CHAPTER NINE

Case History: Eliquis™ (Apixaban), a Potent and Selective Inhibitor of Coagulation Factor Xa for the Prevention and Treatment of Thrombotic Diseases

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1. INTRODUCTION

Despite substantial advances in the prevention and treatment of cardiovascular diseases, they continue to be the leading cause of death in developed countries and are increasingly a major cause of morbidity and mortality in developing countries as well. Although the underlying causes of cardiovascular

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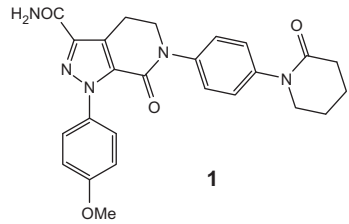

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Figure 9.1 Eliquis™ (Apixaban).

diseases involve multiple, complex mechanisms, a common component of the end stage of these diseases is thrombosis. Safe and effective antithrombotic drugs are therefore critical to effective treatment of cardiovascular diseases. Paradoxically, many patients who are at the highest risk for thromboembolic diseases, including the very elderly, are often less likely to be taking highly effective antithrombotic drugs due to risk of bleeding. Due to the large remaining unmet medical need, there has been intense activity to develop new antithrombotic therapies with improved efficacy and safety. In recent years, the strategy of targeting coagulation factor Xa (FXa) has received substantial clinical validation. Eliquis™ (Apixaban **1**, Fig. 9.1) is one of the first compounds acting by this mechanism to complete late-stage clinical studies and enter clinical practice. Along with other novel oral anticoagulants, it is poised to usher in a new era of antithrombotic therapy.

2. RATIONALE FOR TARGETING FXa

Vitamin K antagonists (VKAs), such as warfarin, are no longer the only available oral anticoagulants. Direct thrombin inhibitors, such as dabigatran etexilate, and FXa inhibitors, such as rivaroxaban and apixaban, have been developed and shown to be effective oral anticoagulants.[1–4] To date, there is no direct clinical evidence favoring one target over the other. However, there is some theoretical and preclinical evidence to support that the FXa mechanism may positively differentiate from thrombin as a preferred antithrombotic target.

First, as blood coagulation involves sequential steps of activation and amplification of coagulation proteins, generation of one molecule of FXa results in the production of hundreds of thrombin molecules.[2] In theory, therefore, inhibition of FXa may be more efficient in reducing fibrin formation than direct inhibition of thrombin activity. This principle is consistent with an

in vitro observation that inhibition of FXa produced a more effective sustained reduction of thrombus-associated procoagulant activity than inhibition of thrombin activity.[3] Second, inhibition of FXa is not thought to affect existing levels of thrombin and its activity. In addition, reversible FXa inhibitors might not completely suppress the production of thrombin. These small amounts of thrombin might be enough to activate high-affinity platelet thrombin receptors to preserve hemostasis. Early work from several laboratories provided experimental evidence from animal studies suggesting that the antithrombotic efficacy of FXa inhibitors is accompanied by a lower risk of bleeding when compared with thrombin inhibitors.[4–8] In summary, inhibition of FXa may represent an attractive approach compared with thrombin inhibition for effective and safe antithrombotic therapy. However, head-to-head clinical studies to validate this hypothesis have not been performed.

3. MEDICINAL CHEMISTRY EFFORTS CULMINATING IN APIXABAN

3.1. Factor Xa program objectives

As we believed that Factor Xa had the hallmarks of a target that would positively differentiate from anticoagulant standard of care, our discovery objective was to continuously deliver compounds until an optimal compound was in full development. This was driven by our strong belief that a high-quality factor Xa inhibitor would be transformational. The objective was to optimize for the right balance of efficacy and safety. The goal of the program was to identify potent, highly selective noncovalent FXa inhibitors with good oral bioavailability ($>20\%$) and a half-life suitable for either twice daily (BID) or once daily (QD) dosing with low peak/trough to minimize the potential for bleeding liabilities. *In vivo*, the compounds were required to demonstrate efficacy in preclinical thrombosis models. The ideal candidate would also not have drug–drug or food interactions, particularly given that these are issues with warfarin.

3.2. Early preclinical leads

When the medicinal chemistry program began in the mid-1990s, the only published FXa inhibitors were dibasic compounds which were not orally bioavailable (Fig. 9.2) such as **2** (DABE), **3** (BABCH), and **4** (DX-9065a) ($K_i = 570$, 13, and 41 nM, respectively).[9–11] The potency of these compounds resulted from a strong interaction between the amidine of

the inhibitor with the S1 Asp189 residue, and a π-cation interaction between the hydrophobic residues in the S4 subsite and the remaining basic functionality.[10]

At the genesis of our program, homology models and the X-ray coordinates for the FXa dimer were used extensively to design a number of dibasic FXa inhibitors.[12,13] An initially designed compound, ketone **5**, though a weak inhibitor of FXa ($K_i = 5100$ nM) was rapidly improved to **6** ($K_i = 34$ nM), by the introduction of an ester group.[12] With the aid of molecular modeling, these compounds evolved into amidine-based benzimidazoles such as compound **7** ($K_i = 140$ nM, Fig. 9.3). The SAR was extended to include several additional scaffolds such as indole, indoline, and phenylpyrrolidine, all conferring improved FXa activity, albeit with poor selectivity over trypsin and no oral bioavailability.[13]

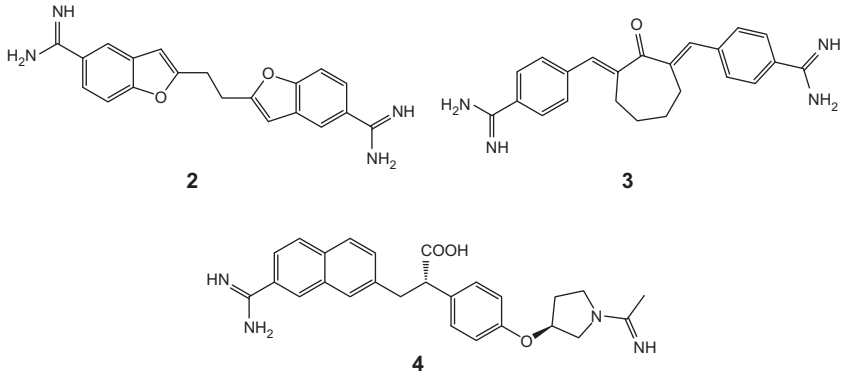


Figure 9.2 Published dibasic benzamidine FXa inhibitors.

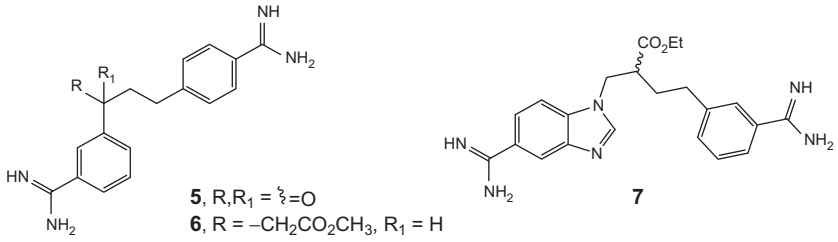


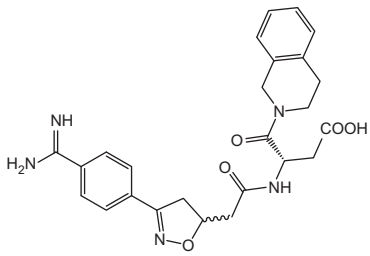
Figure 9.3 Early bis-benzamidine leads.

3.3. Screening library hits—The discovery of the isoxazoline scaffold

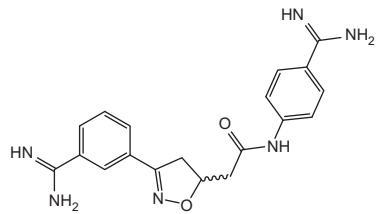
The second and more innovative approach resulted in a "focused screening" strategy which was coupled early with structure-based design to drive affinity. We recognized that the peptide sequence of ligands for the GPIIb/IIIa receptor Arg–Gly–Asp (RGD) and the two prothrombin cleavage sequences for FXa, namely, Glu–Gly–Arg (EGR), though reversed, shared some similarity. Based on these observations, and because known GPIIb/IIIa receptor antagonists contain a benzamidine group which is also found in FXa inhibitors such as **5**–**7**, our internal proprietary collection of small molecule GPIIb/IIIa antagonists was screened against FXa.[14]

This effort led to the identification of a weak isoxazoline inhibitor **8** ($K_i = 38.5$ μM; Fig. 9.4). Not discouraged by the weak affinity of **8**, lead optimization was jump-started by expeditiously improving affinity to subnanomolar levels by enhancing hydrophobic interactions in the S1 and S4 pockets.

Replacement of the aspartate residue in **8** with a second benzamidine afforded compound **9** ($K_i = 1.4$ μM), providing ~30-fold improvement in FXa affinity. Direct substitution of the carboxamide group onto the isoxazoline core, followed by substitution with a geminal carbonyl group designed to hydrogen bond to



8 (screening hit)



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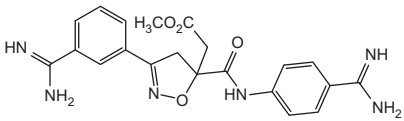

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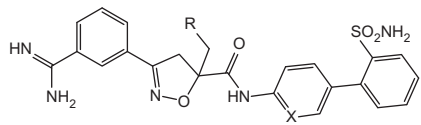

11 R = CO_2CH_3, X = CH (–) enant.
12 R = N-tetrazole, X = N (–) enant.

Figure 9.4 Optimizing isoxazoline analogs.

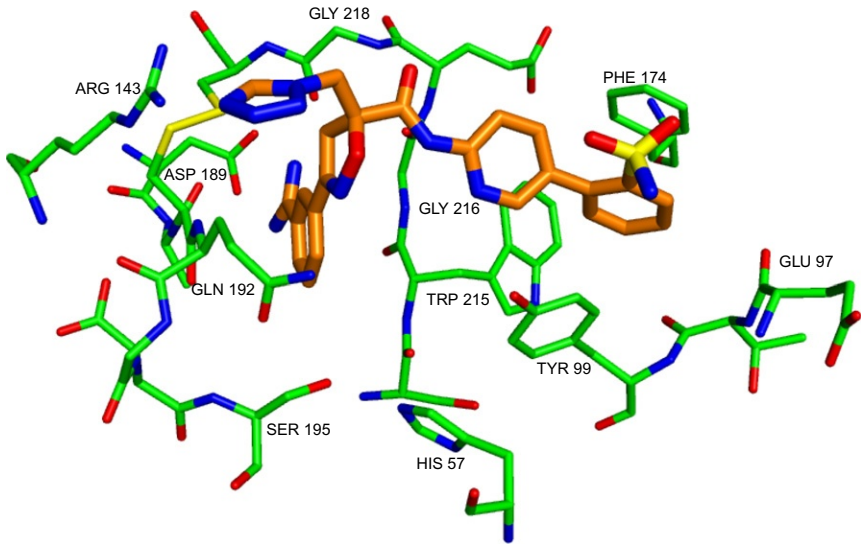

Figure 9.5 Model of isoxazoline **12** in the active site of FXa.

Tyr99 and the backbone of Gln192, led to 5,5-disubstituted isoxazoline **10** ($K_i = 94$ nM). Finally, replacement of the basic P4 amidine with a biaryl moiety resulted in high-affinity inhibitors such as **11** ($K_i = 6.3$ nM) and **12** ($K_i = 0.52$ nM).[15,16] Based on modeling in the active site of FXa, the excellent affinity exhibited by these compounds was rationalized to be the result of bidentate interactions of the benzamidine with Asp189 in the S1 specificity pocket, favorable π stacking of the pendent phenyl ring with Tyr99 and Phe174, and an edge-to-face interaction with Tyr215 in the S4 pocket (Fig. 9.5). The biarylsulfonamide P4 motif represented a major milestone, as for the first time high affinity was achieved with a compound containing a neutral P4 moiety, which was used extensively during lead optimization.[17,18] Interestingly, inhibitors from both the pharmacophore approach and the "focused" screening approach converged on a similar binding motif, where inhibitors bound in the active site in an L-shaped configuration.

3.4. The discovery of pyrazole-based inhibitors

Efforts continued to focus on driving FXa-binding affinity to picomolar levels, recognizing that the ultimate goal of replacing the benzamidine with a less basic/neutral P1 functionality was expected to achieve permeability at the expense of affinity. Scaffold optimization (Fig. 9.6) initially led to vicinally substituted isoxazoline compounds such as **13** (diastereomeric mixture,

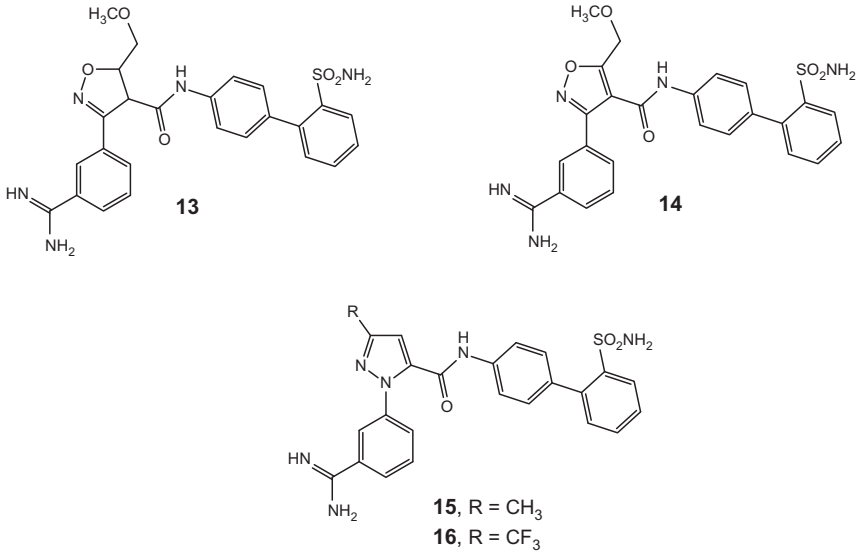

Figure 9.6 The discovery of pyrazole **15** (SN429).

$K_i = 0.5$ nM) with more optimal complementarity with the S1 and S4 pockets.[17] The isoxazole analog **14** ($K_i = 0.15$ nM) lacking the stereogenic centers showed similar affinity. Pyrazole **15** (SN429, $K_i = 0.013$ nM), the result of an independent rational design effort, was a major program milestone based on its picomolar affinity.[18] An X-ray structure of **15** bound to trypsin confirmed the interaction of the carboxamide carbonyl with the Gly216 NH and the biarylsulfonamide P4 group optimally stacked in the hydrophobic S4 region.[18] While extremely potent, **15** had a short half-life, poor oral bioavailability, and lacked selectivity over trypsin-like serine proteases. Evaluation of numerous heteroaryl scaffolds affirmed the pyrazoles to be superior in terms of affinity.[17–19] In fact, the most potent compound synthesized in the program was achieved with the 3-trifluoromethylpyrazole analog **16** ($K_i < 5$ pM). This was a significant development for the program, as we achieved one of our main goals of driving the affinity to a high level thus enabling us to focus on addressing permeability and oral bioavailability for the series.

3.5. Benzamidine mimics—SAR leading to Razaxaban and preclinical properties

Successful replacement (Fig. 9.7) of the benzamidine with the less basic P1 benzylamine ($pK_a \sim 8.8$)[18] and insertion of a fluoro substituent on the inner phenyl ring of the P4 group led to **17** ($K_i = 2.7$ nM, activated partial

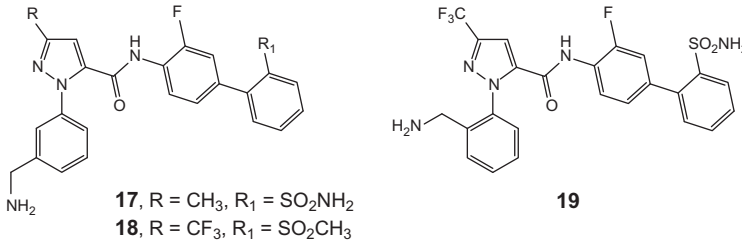

Figure 9.7 Lead optimization of pyrazoles to early clinical candidate **18** (DPC423).

thromboplastin time (APTT) $IC_{2x} = 2.3\ \mu M$). Replacement of the methylsulfonamide P4 aryl substituent with a methylsulfone and the pyrazole methyl substituent with a trifluoromethyl group afforded **18** ($K_i = 0.15$ nM), which exhibited good Caco-2 permeability ($P_{app} = 4.86 \times 10^{-6}$cm/s) and high selectivity over other serine proteases, with the exception of trypsin and kallikrein (both $K_i = 60$ nM).[20] The improved pharmacokinetic (PK) profile ($F\% = 57$, $T_{1/2} = 7.5$ h), and efficacy in rabbit thrombosis models (A–V shunt, $ID_{50} = 1.1$ μmol/kg/h), enabled advancement of **18** (DPC423) to clinical trials as the first clinical candidate from the program.

In Phase I, **18** showed desirable exposure at the doses studied and had a half-life of ~30 h.[20] However, further advancement was curtailed due to preclinical toxicity. With the advancement of **18**, a deep backup strategy for advancing compounds was adopted, which focused on optimizing the selectivity profile to minimize the potential for off-target safety issues. To our delight, the *o*-benzylamine analog, **19** (DPC602) was potent, highly selective (FXa $K_i = 0.9$ nM, trypsin $K_i = 3500$ nM), and demonstrated improved oral bioavailability.[21] Under basic conditions, chemical instability of **19** was observed; the amine cyclized on to the carbonyl of the pyrazole carboxamide, thereby liberating the biarylamine, and development of **19** was discontinued.

At the time this work was done, there was little in the serine protease inhibitor literature describing less basic benzamidine replacements. Therefore, the medicinal chemistry team embarked on a pioneering and comprehensive evaluation of less basic and neutral P1 groups to build in selectivity and achieve oral bioavailability.[22] This work is summarized schematically in Fig. 9.8, along with additional SAR trends. Emphasis was placed on large P1 moieties which could be accommodated in the S1 specificity pocket of FXa that is larger and more lipophilic than in trypsin-like serine proteases owing to the presence of Ala190 in the former, but that might be expected

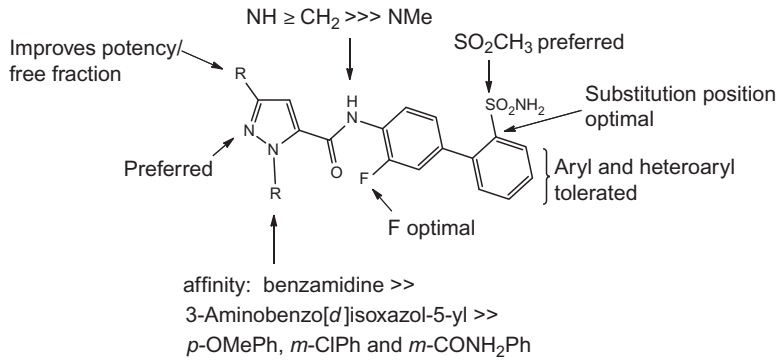

Figure 9.8 SAR trends for the pyrazole series.

to clash with the side chain of Ser190 present in trypsin-like serine proteases. Substitution at the 1-position of the pyrazole with the 3-aminobenzo[*d*] isoxazol-5-yl P1 group was shown to confer desirable affinity and selectivity.

Through a parallel synthesis effort, neutral P1 moieties including 4-methoxyphenyl, 3-carbamoylphenyl, and 3-chlorophenyl were also identified, and although 10- to 20-fold less potent, the compounds were more permeable than similar benzamidines and showed improved selectivity.[22] Success in exploiting the larger S1 pocket of FXa to afford selectivity and oral bioavailability was another highly significant milestone for our program. In addition, a novel and mild cross-coupling methodology (Chan-Lam) was developed in part to facilitate synthesis and scale-up of compounds containing these azole P1 moieties.[23]

Based on its greater potency and improved selectivity, the aminobenzisoxazole P1 series was selected for further optimization.[22,24] To this end, **20** (Fig. 9.9) showed greater affinity and selectivity (FXa $K_i = 0.16$ nM, trypsin $K_i > 3000$ nM) but demonstrated poor clotting activity and permeability. To overcome this, the biaryl P4 group was replaced with a phenylimidazole moiety (**21** $K_i = 0.70$ nM) to increase polarity, solubility, and permeability.[24] Further improvement in affinity and high selectivity (>40,000-fold) was realized with 2-aminomethylimidazole P4 analogs such as **22** ($K_i = 0.17$ nM, Caco-2 $P_{app} = 0.2 \times 10^{-6}$ cm/s) and **23** ($K_i = 0.19$ nM, Caco-2 $P_{app} = 5.56 \times 10^{-6}$ cm/s), with **23** showing improved permeability in the Caco-2 assay. When orally administered to dogs, **23** demonstrated high oral bioavailability, a high volume of distribution, and a moderate half-life ($F\% = 84\%$, $Cl = 1.1$ L/kg/h, $V_{dss} = 3.5$ L/kg, $T_{1/2} = 5.3$ h). A FXa-bound X-ray crystal structure of

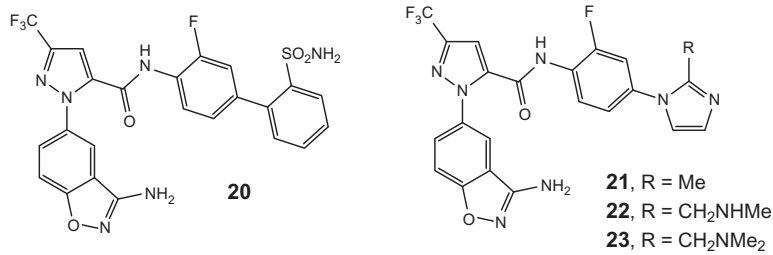

Figure 9.9 Lead optimization of pyrazoles to clinical candidate razaxaban **23**.

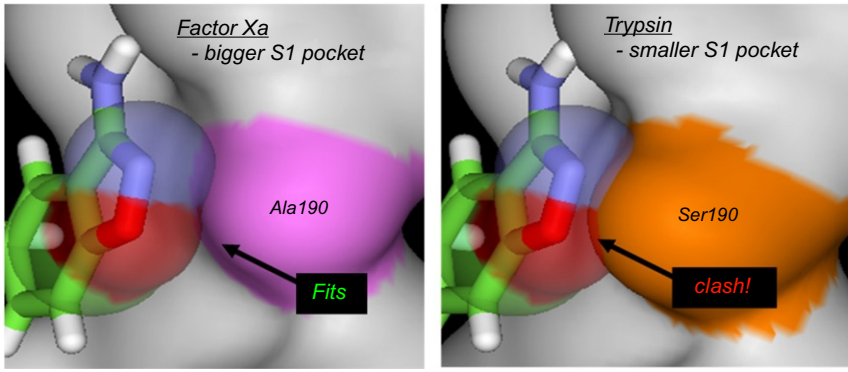

Figure 9.10 Aminobenzisoxazole **23** in the S1 specificity pocket of FXa and trypsin.

23 showed a binding orientation similar to that observed with previous pyrazole candidate **18**. The larger P1 aminobenzisoxazole successfully exploits the differences in the S1 specificity pockets as predicted, resulting in its favorable selectivity profile (>2000-fold, Fig. 9.10). The pendant P4 (dimethylaminomethyl)imidazole nitrogen interacts directly with Glu97 through a network of water molecules in the S4 pocket. Compound **23** was highly efficacious in rabbit models of thrombosis and, given its overall very good profile, was subsequently advanced to clinical development as razaxaban, DPC906 (BMS-562389). Razaxaban was the first small molecule, direct FXa inhibitor to complete a pivotal Phase II proof-of-principle study in deep vein thrombosis (DVT), demonstrating strong efficacy and a favorable bleeding profile.[25] The clinical profile of razaxaban was a major advance for the field and was critical for our program in establishing the dose projections for later clinical studies with apixaban.

3.6. Strategies leading to the dihydropyrazolopyridinone scaffold

Immediately after **23** was advanced, the goal was to identify a structurally diverse backup compound in the event of other unexpected issues. A key backup strategy was to design rigidified pyrazole scaffolds in order to address the potential for metabolic cleavage of the carboxamide moiety given the potential for generation of a mutagenic aniline fragment. Although this was never an issue for the P4 fragment of **23** or its predecessor compound **18**, Ames testing of aniline fragments for all new compounds was implemented, which was resource intensive and rate limiting. A two-pronged approach which leveraged new scaffolds was adopted to eliminate this concern (Fig. 9.11). Strategy 1 focused on tying back the amide NH onto the P4 inner phenyl ring affording compounds with bicyclic scaffolds such as indoline amides. Although these compounds were potent and selective, they had weak clotting activity and poor oral bioavailability.[26,27] Strategy 2 focused on cyclizing the amide moiety onto the pyrazole based on the crucial observation that the 5-pyrazole carboxamide is in a planar orientation with the pyrazole ring in analogs such as razaxaban. Accordingly, several 5,6- and 5,7-bicyclic pyrazole scaffolds were synthesized.[28–30]

From this series, only the pyrazolopyrimidinone **24** and the dihydropyrazolopyridinone **25** maintained a combination of a high level of

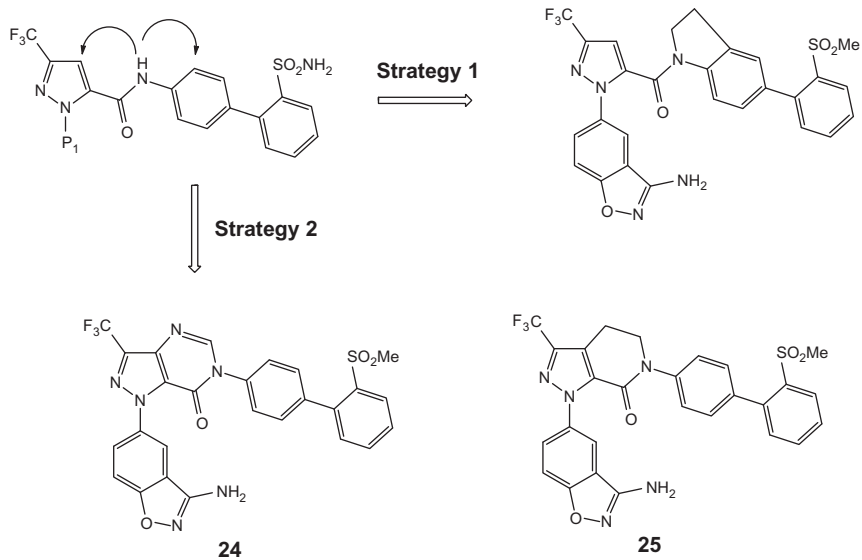

Figure 9.11 Strategies for structural diversification.

FXa affinity, potency in the clotting assay, and high oral bioavailability. Ultimately, instability of the pyrimidinone analogs under acidic conditions precluded them from further consideration. The dihydropyrazolopyridinone scaffold as in **25** showed good stability and versatility in tolerating a broader range of functional groups[29] and, hence, was selected as the scaffold of choice for further optimization.

3.7. SAR in the dihydropyrazolopyridinone series: Optimizing for ideal PK

Initially, the aminobenzisoxazole P1 group was maintained and extensive variation of the P4 moiety was carried out.[30] This led to potent compounds such as **26** and **27** ($K_i = 0.04$ nM, and $K_i = 0.03$ nM, respectively; Fig. 9.12). The overall efficacy and PK profile favored **27**, which was considered for development.

The high FXa affinity exhibited by the bicyclic pyrazole scaffold prompted a reevaluation of the less potent neutral P1 moieties. As part of this effort, the *p*-methoxyphenyl P1 group (Fig. 9.8) which had previously demonstrated high *in vivo* exposure was reintroduced. The 3-position of the pyrazole ring was targeted to optimize for affinity, polarity, and free fraction. This strategy provided compounds such as **28** ($K_i = 0.14$ nM, prolongation of prothrombin time (PT) $IC_{2x} = 1.2$ μM; Fig. 9.12), which demonstrated high affinity and a reasonable free fraction.[31] When administered to dogs, **28** had high oral bioavailability and a long half-life; however, the latter was the result of a high volume of distribution and moderate clearance (Cl = 1.3 L/kg/h, $V_{dss} = 7.4$ L/kg, $T_{1/2} = 7.3$ h, $F\% = 56$). While identifying compounds with a high volume of distribution was a strategy we had

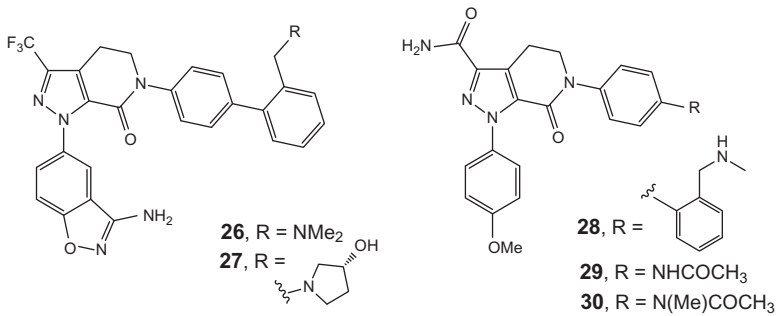

Figure 9.12 Optimization strategies leading to apixaban **1**.

used to achieve long half-life, many compounds with this property were discontinued due to safety issues during preclinical development. An innovative strategy ensued to identify compounds with low volume of distribution (to maintain high drug levels in the bloodstream) and low clearance (to provide acceptable half-lives) while maintaining high affinity, selectivity, and oral bioavailability. Evaluation of metabolic stability in human liver microsomes was included in the early assessment of new compounds to identify compounds likely to have low clearance.

To minimize volume of distribution, we returned to the P4 position, specifically examining polar, nonbasic functionality. Initial analogs such as acetamide **29** showed weak FXa activity ($K_i = 180$ nM). However, the activity was recovered with **30** ($K_i = 0.61$ nM, PT $IC_{2x} = 3.1$ μM). Rigidification of the acetamide group leads to the lactam **1** ($K_i = 0.08$ nM, PT $IC_{2x} = 3.8$ μM).[31] An X-ray structure of **1** bound to FXa (Fig. 9.13) showed an orientation similar to that observed for earlier compounds. The *p*-methoxyphenyl group was deep in the S1 pocket, and the lactam makes the requisite hydrophobic interactions in the S4 pocket.

The overall PK profile of **1** in dogs was favorable, demonstrating high oral bioavailability, extremely low clearance, low volume of distribution, and a moderate half-life (Cl = 0.02 L/kg/h, $V_{dss} = 0.2$ L/kg, $T_{1/2} = 5.8$ h,

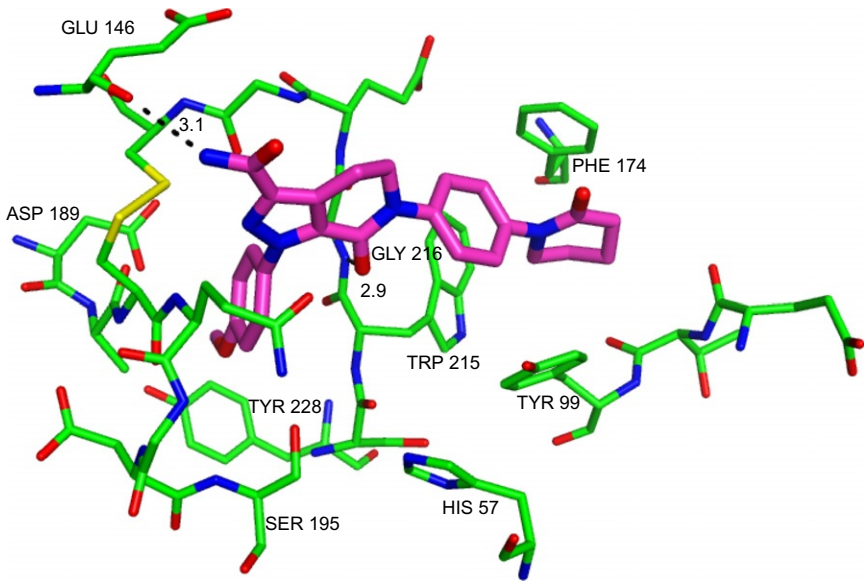

Figure 9.13 FXa-bound X-ray structure of apixaban **1**.

$F\% = 58$).[32] In addition, compound **1** exhibited outstanding selectivity relative to other serine proteases,[33] was highly efficacious in various antithrombotic models and did not demonstrate any liabilities in safety studies.[34] This compound clearly met our predefined criteria for an ideal oral anticoagulant and was selected for clinical development as BMS-562247, apixaban.

4. PRECLINICAL PROPERTIES OF APIXABAN

A review of apixaban describing its preclinical pharmacology as well as its preclinical drug metabolism and PK profile has been published.[34] Apixaban is a potent, reversible, direct, active site inhibitor of FXa, with a K_i of 0.08 nM for human FXa and with greater than 30,000-fold selectivity over other human coagulation proteases. Unlike the indirect FXa inhibitor fondaparinux, apixaban does not require antithrombin III to inhibit FXa. It inhibits free, prothrombinase-bound as well as clot-bound FXa activity and reduces thrombin generation *in vitro*. Apixaban also inhibits FXa from rabbits, rats, and dogs, with K_i values of 0.16, 1.4, and 1.8 nM, respectively, which parallels its antithrombotic potency in these species. Although apixaban has no direct effects on platelet aggregation, it indirectly inhibits platelet aggregation induced by thrombin derived from the upstream proteases in the blood coagulation cascade. In standard clotting assays, apixaban is more potent in the prolongation of prothrombin time than APTT *in vitro* in rats, rabbits, dogs, and humans.

Apixaban given prophylactically caused dose-dependent antithrombotic activity in rats and rabbits, in models of arterial and venous thrombosis and prevented the growth of a preexisting thrombus. Effective concentrations tended to be higher in rats for which the FXa affinity of apixaban was lower. Dose–response studies of apixaban demonstrated a therapeutic window between the dose that inhibits thrombosis and the dose that increases provoked bleeding. When added on top of aspirin or aspirin plus clopidogrel at their clinically relevant doses, apixaban improved antithrombotic activity, without excessive increases in bleeding times.

Apixaban has good oral bioavailability, low clearance, a small volume of distribution in animals and humans, and a low potential for drug–drug interactions. Elimination pathways for apixaban include renal excretion, metabolism, and biliary/intestinal excretion. Although a sulfate conjugate of *O*-demethyl apixaban (*O*-demethyl apixaban sulfate) has been identified as the major circulating metabolite of apixaban in humans, it is biologically

inert and inactive against human FXa. Apixaban was without toxicity in multiple preclinical toxicology studies, including chronic safety studies in rats and dogs, carcinogenic studies in mice and rats, reproductive toxicology studies in rats and rabbits, and mutagenic studies.[35]

Together, these preclinical findings have established the favorable pharmacological and safety profile of apixaban and support the potential use of apixaban in the clinic for the prevention and treatment of various thromboembolic diseases.

5. CLINICAL STUDIES OF APIXABAN

Apixaban has been evaluated in a series of late-stage clinical trials in multiple indications, including prevention and treatment of venous thromboembolism (VTE), secondary prevention of acute coronary syndromes (ACS), and stroke prevention in patients with atrial fibrillation (AF).

After initial studies in healthy human subjects showed a terminal half-life of 8–15 h,[36] it was considered for either QD or BID administration. A dose-ranging study for prevention of VTE in patients undergoing total knee replacement demonstrated that both antithrombotic efficacy and bleeding were dose dependent.[37] Whereas bleeding was similar when the same daily dose was administered QD versus BID, efficacy was consistently better with BID dosing, suggesting that a strategy that minimizes peak to trough fluctuations in concentration provides a more optimal benefit-to-risk profile. Phase 3 studies of apixaban have all utilized a BID regimen.

For prevention of VTE after knee or hip replacement, apixaban 2.5 mg BID demonstrated superior efficacy to enoxaparin 40 mg QD without increasing bleeding.[38,39] In a study versus a 30-mg BID regimen of enoxaparin used in North America, apixaban failed to prove noninferiority, despite similar rates of the primary endpoint, but resulted in less bleeding.[38]

A Phase 2 study for treatment of DVT for 3 months compared apixaban 5 or 10 mg BID, and 20 mg QD with standard treatment with low-molecular-weight heparin (LMWH) followed by warfarin.[40] Results of this study led to the selection of 5 mg BID after 1 week of 10 mg BID for evaluation versus LMWH/warfarin in the initial treatment of VTE in the ongoing AMPLIFY study (ClinicalTrials.gov: NCT00643201), and doses of 2.5 or 5 mg BID versus placebo for extended treatment in the AMPLIFY-EXT study (ClinicalTrials.gov: NCT00633893).

Treatment with apixaban on top of conventional mono or dual antiplatelet therapy was evaluated in patients who had recent ACS. Although promising results were observed in the Phase 2 APPRAISE-1 study,[41] the Phase 3 APPRAISE-2 study was terminated after an observation of a significantly increased risk of major bleeding, especially in patients who were receiving dual antiplatelet therapy.[42]

For prevention of stroke in patients with AF, apixaban 5 mg BID, or 2.5 mg BID in selected patients, was evaluated in two large Phase 3 studies. In AVERROES, apixaban resulted in a 55% reduction in stroke or systemic embolism versus aspirin in patients who were not suitable for treatment with a VKA.[43] Major bleeding was higher in patients treated with apixaban, but the difference was not statistically significant and there was no increase in intracranial or fatal bleeding. When compared with blinded warfarin in the ARISTOTLE study, apixaban demonstrated statistically significant reductions of 21% in stroke and systemic embolism, 31% in major bleeding, and 11% in all-cause death.[2]

6. CONCLUSION

Apixaban was the culmination of a succession of novel and innovative medicinal chemistry discoveries resulting from a structure-based design approach and extensive PK optimization to strike the optimal balance of efficacy and safety. During the lead optimization phase, computer-aided drug design and X-ray crystallography were highly leveraged to drive affinity, selectivity and facilitate oral bioavailability. Potency was achieved by optimizing to extremely high drug affinity (picomolar levels) and oral bioavailability was ultimately achieved by replacing the positively charged group with a neutral group. Selectivity versus other serine proteases resulted from capitalizing on the larger S1 pocket in FXa. The PK profile and favorable safety profile exhibited by apixaban is ideal for an anticoagulant and is due to the low volume of distribution and low clearance. Apixaban was well tolerated in preclinical studies and human clinical trials. Results of the clinical trial program of apixaban have led to its approval in Europe and other countries outside the USA for prevention of VTE after knee or hip replacement. It is currently under review in the USA, Europe, and other countries for stroke prevention in patients with AF.

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Declaration of interest. The authors are employees of Bristol-Myers Squibb Company and were previously involved in factor Xa inhibitor research.

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