A population pharmacokinetic-pharmacodynamic and logistic regression analysis of lotrafiban in patients

Objective: **Our objective was to assess the safety, tolerability, pharmacokinetics, and pharmacodynamics of lotrafiban, an oral glycoprotein IIb/IIIa inhibitor, in patients with a recent myocardial infarction, unstable angina, transient ischemic attack, or stroke.**

Methods: **A 12-week, double-blind, multi-center, placebo-controlled, parallel-group, phase II study of lotrafiban (the Anti-platelet Useful Dose Study) was conducted in patients. Lotrafiban or placebo was administered as a twice daily oral dose at four dose levels (5-100 mg) for 12 weeks with daily doses of aspirin (300-325 mg). The pharmacokinetics of lotrafiban were characterized with the use of a population approach and were described by a two-compartment model with first order absorption and first order elimination. The pharmacodynamic data, ex vivo platelet aggregation, were described with the use of a direct effect inhibitory sigmoidal model with a baseline. The relationship between the severity of bleeding episodes and predicted steady-state lotrafiban exposure was characterized by logistic regression.**

Results: **Pharmacokinetic analysis showed that increasing age and decreasing creatinine clearance resulted in increased exposure to lotrafiban. The concentration-effect relationship was steep, with near complete inhibition of platelet aggregation at lotrafiban concentrations in excess of 20 ng/mL. Logistic regression showed that at exposures that exceeded approximately 835 ng · h/mL, the severity of adverse bleeding events increased considerably; this suggested that dosing recommendations should be generated to minimize the likelihood of patients having an area under the plasma concentration-time curve from 0 to 24 hours in excess of this value.**

Conclusions: **Patients whose age exceeded 65 years or whose creatinine clearance was less than 60 mL/min should be given a lower dose of lotrafiban than younger patients with good renal function. (Clin Pharmacol Ther 2001;69:210-22.)**

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Platelets play a key role in the pathogenesis of atherosclerosis, thrombosis, and acute coronary syndromes.[1,2] Platelet aggregation may be produced by a variety of stimuli, including thrombin, collagen, adenosine diphosphate (ADP), thromboxane A_2, and epinephrine. Inhibition of platelet function by aspirin, an inhibitor of thromboxane A_2–mediated aggregation, has been shown to reduce the incidence of occlusive cardiovascular events in patients, as well as to reduce the risk of nonfatal myocardial infarctions (MIs) and stroke in patients with unstable angina (UA) or a history of MI, transient ischemic attack (TIA), or stroke.[3-5] Agents such as ticlopidine and clopidogrel inhibit ADP-mediated aggregation and have similar effects as aspirin on MI and stroke.[6]

Fibrinogen binding to activated platelets is one of the final steps in platelet aggregation, and there is evidence that the glycoprotein IIb/IIIa (GP IIb/IIIa) receptor complex on the platelet surface is the binding site for fibrinogen.[6-9] Unlike aspirin or ticlopidine, GP IIb/IIIa

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antagonists block the final common pathway of aggregation and inhibit platelet aggregation irrespective of the pathway responsible for initiating the cascade. One drug in this class is abciximab (Reopro, Eli Lilly and Co, Indianapolis, Ind), a mouse/human chimeric monoclonal antibody fragment approved by the Food and Drug Administration as adjunctive therapy for patients undergoing high-risk angioplasty and atherectomy. Abciximab has been observed to prevent acute thrombotic events in patients with percutaneous transluminal coronary revascularization.[10-12]

Lotrafiban is a nonpeptide antagonist of the GP IIb/IIIa receptor that has demonstrated activity in blocking platelet aggregation. After oral administration, peak plasma concentrations of lotrafiban occur within 2 to 3 hours followed by a biphasic decay with a terminal half-life of 12 to 20 hours. Lotrafiban is predominantly cleared by means of renal elimination. Lotrafiban exhibits low protein binding in vitro (<13%). Although the pharmacokinetics exhibit considerable between- and within-subject variability, lotrafiban appears to exhibit dose-proportional pharmacokinetics. Lotrafiban has an estimated bioavailability of approximately 2%, which may account for its high pharmacokinetic variability. Despite the high variability, lotrafiban was found to be safe and well tolerated in early phase studies.

METHODS

Study design. In this phase II study (the Anti-platelet Useful Dose Study [APLAUD]),[13,14] the safety, tolerability, pharmacokinetics, and pharmacodynamics of lotrafiban were examined in patients with recent MI, UA, and TIA. The pharmacokinetic objectives of this clinical trial were to study the pharmacokinetics of lotrafiban in patients and to evaluate the effects of demographics, including age, weight, sex, creatinine clearance, and other patient characteristics on the pharmacokinetics of lotrafiban. A second objective was to characterize the pharmacodynamics of lotrafiban in this patient population with the use of ADP-induced ex vivo platelet aggregation as the indicator of pharmacodynamic effect. The third objective was to correlate patient exposure to lotrafiban with the incidence and severity of bleeding adverse events.

A total of 444 patients with a diagnosis of recent MI, UA, TIA, or stroke as confirmed by medical histories, physical examinations, enzyme levels, electrocardiograms, or computed tomography scans were enrolled in this phase II study. All female patients were either of non-childbearing potential (either post-menopausal with no menstrual period for a minimum of 6 months or surgically sterilized) or capable of bearing children but using an intrauterine device. Patients who did not meet the primary entrance criteria were excluded from participation in this study. Also excluded were patients with endoscopically proven peptic ulceration or frank gastrointestinal bleeding within the last 3 years or who were being treated for peptic ulcer disease, patients with a history of coagulation or hemostatic disorders, patients with known hypercoagulable states, patients with a history of drug-induced hematologic or hepatic abnormalities, and patients with evidence of hepatic or renal disease on laboratory screening (serum creatinine of >2.0 mg/dL; AST, ALT, bilirubin, and alkaline phosphatase of more than twice the upper limits of normal). Finally, patients who had taken experimental medication within the last 30 days, patients who had been treated within 7 days with any drug that affected platelet activity other than aspirin or other nonsteroidal anti-inflammatory drugs or who had been treated within 24 hours with heparin or thrombolytic therapy, and patients who were unable to give written informed consent were excluded from the trial. All patients gave informed consent before participating in this trial. The study was conducted according to Good Clinical Practice and the Declaration of Helsinki.

APLAUD was a randomized, double-blind, placebo-controlled, parallel-group, multiple-dose, multi-center study. Patients were randomly assigned to receive either drug or placebo in a ratio of approximately 4:1. Oral doses of 5, 20, 50, or 100 mg of lotrafiban or placebo were administered twice daily for 12 weeks. All of the patients received 300 to 325 mg of aspirin, which was taken once daily with the morning dose of lotrafiban. The highest dose group (100 mg twice a day) was terminated early because of a high incidence of major bleeding adverse events. There were 113, 98, 105, and 34 patients, respectively, in the 5-, 20-, 50-, and 100-mg dose groups, and there were 94 patients receiving placebo. All of the patients were subjected to a series of laboratory tests before dosing and throughout the study. At each visit, complete blood cell count and platelet counts were assessed and patients were queried about any bleeding incidences and tolerability concerns.

For population pharmacokinetic analysis, blood samples were collected from all of the patients enrolled in the study during the following 3 occasions: day 7 and weeks 3 and 6 of dosing. During each sampling occasion, samples were drawn during one of the following time intervals: 0.5 to 2 hours, 3 to 5 hours, or 6 to 9 hours after administration of the morning dose of lotrafiban. One sample was collected from all patients immediately before dosing during week 12. For patients who provided platelet aggregation data, additional pharmacokinetic samples were drawn at the same times that platelet aggre-

Table I. Patient demographics for population pharmacokinetic data set

Characteristic	*No. of patients*	*Mean*	*Range*
Age (y)	326	61	28-92
Weight (kg)	325	84.2	44-185.3
*CL_{CR} (mL/min)**	317	81.6	24.8-157
Sex	326	—	—
Male	230	—	—
Female	96	—	—
Aspirin dose	326	—	—
300 mg	43	—	—
325 mg	283	—	—
Long-term aspirin use	326	—	—
No	22	—	—
Yes	304	—	—
Hyperlipidemia	306	—	—
No	120	—	—
Yes	186	—	—
Diabetes	306	—	—
No	246	—	—
Yes	60	—	—
Hypertension	306	—	—
No	119	—	—
Yes	187	—	—
Qualifying diagnosis	326	—	—
Recent MI	101	—	—
Recent UA	100	—	—
Recent TIA	51	—	—
Recent stroke	74	—	—
Cigarette smoking status	305	—	—
Nonsmokers	243	—	—
Smokers	62	—	—

*CL_{CR} was calculated on the basis of the equation of Cockcroft and Gault.

Table II. Number of patients in each dose group population pharmacokinetic data set

Lotrafiban dose	*No. of patients*
Group	326
5 mg	105
20 mg	94
50 mg	99
100 mg	28

gation data were collected. The samples were collected in heparinized tubes and immediately centrifuged at 4ºC to separate plasma, which was stored frozen at approximately –20°C until analyzed.

For the pharmacokinetic-pharmacodynamic analysis of platelet aggregation data, additional blood samples for the determination of plasma lotrafiban concentrations and ex vivo platelet aggregation were collected from 76 patients enrolled at selected centers at 0 (predose), 1.5, 3, and 6 hours after the morning dose on days 1 and 14. The samples for ex vivo platelet aggregation were drawn in Vacutainer tubes that contained 3.8% citrate and were spun in a swinging bucket centrifuge at 800 rpm at 20ºC to prepare platelet-rich plasma. The platelet-rich plasma was separated, and ADP was added to a final concentration of 20 μmol/L. Platelet aggregation in platelet-rich plasma was evaluated with the use of a Chrono-Log Whole Blood Aggregometer (Model 560-VS) with 810/DF Aggro/Link Data Reduction System (Chrono-log, Havertown, Pa). The data were obtained as percentage of aggregation.

Assay for lotrafiban. Plasma samples were analyzed for lotrafiban by a validated reversed phase HPLC with mass spectrometry-mass spectrometry detection by means of online solid phase extraction (Prospekt, Spark-Holland Instruments, the Netherlands). Positive ions were produced by electrospray and analyzed with the use of multiple reaction monitoring. The assay methodology used was similar to that used to detect another GP IIb/IIIa inhibitor, tirofiban.[15] The assay range was 0.2 to 100 ng/mL, with a 200-μL aliquot of plasma. The performance of the assay was assessed in a three-run validation, in which 6 replicate samples were analyzed at each of five concentrations on 3 separate days. Average within-run precision ranged from 5.8% to 17.6%, between-run precision ranged from 1.6% to 10.8%, and average bias ranged from –0.6% to 6.8%.

Population pharmacokinetics. Data from 326 of 350 patients who were receiving active drug with a total of 1444 concentrations were included in the final population pharmacokinetic database. The demographics of the patients included in this database are given in Table I, and the number of patients in each dose group in the population data set is given in Table II. For 24 patients, either pharmacokinetice samples were not drawn or there was insufficient dosing or sample information available to include the patients in the database. One patient (0.3% of the patients with available pharmacokinetic data) was removed because both measured concentrations were determined to be outliers (weighted residuals >±8) and no explanation or covariate could be found for this behavior. An additional 10 data points from 10 different patients (0.82% of the total available pharmacokinetic observations) were removed from the database as outliers with weighted residuals of more than ±8. These data were returned to the database at the end of model building to assess the impact of the data on the final model.

Pharmacokinetic data were analyzed with the use of the nonlinear mixed effects modeling program (NONMEM).[16-18] A two-compartment model with a fixed

first-order absorption and first-order elimination from the central compartment (ADVAN 4 Trans 4) was used to fit the concentration-time data. The model was parameterized for oral clearance (CL/F), apparent volumes of distribution of the central compartment (V_2/F) and the peripheral compartment (V_3/F), the intercompartmental clearance, and the absorption rate constant. The potential effects of patient demographics (age, creatinine clearance [CL_{CR}], weight, and sex), concomitant medications (aspirin dose, chronic aspirin use, and smoking status), concurrent diseases (diabetes and hyperlipidemia), and qualifying diagnoses (MI, UA, TIA, or stroke) were examined on the pharmacokinetic parameters. CL_{CR} for all of the patients was estimated from the Cockcroft-Gault equation.[19]

During each step in the model building process, improvements to the model were assessed by evaluation of the agreement between the observed and predicted plasma concentrations, reductions in the range of weighted residuals, uniformity of the distribution of the weighted residuals versus the predicted concentrations about the line of identity, and increases in the precision of the parameter estimates, as well as reduction of the terms for interindividual variability and random residual variability. Assessment of the log likelihood ratio test was also conducted as a means of assessing improvement in the model.

Population pharmacodynamics. Platelet aggregation data were collected from 76 patients at selected study centers. Seven subjects were removed from this database because no corresponding pharmacokinetic observations were available. Thus data from 69 patients with 416 observations were used in the final analysis. The demographics of the patients included in the pharmacodynamic database are given in Table III. No further deletions were made to the data set. An inhibitory sigmoidal maximal change in aggregation (E_{max}) model with a baseline effect characterized the platelet aggregation data with the use of NONMEM.[16-18]

$$\%\text{Aggregation} = E_0 - (E_{max} \cdot Cp^{\gamma})/(IC50^{\gamma} + Cp^{\gamma})$$

The model was parameterized for E_0, the baseline platelet aggregation in the absence of lotrafiban; E_{max}, the maximal change in aggregation from baseline in the presence of lotrafiban and aspirin; IC_{50}, the concentration that elicits 50% of maximal inhibition; and the Hill coefficient, γ, which describes the steepness of concentration-effect relationship. Model building was conducted with the use of the same criteria that were used in the development of the pharmacokinetic model. Covariates examined during the development of the population pharmacokinetic model were also evaluated for the assessment of their effect on lotrafiban pharmacokinetics and pharmacodynamics.

Table III. Patient demographics for population pharmacokinetic-pharmacodynamic data set (N = 69)

Characteristic	*No. of patients*	*Mean*	*Range*
Age (y)		60.7	38-81
Weight (kg)		83.1	45.4-185.3
*CL_{CR} (mL/min)**		78.6	30.7-157
Qualifying diagnosis		—	—
Recent MI (Diag 1)	6	—	—
Recent UA (Diag 2)	26	—	—
Recent TIA (Diag 3)	13	—	—
Recent stroke (Diag 4)	24	—	—
Cigarette smoking status		—	—
Nonsmokers	49	—	—
Smokers	20	—	—

*CL_{CR} was calculated on the basis of the equation of Cockcroft and Gault.

Bleeding adverse events. The severity of the bleeding events versus estimated lotrafiban exposure data from all 444 patients was evaluated by logistic regression analysis[20-22] with a population-based approach with the use of NONMEM.[16-18] Peak lotrafiban concentration was also assessed as a potential predictor of bleeding-related adverse events (data not shown). Graphically, lotrafiban exposure appeared to be predictive of the severity of bleeding adverse events and to be more reliable than peak concentrations. Platelet aggregation was not assessed because there were no aggregation data available for the majority of patients participating in this study.

Lotrafiban exposure (area under the plasma concentration-time curve from 0 to 24 hours [$AUC_{0\text{-}24}$]) was calculated by division of the daily dose by the maximum a posteriori Bayesian estimates of the individual CL/F values from the final population pharmacokinetic model for each patient. Patients in the placebo group (n = 94) were assigned an $AUC_{0\text{-}24}$ value of 0. CL/F values for patients without pharmacokinetic data (N = 24) were based on the equation for CL/F developed from the population model. Bleeding event data were ordered categorical data taking on integer values on a scale of 0 to 3 with the values as follows: 0, no bleeding; 1, minor bleeding with no study withdrawal; 2, minor intolerable bleeding with study withdrawal; and 3, major bleeding event. The baseline probabilities of the occurrence of these bleeding adverse events in order of severity were described as B0, B1, B2, and B3, respectively.

The probability of a patient experiencing a bleeding adverse event was assumed to be independent within

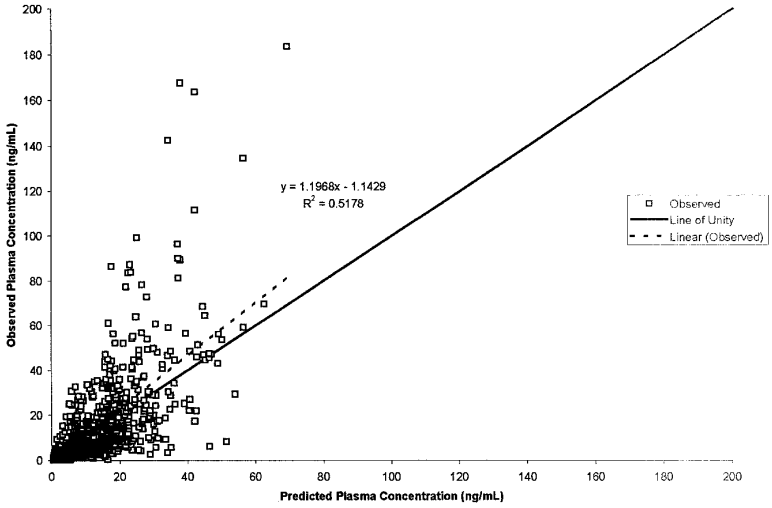

Fig 1. Observed versus predicted plasma lotrafiban concentration. Predicted concentrations were obtained from the final population pharmacokinetic model.

each patient. Thus no predetermined progression in which a patient might experience different grades of adverse events was assumed. Also, after preliminary assessment of the data, the probability of experiencing bleeding adverse events did not appear to change with time; therefore the probability of experiencing an adverse event was modeled only in relation to estimated exposure. Therefore the probability that an adverse event (Y) is less than or equal to an event of grade m (in which m = 1, 2, or 3) is given by the function listed below.

$$g\{P(Y \leq m|\eta)\} = f\text{ASA}(m) + f\text{d}(\text{AUC}) + \eta_Y$$

in which g{x} denotes the logit transformation of the odds of an occurrence of a specific grade of bleeding adverse event, ***f***ASA is the baseline odds of an event in a patient receiving only aspirin (parameterized as Bn), and ***f***d is the probability function describing the drug effect given a particular exposure to lotrafiban and η_Y, which is a scalar random individual effect parameter that accounts for the fact that patients may experience different grades of adverse events at the same exposure. The probability function for ***f***d(AUC) described a nonlinear E_{max} relationship, which is given below.

$$f\text{d}(\text{AUC}) = \frac{AE_{\max} \cdot AUC(0-24)}{AE_{50} + AUC(0-24)}$$

in which AE_{max} is the maximum increase in probability of occurrence that can be caused by lotrafiban and AE_{50} is the exposure at which half maximal increase in the probability odds ratio is reached.

RESULTS

Population pharmacokinetics. The plasma lotrafiban concentration versus time data were well described by a 2-compartment model with first-order input and first-order elimination from the central compartment. The distribution of random residual errors was best described by a constant coefficient of variation model, and an exponential model best described interpatient variability. The best parameter estimates were obtained with the use of the first order with conditional estimation method. The final population model retained age, CL_{CR}, and sex as covariates on CL/F, as shown in the following equation.

$$\frac{CL}{F} = \Theta_1 \cdot (1 - GENDER) + \Theta_2 \cdot GENDER + \Theta_3 \cdot \left(\frac{CLCR}{78}\right) - \Theta_4 \cdot \left(\frac{AGE}{61}\right)$$

in which CL_{CR} and AGE were normalized to their respective median values determined from the database (78 mL/min and 61 years, respectively), male patients were designated with 0, and female patients were designated with 1. In addition, age was retained as a covariate on V_2/F and V_3/F. The formulas for these parameters follow.

$$\frac{V_2}{F} = \Theta_5 - \Theta_6 \cdot \left(\frac{AGE}{61}\right)$$

$$\frac{V_3}{F} = \Theta_8 - \Theta_9 \cdot \left(\frac{AGE}{61}\right)$$

The population pharmacokinetic parameters estimated from the final model are listed in Table IV. For a typi-

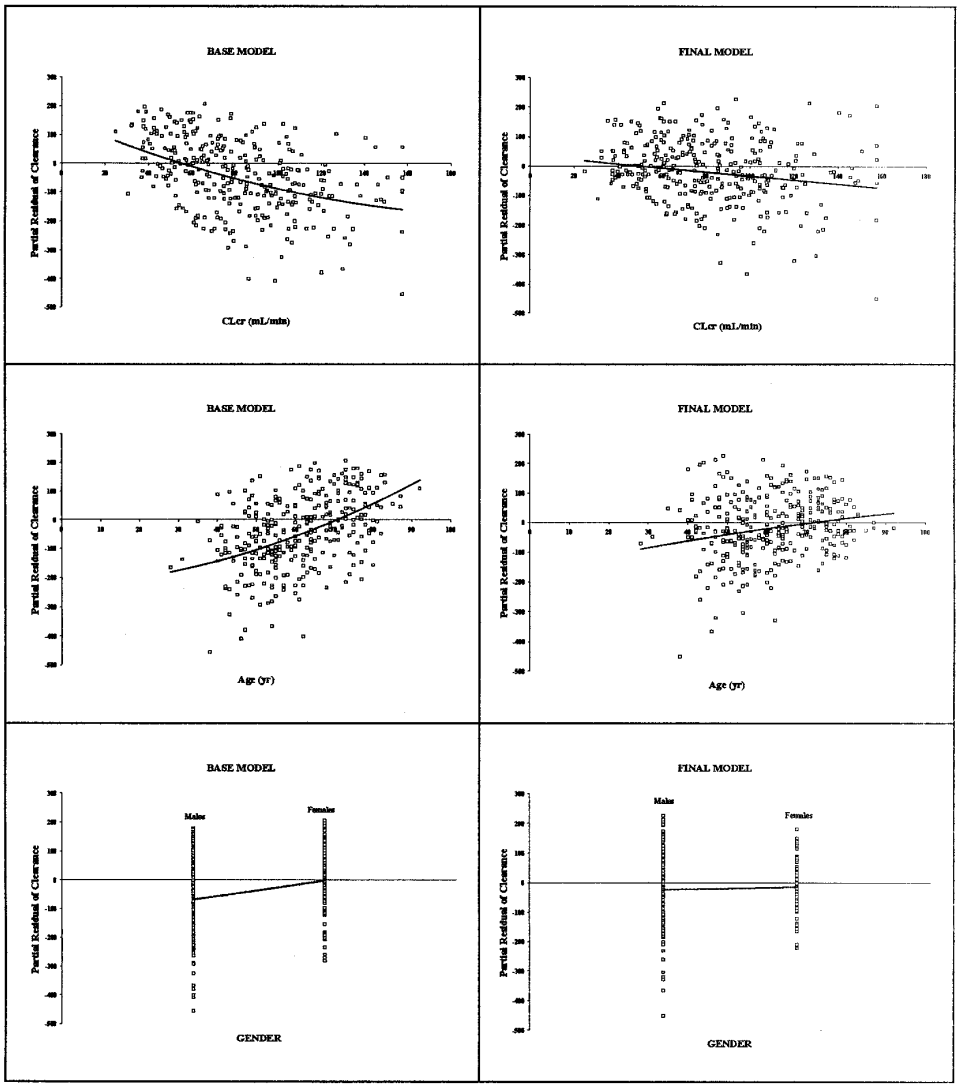

Fig 2. Partial residuals of clearance versus the key covariates (CL_{CR}, age, and sex) for the base and final pharmacokinetic models.

cal 61-year-old male patient with a CL_{CR} of 78 mL/min, the estimates for CL/F, V_2/F, and V_3/F were 299 L/h, 1660 L, and 29800 L, respectively. The standard error of estimate for the parameters was reasonable; it ranged between 21% and 66%. The interpatient variability in CL/F was approximately 50%, and for V_2/F and V_3/F it was approximately 78%. The quality of the final model fit is represented graphically in Fig 1. The visual agreement between predicted and observed concentrations indicates that the model adequately describes the data, although peak concentrations from several individuals who received the highest dose were underestimated. Attempts to model these higher than expected concentrations with the use of a covariate for this dose group were not successful. The partial residuals for the CL/F of lotrafiban versus several key covariates (CL_{CR}, age, and sex) from the base and the final models are shown in Fig 2. The trends in the parameter estimates observed in the base model (left panels) are largely accounted for in the final model (right panels).

When the outliers were returned to the database and the final model was rerun, the covariance step failed, although the final parameter estimates were generally unchanged (Table V); this suggested that the removal of the data points had not greatly influenced the results. Because the covariance matrix was required for future modeling work, the final model output with outliers removed was retained.

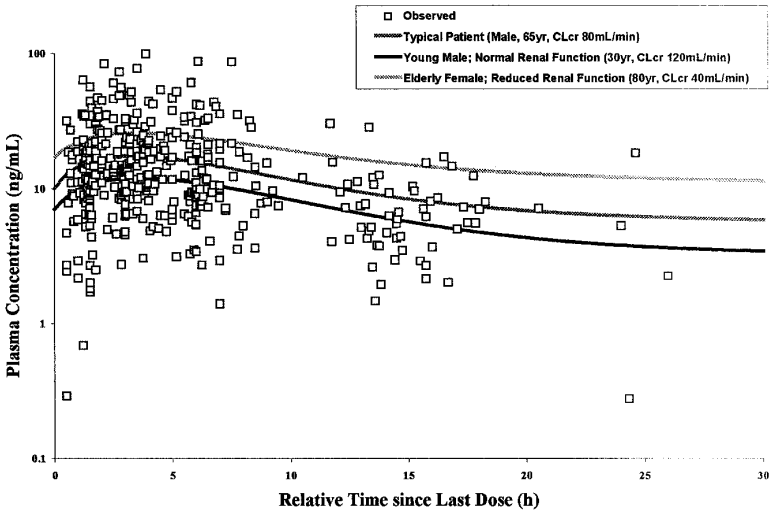

Fig 3. Plasma lotrafiban concentration versus time profile for the 50-mg twice daily regimen for demographics for 3 representative patients. Symbols are observed data, and the lines represent the results of nonlinear regression fitting of the data to the population pharmacokinetic model.

Table IV. Population pharmacokinetic parameters of lotrafiban with outliers removed

Parameter		*Estimate (%SE)*	*Interpatient variability (%)*
*CL/F (L/h)**			
For men	Θ1	409 (26.2)	49.7
For women	Θ2	350 (30.3)	
Effect of CL_{CR}	Θ3	58.9 (66.4)	
Effect of age	Θ4	169 (43.4)	
V_2/F *(L)*†	Θ5	3440 (28.8)	77.7
Effect of age	Θ6	1780 (48.4)	
Q (L/hr)	Θ7	215 (20.6)	NE
V_3/F *(L)*‡	Θ8	81,000 (44.7)	77.7
Effect of age	Θ9	51,200 (44.5)	
K_a (h^{-1})	Θ10	0.21 FIXED	NE
Random residual variability		54.1	

%SE, Percent standard error of the parameter estimate expressed as a percentage of the coefficient of variation; Q, Q-07; NE, not evaluated; K_a, absorption rate constant.

*$CL/F = Θ1 \cdot (1 – GEND) + Θ2 \cdot (GEND) + Θ3 \cdot (CL_{CR}/78) – Θ4 \cdot (AGE/61)$, in which GEND = 0 for men and GEND = 1 for women.

†$V_2/F = Θ5 – Θ6 \cdot (AGE/61)$.

‡$V_3/F = Θ8 – Θ9 \cdot (AGE/61)$.

Table V. Population pharmacokinetic parameters of lotrafiban with outliers restored

Parameter		*Estimate*	*Interpatient variability (%)*
*CL/F (L/h)**			
For men	Θ1	405	53.7
For women	Θ2	364	
Effect of CL_{CR}	Θ3	59.2	
Effect of age	Θ4	134	
V_2/F *(L)*†	Θ5	5670	111
Effect of age	Θ6	3450	
Q (L/h)	Θ7	191	NE
V_3/F *(L)*‡	Θ8	84,700	109
Effect of age	Θ9	50,800	
K_a (h^{-1})	Θ10	0.21 FIX	NE
Random residual variability		66.8	

Q, Q-07; NE, not evaluated; K_a, absorption rate constant.

*$CL/F = Θ1 \cdot (1 – GEND) + Θ2 \cdot (GEND) + Θ3 \cdot (CL_{CR}/78) – Θ4 \cdot (AGE/61)$, in which GEND = 0 for men and GEND = 1 for women.

†$V_2/F = Θ5 – Θ6 \cdot (AGE/61)$.

‡$V_3/F = Θ8 – Θ9 \cdot (AGE/61)$.

Effect of age, creatinine clearance and sex. From the population pharmacokinetic modeling it was evident that age and CL_{CR} have a significant effect on the pharmacokinetics of lotrafiban. The CL/F of lotrafiban decreased with increase in age or decrease in CL_{CR} (Table VI). Lotrafiban exposure was significantly higher in elderly patients (patients who were older than 65 years) and in patients with low CL_{CR} (<60 mL/min) compared with that in young patients with normal CL_{CR}. In general, sex appeared to have a minor effect on the pharmacokinetics of lotrafiban; female patients had a slightly lower CL/F than male patients had (Table VI). Only in elderly patients did the difference between sexes become notable, with the difference increasing from 14% to 24% in patients with good renal function and from 17% to 32% in patients with poor renal function.

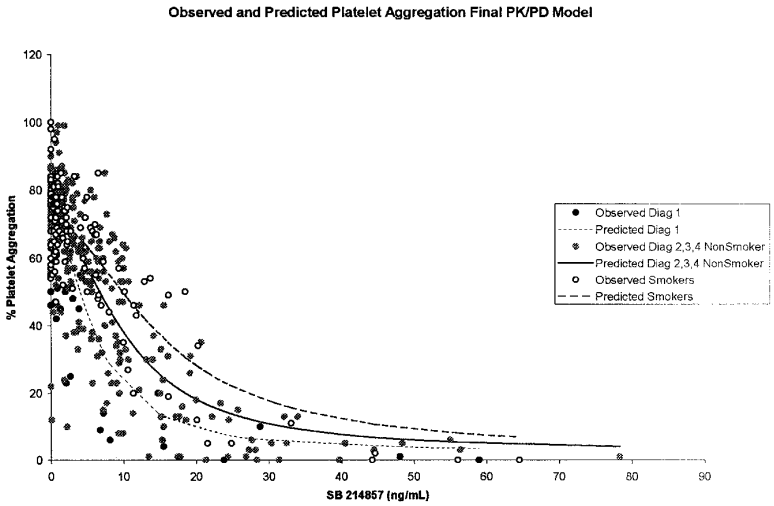

Fig 4. Platelet aggregation versus plasma lotrafiban concentration profiles. Symbols are observed data, and the lines represent the results of nonlinear regression fitting of the data to the population pharmacodynamic model. Diagnosis 1 included patients with recent MI; diagnoses 2, 3, and 4 included patients diagnosed with UA, TIA, and stroke, respectively.

Observed and predicted plasma lotrafiban concentration versus time profiles during multiple oral dosing at 50 mg twice daily for three different patient demographics are shown in Fig 3. The predicted profiles show that the model adequately describes the central tendency of observed plasma concentration-time data obtained from this regimen and suggests that a relatively wide range of plasma concentrations might be expected in a diverse patient population receiving the same dose of lotrafiban.

Population pharmacodynamics. In the dose groups of 20, 50, and 100 mg, the platelet aggregation decreased in a concentration-dependent manner after dosing on day 1. However, for the placebo and 5-mg dose groups, platelet aggregation did not differ significantly from the baseline in general. The concentration-effect relationship was steep, and approximately 50% of the maximum decrease in platelet aggregation occurred at lotrafiban concentrations of 10 to 12 ng/mL (Fig 4). Platelet aggregation reached nearly minimum values at lotrafiban concentrations approaching 20 ng/mL, and there was no further decrease in aggregation at higher concentrations.

The relationship between plasma lotrafiban concentration and platelet aggregation was described with the use of an inhibitory sigmoidal E_{max} model with a baseline. The fitted line shows that the plasma lotrafiban concentration versus platelet aggregation data were well characterized by the proposed model (Fig 4). The estimates for the population pharmacodynamic model are listed in Table VI. The typical values for platelet aggregation at baseline (ADP induced) and maximum change from baseline (E_{max}) caused by lotrafiban were 71% and 69%, respectively; the latter corresponded to a maximum of 97% inhibition of platelet aggregation. The model parameters were well defined with standard errors of less than 24%. The interpatient variability was low (<33%).

Table VI. Effect of age, CL_{CR}, and sex on typical value of CL/F of lotrafiban

		CL/F (L/h)	
Age (y)	*CL_{CR} (mL/min)*	*Men*	*Women*
30	120	417	358
	30	349	290
90	80	220	161
	30	182	123

The IC_{50} of lotrafiban was 10.4 ng/mL for nonsmokers with a history of UA, TIA, or stroke (Table VII). Smoking and a qualifying diagnosis of recent MI appeared to have a significant effect on the IC_{50} of lotrafiban. The IC_{50} was 46% higher in smokers (15.2 ng/mL); this suggests that smokers are less sensitive to the effect of lotrafiban than are nonsmokers. In contrast, patients with a qualifying diagnosis of recent MI had an IC_{50} of 6.23 ng/mL, which was approximately

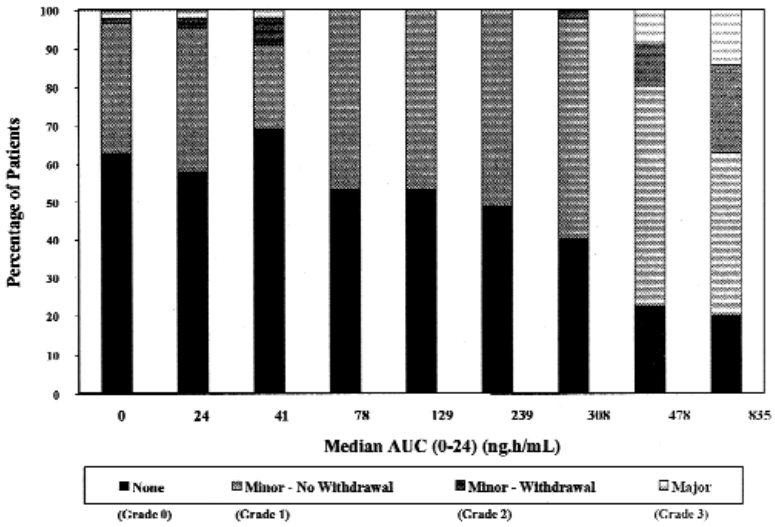

Fig 5. Percentage of patients with incidence of bleeding versus median steady-state systemic lotrafiban exposure ($AUC_{0\text{-}24}$). The $AUC_{0\text{-}24}$ values were estimated with the use of the maximum a posteriori Bayesian estimates of clearance determined from the final population pharmacokinetic model.

Table VII. Population pharmacodynamic parameters for lotrafiban

Parameter	*Estimate (%SE)*	*Interpatient variability*
E_o *(% aggregation)*	71.2 (2.11)	12.9%
E_{max} *(% aggregation)*	68.7 (6.87)	4.32%
IC_{50} *(ng/mL)*		
Recent MI	6.23 (23.8)	NE
Nonsmokers with UA, TIA or stroke	10.4 (11.2)	32.6%
Smokers with UA, TIA or stroke	15.2 (13.0)	21.6%
γ *(%SE)*	1.85	
Random residual variability (additive [% aggregation])	9.42	

%SE, Percent standard error of the parameter estimate expressed as the percentage of the coefficient of variation; NE, not evaluated.

40% lower than that for other patients. Five of 6 patients with recent MIs had been treated with other agents that inhibit platelet aggregation, including ticlopidine, heparin, abciximab, or tissue plasminogen activator; this may have contributed to the apparent lower concentration of lotrafiban needed to maximally inhibit platelet aggregation. Twenty-six of the 69 patients included in the data set were concurrently taking one of the medications listed. The mean value of the individual IC_{50} estimates for these 26 patients was 8.20 ng/mL, which was lower than the average IC_{50} for patients with UA, TIA, or stroke (10.4 ng/mL) who were not taking these medications. Therefore it appears that the medications listed may have some additional effect on the inhibition of platelet aggregation caused by lotrafiban.

Bleeding adverse events. For graphical purposes, data on bleeding adverse events from all 444 patients were divided into groups based on predicted lotrafiban exposure ($AUC_{0\text{-}24}$). A frequency distribution plot of the severity of all grades of bleeding events by group showed that increasing systemic exposure of lotrafiban resulted in an increase in the severity of bleeding events (Fig 5). The relationship between calculated lotrafiban exposure and severity of bleeding event was described with the use of a nonlinear E_{max} logistic regression model. The estimates of model parameters are listed in Table VIII. The standard errors of the baseline odds ratio of a bleeding event of grade 0 to 2 (B0, B1, and B2) were reasonable (~23%). The standard errors for AE_{max} and AE_{50} were higher (42% and 59%) but not unexpected because of the nonlinearity of the function used to describe these data. The interpatient variance was low (11%).

The data and model-based probabilities of a bleeding event were plotted against median lotrafiban exposure ($AUC_{0\text{-}24}$; Fig 6). Up to a median $AUC_{0\text{-}24}$ value of approximately 300 ng · h/mL, the probability of experiencing a grade 2 (minor but intolerable) or grade 3 (major) bleeding event was low, as predicted by the model. As the $AUC_{0\text{-}24}$ values increased, the probabil-

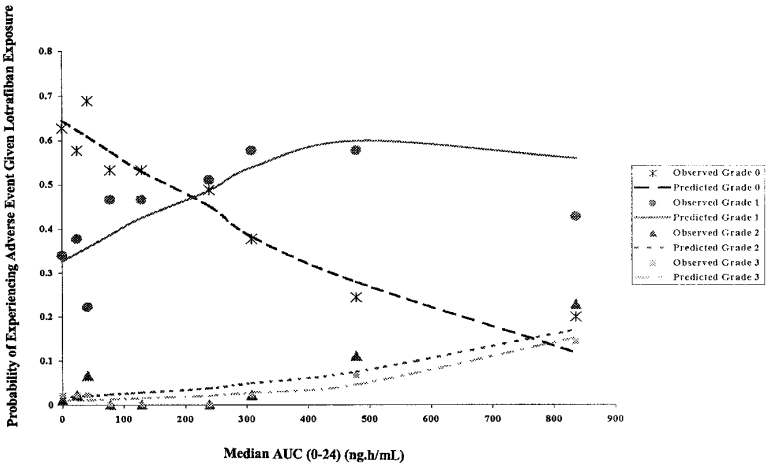

Fig 6. Probability of severity of bleeding event versus median steady-state systemic lotrafiban exposure. Symbols are observed data, and the lines represent the results of logistic regression analysis of the data. Adverse event grades 0, 1, 2, and 3 were no bleeding event, minor tolerable bleeding event, minor intolerable event, and major bleeding, respectively.

ity of experiencing a grade 2 or 3 bleeding adverse event increased steadily. At a median $AUC_{0\text{-}24}$ value of 835 ng · h/mL, the predicted probability of a grade 2 or grade 3 event approached 15%. The majority of patients with $AUC_{0\text{-}24}$ values of 835 ng · h/mL or higher were elderly patients with impaired renal function or were patients randomly assigned to the dosing regiment of 100 mg twice a day.

DISCUSSION

Lotrafiban is an inhibitor of platelet aggregation that could potentially be useful in the secondary prevention of cardiovascular and cerebrovascular diseases such as deaths, MIs, TIAs, UAs, and strokes. In this study, the population pharmacokinetics and pharmacodynamics of lotrafiban were evaluated in the target patient population.

The selection of an appropriate dose regimen for evaluation in phase III trials is generally difficult because phase II trials often are not powered to evaluate and compare safety and efficacy at each dose group studied. Population-based approaches were used to evaluate data from APLAUD. Covariates were identified for the pharmacokinetics of lotrafiban, and the resulting pharmacokinetic model was used to estimate patient exposure. Age and CL_{CR} appeared to have the greatest effect on the pharmacokinetics of lotrafiban; with increasing age and decreasing renal function the systemic exposure increased significantly. Because systemically available lotrafiban is primarily eliminated as unchanged drug in urine, the effect of renal function on the clearance is not unexpected. In this database, age and CL_{CR} were highly correlated, as might be expected because the Cockcroft-Gault equation used to estimate patient CL_{CR} in this study utilizes age in the formula. Therefore the effect of age on the pharmacokinetics of lotrafiban may be partly related to the decrease estimated CL_{CR}. However, in a separate study of normal volunteers whose age and measured CL_{CR} were not as highly correlated, these characteristics were again found to impact the pharmacokinetics of lotrafiban (data on file). Thus, renal function and age appear to have separate effects on lotrafiban pharmacokinetics. Furthermore, the effect of age was seen on both V_2/F and V_3/F as well as CL/F; this suggested that age affected all parameters in a similar fashion. This could indicate that the extent of oral absorption of lotrafiban might be higher in elderly patients than in young patients (patients younger than 65 years). Because the oral bioavailability of lotrafiban is low as a result of poor permeability, the difference in absorption in elderly patients may be partly attributed to slow gut transit times or to altered gut permeability, which potentially allow elderly patients to absorb a greater percentage of the dose than younger patients. Sex was found to play a minor role in affecting the pharmacokinetics of lotrafiban; women had slightly greater systemic exposure values than did men.

The relationship between exposure and bleeding adverse events was evaluated with the use of logistic regression analysis, which helped to identify a lotrafiban exposure above which the tolerability of bleeding events was not acceptable. The use of the population pharmacokinetic model to predict patient expo-

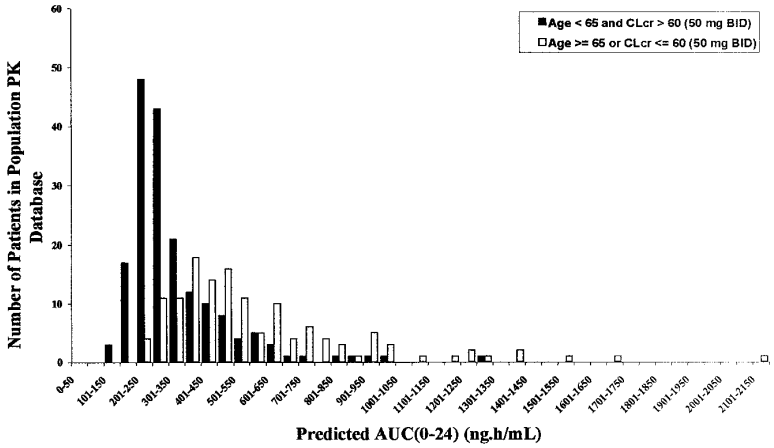

Fig 7. Distributions of individual predicted steady-state systemic lotrafiban exposure for the 50-mg twice a day regimen in patients younger than 65 years with normal renal function compared with elderly renally impaired patients ($CL_{CR} \leq 60$ mL/min).

Table VIII. Parameter estimates for bleeding event model with the use of logistic regression analysis

Parameter	*Estimate (%SE)*
B0	0.615 (23.1)
B1	2.95 (7.40)
B2	1.05 (23.3)
AE_{max}	8.67 (41.6)
AE_{50}	2170 (59.0)
Interpatient variability (%)	10.5

%SE, Percent standard error of the parameter estimate expressed as the percentage of the coefficient of the variation.

sures with varying dose regimens helped to identify potential regimens that would reduce the chance of a patient exposure exceeding the identified upper level.

Although platelet aggregation had not been validated as a surrogate marker for clinical efficacy, the pharmacodynamic model nonetheless provided some feedback on the likely platelet aggregation profile with the use of the selected dose regimens and provided additional confirming information on the dose selection. The pharmacokinetic-pharmacodynamic analysis of this phase II study indicated that the relationship between platelet aggregation and plasma lotrafiban concentrations is steep; small changes in concentration may result in large changes in platelet aggregation. The effect of patient demographics on the pharmacodynamic activity of lotrafiban was also evaluated. Smokers were found to have a higher average IC_{50}, which suggests that a greater concentration of drug is needed to reach the same effect on platelet aggregation as in nonsmokers. This would suggest that patients who smoke are less sensitive to the effects of lotrafiban on platelet aggregation than nonsmokers. In contrast, patients with a recent MI had a lower average IC_{50} value compared with other patients who were nonsmokers. The increased sensitivity to lotrafiban in these patients may be related to the previous administration of medications that inhibit platelet aggregation. The IC_{50} for nonsmokers without a recent MI estimated in this study was similar to that found previously for healthy subjects (10.4 versus 10.6 ng/mL; data on file).

The distribution of estimated exposure ($AUC_{0\text{-}24}$) at 50 mg of lotrafiban twice a day is shown in Fig 7. Relative to the young patients, the exposure distribution for elderly and renally impaired patients was shifted to the right with an extended tail. Therefore in elderly patients or patients with impaired renal function a dose of 50 mg twice a day may result in higher than desirable lotrafiban exposure (ie, $AUC_{0\text{-}24} > 835$ ng · h/mL). As was seen in the logistic regression analysis, this exposure may result in a higher incidence of minor intolerable and major bleeding events. Therefore elderly and renally impaired patients may require a lower dose of lotrafiban than 50 mg twice a day. Simulations of a dose of 30 mg twice a day in elderly patients (age, ≥65 years) and renally impaired patients (CL_{CR}, ≤60 mL/min) generated a distribution of estimated exposures similar to those in patients who were younger than 65 years with normal renal function who were given 50 mg twice a day (Fig 8). Furthermore, at these doses, a majority of patients (60%-80%) are predicted to sustain 40% to 80% inhibition of platelet

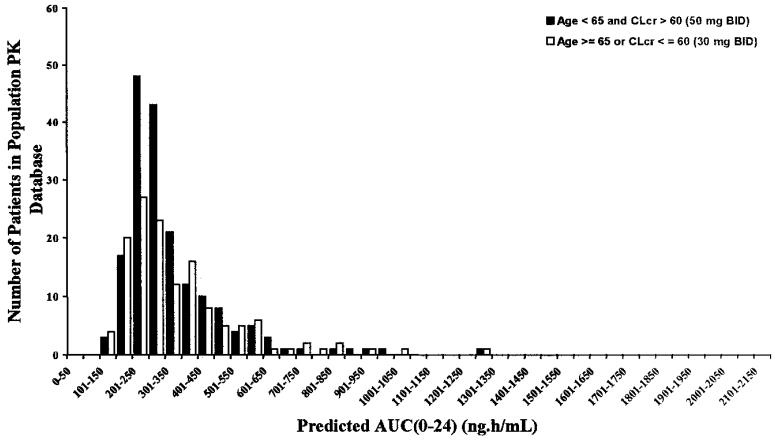

Fig 8. Distributions of individual predicted steady-state systemic lotrafiban exposure for the 30-mg twice a day regimen in elderly or renally impaired patients compared with the 50-mg twice a day regimen in patients younger than 65 years of age and with normal renal function.

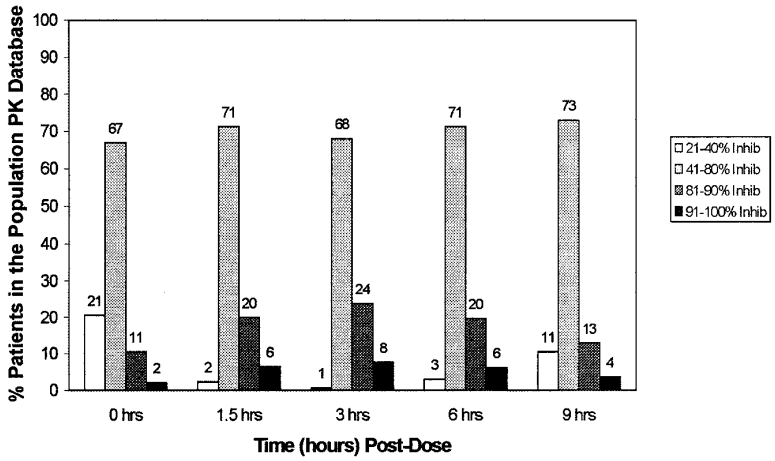

Fig 9. Predicted ex vivo steady-state percentage of inhibition of platelet aggregation (20 μmol/L of ADP) at 0, 1.5, 3, 6, and 9 hours after the dose. The 30-mg twice a day dose regimen was used to simulate response for elderly or renally impaired patients, and the 50-mg twice a day regimen was used with young patients with normal renal function. The predicted percentage of inhibition of platelet aggregation was categorized.

aggregation during a 12-hour dosing interval at steady-state (Fig 9), a range of inhibition that is thought to be safe and potentially therapeutically effective.

Because the range of predicted lotrafiban exposures with the recommended dose regimen was anticipated to be wider, the number of patients expected to have greater than optimal inhibition of platelet aggregation was also scrutinized. Approximately 2% to 8% of the patients were expected to have platelet aggregation inhibition between 91% and 100% at some point during the dosing interval (Fig 9), although the clinical relevance of this is not clear because platelet aggregation is not a surrogate marker for either safety or for efficacy. However, the fraction of patients with a predicted $AUC_{0\text{-}24}$ in excess of 834 ng · h/mL at the recommended dose was very low; this suggests that the majority of patients would not be at undue risk for serious bleeding events. The consequences of having low exposure were not known.

On the basis of these results, a dose regimen of 30 mg twice a day was proposed for use in the phase III trial (Blockade of the GP IIb/IIIa Receptor to Avoid

Vascular Occlusion study) in elderly patients (age, ≥ 65 years) and renally impaired patients (CL_{CR}, ≤60 mL/min), and a dose regimen of 50 mg twice a day was proposed for young patients with normal renal function. The actual safety and efficacy of this regimen was tested during this study; approximately 4% of the patients enrolled in this study experienced serious bleeding adverse events.

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