrombosis and/or Pulmonary Embolism, HOKUSAI)\(^81\) and for stroke prevention in patients with AF (Effective Anticoagulation with Factor Xa Next Generation in Atrial Fibrillation, ENGAGE-AF).\(^82\) The dose of edoxaban used for VTE treatment is 60 mg once daily. For stroke prevention in AF, edoxaban is being evaluated at doses of 30 or 60 mg once daily and a dynamic dose-adjustment regimen is being used to maintain edoxaban drug exposures at a lower or higher level. Results from both trials are expected in 2013.

**Conclusions**

Vitamin K antagonists have been the mainstay oral anticoagulants since 1945. Now, because of the determined efforts of several major pharmaceutical companies, we have options. Thus, dabigatran etexilate, an oral thrombin inhibitor, was licensed in 2010, rivaroxaban was licensed in 2011, and apixaban will likely soon follow (Figure 4). The proof-of-principle that direct FXa inhibition was a viable strategy for anticoagulation came with studies of antistasin and TAP isolated from hematophagous ticks, whereas clinical evidence came from studies of fondaparinux. Synthesis of the orally available direct FXa inhibitors involved structure-based design, and the latest “brute force” technology was used to maintain potency with good oral bioavailability. The success of these drug development programs is highlighted by the results of the clinical trials (Table 3). Dabigatran, rivaroxaban, and apixaban, the agents that have completed phase III evaluation in the AF setting, have shown robust noninferiority to warfarin for prevention of stroke and systemic embolism, and higher-dose dabigatran (150 mg twice daily) and apixaban have even shown superiority. Furthermore, there is a consistent trend for a reduction in all-cause mortality and cardiovascular mortality with all three drugs. These efficacy advantages have been achieved without an increase in major
bleeding, and all of the new oral anticoagulants have been associated with a significant reduction in intracranial bleeding compared with warfarin. To be able to achieve these beneficial effects with drugs that are administered in fixed doses without routine coagulation monitoring represents an enormous step forward. The convenience of these agents over warfarin will enable stroke prevention in a greater proportion of AF patients, thereby reducing death and disability from stroke.

Although clinicians are tempted to compare results across trials, such comparisons are problematic because of differences in study design (open-label versus double-blind), risk profiles of patients enrolled in the trials, duration of follow-up, and quality of warfarin management. Instead, the most important decision a clinician can make is to provide anticoagulant prophylaxis to AF patients at risk. The choice of anticoagulant can then be made based on patient characteristics. Whether one agent is superior to another requires head-to-head trials, which are unlikely to be performed in the near future.

Where do we go from here? Despite the advances afforded by the new oral anticoagulants, questions remain. Data on how the new agents will perform in general use are lacking. This information is critical because adherence is difficult to assess in the absence of routine coagulation monitoring.

There are no specific antidotes for the new oral anticoagulants, which can be problematic if urgent reversal is necessary in preparation for surgery or in case of serious bleeding. The relatively short half-life of the oral FXa inhibitors obviates the need for an antidote in most situations. This is an advantage because if there is serious bleeding, the clinical utility of procoagulants, such as unactivated or activated prothrombin complex concentrates or recombinant activated FVII, remains uncertain. Although reversal agents are in development, these are several years away from approval. Until these are available, however, more information is needed about the optimal management of bleeding in patients using oral FXa inhibitors.

Monitoring the anticoagulant activity of the new oral anticoagulants is more problematic than monitoring warfarin. Warfarin is routinely monitored using the international normalized ratio, a coagulation test that is widely available and well-standardized. In contrast, the oral FXa inhibitors have variable effects on routine tests of coagulation and likely will need specialized monitoring assays to measure drug levels. The lack of simple monitoring tests may complicate management of patients before surgery or procedures or identification of the mechanisms of bleeding or thrombosis in patients using oral FXa inhibitors.

Although we still have much to learn, the oral FXa inhibitors are here to stay. From proof-of-principle to licensed drugs, these agents represent a translational science success story.

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References
Group. Effects of fondaparinux on mortality and reinfarction in patients
Lam PY, Clark CG, Li R, et al. Structure-based design of novel gua
Lynch JJ Jr, Sitko GR, Mellott MJ, Nutt EM, Lehman ED, Friedman
Hofman M, Monroe DM. Coagulation 2006: a modern view of hemo
Eisenberg PR, Siegel JE, Abendschein DR, Miletich JP. Importance of
Kubitza D, Becka M, Roth A, Mueck W. Dose-escalation study of the
Kubitza D, Becka M, Voith B, Zuehlsdorf M, Wensing G. Safety, phar
Quan ML, Ellis CD, He MY, Liauw AY, Lam PY, Rossi KA, Knabb
Maignan S, Mikol V. The use of 3D structural data in the design of spe
Nazaré M, Will DW, Huber R, Blankenship DT, Cardin AD, Kisiel W. Structure of human
drug disposition of factor Xa during thrombotic therapy markedly ie
Yeh et al Oral Direct Factor Xa Inhibitors 1077
Lam PY, Clark CG, Li R, et al. Structure-based design of novel gua
Nadarar M, Will DW, Huber R, Blankenship DT, Cardin AD, Kisiel W. Structure of human


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