Pharmacological Interruption of Acute Thrombus Formation with Minimal Hemorrhagic Complications by a Small Molecule Tissue Factor/Factor VIIa Inhibitor: Comparison to Factor Xa and Thrombin Inhibition in a Nonhuman Primate Thrombosis Model

OSMAN D. SULEYMANOV, JAMES A. SZALONY, ANITA K. SALYERS, RHONDA M. LACHANCE, JOHN J. PARLOW, MICHAEL S. SOUTH, RHONDA S. WOOD, AND NANCY S. NICHOLSON
Pfizer Corporation, Department of Cardiovascular Pharmacology, Skokie, Illinois
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ABSTRACT
This study was designed to evaluate the antithrombotic efficacy and bleeding propensity of a selective, small-molecule inhibitor of tissue factor/factor VIIa (TF/VIIa) in comparison to small-molecule, selective inhibitors of factor Xa and thrombin in a nonhuman primate model of thrombosis. Acute, spontaneous thrombus formation was induced by electrolytic injury to the intimal surface of a femoral blood vessel, which results in thrombus propagation at the injured site. The TF/FVIIa inhibitor 3-amino-5-[1-[2-{4-[amino(imino)methyl]benzyl}amino]-2-oxoethyl]-3-chloro-5-(isopropylamino)-6-oxo-1,6-dihydropyrazin-2-yl]benzoic acid dihydrochloride (PHA-927F) was fully effective in prevention of thrombosis-induced vessel occlusion at a dose of 400 μg/kg/min, i.v., in the arterial vasculature (femoral artery). Neither the effective dose nor multiples up to 4.4-fold the effective arterial plasma concentration elicited any significant effect on bleeding time or blood loss from either the bleeding time site or the surgical (femoral isolation) site. Small-molecule inhibitors of factor Xa or thrombin were effective arterial antithrombotic agents; however, in contrast to the TF/FVIIa inhibitor, they both elicited substantial increases in bleeding propensity at the effective dose and at multiples of the effective plasma concentration. These data indicate that TF/VIIa inhibition effectively prevented arterial thrombosis with less impact on bleeding parameters than equivalent doses of factor Xa and thrombin inhibitors.

The most common cause of mortality in the United States and Western countries is cardiovascular disease, predominantly associated with acute coronary syndromes consisting of unstable angina, myocardial infarction, and sudden death (Braunwald et al., 1989). Acute coronary syndromes are associated with acute thrombus formation, often as the result of rupture of the thin fibrous cap of a vulnerable plaque. Upon plaque rupture, platelet activation occurs followed by a cascade of coagulation reactions as tissue factor is exposed to soluble factor VIIa in the blood (Moreno et al., 1996). Tissue factor initiates the extrinsic pathway of blood coagulation and is the obligatory cofactor for activation of zymogen coagulation factor VII to the serine protease factor VIIa. The tissue factor/factor VIIa complex (TF/VIIa) initiates coagulation by activating the physiological substrates factor IX and factor X, ultimately leading to thrombin generation and fibrin deposition. Tissue factor is a cell surface-expressed glycoprotein that is composed of a 219-amino acid residue extracellular domain, a single transmembrane sequence, and a short cytoplasmic domain (Edgington et al., 1991). Tissue factor is primarily expressed in the media and adventitia of blood vessels forming a hemostatic envelope that surrounds the vessel.

Following plaque rupture or vascular damage, such as occurs in hypertension, atherosclerosis, diabetes, smoking, and interventional procedures, such as balloon angioplasty, tissue factor is exposed to the blood and soluble factor VIIa with which it readily complexes and triggers the coagulation cascade. Ultimately the proteolytic enzyme thrombin is generated and efficiently converts plasma fibrinogen to fibrin, which stabilizes newly formed thrombi or blood clots. Throm-
bin is also a potent platelet agonist, inducing platelet aggregation and possessing smooth muscle cell proliferative properties as well (Coughlin, 2000). Pharmacological attempts have been made at various points of potential intervention in the coagulation cascade, ranging from nonspecific inhibitors such as warfarin, unfractionated heparin, and low-molecular weight heparins, to specific inhibitors of factor Xa or direct acting thrombin inhibitors (Hara et al., 1995; Sanderson et al., 1998; Hauptmann and Sturzebecher, 1999; Pinto et al., 2001). Previous reports suggest that inhibitors of the TF/VIIa complex may prevent thrombosis with a lower bleeding risk than other types of coagulation inhibitors (Harker et al., 1996; Himber et al., 1997; Zoldhelyi et al., 2000).

The intent of the present study is to compare, in a nonhuman primate model of acute thrombus formation, the antithrombotic efficacy and propensity for bleeding diatheses of a small-molecule inhibitor of TF/VIIa (PHA-927F) to a selective, small-molecule inhibitor of factor Xa (Pinto et al., 2001) or a selective, small-molecule, direct-acting inhibitor of thrombin (Sanderson et al., 1998).

In the discovery of PHA-927, structure-based drug design techniques were used to create a molecule that is a potent inhibitor of the TF/VIIa complex but maintains selectivity against the highly homologous serine proteases in the coagulation cascade (factor Xa and thrombin). Using the crystal structure of TF/VIIa bound to a tripeptide-α-ketohiazole, hundreds of pyrazinone TF/VIIa inhibitors were prepared and tested in several serine protease enzyme assays (Parlow et al., 2003; South et al., 2003). The structure activity relationship of the pyrazinone inhibitors was used to develop PHA-927F, a potent, selective, nonpeptide inhibitor of TF/VIIa. This reversible inhibitor binds to the active site in a noncovalent manner and does not interact with the catalytic apparatus of the enzyme. PHA-927F was designed for use in preclinical proof of concept studies (such as those described in this article) to examine the separation of antithrombotic efficacy and bleeding side effects.

In the present study, antithrombotic dose-response relationships were determined for each inhibitor, and bleeding propensity was measured at the fully efficacious antithrombotic doses and at multiples of the plasma concentration observed at the efficacious dose. Bleeding was assessed by determining forearm bleeding time as well as measuring acute blood loss from the bleeding time site and from the surgical (femoral isolation) site at specified times during the course of the experiment.

The monkeys were placed on a thermostatically controlled heating pad to maintain constant body temperature. Blood pressure, heart rate, and blood gases were monitored to maintain normal vital signs. Experiments were conducted in a primate procedure room, equipped with Herpes B exposure kits for emergency use. The left carotid artery was cannulated for continuous measurement of blood pressure and heart rate. A catheter was placed in the right jugular vein for administration of test compounds. The left femoral artery was exposed by blunt dissection, clearing 2 to 3 cm of the vessel to place an ultrasonic flow probe (Transonic Systems, Inc., Ithaca, NY), connected to a Gould PONEMAH data acquisition interface (Valley View, OH) for monitoring of the blood flow. An anodal stimulation electrode consisting of a 2.5-mm long, 25-gauge needle attached to 30-gauge Teflon insulated silver-coated copper wire supplying 150 μA current to the intima of the femoral artery was placed proximal to the flow probe. Electrical stimulation started after a 30-min test compound or vehicle infusion and continued for 120 min (in case of no occlusion) or 5 min past occlusive thrombus formation (zero blood flow). To assure that zero flow resulted from thrombus formation, the injured vessel segment was excised, opened lengthwise, and the thrombus visualized. For the animals treated with PHA-927, after zero flow had persisted for 15 min, electrolytic stimulation was stopped, the injured vessel segment was excised, and the thrombus was visualized and weighed. The thrombus weight was not used for comparison purposes in these studies.

The percentage of theoretical maximum blood flow was also monitored in these studies. This measure compares the area under a theoretical ideal curve, which assumes that the initial rate of flow is maintained throughout the study, to the area under the curve for actual blood flow. This measure provides a basis for comparison across compounds by eliminating the variations in initial blood flow that may occur in individual animals.

The major endpoint derived in this study was time to thrombotic occlusion. In addition, test animals were subjected to periodic bleeding time determinations, before and during test compound or vehicle infusion. Bleeding time measurement was accomplished by placing a blood pressure cuff on the upper arm and inflating the cuff to 40 mm Hg pressure. A Surgicutt device (Simplate bleeding time device) was used to make two incisions below the cuff in the forearm. The device was modified by raising the blade 3.5 mm and fixing it to accommodate the thick and tough skin of primates. Blood loss was determined by placing all surgical field and bleeding time site sponges into flasks containing aliquots of Drabkin's solution. Total content of shed he moglobin (from hemolyzed red blood cells) was determined spectrophotometrically and blood volume calculated by adjusting for whole-blood hemoglobin concentration (Technicon, Tarrytown, NY). Blood samples were obtained throughout the course of the experiment for determination of drug plasma concentration and ex vivo determination of coagulation parameters [prothrombin time (PT), activated partial thromboplastin time (aPTT), and activated clotting time (ACT)]. For PT measurements, a standard clotting assay was performed using an Electra 900 automated coagulation timer (Medical Laboratories Automation, Pleasantville, NY) with recombinant human tissue factor (Innovin; Dade Behring, Inc., Newark, DE). aPTT was determined using reagent SynthASil (Hemoliance, Raritan, NJ). For measurement of ACT, an i-STAT portable clinical analyzer (Sensor Devices, Inc., Waukesha, WI) was used.

**Dosing Solutions and Procedures for Primate Efficacy and Bleeding Studies.** Blood samples for ACT, PT, aPTT, or plasma levels of test compound (PHA-927F) were obtained at baseline (30 min following completion of all surgical and instrumentation procedures), 30 min after initiation of drug infusion (prestimulation), and 60 and 120 min postelectrical stimulation to the intimal surface of the blood vessel, or at the time of zero flow. Bleeding time and blood loss were measured at baseline and 60 and 120 min poststimulation or at the time of zero flow.

**Vehicle Control.** Sterile isotonic saline was infused into right jugular vein at 0.2 ml/min. Electrical stimulation with a 150-μA

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**Materials and Methods**

Male cynomolgus (*Macaca fascicularis*) monkeys with body weights ranging between 3.1 and 10.5 kg were selected for the study. The monkeys were housed in primate cages on a 14/10-h light/dark cycle with ad lib access to conditioned (by reverse osmosis) tap water. They were fed Purina Certified Primate Diet no. 5048 and participated fully in the primate environmental enrichment program (including paired housing if compatible). All studies were conducted under a protocol approved by the site Institutional Animal Care and Use Committee. The test animals were fasted for 16 to 18 h and were sedated by intramuscular administration of 20 mg/kg ketamine hydrochloride to facilitate placement of an endotracheal tube and an intravenous catheter for administration of 15 mg/kg (with maintenance doses of 5 mg/kg when needed) pentobarbital sodium to achieve a surgical plane of anesthesia throughout the experiment.
anodal current was started after a 30-min infusion period and continued for 5 min following thrombotic occlusion.

**Factor Xa Inhibitor.** The small molecule factor Xa inhibitor 1-(3-(aminomethyl)phenyl)-N-{2-(aminosulfanyl)-1,1'-biphenyl}-4-y]-3-methyl-1H-pyrazole-5-carboxamide (Pinto et al., 2001) was synthesized in the Pfizer laboratories. Pinto et al. (2001) reported $K_V$ values for this compound of 0.013 nM against factor Xa, 300 nM against thrombin, and 16 nM against trypsin. The inhibitor was dissolved in a combination of 10% ethanol, 10% PEG-400, and 80% saline to a final concentration of 3 mg/ml. The stock solution was diluted with saline to appropriate concentration to administer doses of 1 or 3 μg/kg/min at an intravenous infusion rate of 0.2 ml/min. The compound was infused for 30 min before starting electrical stimulation and for 5 min after zero flow or a maximum of 150 min in those animals in which no occlusion was observed.

**Thrombin Inhibitor.** The small molecule thrombin inhibitor 3-(2-phenethylamino)-6-methyl-1-(2-amino-6-methyl-5-methylencycarboxamidomethyl)pyridinyl]pyrazine dihydrochloride (Sanderson et al., 1998) was synthesized in the Pfizer laboratories. Sanderson et al. (1998) reported $K_V$ values for this compound of 0.8 nM against thrombin and 1800 nM against trypsin. The compound was dissolved to provide a stock solution with a concentration of 20 mg/ml in a combination of 10% ethanol, 10% PEG-400, and 80% sterile saline. The stock solution was diluted with saline to appropriate concentration to administer doses of 30 or 60 μg/kg/min at an intravenous infusion rate of 0.2 ml/min. The compound was infused for 30 min before starting electrical stimulation and for 5 min after zero flow or a maximum of 150 min in those animals in which no occlusion was observed.

**PHA-927F (Tissue Factor/FVIIa Inhibitor).** PHA-927F is a specific TF/VIIa inhibitor with an in vitro IC$_{50}$ of 0.025 μM (Fig. 1). PHA-927F exhibits high selectivity for the TF/VIIa complex, with greater than 3500-fold selectivity versus factor Xa and thrombin (South et al., 2003). For in vivo studies to determine antithrombotic efficacy, PHA-927F was dissolved in sterile saline at a concentration of 40 mg/ml, pH 2.7. The stock solution was diluted with saline to an appropriate concentration to administer doses of either 200 or 400 μg/kg/min at an intravenous infusion rate of 0.2 ml/min. The compound was infused for 30 min before starting electrical stimulation and for 5 min after zero flow or a maximum of 150 min in those animals in which no occlusion was observed.

**Multiple Dose Bleeding and Blood Loss Studies.** Each test compound was administered by intravenous infusion in separate studies in escalating doses (multiples of the effective dose) to determine the effect on forearm bleeding time and blood loss at the bleeding time site and at a surgical incision site (femoral) in the groin. Multiples of $3 \times$ and $5 \times$ the efficacious dose were evaluated for each of the inhibitors (note: subsequent evaluation of the plasma concentrations of the inhibitors revealed that multiples of $3 \times$ and $7 \times$ the efficacious plasma level of the factor Xa inhibitor, multiples of $3 \times$ and $8 \times$ the efficacious plasma level of the thrombin inhibitor, and multiples of $3 \times$ and $4.4 \times$ the efficacious plasma level of the TF/VIIa inhibitor, PHA-927F were achieved; only the $1 \times$ and $3 \times$ groups were used for intercompound statistical analysis). All infusions at the multiple dosing levels were 60 min in duration, and bleeding time and blood loss were determined at the midway point (30 min) of the infusion.

**Reagents.** Experimental reagent PHA-927F (TF/FVIIa inhibitor) is a new chemical entity that was synthesized in the laboratories of Pfizer Corp. (St. Louis, MO). The small molecule thrombin inhibitor and the small molecule factor Xa inhibitor were prepared in the laboratories of Pfizer Corp. (St. Louis, MO) based on previously published information (Sanderson et al., 1998; Pinto et al., 2001).

**Statistics.** All data are expressed as the mean ± S.E.M. Significance was at $p < 0.05$. A transformation was needed to stabilize the variance across groups for all four responses. The transformations were reciprocal for bleeding time, square root reciprocal for blood loss at the bleeding time site and log for activated clotting time. Since the measurement for blood loss at the surgical site was cumulative over time, the response was time adjusted to scale the data so the amount of blood measured was over the same period of time. A log transformation was performed on the adjusted response.

For each treatment, a contrast-based trend test was implemented within a mixed model analysis of variance to indicate values that were significantly different from their respective control values. For the pairwise comparison of each compound to PHA-927F at doses 1.0$\times$, 2.0$\times$, and 3.0$\times$, a t test within an analysis of covariance was used.

The mixed model took into account the repeated dose, random effect of primate, Satterthwaite approximation, and compound symmetry for the variance-covariance matrix. The random effect of primate identifies the subjects in the mixed model. Complete independence of the data.

**Results**

**Saline-Infused Control Animals.** Untreated control animals were infused with saline concomitant with electrolytic injury (150 μA) to the intimal surface of the femoral artery. Cessation of blood flow occurred in all control animals (six of six) with a mean time to thrombotic occlusion of 67 ± 15 min (Fig. 2). All animals exhibited cyclical flow variations periodically as platelet thrombi formed and subsequently dislodged...
until complete thrombotic occlusion occurred as determined by elimination of blood flow through the injured arterial segment of the femoral artery. The percentage of maximum theoretical blood flow in these animals was 36 ± 6%.

Effect of Factor Xa Inhibition on Occlusive Thrombus Formation. A specific small-molecule factor Xa inhibitor was infused in animals undergoing electrolytic-induced arterial injury and the time to occlusive thrombus formation determined. Doses of the factor Xa used were 1 and 3 μg/kg/min by intravenous infusion. Both animals (two of two) occluded at the low dose of 1 μg/kg/min with a mean time of 36 min. At the higher dose of the factor Xa (3 μg/kg/min), all animals (six of six) were occlusive thrombus-resistant and did not occlude during the 120 min observation period (Fig. 3a). Percent maximum theoretical blood flow in the efficacious dose group was 64 ± 7% but only averaged 25% in the lower dose group. Despite the apparent decrease in time to thrombus formation at the 1 μg/kg/min dose, one of the two animals had an occlusion time (59 min) similar to control (67 min), whereas the other had a time that was very short (14 min). In the control group, times to zero flow as short as 27 and 43 min were observed indicating that rapid thrombus formation can occur and that it is unlikely that the compound was acting in a prothrombotic manner. Because this dose of the factor Xa inhibitor was clearly not efficacious, it was not tested in any additional animals.

Effect of Thrombin Inhibition on Occlusive Thrombus Formation. A specific direct-acting thrombin inhibitor was also used for comparative purposes in the primate electrolytic injury model. Doses of the thrombin inhibitor infused were 30 and 60 μg/kg/min, i.e., which resulted in occlusive thrombus formation in three of four animals at the lower dose, with mean time to occlusion of 94 ± 19 min (n = 3) and complete protection from thrombotic occlusion at the higher dose in six of six animals (Fig. 3b). Percent maximum theoretical blood flow in the efficacious dose group was 70 ± 5% and averaged 41 ± 14% in the lower dose group.

Effect of Tissue Factor/Factor VIIa Inhibition on Occlusive Thrombus Formation. The specific TF/VIIa inhibitor PHA-927F was evaluated in the primate electrolytic femoral artery injury model at intravenous doses of 200 and 400 μg/kg/min. The lower dose resulted in thrombotic occlusion in 67 ± 11 min (n = 4), whereas at the higher dose thrombotic occlusion was prevented in four of four animals (Fig. 4). Percent maximum theoretical blood flow in the efficacious dose group was 81 ± 6% similar to that observed with the other inhibitors at efficacy. The value in the lower dose groups was 33 ± 7 also similar to the values in the nonefficacious groups of the other inhibitors.

Forearm Bleeding Time Evaluations. Forearm bleeding times were evaluated for all animals at the effective dose (1×) and multiples of the effective plasma concentration for 1) the TF/VIIa inhibitor PHA-927F, 2) the factor Xa inhibitor, and 3) the thrombin inhibitor. The effective dose, as determined in the primate thrombosis model, was the minimal dose that prevented arterial thrombus formation in all animals tested. For PHA-927F, the factor Xa inhibitor and the thrombin inhibitor these effective intravenous doses were determined to be 400, 3, and 60 μg/kg/min, respectively. The plasma concentrations at these doses were 107, 0.09, and 3.0 μg/ml for PHA-927F, the factor Xa inhibitor, and the thrombin inhibitor, respectively.

Forearm bleeding time, under 40 mm Hg pressure, was determined for PHA-927F. At all doses tested, up to that which resulted in a multiple of 4.4× of the effective plasma level, bleeding time never increased more than 2-fold over baseline (Fig. 5). In fact, at the effective plasma level of PHA-927F, no significant increase in bleeding time was determined. In contrast, the factor Xa inhibitor and the thrombin inhibitor both induced moderate increases in bleeding time at the effective plasma level and substantial and significant increases in bleeding time at multiples of the effective plasma level.

Blood Loss at the Bleeding Time Site. Similarly, blood loss from the bleeding time site was determined at the effective dose and at multiples of the effective doses for all compounds evaluated in this study. PHA-927F elicited little or no blood loss from the bleeding time site at the effective plasma level (1×) or multiples up to 4.4×, whereas both the factor Xa inhibitor and the thrombin inhibitor induced substantial and significant blood loss from the bleeding time site at both the effective plasma levels and at the multiples thereof (Fig. 6a).

Blood Loss at the Surgical Site. Blood loss was also determined in this study at the femoral arterial surgical site in each animal evaluated at the effective plasma concentran-
increases in blood loss from the surgical site were noted with both the factor Xa and the thrombin inhibitors (Fig. 5b).

**PT and aPTT.** PT and aPTT were determined in the efficacy studies for all animals, and the values after 90 min of compound infusion are reported in Table 1. Both PT and aPTT were largely unaffected by the factor Xa inhibitor indicating that compounds acting by this mechanism of action are not easily monitored by these common coagulation assays. The thrombin inhibitor prolonged both PT and aPTT in a dose-dependent manner with PT at 81 ± 9 s and aPTT at 101 ± 4 s (8 and 5 fold increases, respectively) at the efficacious dose. The TF/VIIa inhibitor had the expected effect of prolonging PT while not altering aPTT. PT was prolonged in a dose-dependent manner reaching 27 ± 1 and 55 ± 5 s (3- and 5-fold increases) at 200 and 400 μg/kg/min, respectively.

**ACT.** ACT was determined in all animals and PHA-927F elicited a significant but small, dose-responsive increase in the ACT (Fig. 7). However, the ACT never increased to over 400 s, which is the general standard used for full heparin-induced anticoagulation. In contrast, both the factor Xa and thrombin inhibitors induced substantial and significant increases in the ACT at the effective dose and at multiples of the effective dose to levels that indicated the potential for bleeding risk to occur.

**Plasma Concentrations of PHA-927F.** Plasma concentrations of PHA-927F were determined in all animals, as were PT values so that the correlation between plasma concentration and PT could be determined. Figure 8 represents this correlation, and the calculated correlation coefficient is 0.95, which indicates that the PT is an excellent predictor of plasma concentration. The plasma concentration at the effective dose in the efficacy studies was 107 μg/ml. In the multiple dose bleeding and blood loss studies, the plasma concentrations were 120, 337, and 472 μg/ml for the 1×, 3×, and 4.4× plasma levels, respectively.

**Hemodynamic Parameters.** Heart rate and mean arterial blood pressure were measured throughout the efficacy and bleeding studies for all compounds. No changes were observed during any of the infusions.

### Discussion

The results of this study indicate that selective inhibition of TF/VIIa activity with the selective, small-molecule inhibitor PHA-927F is a highly effective antithrombotic strategy with little propensity to elicit bleeding in the experimental model used herein. Small molecules used to inhibit factor Xa and thrombin also resulted in complete antithrombotic efficacy. In contrast to TF/VIIa inhibition, however, efficacy was associated with substantial bleeding propensity. Increased risk of bleeding is a serious complication with almost all antiplatelet, anticoagulant, and fibrinolytic drugs, and thus, some risk/benefit analysis must also be taken into account when using such agents.

The studies described demonstrate the ability of a small-molecule inhibitor of TF/VIIa to inhibit or prevent thrombosis while having minimal effects on bleeding parameters. Previous reports (Harker et al., 1996; Himber et al., 1997; Zoldhelyi et al., 2000), using various approaches to inactivate the TF/VIIa axis, corroborate the current findings. Taken together, these studies indicate that pharmacological interruption of coagulation at this very early point in the cascade
TABLE 1

Effects of saline, the factor Xa inhibitor thrombin inhibitor, or tissue factor/factor VIIa inhibitor infusion on prothrombin time and activated partial thromboplastin time.

Data are presented as the mean ± S.E.M.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Dose (μg/kg/min)</th>
<th>Prothrombin Time [Fold Increase]a</th>
<th>Activated Partial Thromboplastin Time [Fold Increase]a</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>11 ± 1 [1.0]</td>
<td>22 ± 1 [0.9]</td>
<td></td>
</tr>
<tr>
<td>Factor Xa Inhibitor</td>
<td>12 ± 1 [1.1]</td>
<td>30 ± 1 [1.3]</td>
<td></td>
</tr>
<tr>
<td>Thrombin Inhibitor</td>
<td>14 ± 0 [1.3]</td>
<td>32 ± 1 [1.3]</td>
<td></td>
</tr>
<tr>
<td>PHA-927 (Tissue Factor/Factor VIIa Inhibitor)</td>
<td>40 ± 4 [3.9]</td>
<td>65 ± 5 [2.9]</td>
<td></td>
</tr>
<tr>
<td></td>
<td>40 ± 4 [3.9]</td>
<td>65 ± 5 [2.9]</td>
<td></td>
</tr>
<tr>
<td></td>
<td>60 ± 8 [7.8]</td>
<td>101 ± 4 [4.7]</td>
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<tr>
<td></td>
<td>27 ± 1 [2.8]</td>
<td>22 ± 2 [1.1]</td>
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</tr>
<tr>
<td></td>
<td>55 ± 5 [5.3]</td>
<td>29 ± 1 [1.3]</td>
<td></td>
</tr>
</tbody>
</table>

a: Fold increase over baseline value at 90 min of compound infusion (saline infusion in control animals). For PHA-927 200 μg/kg/min animals, the treated value is from a sample collected at the time zero flow occurred.

b: This group had only two animals and therefore no S.E.M.

(TF/VIIa inhibition), results in desirable antithrombotic efficacy that is accompanied by little or no change in bleeding diatheses. This is true despite changes in traditional anticoagulant parameters such as the aPTT and the PT. The separation of efficacy and bleeding is an important distinction and may be related to the biology of TF/VIIa inhibition in part owing to the membranous cell surface anchored nature of tissue factor, whereas most other traditional factors that are inhibited in coagulation reactions are principally soluble factors like factor Xa and thrombin. Furthermore, it has been demonstrated that blood-borne tissue factor activity may also play an important role in the etiology of acute thrombus formation (Giesen et al., 1999) and possibly soluble tissue factor interaction with soluble factor VIIa is inhibited while maintaining the integrity of the membrane-associated forms of tissue factor to preserve hemostasis.


Address correspondence to: John J. Parlow, Research Advisor Chemistry, Associate Fellow, Department of Medicinal and Combinatorial Chemistry, Pfizer Corporation, 800 N. Lindbergh Blvd., St. Louis, MO 63167. E-mail: john.j.parlow@pfizer.com