A Multicenter, Randomized, Double-Blind, Placebo-Controlled Study to Evaluate the Safety of PZ-128 in Subjects Undergoing Non-Emergent Percutaneous Coronary Intervention

Thrombin Receptor Inhibitory Pepducin in PCI (TRIP-PCI)

DRUG NAME:	PZ-128
IND NUMBER:	110,931
PROTOCOL NUMBER:	TMC-PZ128-02 NCT02561000
DRUG DEVELOPMENT PHASE:	2
TRIAL SPONSOR:	Tufts Medical Center, Inc. 800 Washington Street Boston, MA 02111
SPONSOR REPRESENTATIVE & MEDICAL DIRECTOR:	[redacted]
CO-PRINCIPAL INVESTIGATORS:	[redacted]
PROTOCOL DATE:	04 AUG 2016

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STUDY PROTOCOL AMENDMENT HISTORY

A Multicenter, Randomized, Double-Blind, Placebo-Controlled Study to Evaluate the Safety of PZ-128 in Subjects Undergoing Non-Emergent Percutaneous Coronary Intervention-**T**hrombin **R**eceptor **I**nhibitory **P**epducin in **PCI** (TRIP-PCI) (Protocol No. TMC-PZ128-02)

Amendment Number	**Protocol Version Date**
Initial preparation(s)	07 JUL 2015 15 SEP 2015 09 OCT 2015
Final	04 JAN 2016
#1	**04 AUG 2016**

1.0 SYNOPSIS

Name of Sponsor/Institute: Tufts Medical Center, Inc.

Funding grant provided by the National Heart, Lung and Blood Institute, National Institutes of Health (NHLBI/NIH)

Name of Investigational Product: PZ-128	**Name of Active Ingredient:** PZ-128

Title of Study: A Multi-center, Randomized, Double-Blind, Placebo-Controlled Study to Evaluate the Safety of PZ-128 in Subjects Undergoing Non-Emergent Percutaneous Coronary Intervention

Thrombin **R**eceptor **I**nhibitory **P**epducin in **PCI** (TRIP-PCI) (Protocol No. TMC-PZ128-02)

Study Sites: Inova Heart and Vascular Institute, Inova Fairfax Hospital, Falls Church, VA
Tufts Medical Center, Boston, MA
UMass Memorial Medical Center, Worcester, MA

Projected Study Timelines:		**Clinical Phase:** 2
Actual date first subject enrolled:	27 May 2016	
Estimated date last subject enrolled:	December 2018	
Estimated date last subject completed:	March 2019	

Indication: Non-emergent percutaneous coronary intervention (PCI)

Primary Objective: The primary objective is to evaluate the safety of single dose PZ-128, in addition to the standard of care, with respect to the incidence of major and minor bleeding events, as assessed by the TIMI (Thrombolysis in Myocardial Infarction) classification, in subjects undergoing non-emergent PCI or non-emergent cardiac catheterization with the intent to perform PCI.

Key Secondary Objective: The key secondary objective is to evaluate the potential clinical benefit of PZ-128 with respect to the composite endpoint of major adverse cardiac events (MACE) - any of cardiovascular death, non-fatal myocardial infarction (MI), non-fatal stroke, recurrent ischemia requiring hospitalization or urgent coronary revascularization.

Other Secondary Objectives Related to Safety: Other secondary safety objectives include evaluation of the incidences of the following:

1. bleeding that does not meet the TIMI criteria for major or minor (i.e., minimal bleeding);
2. "clinically significant" bleeding;
3. TIMI bleeding related to coronary artery bypass grafting (CABG); and

Other Secondary Objectives Related to Efficacy: Other secondary efficacy objectives will include evaluation of the following:

1. incidence of the individual components of the MACE composite as measures of potential clinical benefit;
2. incidence of definite or probable stent thrombosis as a measure of potential clinical benefit;
3. inhibition of platelet aggregation induced by SFLLRN (PAR1 agonist) and thrombin as an indicator of the desired pharmacodynamic effect, and ADP (adenosine diphosphate), collagen and AYPGKF (PAR4 agonist) to evaluate the specificity of PZ-128;
4. effect of the investigator choice of $P2Y_{12}$ inhibitor on the individual components of the MACE composite; and
5. population pharmacokinetics.

Exploratory Objectives: Exploratory objectives will include the following:

1. incidence of bleeding events according to the BARC (Bleeding Academic Research Consortium) classification;
2. effect on expression of systemic markers of thrombosis and platelet reactivity, inflammation, and metabolomics; and
3. population pharmacogenomics.

Methodology: Multicenter, prospective, randomized, double-blind, placebo-controlled, balanced-parallel-groups, non-inferiority investigation.

Number of Subjects (planned): Approximately 600 (400 to receive PZ-128, 200 to receive placebo). There is no pre-specified minimum or maximum enrollment for each site.

Diagnosis and Main Criteria for Inclusion: Adult subjects (≥18years), meeting all inclusion criteria and none of the exclusion criteria, scheduled to undergo non-emergent coronary angiography +/- PCI (if indicated) for either stable coronary artery disease or non-ST-segment elevation acute coronary syndromes (NSTE-ACS) (i.e., candidates for elective or urgent PCI), and for whom elective use of a GP IIb/IIIa inhibitor (GPI) is not intended, will be enrolled.

Investigational Product, Dose and Mode of Administration: PZ-128 lyophilized powder for reconstitution in Sterile Water for Injection and dilution in 5% Dextrose. Subjects receive 0.3 mg/kg or 0.5 mg/kg as a single-dose via a continuous, 2-hour, intravenous infusion.

Reference therapy, Dose and Mode of Administration: Matching placebo lyophilized powder for reconstitution in Sterile Water for Injection and dilution in 5% Dextrose. Subjects receive placebo corresponding to PZ-128 0.3 mg/kg or 0.5 mg/kg as a single-dose via a continuous, 2-hour, intravenous infusion.

Duration of Treatment: Single dose infusion to start within one hour before the diagnostic angiography begins. Subjects are expected to receive the entire 2-hour infusion regardless of whether they proceed to undergo PCI, CABG or medical management.

Criteria for Evaluation

Primary Safety Endpoint: The primary safety endpoint of the study will be the incidence of TIMI major plus minor bleeding not related to CABG through 30 days after treatment. TIMI Major bleeding is defined as (1) intracranial hemorrhage or (2) clinically significant overt signs of bleeding associated with a decrease in hemoglobin concentration of ≥5 g/dL (or hematocrit ≥15%) or (3) fatal bleeding within 7 days. TIMI Minor bleeding is defined as clinically overt signs of bleeding (including imaging) associated with a decrease in hemoglobin concentration of 3 to <5 g/dL (or hematocrit of 9 to <15%) that does not otherwise meet criteria for major bleeding. Hemoglobin concentration and hematocrit will be adjusted for any transfusion of packed red blood cells or whole blood given between enrollment and post-transfusion measurements.

Key Secondary Efficacy Endpoint: The key secondary efficacy endpoint is the incidence of any component of the MACE composite through 30 days and 90 days after treatment.

Other Secondary Endpoints Related to Safety: Other secondary safety endpoints include the incidences of the following:

1. TIMI minimal bleeding not related to CABG (defined as clinically significant overt signs of bleeding associated with a drop in hemoglobin concentration of <3 g/dL (or hematocrit of <9%) that does not otherwise meet criteria for major or minor bleeding) through 30 days after treatment;
2. "clinically significant" bleeding (defined as TIMI major or minor bleeding or bleeding requiring medical attention) through 30 days after treatment;
3. TIMI bleeding related to CABG through 30 days after treatment (subjects who undergo CABG will specifically have assessment of (a) surgical wound bleeding using quantitative total chest tube drainage in the first 24 hours; (b) need for transfusion of blood products in the first 48 hours; and (c) need for surgical re-exploration).

Other Secondary Endpoints Related to Efficacy: Other secondary efficacy endpoints will include the following:

1. incidence of the individual components of the MACE composite through 30 days and 90 days after treatment;

2. incidence of definite or probable stent thrombosis through 30 days and 90 days after treatment;
3. inhibition of platelet aggregation induced by SFLLRN, thrombin, ADP, collagen and AYPGKF, relative to baseline at several time points following treatment at select sites;
4. population pharmacokinetics at baseline and at several time points following treatment; and
5. effect of the investigator choice of P2Y12 inhibitor (i.e., clopidogrel versus prasugrel/ticagrelor) on the individual components of the MACE composite.

Exploratory Endpoints: These endpoints are not part of the formal objectives and are included as additional ways to evaluate bleeding and to gather information for future research.

1. incidence of bleeding events according to the BARC (Bleeding Academic Research Consortium) classification through 30 days after treatment;
2. change from baseline in select systemic markers of thrombosis and platelet reactivity, inflammation, and metabolomics at select time points;
3. population pharmacogenomics at baseline.

Clinical evaluations including physical examinations, vital signs, laboratory tests, electrocardiograms and the assessment of adverse events will be performed for all subjects during hospitalization and at follow-up time points as measures of the safety profile.

Randomization: Randomization will be conducted prior to the diagnostic angiography.

Stratification: Randomized assignment of PZ-128 and placebo will be stratified for whether the cardiac catheterization ± PCI is being done on an elective or urgent basis at the time the operator decides to perform the procedure.

Sample Size/Power: The current design anticipates a sample size of 600 subjects who will undergo non-emergent cardiac catheterization ± PCI. The sponsor anticipates that the incidence of major plus minor bleeding among placebo-treated subjects will range from 2% to 6% on the basis of historical experience. Given the anticipated sample sizes and estimated range of incidences associated with placebo, the study will be able to demonstrate that the bleeding risk of PZ-128 is non-inferior to the risk associated with placebo with non-inferiority margins in the range of 2.6 to 7.6%, depending on the underlying bleeding risk of placebo. Given the anticipated benefits of PZ-128, these non-inferiority margins are reasonable for the planned safety decisions generated from this trial.

Statistical Methods: All subjects who are randomized will be considered evaluable for the analyses. The following statistical analysis will be provided for the primary endpoint. The point estimate of the difference in risk of any event of non-CABG TIMI major plus minor bleeding through 30 days post treatment for between PZ-128 and placebo (e.g., Pr (Event in PZ-128 patient)-Pr (Event in placebo patient)), and corresponding lower one-sided 95% confidence interval will be presented. These estimates will be generated using subjects who underwent diagnostic angiography for all PZ-128 treatment arms pooled across both doses (~400 subjects) and all placebo (~200 subjects) and descriptive results will also be generated for each individual dose. The proposed phase 2 study is not of sufficient size or of sufficient follow-up duration to allow formal analysis of clinical efficacy.

Sub-analyses of the endpoints will be performed across subject cohorts (i.e., PCI, CABG and medical management). Comparisons for primary and secondary endpoints will be done for PZ-128 in aggregate and per dosing group. The potential influence of baseline risk factors and concomitant therapies such as statins, choice of $P2Y_{12}$, and aspirin dosing on the incidences of the endpoints will also be explored using generalized linear models and survival models.

Subgroup analyses will be conducted for baseline characteristic variables such as age, sex, and race.

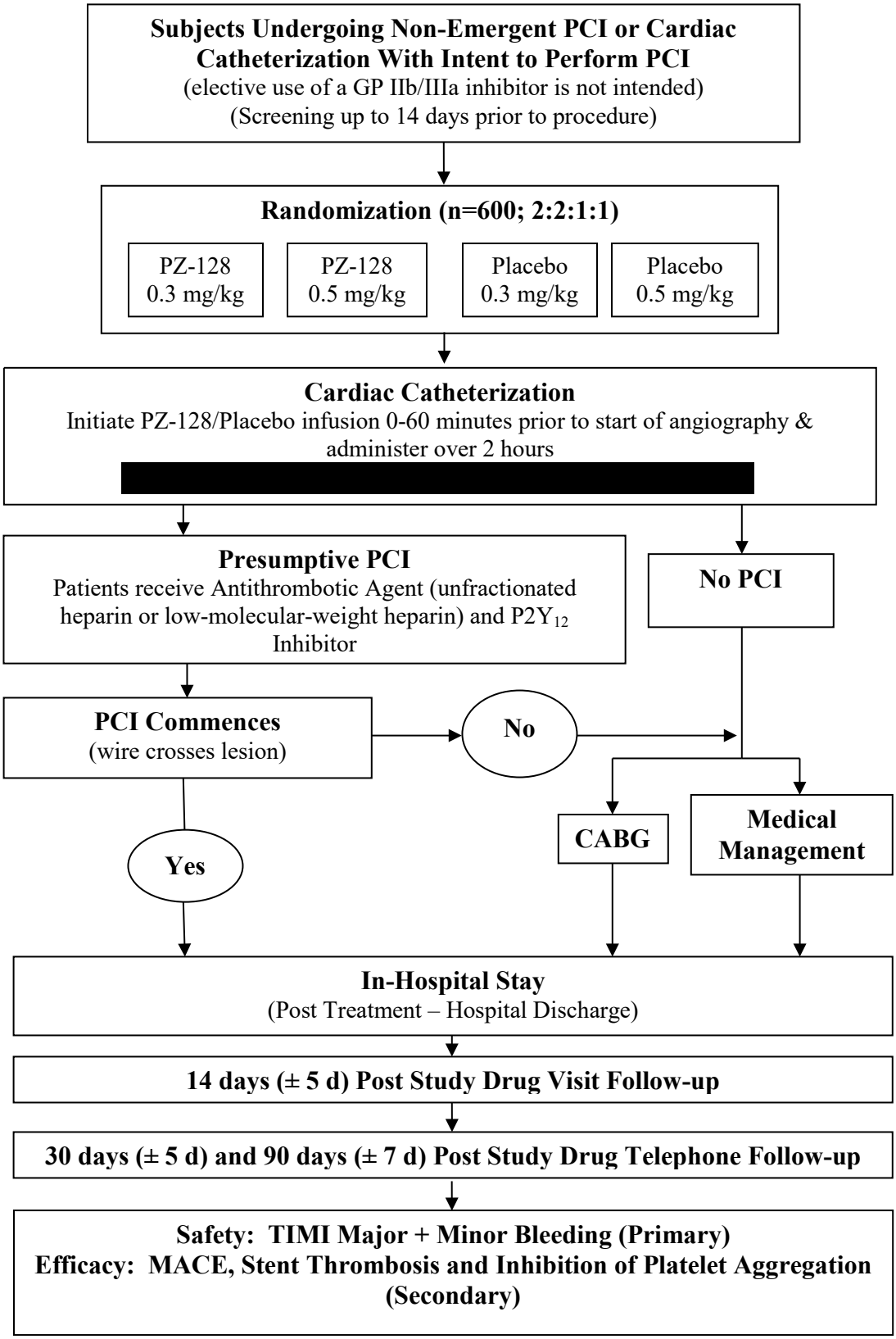
All bleeding events will be characterized as non-CABG or CABG related and the following bleeding rates will be reported: total, CABG-related and non-CABG-related.

Tabulations of descriptive statistics will be provided for TIMI major plus minor bleeding. In addition, tabulations of descriptive statistics will also be provided on the incidence of all other relevant variables.

Although no formal stopping boundaries will be utilized, one interim safety analysis will be performed at approximately one year following the enrollment of the first subject, or after enrollment of 50% of the study population, whichever occurs first. The Data and Safety Monitoring Board (DSMB) will be provided point and interval estimates of PZ-128 benefit at the time of that analysis based on survival estimates, and will evaluate evidence of bleeding risk in light of the potential efficacy benefits in making recommendations about stopping the trial.

Among the committees formed to oversee the conduct of the study will be an independent DSMB to protect further the rights, safety and well-being of subjects who will be participating in this study by monitoring the progress and results of the trial, and an independent Clinical Events Committee (CEC) to review and adjudicate each suspected bleeding and efficacy endpoint event while blinded to treatment.

1.1 Study Design Diagram



1.2 Study Flow Chart

Study Assessment/Procedure	Screening[10]	Enrollment	Always in Hospital				Post-Treatment Follow-up		
			Immediately Prior to Study Drug Infusion[11]	Immediately Post Study Drug Infusion	Post Cardiac Cath ± PCI[14]	24 Hours (±2) Post Start of Study Drug or Discharge[17, 18]	14 days (± 5) Post Study Drug Infusion Office Visit	30 days (± 5) Post Study Drug Infusion Phone Call	90 days (± 7) Post Study Drug Infusion Phone Call
Explain Study / Written Informed Consent(s)[1]	X								
Review of Inclusion/Exclusion Criteria	X								
Medical History[2] and Demographics	X								
Physical Examination	X					X[19]	X		
Electrocardiogram (12-Lead ECG)	X				X		X		
Vital Signs[3] Assessment	X		X	X	X	X	X		
Previous and Concomitant Medications	X →								
Adverse Events Assessment (Bleeding and Non-Bleeding)		← X →							
Serious Adverse Events Assessment (Bleeding and Non-Bleeding)		← X →							
Clinical Efficacy Endpoint Events Assessment (MACE, Stent Thrombosis)		← X →							
Standard Local Laboratory Tests:									
Hematology[4]	X				X	X	X		
Serum Chemistry[5]	X					X			
PT/INR, aPTT	X					X			
Activated Clotting Time					X[15]				
Lipid Panel[6]	X								
Hemoglobin A1c	X								
Pregnancy Test[7] (urine)	X		X[7]				X		

Study Assessment/Procedure	Screening[10]	Enrollment	Always in Hospital				Post-Treatment Follow-up		
			Immediately Prior to Study Drug Infusion[11]	Immediately Post Study Drug Infusion	Post Cardiac Cath ± PCI[14]	24 Hours (±2) Post Start of Study Drug or Discharge[17, 18]	14 days (± 5) Post Study Drug Infusion Office Visit	30 days (± 5) Post Study Drug Infusion Phone Call	90 days (± 7) Post Study Drug Infusion Phone Call
Sample Collections For Research (required):									
Cardiac Troponin I[8] Serum			X		X[16]	X[16]			
Sample Collections For Research (optional[1]):									
Pharmacodynamics (PD)-Platelet Aggregation[9] Whole Blood			X	X[12]		X[12, 20]	X		
Pharmacokinetics (PK)[9] Plasma			X	X[12]		X[12, 20]	X		
Exploratory Biomarkers Plasma			X	X[13]		X[13, 20]	X		
Pharmacogenomics (DNA/RNA) Whole Blood			X						
Randomization		X							
[redacted]									

1. Written informed consent for the optional sampling: pharmacogenomics (DNA/RNA), pharmacodynamics-platelet aggregation, pharmacokinetics, and exploratory biomarkers may be contained in the same instrument as written informed consent for the rest of the study, or may be a separate document, at the discretion of the institutional review board (IRB). Regardless, separate signatures of informed consent are required to collect each of the optional samples.
2. Including subject's cardiac history, the clinical evaluation leading to the cardiac catheterization procedure (i.e., CAD presentation, Canadian Cardiovascular Society Classification for angina, Thrombolysis in Myocardial Infarction Risk Score for UA/NSTEMI, and New York Heart Association Classification of Functional Status for heart failure), bleeding history and allergy history (drugs, IV contrast media, food, stings and atopic conditions and any required treatment/ medical intervention).
3. Vital signs to include heart rate, blood pressure, and at Screening only, height and weight.
4. Hematology includes a complete blood cell count, differential WBC count and platelet count.
5. Chemistry includes sodium, potassium, chloride, carbon dioxide, glucose, BUN, creatinine, alkaline phosphatase, total bilirubin, ALT and AST.
6. Lipid Panel includes total cholesterol, high-density lipoprotein cholesterol, calculated low-density lipoprotein cholesterol and triglycerides (does not have to be fasting).
7. Urine pregnancy test (urine beta-HCG) must be performed on all women of child-bearing potential prior to randomization and must also be repeated prior to study drug administration if screening assessment >24 hours.

8. Cardiac Troponin I (cTnI) analysis to be conducted by a centralized lab at Tufts Medical Center. Additional cardiac biomarkers (i.e., Troponin I or T, CK-MB) analyzed locally at the clinical site for routine management of the subject will be guided by standard of care/local hospital clinical practice and the results will be collected when applicable.
9. At select sites only in the Sub-study.
10. Screening evaluations must be performed prior to Enrollment/Randomization and must be within 14 days prior to cardiac catheterization and study drug treatment (may include standard of care preadmission testing for the planned procedure).
11. Study drug infusion must be started within 0-60 min prior to the commencement of the cardiac catheterization procedure (diagnostic angiography).
12. Samples will be collected at 30 min ± 5 min, 1 h ± 5 min, 2 h + 15 min (immediately after completion of infusion), 6 h – 2 h (4 to 6 h) and 24 h ± 2 h (or at hospital discharge, whichever occurs sooner) after the start of the study drug infusion (T=0).
13. Samples will be collected at 2 h + 15 min (i.e., immediately after completion of infusion), 6 h – 2 h (4 to 6 h) and 24 h ± 2 h (or at hospital discharge, whichever occurs sooner) after the start of the study drug infusion (T=0).
14. After cardiac catheterization, if PCI does not commence or after PCI commences, regardless of outcome. Except as specifically noted, timing should be consistent with standard practice at the clinical site.
15. Following administration of the initial bolus of anticoagulant (UFH/LMWH), activated clotting time will be measured locally at "peak dose" and just prior to sheath removal, as well as according to the standard of care and applies only to PCI subjects.
16. Collect samples for research cTnI at 4 to 8 h and 12 to 24 h after completion of PCI. In addition, collect samples immediately after occurrence of symptoms suggestive of acute coronary syndrome, and again 4 to 8 h and 12 to 24 h after symptoms if the subject has not yet been discharged from the hospital.
17. Assessments should be performed at 24 Hours (± 2 h) post start of study drug infusion or just prior to Discharge, whichever occurs sooner.
18. If a subject remains in the hospital for >24 hours, additional testing/evaluation will be guided by standard of care and local guidelines.
19. The clinical site's standard of care, cardiac catheterization nursing assessment may be performed to evaluate the subject's clinical status in lieu of a complete physical exam at the 24 hour/Discharge (whichever occurs sooner) time point.
20. After the 24 hour collection, if a subject remains in the hospital for ≥48 h, collect an additional sample at discharge.

2.0 TABLE OF CONTENTS

2.1 List of Appendices

3.0 LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

ACC	American College of Cardiology
ACS	Acute coronary syndrome(s)
ACT	Activated clotting time
ADP	Adenosine diphosphate
AE	Adverse event
AHA	American Heart Association
ALT	Alanine aminotransferase
AMI	Acute myocardial infarction
API	AmbioPharm, Inc.
aPTT	Activated partial thromboplastin time
ARC	Academic Research Consortium
ASA	Acetyl salicylic acid (aspirin)
AST	Aspartate aminotransferase
AUC	Area under plasma concentration-time curve from zero to infinity
AYPGKF	Ala-Tyr-Pro-Gly-Lys-Phe (PAR4 agonist peptide)
BP	Blood pressure
BUN	Blood urea nitrogen
CABG	Coronary artery bypass graft surgery
CAD	Coronary artery disease
cAMP	Cyclic adenosine monophosphate
CBC	Complete blood count
CEC	Clinical Events Committee
CFR	USA Code of Federal Regulations
CHF	Congestive heart failure
CI	Confidence interval
CK	Creatine kinase
CKD	Chronic kidney disease
CK-MB	Creatine kinase-myocardial band
C_{max}	Maximum plasma (peak) drug concentration
CRF	Case report form
CRS	Central Randomization Service

CSR	Clinical study report
cTn	Cardiac troponin
CVA	Cerebrovascular accident
CXR	Chest x-ray
DAPT	Dual anti-platelet therapy
DSMB	Data and Safety Monitoring Board
ECG, EKG	Electrocardiogram
eCRF	Electronic case report form
EDC	Electronic data capture
Endpoint	A status of the subject that constitutes the 'endpoint' of a subject's participation in a clinical study and that is used as the final outcome
FACS	Fluorescence activated cell sorter
FAX	Telefacsimile
FDA	Food & Drug Administration, USA
GCP	Good Clinical Practice
GLP	Good Laboratory Practices
GMP	Good Manufacturing Practices
GPI	Glycoprotein IIb/IIIa inhibitors
GPCR	G protein coupled receptor
GPRP	Gly-Pro-Arg-Pro (fibrinogen inhibitor)
h	Hour(s)
Hct	Hematocrit
HDL-C	High-density-lipoprotein cholesterol
Hgb	Hemoglobin
HPR	High platelet reactivity
HR	Heart rate
IATA	International Air Transport Association
ICH	International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use
IFU	Instructions for Use
IND	Investigational New Drug Application
INDSR	Investigational New Drug Safety Report
INR	International Normalized Ratio

IPA	Inhibition of platelet aggregation
IP	Investigational Product(s)
IRB	Institutional Review Board
IPA	Inhibition of platelet aggregation
ITT	Intent-to-treat- refers to a study population for statistical analysis
IUD	Intrauterine device
IV	Intravenous, intravenously
IVRS	Interactive voice response system
Kg	Kilogram
LBBB	Left bundle branch block
LC/MS/MS	Liquid chromatography/tandem mass spectrometry
LDL-C	Low-density-lipoprotein cholesterol
LMWH	Low molecular weight heparin
LTI	Lyophilization Technology, Inc.
LVEF	Left ventricular ejection fraction
MACE	Major adverse cardiac events
MedDRA	Medical Dictionary for Regulatory Activities
mg	milligram
MI	Myocardial infarction
min	Minute(s)
nM	Nanomolar
MMP-1	Matrix metalloprotease-1
NSTE-ACS	Non-ST segment elevation acute coronary syndrome
NSTEMI	Non-ST segment elevation myocardial infarction
NYHA	New York Heart Association
PAR	Protease Activated Receptor
PAR1	Protease Activated Receptor-1
PAR4	Protease Activated Receptor-4
$P2Y_{12}$	A sub-type of P2 receptor found on platelets
PCI	Percutaneous coronary intervention
PD	Pharmacodynamic
PE	Physical exam

PK	Pharmacokinetic
PPP	Platelet poor plasma
PRBCs	Packed red blood cells
PRN	Pro re nata (as needed, as necessary)
PRP	Platelet rich plasma
Principal Investigator	A person responsible for the conduct of a clinical study at an investigational study site
PSI	Post start of the infusion
PT	Prothrombin time
Qualified designee	A person designated by the investigator who is qualified by training and/or experience to perform the specified tests, evaluation, measurement, recording, procedure, or other technique, for the purpose of a physical examination, a qualified designee is a medical professional licensed and/or approved per local requirements to performed the required portions of a physical examination
RAVE	Proprietary name for Medidata web based data capture software
RBC	Red blood cell
SAE	Serious adverse event
SAP	Statistical analysis plan
SD	Standard deviation
SFLLRN	Ser-Phe-Leu-Leu-Arg-Asn (PAR1 agonist peptide)
SGOT	Serum glutamic oxaloacetic transaminase (AST)
SGPT	Serum glutamic pyruvic transaminase (ALT)
SNP	Single nucleotide polymorphism
SOC	Standard(s) of care
SOP	Standard operating procedure(s)
STEMI	ST-segment-elevation myocardial infarction
$t_{1/2}$	Half-life
TAT	Thrombin-anti thrombin III complex
TEG	Thromboelastography
TIA	Transient ischemic attack
TIMI	Thrombolysis in Myocardial Infarction
TNFα	Tumor necrosis factor-α
TRAP	Thrombin receptor activating peptide (SFLLRN)

TRA-PCI	Thrombin Receptor Antagonist-Percutaneous Coronary Intervention
TXA2	Thromboxane A2
TRIP-PCI	Thrombin Receptor Inhibitory Pepducin-Percutaneous Coronary Intervention
UA	Unstable angina
UFH	Unfractionated heparin
ULN	Upper limit of normal
URL	Upper reference limit
US, USA	United States of America
WBC	White blood cell count
WHO	World Health Organization
WHODD	World Health Organization drug dictionary
µg	Microgram
µM	Micromolar

4. INTRODUCTION

The investigator should refer to the current version of the Investigator's Brochure (IB) for more detail.

4.1 Background

4.1.1 Clinical Overview

Ischemic complications during and after percutaneous coronary interventions (PCIs) are strongly influenced by platelet function [1, 2]. Platelets play a critical role in normal hemostasis after injury, as well as in the formation of an arterial thrombus that causes acute closure of vessels in the cardiovascular system [3] and in atherogenesis [4]. Platelet activation is initiated and perpetuated by binding of multiple agonists to specific G-protein–coupled receptors (GPCRs) [5]. Protective hemostasis involves platelet adhesion to sites of vascular injury mediated by von Willebrand factor and collagen in the vessel wall, activation and recruitment of additional platelets (mediated primarily by collagen-stimulated release of thromboxane A2 (TXA2) and adenosine diphosphate (ADP) from adherent platelets), and stabilization of the fibrin-mesh clot mediated by thrombin generation [4].

Currently, the only effective agent preventing thromboxane A2 generation is aspirin (ASA). Although ASA does not impact strong agonists like thrombin or collagen-dependent platelet activation under high arterial shear stress, it lowers the risk of cardiovascular events in subjects with established cardiovascular disease by approximately 25%. ASA is first-line antiplatelet therapy [6]. ADP is an agonist for the platelet $P2Y_1$ and $P2Y_{12}$ receptors and is a key cofactor in platelet activation induced by thrombin and collagen, which results in release of more ADP. Clopidogrel is a pro-drug yielding an active metabolite that binds irreversibly to the $P2Y_{12}$ receptor. In the Clopidogrel in Unstable angina to prevent Recurrent Events (CURE) study, the addition of clopidogrel to ASA reduced the incidence of the composite of cardiovascular death, non-fatal MI, or stroke by 20% relative to ASA alone (9.3% vs 11.4%) in subjects mainly receiving medical treatment for non-ST-segment-elevation acute coronary syndrome (NSTE-ACS); however, the combination also resulted in a modest increased risk of major bleeding (3.7% vs 2.7%) [7]. Among the subset of subjects who received PCI, the incidence of minor bleeding was greater among those who received clopidogrel (3.5%) rather than placebo (2.1%) [8]. Pharmacodynamic studies evaluating antiplatelet responses in subjects undergoing stenting have revealed various limitations of $P2Y_{12}$ inhibition such as a delayed or irreversible pharmacodynamic response; distinct subject-to-subject response variability with a substantial percentage of subjects exhibiting non-responsiveness; and a potential influence of drug-drug interactions [9]. Compared with clopidogrel, new and more potent oral $P2Y_{12}$ inhibitors such as prasugrel and ticagrelor reduce the risk of ischemic events by ~20% in subjects with acute coronary syndrome (ACS) [10, 11]. However, intensified blockade of $P2Y_{12}$ with new $P2Y_{12}$ agents may be associated with elevated bleeding and potentially other side effects such as dyspnea [12]. In this regard, ASA and $P2Y_{12}$ receptor antagonists inhibit platelet aggregation induced by collagen, TXA_2 and ADP, properties that may interfere with normal hemostasis. The slow offset of $P2Y_{12}$ inhibition by all currently

approved oral agents may be problematic in subjects requiring coronary artery bypass graft surgery [13].

Moreover, despite improvements in morbidity and mortality, subjects with cardiovascular disease remain at a substantial risk of future ischemic events. Thus, there exists a need for new antiplatelet strategies that will provide additional efficacy with minimum to no added risks of bleeding [14].

Thrombin, a plasma serine protease, plays a key role not only in coagulation and hemostasis, but also in the activation of a variety of cell types [15]. Three thrombin responsive, G-protein-coupled, protease-activated receptors (PARs) are known (PAR1, PAR3, and PAR4); however, many of the cellular actions of thrombin in the human body appear to be mediated through the high-affinity PAR1 thrombin receptor on the surface of these target cells [15]. PAR1 is activated through proteolysis and cleavage of the N-terminal extracellular domain of the receptor [16]. This cleavage leads to the generation of a new amino terminus, which functions as a tethered ligand that binds to extracellular loops of the receptor, leading to a conformational change and transmembrane signaling to intracellular G proteins [17]. Many of the resulting cellular effects are thought to be mediated via the induction and release of a host of secondary intracellular mediators, including the raising of intracellular calcium [18]. The high affinity PAR1 thrombin receptor has been implicated in a variety of cardiovascular disorders including atherosclerosis, thrombosis, and restenosis following PCI [15]. The generation of thrombin is markedly increased at the sites of vascular injury [19]. Both atherosclerotic lesions and angioplasty specimens express tissue factor, PAR1 and actively generate thrombin. PAR1 is also expressed on smooth muscle cells and monocytes/ macrophages isolated from atherectomy samples of human blood vessels and on the surface of smooth muscle cells following angioplasty procedures. These facts suggest that both thrombin and its receptor are involved in the pathology of thrombotic disorders [15, 20, 21]. By selectively interfering with the cellular actions of thrombin, an antagonist would have an antiplatelet effect under conditions in which thrombin-stimulated platelet activation is critical, including PCI, acute coronary syndromes such as unstable angina and acute myocardial infarction, and stroke. Hence, a thrombin-receptor antagonist should have an improved safety profile over direct and indirect thrombin inhibitors.

Contrary to thrombin inhibitors (bivalirudin, hirudin, argatroban, and dabigatran), PAR1 antagonists do not directly interfere with the procoagulant activity of thrombin cleavage of fibrinogen. Because thrombin-dependent fibrin generation is largely unaffected by inhibition of PAR1, a thrombin-receptor antagonist should be more specific than a thrombin inhibitor in treating arterial thrombosis. Indeed, early preclinical studies demonstrated that PAR1 antagonism did not prolong bleeding time or coagulation-related time (activated partial thromboplastin time (aPTT), prothrombin time (PT), or thrombin time) [22, 23]. PAR1 antagonism also did not inhibit ADP- or collagen-induced platelet aggregation. Taken together, these findings suggest that PAR1 inhibition can permit the formation of initial platelet monolayer necessary for control of bleeding, but block pathological thrombus propagation that occurs at the site of plaque rupture or endothelial denudation [24].

Approved by the U.S. Food and Drug Administration (FDA) in 2014 for secondary prevention of cardiac events, the PAR1 antagonist, vorapaxar (SCH 530348) is marketed by Merck & Co, Inc. (Whitehouse Station, NJ) and E-5555 (atopaxar) was developed by Eisai Company, Ltd. (Japan). Vorapaxar, an orally active, synthetic analog of himbacine with high affinity (K_i = 3-8 nM) [25] to PAR1, has an exceptionally long elimination half-life of 6.6-13 days [26] and a functionally irreversible binding mode [27]. The pharmacodynamic half-life typically exceeds the entire lifespan of a circulating platelet with 50% recovery of platelet function by 4-8 weeks after a single 20 or 40 mg loading dose of vorapaxar [28]. The onset of inhibition of vorapaxar on PAR1 activity occurs by 2 h after receiving the loading dose. A phase 2 randomized clinical trial, TRA-PCI, tested the safety, tolerability and preliminary efficacy of this compound in 1030 subjects undergoing non-emergent cardiac catheterization with intent to perform percutaneous revascularization [29]. The trial was designed as a sequential randomization scheme. First, prior to angiography all subjects were randomized 3:1 to receive one of three loading doses (10, 20 or 40 mg) or matching placebo. Subsequently, only those who actually underwent PCI (n = 573; 56% of the total cohort) – representing the primary cohort of interest – were further randomized to a 60-day maintenance treatment of 0.5, 1 or 2.5 mg or matching placebo. The PCI cohort also received unfractionated heparin, low-molecular-weight heparin, or bivalirudin, and a loading dose of clopidogrel (300-600 mg) and aspirin (162-325 mg oral or iv 150-500 mg). The incidence of the primary endpoint of TIMI major plus minor bleeding was low overall (3% in both the treatment and placebo groups) and no significant differences were observed in the primary cohort between the placebo and study drug groups. Similarly, TIMI major plus minor bleedings were not significantly different in both of the secondary cohorts of subjects medically managed and surgically revascularized. Although the study was not powered to detect these differences, the composite endpoint of death, major cardiac adverse event or stroke was proportionately less in subjects treated with any vorapaxar dose (n = 24; 6%) than with placebo (n = 13; 9%; odds ratio 0.67; 95% CI 0.33, 1.34), a difference mainly driven by a reduction of the myocardial infarction (MI) component in vorapaxar-treated subjects. Also, a dose-effect relationship was observed, with the 40 mg loading dose achieving the strongest reduction in the rate of MI. This study suggested a potential benefit with trends towards lower thrombotic events, without an increased bleeding risk and encouraged the development of phase 3 trials to evaluate the clinical benefit of vorapaxar in combination with current standard-of-care dual antiplatelet therapy in high risk subjects with coronary artery disease.

The safety and efficacy of vorapaxar was also conducted in Japanese subjects (n=117) with a history of non-ST-segment elevated (NSTE) ACS who were receiving standard-of-care therapy (aspirin, ticlopidine and heparin) [30]. Subjects were randomized to receive a loading dose (20 or 40 mg) or placebo and daily oral maintenance doses (1 or 2.5 mg) or placebo for 60 days post PCI. The primary endpoint was bleeding (TIMI criteria) and the exploratory endpoint included all-cause death and MACE (non-fatal MI, non-fatal stroke, hospitalization for recurrent ischemia or urgent coronary revascularization). Periprocedural MI was defined as an elevated CK-MB or troponin-I (above 3 times the upper limit of normal with >50% increase above baseline) was measured at baseline, 8, 16 and 24 h after PCI. Vorapaxar did not result in excess bleeding in the Japanese subjects with NSTE ACS and significantly (p=0.013) reduced the incidence of periprocedural MI by 2.5-fold in subjects undergoing PCI. The

majority of MIs that did occur were asymptomatic elevations of CK-MB and troponin-I that were documented during the periprocedural period shortly after PCI. The safety of vorapaxar was also evaluated in Japanese subjects (n=90) with a history of ischemic cerebral infarction [31]. All subjects received 1.0 mg or 2.5 mg vorapaxar or placebo once daily for 60 days plus aspirin (75-150 mg/day). The primary endpoint was overall incidence of adverse events (AE excluding MACE). The AE rate was not significantly different with the dual vorapaxar/aspirin regimen at either dose. The secondary endpoint of bleeding (TIMI categorized) was similar between placebo and vorapaxar [31].

The efficacy and long-term safety of vorapaxar over 1-2.5 year treatment periods was evaluated in two large phase 3 randomized trials. The effects of vorapaxar in preventing MI and stroke in subjects (n=26,449) with atherothrombotic disease (either post-MI, a history of stroke, or peripheral arterial disease) was assessed in the Thrombin Receptor Antagonist TRA 2°P-TIMI 50 study [32]. In addition, a second trial assessed the ability of vorapaxar to prevent MI and stroke in subjects (n=12,944) with chronic ACS (TRA-CER) [26]. Due to elevated bleeding rates, TRA 2°P-TIMI 50 was terminated early in subjects that had experienced a stroke before or during the trial (17% of the enrolled subjects) [32], and TRA-CER was terminated in all subjects due to significantly increases in the risk of moderate and severe bleeding, including intracranial hemorrhage [33]. When added to standard of care in subjects with non-ST elevation ACS and high use of aspirin and $P2Y_{12}$ inhibition, vorapaxar did not significantly reduce the composite of cardiovascular death, MI, stroke, hospitalization for ischemia, or urgent revascularization. Vorapaxar therapy was associated with a reduction in recurrent myocardial infarction at the expense of more bleeding, including more intracranial bleeding. Vorapaxar reached ≥80% inhibition of SFLLRN-induced aggregation at 2 h after a 40 mg loading dose in 96% of subjects [29]. Unlike the relation of ADP-induced platelet aggregation to clinical thrombotic event occurrence, little is known about whether the magnitude of SFLLRN-induced aggregation affects clinical event occurrence. In the PREPARE POST-STENTING trial, we demonstrated that high thrombin-induced platelet – fibrin clot strength was a marker for recurrent ischemic event occurrence in subjects treated with PCI [34]. The effects of vorapaxar and PZ-128 on thrombin activation on platelets in subjects are unknown, as we documented in PCI subjects receiving the direct thrombin inhibitor bivalirudin [19]. Vorapaxar has a suboptimal onset of pharmacodynamic effects with very long pharmacodynamic half-lives of ≥3 weeks. Both of these properties are important limitations for emergent ACS subjects or subjects requiring unscheduled surgery.

PZ-128 is a first-in-class ‘pepducin’ inhibitor of PAR1 intended for use in PCI and ACS subjects [35, 36]. Pepducins are lipidated peptides which selectively target the cytoplasmic surface of their cognate GPCR to inhibit G protein signaling both in vitro and in vivo [37-41]. The PAR1-based pepducin, PZ-128 (P1pal-7), is administered by 2 h duration intravenous injection as a single dose. PZ-128 consists of a seven amino acid peptide (KKSRALF) that is conjugated to palmitate lipid to form an N-palmitoylated peptide which is formulated in a 5% Dextrose USP solution.

PZ-128 has been extensively tested in human blood samples and animals for the ability to block thrombin-PAR1-dependent platelet activation, arterial thrombosis and atherosclerosis.

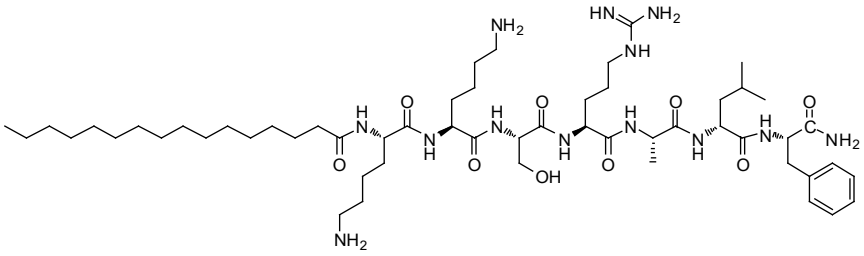
PZ-128 (IV; $t_{1/2}$~1-2 h) has proven to be highly effective in inhibiting platelet function and arterial thrombosis in baboons and guinea pigs and is safe and tolerated up to 30 mg/kg iv (1 h infusion) in non-human primates in 7-day repeat dose GLP studies. PZ-128 has had no effects on coagulation parameters, bleeding time and/or platelet counts in baboon and monkey. A PAR1-dependent blood clotting mechanism has also been identified that is driven by matrix metalloprotease-1 (MMP-1) after platelets are exposed to collagen within the blood vessel wall [42]. PZ-128 also blocked the MMP1-PAR1 pathway and prevented occlusive thrombi from forming [42], suggesting that drugs targeting PAR1 on the inside surface of the plasma membrane such as PZ-128 could offer a new method to treat subjects with ACS and other manifestations of atherothrombotic disease.

There are three areas in which an intracellular PAR-1 antagonist may improve on small molecule inhibitors: 1) effective inhibition of both thrombin and MMP-1 activation of PAR1 by blocking downstream of the receptor, 2) rapid-onset and 3) reversible inhibition of PAR1-induced aggregation, which limits potential side effects such as bleeding when used in combination with standard-of-care ASA and a $P2Y_{12}$ blocker.

4.1.2 Investigational Agent: PZ-128

Pepducins are the first agents to target the cytoplasmic surface of their cognate receptor. PZ-128 is a first-in-class, cell-penetrating lipopeptide pepducin that selectively inhibits PAR1-G protein signaling on the inner leaflet of the lipid bilayer [37, 43, 44]. PZ-128 (P1pal-7) closely resembles the off-state of the corresponding juxtamembrane region of the PAR1 third-intracellular loop and TM6 region. The pepducin rapidly flips across the plasma membrane to target the PAR1-G protein interface to block thrombin activation of the receptor in platelets [43, 45]. The chemical structure of PZ-128 is depicted below.

Chemical Structure of PZ-128



PZ-128 drug substance consists of 7 amino-acid residues (KKSRALF) conjugated to palmitate lipid to form an *N*-palmitoylated peptide. One batch of the active drug substance was manufactured for the phase 2 trial by Ambiopharm, Inc. (API, North August, SC) according to current Good Manufacturing Practices (cGMP) as a sterile white to off-white powder consisting of [redacted]

One batch of PZ-128 active drug product for injection was manufactured for the phase 2 trial by Lyophilization Technology, Inc. (LTI, Ivyland, PA) according to current cGMP as a sterile white to off-white solid lyophilized powder cake contained in 20 cc glass vials stored at -20 ºC ± 10 ºC. [redacted] One batch of PZ-128 matching placebo for injection (inactive components only) was manufactured for the phase 2 trial by LTI, Inc. according to current cGMP as a sterile white to off-white solid lyophilized powder cake contained in 20 cc glass vials stored at -20 ºC ± 10 ºC. PZ-128 and placebo reconstituted in SWFI are diluted into a final IV bag of sterile Dextrose 5% in Water, USP (D5W).

4.1.2.1 Preclinical Studies

An overview of the preclinical studies is provided in the current version of the Investigator's Brochure (IB).

4.1.2.2 Clinical Studies

A phase 1 first-in-human safety study was conducted in 32 subjects who had documented vascular disease or 2 or more risk factors for coronary artery disease. The goal was to test the safety of the PAR1 pepducin in a population which closely approximated the eventual clinical population, namely patients with a risk of developing life-threatening acute arterial thrombosis during PCI. Thirty-one subjects received a single intravenous dose of PZ-128 (1-2 h duration) with escalating dose levels ranging from 0.01 mg/kg to 2 mg/kg, 1 subject received saline only. Subjects either had documented vascular artery disease (22%) or had a variety of risk factors for coronary disease including prior MI (16%), prior PCI and/or CABG (22%), obesity (53%), smoking (41%), hypertension (69%), diabetes (34%), dyslipidemia (81%), and/or age >45 for males and >55 for females. Consistent with the CAD population in the US, the subjects were 57 ± 8 years old, 50% white, 47% black, 59% male, 91 ± 17 kg with a BMI of 31 ± 6. In addition to CAD or multiple CAD risk factors, many of the subjects had a number of co-morbid conditions and were taking multiple medications. Safety evaluations consisted of vital signs, physical exams, ECG, pulmonary function tests, blood tests (clinical chemistry, hematology), bleeding time and coagulation tests, urinalysis, and exploratory tests for allergic markers.

Maximal and final Light Transmission Aggregation (LTA) to the PAR1 agonist SFLLRN versus agonists for other platelet receptors (PAR4, ADP and collagen), was determined for all subjects at several time points. The inhibitory effects of PZ-128 on PAR1 platelet aggregation were dose-dependent with 20-40% inhibition at 0.3 mg/kg, 40-60% at 0.5 mg/kg and up to 80-100% at 1-2 mg/kg. There were no significant effects on aggregation to any other platelet agonists including 160 µM AYPGKF, 5 µM ADP, 20 µM ADP, 4 µg/mL collagen, or 20 µg/mL collagen in any dose cohort.

Aspirin (ASA) appeared to enhance the ability of PZ-128 to block PAR1 platelet aggregation, especially at the 0.5 mg/kg dose. This was expected as ASA monotherapy has been shown to inhibit PAR1 responses in platelets [46] and is used as a concomitant standard-of-care anti-

platelet agent during PCI as discussed above. The subgroup of subjects on concomitant ASA therapy revealed stronger apparent effects of PZ-128 on inhibition of maximal and final aggregation to 8 μM (P=0.084) and 12 μM (P=0.06) SFLLRN in the 0.5 mg/kg dose cohort. The subgroup of subjects receiving ASA in the 0.5 mg/kg dose cohort had 60-80% average inhibition of final aggregation to 8 μM SFLLRN at 30 min-2 h and 100% inhibition by 6 h. There was 40% of recovery to aggregation to 8 μM PAR1 agonist by 24 h and 60% recovery by 192 h (the next sampling time point). Higher concentrations of PAR1 agonist (e.g., 20 μM SFLLRN) showed 80% recovery of aggregation by 24 h and 95% by 192 h in the 0.5 mg/kg dose cohort receiving concomitant ASA. At high doses of 1-2 mg/kg, PZ-128 gave significant mean ~90-100% inhibition at 1-2 h time points regardless of whether the subjects received ASA or not.

At 0.3-0.5 mg/kg doses where anti-PAR1 effects on platelet aggregation were observed at 30 min-2 h, drug levels of PZ-128 achieved therapeutic concentrations of 1-3 μM (~920-2800 μg/mL). Peak drug levels (C_{max}) occurred at the end of the 1 h or 2 h infusion in all dose cohorts with a terminal $t_{1/2}$ of elimination of 1.3-1.8 h. Drug was undetectable in plasma at 24-192 h in all subjects. There was little (≤1-2.6 ng/mL) or no PZ-128 detected in urine at any time point in all subjects consistent with the notion that the pepducin is not eliminated by the kidneys. The drug concentrations were highly linearly correlated with increasing dose at 0.25-8 h time points with R values of 0.980-0.998. Likewise, AUC and C_{max} of PZ-128 in plasma were linearly correlated with dose.

In agreement with pre-clinical studies in non-human primates, persistent anti-platelet effects of 60-100% inhibition were still observed at the 6 h time point with the 0.5 mg/kg dose where plasma PZ-128 drug levels had fallen to <0.5 μM. This is highly consistent with the mechanism of action of the pepducin which flips across the cell membrane where it associates with its cognate receptor on the inner leaflet of the lipid bilayer to give prolonged and selective inhibition of PAR1 activity. Much higher, super-therapeutic drug concentrations were achieved at the 1 mg/kg and 2 mg/kg dose levels with mean peak PZ-128 levels reaching 5 μM and 12 μM, respectively. Accordingly, PZ-128 blocked 85-100% of aggregation to 8 μM SFLLRN agonist at the 1-2 h time points in the 1-2 mg/kg dose cohorts. The drug was well tolerated at 0.01, 0.03, 0.1 and 0.3 mg/kg dose cohorts using a 1 h intravenous infusion with no significant AEs attributable to PZ-128.

[redacted]

No significant effects on coagulation, hemostasis parameters or bleeding were seen at any dose, despite the fact that 65% of the subjects (20/31) who received PZ-128 were also taking aspirin. Of all the laboratory tests, only BUN levels gave a trend towards transiently increasing at the high doses of PZ-128. As creatinine levels were completely unaffected by PZ-128, it is likely that the non-significant increase in BUN was due to transient hypovolemia in certain subjects at the high dose cohort secondary to the drug allergic effect or NPO status. All other laboratory outcomes including ECG parameters, pulmonary function, and urinalysis were not affected by PZ-128.

[redacted]

In summary, PZ-128 is a fast-onset PAR1 pepducin which provides significant anti-platelet efficacy without affecting bleeding or coagulation parameters even in the setting of concomitant aspirin use. Safety and tolerability analysis indicates that PZ-128 should be able to be safely administered at doses of ≤0.5 mg/kg using a 2 h intravenous infusion to subjects with CAD and significant comorbidities typically associated with the PCI population. The information gathered to address the objectives of this study will be used to determine whether the clinical program will proceed to Phase 3, and if so, the dosing regimen to be used.

4.2 Rationale

4.2.1 Rationale for Study Design and Subject Population

This study will be a multicenter, prospective, randomized, double-blind, placebo-controlled, balanced-parallel-groups, non-inferiority investigation to be conducted in conformance with Good Clinical Practice (GCP).

In this phase 2 trial named TRIP-PCI, two intravenous dose levels of PZ-128 (0.3 mg/kg and 0.5 mg/kg) and matching placebo will be studied to determine the safety and efficacy of PZ-128 when added to standard antiplatelet therapy in subjects undergoing non-emergent PCI or non-emergent cardiac catheterization with intent to perform PCI. Standard-of-care procedures outlined in the protocol are based upon practice guidelines for cardiovascular angiography and interventions [47].

Dosing was determined by the safety profile observed in previous GLP animal studies (non-human primates and rodents) and the recently completed phase 1 study which examined the safety, tolerability, pharmacokinetics and pharmacodynamics of PZ-128 in subjects with multiple coronary artery disease risk factors. The phase 1 population closely approximated the eventual clinical population, namely subjects with a risk of developing life-threatening acute arterial thrombosis during PCI.

The phase 2 subject population will consist of approximately 600 low-to-moderate risk subjects including those undergoing elective PCI and subjects with NSTE-ACS. Outcomes will be assessed over 30 day and 90 day time periods. Higher risk subjects with STEMI or those requiring GPI therapy will be excluded. Based on the phase 1 data with PZ-128 which documented anaphylactoid/histaminic reactions in some subjects at high doses of 1-2 mg/kg, subjects with baseline hypotension (systolic blood pressure <95 mm Hg) or history/high risk of drug allergies will be excluded, and a prophylactic anti-histamine regimen will be incorporated. Based on the extensive clinical data with vorapaxar, subjects with a prior history of stroke or transient ischemic attack, or body weight of <60 kg will also be excluded. Bleeding is the most common and severe side effect of anti-thrombotic and/or anti-platelet agents in general. In standard-of-care treatment, subjects undergoing PCI may be exposed to several of these medications (e.g., ASA, $P2Y_{12}$ inhibitor, heparin). When introducing additional potent anti-platelet agents in this setting such as PZ-128, potential efficacy must be balanced against a possible increase in the frequency of bleeding events. The design of the primary outcome measure in this study (relative bleeding rate of PZ-128 vs placebo) is consistent with established practice for evaluation of bleeding events in subjects undergoing cardiac catheterization with intent to perform PCI in phase 2 studies such as TRA-PCI. Addition of an antiplatelet agent to existing standard-of-care in this setting, so-called dual anti-platelet therapy (DAPT), has been studied extensively and found to be beneficial [7]. As PZ-128 is an effective platelet aggregation inhibitor with a unique intracellular mechanism, consequently assessing bleeding (hemostasis) will be the primary endpoint in the present study from a safety and feasibility perspective. Although not sufficiently powered, an early efficacy signal (e.g. reduction in MACE or stent thrombosis) may be detected in this 600 subject study as occurred in earlier studies with oral PAR1 inhibitors (vorapaxar, atopaxar). Thus, this design offers the opportunity to assess the safety and potentially early efficacy of PZ-128 in a setting of current clinical practice.

The study will incorporate extensive PK and PD assessments in a large subset of subjects at the various clinical sites. Inhibition of platelet aggregation will be measured by optical aggregometry (maximal and final LTA) in response to the PAR1 agonist SFLLRN (5-20 μM) and thrombin (1 nM-1 μM) versus agonists for other platelet receptors PAR4, ADP and

collagen (160 μM AYPGKF, 5-20 μM ADP, 4-20 μg/mL collagen) for subjects at select time points. The PK/PD data generated will facilitate the continuing evaluation of the relationship between the pharmacokinetics and IPA and will assess these parameters in this subject population for the first time before progressing into the large Phase 3 program. Other relevant biomarkers including systemic levels of the two PAR1 agonists, thrombin (in complex with anti-thrombin III: TAT) and MMP-1, will be assessed and correlated with outcomes. Pharmacogenomic analysis will be undertaken to investigate whether DNA polymorphisms, RNA levels and microRNAs correlate with the expression of genes involved in thrombosis, inflammation, and cardiometabolic diseases and whether these genetic components correlate with PZ-128 outcomes.

4.2.2 Rationale for the PZ-128 Doses

The design consists of a single dose regimen of PZ-128 0.3 mg/kg, or PZ-128 0.5 mg/kg, or Placebo 0.3 mg/kg, or Placebo 0.5 mg/kg in 2:2:1:1 ratio using a 2-hour continuous IV infusion started within 1 hour prior to commencement of the cardiac catheterization procedure (i.e., diagnostic angiography). This regimen is supported by the:

- short half-life of PZ-128 (1-2 h) with reversibility by higher concentrations of PAR1 agonist;
- inhibition of up to 50-100% PAR1 platelet aggregation with quick onset (by 30 min) and prolonged inhibition of PAR1 platelet activation for at least 6 h after initiation of IV infusion; and
- synergy of anti-PAR1 effects with ASA and $P2Y_{12}$ inhibitors

A single-dose infusion of PZ-128 0.3 mg/kg or 0.5 mg/kg will be administered in this study because patients undergoing non-emergent cardiac catheterization (especially those with NSTE-ACS) are in an acute-care situation that requires rapid and reversible inhibition of the ability of platelets to become activated by thrombin, which can be accomplished by these doses. The dose levels of PZ-128 to be evaluated in this study have been selected based on pharmacodynamic, pharmacokinetic, safety and tolerability data from the recently completed phase 1 study in 32 subjects with known cardiovascular disease or multiple coronary artery disease risk factors. The two dose levels will be studied concurrently.

5.0 STUDY OBJECTIVES

5.1 Primary Objective

The primary objective is to evaluate the safety of a single dose of PZ-128, in addition to the standard of care, with respect to the incidence of major and minor bleeding events, as assessed by the TIMI (**T**hrombolysis **I**n **M**yocardial **I**nfarction) system of classification, in subjects undergoing non-emergent PCI or non-emergent cardiac catheterization with the intent to perform PCI.

5.2 Key Secondary Objective

The key secondary objective is to evaluate the potential clinical benefit of PZ-128 with respect to the composite endpoint of major adverse cardiac events (MACE) - any of cardiovascular death, non-fatal myocardial infarction (MI), non-fatal stroke, recurrent ischemia requiring hospitalization or urgent coronary revascularization.

5.3 Other Secondary Objectives Related to Safety

Other secondary safety objectives will include evaluation of PZ-128 in terms of the incidences of the following:

1. bleeding that does not meet the TIMI criteria for major or minor (i.e., minimal bleeding);
2. “clinically significant” bleeding;
3. TIMI bleeding related to coronary artery bypass grafting (CABG); and

5.4 Other Secondary Objectives Related to Efficacy

Other secondary efficacy objectives will include evaluation of PZ-128 in terms of the following:

1. incidence of the individual components of the MACE composite as measures of potential clinical benefit;
2. incidence of definite or probable stent thrombosis as a measure of potential clinical benefit;
3. inhibition of platelet aggregation induced by SFLLRN (PAR1 agonist) and thrombin as an indicator of the desired pharmacodynamic effect, and ADP (adenosine diphosphate), collagen and AYPGKF (PAR4 agonist) to evaluate the specificity of PZ-128;
4. effect of the investigator choice of $P2Y_{12}$ inhibitor on the individual components of the MACE composite; and

5. population pharmacokinetics.

5.5 Exploratory Objectives

Exploratory objectives will include evaluation of PZ-128 in terms of the following:

1. incidence of bleeding events according to the BARC (Bleeding Academic Research Consortium) classification;
2. effect on expression of systemic markers of thrombosis and platelet reactivity (e.g., thrombin and MMP-1), inflammation, and metabolomics; and
3. population pharmacogenomics.

6.0 INVESTIGATIONAL AND ANALYSIS PLAN

6.1 Overall Study Design and Plan: Description

The study will be a multicenter, prospective, randomized, double-blind, placebo-controlled, balanced-parallel-groups investigation to assess the safety of two dose levels of PZ-128 given as a single intravenous (IV) infusion over 2 hours in addition to standard of care dual-antiplatelet therapy (DAPT) in subjects undergoing non-emergent PCI or non-emergent cardiac catheterization with the intention to perform PCI. The study will be conducted at approximately 3 US clinical sites in conformance with GCP. During this trial of PZ-128, investigators are encouraged to follow current applicable standard of care guidelines that outline appropriate medical therapy for subjects with established atherosclerosis, including antiplatelet therapy (e.g., aspirin, $P2Y_{12}$), lipid-modification therapy, antianginal therapy and invasive cardiac procedures/interventions (e.g., cardiac catheterization, cardiac revascularization). Refer to the Study Design Diagram (**Section 1.1**) for an illustration of the overall design, the Study and Treatment Schema (**Figure 1, Section 6.5**) for an illustration of the treatment and follow-up plan, and the Study Flow Chart (**Section 1.2**) for information on tests and measurements to be performed in the main study, and in a sub-study at selected site(s) among those anticipated above.

Eligible subjects will include men and women at least18 years of age who are scheduled to undergo non-emergent PCI, or non-emergent cardiac catheterization with the intent to undergo PCI, and for whom elective use of a GP IIb/IIIa inhibitor is not intended. Subjects will receive aspirin; if PCI proceeds, subjects will receive an anticoagulant and loading dose of a $P2Y_{12}$ inhibitor. The study design specifies randomized assignment of PZ-128 or matching placebo in a 2:1 ratio and the initiation of the study drug infusion prior to the start of the coronary angiography portion of the cardiac catheterization. The sponsor estimates that 600 subjects, 400 to receive PZ-128 and 200 to receive placebo, will be adequate for clinical evaluation.

Two central committees will participate in the trial:

- An independent Clinical Events Committee (CEC) will be established to review and adjudicate in blinded fashion all bleeding events and MACE and stent thrombosis events identified by investigators or triggered from specific criteria on a rolling basis as they occur. The adjudicated findings will be used to address study objectives.
- An independent Data and Safety Monitoring Committee (DSMB) will be established to further the rights, safety and well-being of subjects who will be participating in this study by monitoring the progress and results. The DSMB will review in blinded fashion:
    - unadjudicated bleeding and associated data (aggregated) through 30 days, primarily to determine whether the type and occurrence of bleeding is in accord with anticipated results (i.e., no potential meaningful additional risk), and
    - unadjudicated MACE and stent thrombosis results.

Data may be unblinded if the DSMB believes that this is warranted. The DSMB may elect to review data on platelet aggregation, if deemed necessary and will review other SAE/AE data as indicated. The DSMB will perform one interim safety analysis to evaluate evidence of bleeding risk in light of any potential efficacy benefit(s) and to allow the DSMB to make recommendations to the sponsor (i.e., continuing the study under the current protocol, amending the current protocol, or stopping the study). Otherwise, the DSMB will meet quarterly or as required to perform its function.

Separate charters will be prepared to describe the composition and operating procedures of the two committees.

After written informed consent is obtained, eligibility tests/evaluations will be performed. All screening assessments must be completed prior to randomization and be within 14 days of the study drug treatment (this may include standard of care preadmission testing for the planned procedure). For the purposes of this study, a subject is considered enrolled into the trial upon successful randomization and an assignment of study drug and a unique subject randomization number. On the day of the procedure, subjects will receive a dose of aspirin (81-325 mg) orally. Subjects will receive the randomized assignment of PZ-128 or matching placebo as a single, 2-hour continuous IV infusion, to be initiated within 1 hour prior to the start of the cardiac catheterization procedure (defined as the time of the arterial sheath placement prior to the diagnostic angiography) in a 2:2:1:1 parallel, double-blind fashion of either: **1) PZ-128 0.3 mg/kg, 2) PZ-128 0.5 mg/kg, or 3) Placebo 0.3 mg/kg, or 4) Placebo 0.5 mg/kg.**

If PCI is deemed necessary and appropriate during catheterization, an anticoagulant (unfractionated heparin or low-molecular-weight heparin) will be added and a loading dose of an oral $P2Y_{12}$ inhibitor (clopidogrel, prasugrel or ticagrelor) will be administered per investigator choice and standard of care guidelines before PCI. All other subsequent therapy or care provided and not specified by the protocol will be in accordance with the local standard.

PCI successfully begins when the guide wire crosses the lesion, regardless of any subsequent event or process, including completion of the intended procedure. The planned treatment with any GP IIb/IIIa Inhibitor (GPI) during the PCI will not be allowed, but investigators may use these agents as a thrombotic bailout if indicated. Following the procedure, subjects will be advised regarding continuing/maintenance treatment with aspirin and a $P2Y_{12}$ inhibitor according to local standard of care.

If a PCI is not indicated during catheterization, or is indicated but does not commence (guide wire does not cross the lesion) for any reason, the subject will undergo medical management or surgical revascularization (i.e., CABG) according to the local standard of care.

All subjects will complete the protocol-specified in-hospital tests, collections, and evaluations up to the time they are discharged from the hospital. The duration of hospitalization will vary depending upon the clinical circumstances (e.g., subject comorbidities, type of procedure(s), complications from procedure(s), and need for further testing /procedures) and local standards

of care and may include same day discharge, overnight stay for observation or inpatient admission.

Prior to discharge from the hospital, the return visit will be scheduled for 14 days (± 5 d) after the administration of the study drug, and follow-up telephone contacts should be scheduled for 30 days (± 5 d) and 90 days (± 7 d) after the administration of the study drug.

Appropriate assessments will be used to characterize the overall effects of PZ-128. In particular, selected center(s) will participate in a sub-study that will include specific testing to assess inhibition of platelet aggregation and the pharmacokinetic/pharmacodynamics (PK/PD) relationship of PZ-128.

6.2 Participation in and Completion of the Study

The subject is considered to be enrolled in the study when the subject is successfully randomized and is assigned a study drug treatment regimen and a unique subject randomization number. A subject who receives a randomization assignment without ever receiving any amount of study drug treatment ("randomized but not treated") is still part of the intent-to-treat (ITT) data set for efficacy endpoints and must be followed thereafter per protocol (a telephone contact may be used in lieu of the 14-day follow-up visit for those not having taken any amount of study drug). A subject who signs the informed consent to participate in the study but does not undergo a successful randomization (i.e., screen failure, withdrew consent) will not be followed beyond the point it is determined that the subject will not be randomized- no further data will be collected and the subject will not be included in safety or efficacy analyses. Information on subjects who are screened and consented for the study but fail to undergo randomization will be submitted to the sponsor as per **Section 8.2** for regulatory purposes and to establish that the study population was selected without bias.

The subject is considered to have completed the study drug treatment following cessation of the study drug infusion. Given the single-dose design of the study, once the subject has received the study drug infusion (or once a subject permanently discontinues the study drug infusion), the study drug will no longer be available to the subject.

The subject is considered to have completed the entire study when he/she has undergone the final protocol-specified contact (i.e., study telephone call at 90-days following the study drug administration). Subject participation may be terminated prior to completion for reasons described in **Section 6.3.3**. For those subjects who do not complete the entire study protocol, subject participation will be considered terminated upon the completion of the last visit or phone contact.

The overall study ends when the last remaining subject has completed or has been discontinued/ terminated from the study as defined above. The sponsor, Tufts Medical Center, the sponsor's Institutional Review Board (IRB), the FDA, or the National Heart, Lung and Blood Institute/National Institutes of Health (NHLBI/NIH) may terminate the study at any time. If the study is terminated prior to the completion of expected enrollment for any

reason, all participating centers will be notified within 5 business days. All subjects already enrolled will continue to be followed for the planned course of study described in the protocol, unless otherwise indicated by the sponsor.

6.3 Study Population

This study will be conducted at approximately 3 clinical sites in the United States (US) without any pre-specified minimum/maximum enrollment target per site or per the study stratum:

- Inova Heart and Vascular Institute, Inova Fairfax Hospital (Falls Church, VA)
- Tufts Medical Center (Boston, MA)
- UMass Memorial Medical Center (Worcester, MA)

This study will enroll a total of approximately 600 subjects. Subjects who are randomized, but who discontinue from the study prior to receiving any amount of study drug may be replaced.

The sponsor estimates a maximum screen failure rate of 20% (i.e., those who sign consent but fail to be randomized for any reason). Therefore, the study will require a maximum of 720 prospective participants to sign consent in order to successfully randomize 600 subjects.

Subjects must meet all inclusion criteria and none of the exclusion criteria to be enrolled/randomized and receive study drug assignment.

6.3.1 Subject Inclusion Criteria

The subject must meet **all** of the following criteria listed below for entry:

1. The subject is at least 18 years of age and may be of either sex/gender and of any race and ethnicity.

2. The subject is scheduled to undergo non-emergent PCI or non-emergent cardiac catheterization with the intention of performing PCI. The following classifications of the urgency of the procedure at the time the operator decides to perform it will be used for randomization stratification [54]:

   a. Elective: The cardiac catheterization procedure ± PCI can be performed on an outpatient basis or during a subsequent hospitalization without significant risk of MI or death. For stable inpatients, this is a procedure that is performed during the hospitalization for convenience and ease of scheduling only, and not because the subject's clinical situation demands that the procedure be performed prior to discharge.

   OR

   b. Urgent: The cardiac catheterization ± PCI procedure should be performed on an inpatient basis and before discharge because of significant concerns about the risk of myocardial ischemia, MI and/or death. For subjects who are outpatients or in the emergency department at the time that the cardiac catheterization is requested, this is a procedure that would warrant hospital admission based on clinical presentation.

3. There is no anticipation that the subject would require treatment with a GP IIb/IIIa inhibitor prior to the initiation of the cardiac catheterization ± PCI procedure if the subject were not a participant in the current research study, and no anticipation of use during the procedure.

4. The subject is willing and able to give appropriate informed consent and complete all study-related procedures, and able to adhere to dosing and visit schedules (i.e., subject signs an approved informed consent document(s) and provides HIPAA authorization).

5. The subject will undergo all of the pre-enrollment parameters according to **Section 1.2** prior to randomization and have them completed within 14 days prior to the scheduled cardiac catheterization ± PCI procedure and study drug administration.

6. Women of childbearing potential (all postmenarchal women who are <1 year menopausal or who have not had surgical sterilization or a hysterectomy are considered to be women of child-bearing potential) must agree to use a medically accepted method of contraception from the time written informed consent is given up until 90 days following the study drug administration.

(Injectable, implantable, patch or oral ("the pill") hormonal contraceptives, medically prescribed intrauterine device (IUD) or partner vasectomy are medically accepted methods of contraception. Double barrier methods are acceptable although the risk of pregnancy is higher. Examples of double barrier methods are diaphragm with spermicidal gel or condoms with contraceptive foam.)

6.3.2 Subject Exclusion Criteria

The subject will be excluded from entry if **any** of the criteria listed below are met:

(General Exclusions)

1. Subject is pregnant, intends to become pregnant or is breast-feeding (all women of child-bearing potential must have a negative pregnancy test result confirmed prior to randomization and it must be repeated to be within 24 hours prior to the study drug administration if necessary).

2. Any of the following allergy history(s):
   - History of an allergic reaction* or contraindication to any of the following protocol-directed drugs: aspirin, heparin, $P2Y_{12}$ inhibitor (clopidogrel, prasugrel, ticagrelor), antihistamines (benadryl, famotidine); or
   - History of an allergic reaction* to contrast media; or
   - History of an allergic reaction* to a drug which required emergency medical treatment;
   - History of an allergic reaction* to a *Hymenoptera* sting which currently necessitates the subject to carry an EpiPen/injector or the subject has been prescribed one to treat an allergic reaction to a sting.

   *An allergic (anaphylactic) reaction is characterized by an adverse local or general response from exposure to an allergen involving skin/mucosal tissue manifestations (hives, pruritus, flushing, angioedema), and/or respiratory compromise (dyspnea, wheeze-bronchospasm, stridor, reduced peak expiratory flow, hypoxemia), and/or hemodynamic effects (hypo/hypertension, hypotonia, syncope).

   .
3. Participation in another research study of investigational therapy (drug or device) within the past 30 days prior to randomization or planned use of other investigational therapy(s) during this research study (until 90 days following the study drug administration).

4. Subject is part of the study staff personnel directly involved with this trial, or is a family member of the study staff (clinical site or sponsor).

5. Prior enrollment (randomization) in this research study.

6. Any condition which could interfere with, or the treatment for which might interfere with, the conduct of the research study or which would, in the opinion of the

investigator, unacceptably increase the subject's risk by participating in the research study. This would include, but is not limited to alcoholism, drug dependency or abuse, psychiatric disease, epilepsy or any unexplained blackouts.

(Exclusionary Prior/Concomitant Conditions)

7. Evidence of an ST-segment elevation myocardial infarction (STEMI) on presentation or during current hospitalization or a history of STEMI within the past 30 days prior to randomization.

8. Subject is scheduled to undergo PCI for known unprotected left main coronary artery (LMCA) disease (i.e., left main stenosis ≥50% not protected by at least 1 patent bypass graft).

9. Any history of a prior stroke (hemorrhagic or ischemic) or transient ischemic attack (TIA) of any etiology.

10. Cardiogenic or any type of shock on presentation or during current hospitalization (i.e., systolic blood pressure <90 mm Hg requiring vasopressor or hemodynamic support).

11. History of heparin-induced thrombocytopenia (HIT).

12. Any active bleeding within the past 30 days prior to randomization.

13. Any condition or personal belief (e.g., Jehovah's Witness) which would interfere with the subject's ability or willingness to undergo a blood transfusion.

14. Any of the following conditions associated with increased risk of bleeding:

    a. history of intracranial, intraocular, spinal, retroperitoneal or atraumatic intra-articular bleeding;

    b. gastrointestinal bleeding within the past 30 days prior to randomization;

    c. gastric or duodenal ulcer disease verified by endoscopy or barium meal contrast technique within the past 6 months prior to randomization;

    d. history of bleeding disorder or diathesis;

    e. major surgical procedure or trauma within the past 60 days prior to randomization or a planned surgical procedure to take place within 30 days following the study drug administration;

    f. history or suspicion of intracranial neoplasm, arteriovenous malformation, or aneurysm; or

g. clinical finding(s) in the judgment of the investigator that poses an increased risk of bleeding.

15. Sustained severe hypertension: systolic blood pressure >185 mm Hg or diastolic blood pressure >105 mm Hg with or without anti-hypertensive treatment (as demonstrated by repeated BP measurements >185/105 mm Hg including the final BP measurement before randomization).

16. Hypotension: systolic blood pressure <95 mm Hg (as demonstrated by repeated systolic BP measurements <95 mm Hg including the final systolic BP measurement prior to randomization).

17. Known active hepatobiliary disease, or known unexplained persistent increase in serum alanine aminotransferase (ALT) or aspartate aminotransferase (AST) activity to ≥2.5 times the upper limit of the reference range within the past 30 days prior to randomization.

18. Hemoglobin <10 g/dL or hematocrit <30%.

19. Platelet count <75,000/mm3.

20. Stage 4-5 Chronic Kidney Disease (National Kidney Foundation) or on dialysis.

21. Active sepsis or suspected sepsis.

22. Body weight <60 kg or >175 kg.

23. Current evidence of invasive cancer (persistent disease excluding basal cell carcinoma of the skin) or treatment for invasive cancer within the past 6 months prior to randomization.

24. Left ventricular ejection fraction <25% if known (any imaging technique) or New York Heart Association (NYHA) Class IV congestive heart failure.

(Exclusionary Prior/Concomitant/Anticipated Medication/Therapy)

25. Coronary interventional procedure of any kind within the past 30 days prior to randomization.

26. Anticipated subsequent staged multi-vessel PCI within 30 days following the study drug administration.

27. History of treatment with any parenteral GP IIb/IIIa inhibitor (GPI) within the past 30 days prior to randomization. (As stated in the Inclusion section, the planned treatment with a GPI prior to initiation of the cardiac catheterization ± PCI is <u>not</u> allowed;

however, GPI for thrombotic bailout may be used during the PCI at the investigator's discretion).

28. Concurrent or anticipated treatment with a parenteral direct thrombin inhibitor (e.g., bivalirudin) for the cardiac catheterization ± PCI procedure.

29. History of treatment with another PAR1 inhibitor within the past 60 days prior to randomization or the concurrent/anticipated use after randomization up until 30 days following the study drug administration.

30. History of treatment with another IV anti-platelet drug within 30 days prior to randomization or the concurrent/anticipated use after randomization up until 30 days following the study drug administration.

31. Any of the following anticoagulant or thrombolytic/fibrinolytic treatment(s):
    - History of treatment with warfarin within 5 days prior to randomization or the concurrent/anticipated use after randomization up until 2 days following the study drug administration; or
    - History of treatment with oral Factor Xa or direct thrombin inhibitors within 2 days prior to randomization or the concurrent/anticipated use after randomization up until 2 days following the study drug administration; or
    - History of treatment with thrombolytic/fibrinolytic agents within 7 days prior to randomization or the concurrent/anticipated use of any of those agents after randomization up until 30 days following the study drug administration.

Subjects excluded for any of the above reasons may be re-screened for participation at any time if the exclusion characteristic has changed.

6.3.3 Study Drug Discontinuation and Study Termination Criteria

Treatment with the study drug may be terminated/ discontinued for any of the following reasons:

- Serious adverse event (SAE);
- Failure to comply with the dosing, evaluations, or other requirements of the study;
- Unusual or excessive bleeding/signs or symptoms of abnormal bleeding from any source that cannot be controlled without discontinuation of the study drug administration (hematoma at the site of vascular sheath insertion may normally occur and should not serve as a reason to discontinue study drug unless bleeding cannot be controlled with normal measures).

Treatment with the study drug must be terminated/ discontinued for any of the following reasons:

- Request of the subject (subjects have the right to discontinue study drug administration at any time for any reason);

- There is a need for concomitant medication(s), which makes the subject ineligible to receive the study drug;
- Pregnancy; or
- Anaphylaxis.

In addition, if a subject who does not meet enrollment criteria is inadvertently randomized in the study, the sponsor must be notified. Although the subject should be discontinued from receiving study drug treatment, an exception may be granted by the sponsor if there is a compelling reason to continue with the administration of the study drug, but the investigator must obtain documented approval from the sponsor.

It is the right and duty of the investigator or sub-investigator to interrupt/stop study drug treatment of any subject if he/she feels that it is necessary to protect the subject, or that there are unmanageable factors, that may interfere significantly with the study procedures and/or the interpretation of results. If the study drug is discontinued prior to the completion of the infusion, the reason for the discontinuation will be obtained. This information will be documented in source records and in the appropriate section of the electronic case report form (eCRF) and the Sponsor should be notified promptly. Every effort should be made to ensure all activities outlined in the **Section 1.2**, Study Flow Chart and **Section 6.5**, Study Schedule are performed as usual.

Normal study termination/discontinuation is defined as the completion of all visits and procedures listed under **Section 1.2**. Early study termination/discontinuation occurs if the subject fails to complete the entire study, through to the 90-day telephone follow-up. Early termination from the study may occur for the following reasons:

- Subjects are free to withdraw from the study at any time for any reason- formal withdrawal of the subject's consent to participate further in any of the remaining study procedures must be documented by the investigator/qualified designee;
- In the opinion of the investigator, the subject cannot safely perform the procedures required by the protocol (e.g., adverse event, concomitant treatment);
- Subject does not comply with the protocol instructions given by the investigator/ qualified designee;
- The subject becomes pregnant (pregnancy will be followed as per **Section 6.7.2.2.7**);
- The sponsor, Tufts Medical Center may terminate the study for any reason;
- The study may be terminated at any time by the national or local regulatory authorities overseeing and/or funding the trial (e.g., FDA, NHLBI/NIH, or IRB).

A subject has the right to discontinue participation in the study at any time for any reason without prejudice to his/her future medical care by the investigator or at the institution. Withdrawal of partial consent means that the subject does not wish to receive the study drug any longer but is still willing to collaborate in providing further data by continuing on study (e.g., participate in subsequent study visits/contacts or procedures, allow release of follow-up

information). Withdrawal of full consent means that the subject does not wish to receive further study drug and does not wish to or is unable to continue further study participation; subject data up to withdrawal of consent will be included in the subjects' study data. Withdrawal of partial or full consent by the subject must be documented in writing by the investigator/qualified designee. The investigator will discuss with the subject appropriate procedures for withdrawal from the study drug and/or study. Should a subject request or decide to withdraw from the study, all efforts will be made to complete and report the observations as thoroughly as possible up to the date of withdrawal. All information should be reported on the applicable eCRFs and the Sponsor should be notified promptly.

6.3.4 Replacement of Subjects

Subjects who are randomized, but who discontinue from the study prior to receiving any amount of study drug may be replaced.

6.4 Study Treatments

The investigator or qualified designee is responsible for delivering the investigational therapy and any required co-administered therapy and the standard of care treatment(s) outlined according to the protocol and verifying compliance in source documentation and on the eCRF, and ensuring that any other therapy not specified in the protocol is delivered according to the local standard of care.

6.4.1 Blinded Investigational Study Drugs

Approximately 600 subjects will be randomized in a 2:1 (PZ-128: PZ-128 placebo) ratio to receive 1 of the following 4 treatment assignments:

- Single, 2-h IV infusion of PZ-128 0.3 mg/kg
- Single, 2-h IV infusion of PZ-128 0.5 mg/kg
- Single, 2-h IV infusion of Placebo 0.3 mg/kg
- Single, 2-h IV infusion of Placebo 0.5 mg/kg

The PZ-128 active drug and matching placebo are provided as vials of solid lyophilized powder cake for reconstitution and will be indistinguishable. The individualized dose will be based upon the baseline (i.e., Screening) weight of the subject and the dose level assignment by the centralized randomization system used as part of the method of treatment assignment (**Section 6.4.4**). The type and number of vials of study drug will be selected by an unblinded investigational pharmacist (**Section 6.4.6**) at the clinical site who is able to obtain the subject's unmasked randomized treatment assignment via a restricted access login within the randomization system. Each vial of study drug will be reconstituted with 10 mL of Sterile Water for Injection, USP (SWFI) and the calculated amount of reconstituted volume of study drug will be mixed and diluted with 250 mL (plus the manufacturer's overfill volume) of Dextrose 5% in Water, USP (D5W) to yield a final IV bag (blinded) for administration.

PZ-128 or matching placebo, as determined by the randomization scheme, will be administered by a qualified member of the study staff or institution's clinical staff over a 2-hour continuous IV infusion and the infusion must begin 0-60 min prior to the start of the cardiac catheterization (i.e., diagnostic coronary angiography). The infusion will run through completion regardless of whether the subject progresses to PCI, CABG or medical management.

[redacted] The peripheral IV line for the drug infusion should not be smaller than 20-gauge. Flushing of the IV access site (not the line) will be done using 0.9% Sodium Chloride for Injection (Normal Saline).

[redacted]

6.4.3 Other Required and Potential Rescue Treatment(s) (Standard of Care)

Investigators will use current applicable standard of care guidelines, the individual's clinical circumstances, and the local practices for the procedures surrounding the cardiac catheterization ± PCI and follow-up care post cardiac catheterization ± PCI [47].

Standard of care treatments/parameters in the context of this study will include oral anti-platelet therapy- aspirin and $P2Y_{12}$ inhibitor (i.e., clopidogrel, prasugrel, or ticagrelor), an anti-coagulant (i.e., unfractionated heparin or low-molecular-weight heparin) and potential rescue therapy (thrombotic bailout) with a GP IIb/IIIa inhibitor (e.g., eptifibatide). These treatments administered/taken within 30 days prior to randomization and at any time during the study must be documented in the subject's source documents/medical records and recorded in the "Previous and Concomitant Medication" module of the eCRF.

- **Aspirin:** Subjects not on aspirin therapy should be given nonenteric aspirin 325 mg before the study drug administration (subjects already taking daily aspirin therapy should take 81-325 mg before the study drug administration). After the cardiac

catheterization/PCI aspirin 81-325 mg daily may be continued indefinitely. ASA for pain relief should where possible be discouraged and acetaminophen given.

- **$P2Y_{12}$ Inhibitor(s):** A loading dose of an FDA-approved oral $P2Y_{12}$ inhibitor should be given to subjects before PCI with stenting (dosing may be adjusted depending upon prior chronic use of oral $P2Y_{12}$). Investigator discretion will be used and options include the following:
    - Clopidogrel (Plavix®) 600 mg
    - Prasugrel (Effient®) 60 mg
    - Ticagrelor (Brilinta®) 180 mg

  The choice and duration of oral $P2Y_{12}$ inhibitor therapy after stent implantation will be as per investigator discretion. Options include the following FDA-approved drugs:
    - Clopidogrel (Plavix®) 75 mg daily
    - Prasugrel (Effient®) 5-10 mg daily
    - Ticagrelor (Brilinta®) 90 mg twice daily

- **Anticoagulant:** An anticoagulant should be administered to subjects undergoing PCI. Following administration of the initial bolus of anticoagulant, activated clotting time (ACT) will be measured locally at "peak dose" and just prior to sheath removal, as well as according to the standard of care.
    - Administration of IV Unfractionated Heparin (UFH) is preferred in this study and should be dosed per standard of care and routine hospital practice (i.e., weight-based and adjusted to attain target ACT).
    - Low-molecular-weight heparin (LMWH) is an acceptable alternative to UFH within the context of this trial (weight adjusted in accordance with approved dosage and administration).
    - It is strongly recommended to use only one anti-coagulant drug consistently and not switch between agents before or during the PCI procedures.
    - A parenteral direct thrombin inhibitor (e.g, bivalirudin) is not to be administered.
    - Anticoagulant should not be restarted after access sheath removal unless specifically indicated for another condition such as anticoagulation for a deep venous thrombosis, atrial fibrillation, or a mechanical prosthetic heart valve **(Section 6.3.2)**.

- **Rescue Therapy (thrombotic bailout):** The planned administration of GP IIb/IIIa inhibitors (GPI) for a subject excludes that subject from participation in this study and therefore should not be an anticipated part of the regimen. During PCI, bail-out with an FDA-approved GPI such as eptifibatide (Integrilin®) may be used, at the investigator's

discretion, in subjects with new ischemia on telemetry or ECG, worsening of angiographic coronary flow during procedure or thrombotic complication(s) related to stenting during or after PCI.

- **Coronary Stents:** During PCI, the number and type of coronary stenting will be determined by the investigator and will include only FDA-approved devices (bare metal and drug-eluting) which are indicated for PCI. The stent implant procedure will be performed in accordance with the device Instructions for Use (IFU).

- **Vascular Access and Closure Devices:** The use of radial, femoral or brachial artery access will be allowed. Timing of the arterial sheath removal will be performed as per standard of care and local routine practice (e.g., target ACT). FDA-approved vascular closure devices (e.g., Angioseal™, Vasoseal™, Perclose™) may be used as an alternative to manual compression to attain vascular hemostasis. Pressure on the access site (either manual or by a mechanical device) should be applied for a minimum of 30 minutes.

6.4.4 Method of Study Drug Assignment, Randomization and/or Stratification

A centralized randomization computerized program will provide randomized study drug assignment at enrollment through an interactive, web-based electronic data capture (EDC) system (i.e., integrated randomization within the Medidata® platform), using randomization schemes (permuted block method) provided by the sponsor's designee. After a subject qualifies for enrollment in the study, the designated staff at the clinical site will enter confirmation that the subject meets all of the inclusion criteria and none of the exclusion criteria into the system (available 24 hours a day, 7 days a week). Upon successful randomization, the subject will be assigned a unique subject randomization number.

Subjects will be randomized via the interactive web-based system in the order that they qualify, with stratification based on clinical site as well as by the urgency of the cardiac catheterization/PCI procedure at the time the operator decides to perform it- Elective versus Urgent- to obtain balance across treatment groups. Subject randomization numbers that are assigned to subjects who do not receive study drug will not be re-utilized. No enrollment caps will be set for the individual clinical sites or stratum.

6.4.5 Selection and Timing of Dose for Each Subject

All subjects who fulfill study entry criteria as specified in **Sections 6.3.1-6.3.2** and undergo successful randomized treatment assignment will receive PZ-128 or matching placebo, as determined by the randomization scheme: a single, weight-adjusted IV dose of 0.3 mg/kg or 0.5 mg/kg. The infusion must be started within 1 hour before the commencement of the cardiac catheterization procedure (i.e., coronary angiography). [redacted]

There is no provision for delaying the start of the study drug administration outside of the pre-diagnostic angiography window.

There is no provision for dose modification (increase or decrease), and this is not allowed. See **Sections 6.4.7.7 and 6.7.2.3**, however for information concerning temporary study drug infusion interruption or permanent discontinuation. The single dose of PZ-128 will be one of two possible dose levels being studied in a parallel manner- 0.3 mg/kg and 0.5 mg/kg- and the dose groups will enroll in parallel until the appropriate number of subjects have been treated.

6.4.6 Blinding of Study Drug

This will be a double-blind study. Study drug treatment assignment (activity of IP) will be blinded for subjects, investigators and staff who evaluate subjects and their responses or make decisions about subject care, and sponsor personnel who are involved in the data analysis. Statisticians and programmers at the sponsor's designated partner (Research Triangle Institute, International- RTI) will prepare all randomization schemes which will then be loaded into the randomization system by Medidata Solutions, Inc. In addition, copies of the randomization schemes will be made available to the following within the sponsor and/or its designee:

- Independent statistician(s) at RTI to support requests by the DSMB
- Study lead(s) for the analysis of the pharmacokinetic and pharmacodynamics samples, exploratory biomarker samples and pharmacogenomics samples.

Appropriate personnel within Clinical Supplies Management, Inc® (CSM) will have access to vial sequencing assignment for proper labeling, packaging and shipment of study drug.

Unblinding of the study drug assignment for a subject at a clinical site should occur only in the event of a medical emergency or serious adverse event for which it is necessary to know the study treatment to determine the appropriate medical management for the subject. This circumstance is extraordinary and will likely impact a minor fraction of the enrolled subjects. Unblinding at the study site for any other reason, including selection of subsequent anti-platelet therapies, will be considered a protocol violation. The investigator is strongly encouraged to contact the Medical Director at Tufts Medical Center before unblinding any subject's study drug assignment, but must do so within 1 working day after the event and must document the unblinding and its rationale in the subject's electronic case report form (eCRF). For emergency unblinding, the investigator should be the individual who authorizes the designated investigational pharmacy staff to proceed with unblinding. When the ability to manage the health or welfare of the study subject is dependent upon the requested information, the information may be revealed by the unblinded designated investigational pharmacy staff immediately and independent of the investigator's authorization. However, this delegation of authority must be documented in advance (i.e., pharmacy policy and procedure manual, study-specific delegation of authority log). In case of any accidental unblinding, the procedures noted above should be followed.

The sponsor, Tufts Medical Center, retains the right to unblind the study drug assignment for

SAEs that potentially require expedited reporting to regulatory authorities.

The external, independent DSMB (**Section 9.3.1**) will be allowed to inspect unblinded results for bleeding, platelet aggregation, MACE and stent thrombosis and other SAEs if it is felt necessary to make decisions about enrolling subjects into any dose group. The operating procedures of the DSMB will be defined in a separate charter.

The sponsor and DSMB will monitor all episodes of unblinding.

6.4.7 Investigational Product(s)

The investigator shall take responsibility for and shall take all steps to maintain appropriate records and ensure appropriate supply, handling, storage, distribution, and usage of these materials in accordance with the protocol and any applicable laws and regulations. The investigator will designate a qualified (i.e., licensed/registered) member of the institution's research/investigational pharmacy staff to perform the day-to-day pharmacy activities and investigational product management including, but not limited to, the procurement, storage, preparation, dispensing, and final disposition of investigational products, keeping in mind that all aspects of the investigational trial, including conduct, management, and record maintenance, are ultimately the responsibility of the investigator. Neither the investigator nor any member of the clinical research team should have access to the investigational product supply or accountability records in a manner that would jeopardize the maintenance of the blind. See the *Investigational Product Instruction Manual (IPIM)* for complete details and requirements for study drug packaging, labeling, storage, preparation, administration and accountability

6.4.7.1 Identity of Investigational Product(s)

Investigational Products (IP) are active PZ-128 and its matching placebo. The PZ-128 is available as a sterile, preservative-free, white to off-white, solid lyophilized powder cake for reconstitution. [redacted]

Each PZ-128 or placebo vial is intended for single-dose/single-use only and is not to be used to treat more than 1 subject.

6.4.7.2 Source

PZ-128 and matching placebo will be provided free of charge by the sponsor, Tufts Medical Center, and will be distributed by the sponsor's designee, Clinical Supplies Management, Inc®

(CSM, Fargo, ND).

6.4.7.3 Labeling

Information provided on the labels for the IP will comply with ICH, GCP and local regulatory requirements. All vial labels will contain a unique vial sequence number, lot number, study number, product identity (i.e., PZ-128 48 mg/vial or Placebo), vial size (20 cc), reconstitution/ dilution instructions, storage conditions, sponsor identification, IP form (powder for solution for infusion), method of administration (for intravenous use) and investigational statement. All box/carton labels will contain the study number, product identity, number of vials/box, storage conditions and sponsor identification. In lieu of an expiration date, stability information will be provided by the sponsor to the clinical site when the investigational product (IP) is shipped from CSM and will be updated during the clinical investigation.

6.4.7.4 Packaging

Multi-vial boxes/cartons will contain either 20 vials of active PZ-128 or 20 vials of placebo. The sponsor will request the initial IP supply shipment from CSM after the clinical site has completed the appropriate regulatory requirements and prior to the screening of the first subject for study participation. Subsequent IP supply will be requested by each site's designated pharmacy staff according to its ongoing inventory management.

6.4.7.5 Storage

Vials of the investigational products must be stored at a controlled freezing temperature of -20 °C ± 10 °C (i.e., -30 °C to -10 °C [-22 °F to 14 °F]) and protected from direct sunlight, unless otherwise authorized by the sponsor. The IP supply will be stored in a securely locked location in the site's pharmacy and access should be strictly limited to the designated pharmacy staff.

6.4.7.6 Reconstitution, Dilution and Dispensing

The investigator agrees neither to dispense the study drug from, nor store it at any site(s) other than those listed on the Form FDA 1572. The investigator agrees that study drug(s) will be dispensed by the investigator or subinvestigator(s) named on the Form 1572, or their qualified designee (i.e., prepared for administration and dispensed by registered pharmacy staff). The investigator, subinvestigator(s), or qualified designee(s) also agree that the study drug(s) will be dispensed with a signed medical order from an authorized prescriber only to study subjects who have provided written informed consent, have met all entry criteria and have completed successful randomization (allocation of unique randomization number and assigned study treatment). Investigational product(s) may not be used for any purpose other than that stated in the research protocol.

The designated, unblinded investigational pharmacist at the clinical site will access the

subject's randomized study drug assignment through a password-protected and restricted role login within the Medidata Rave® EDC system prior to the preparation of the blinded IV bag for infusion. The pharmacy staff will select 1 to 2 vials from either the PZ-128 or placebo supply depending upon the subject's assigned IP and dose level (0.3 mg/kg or 0.5 mg/kg) and the subject's actual body weight at the time of Screening (rounded to the nearest kg) in order to provide individual dosing.

[redacted]

The final IV bag for administration will be labeled as per the institution's investigational drug policy(s) for blinded products and should include the following: subject name/medical record number/study ID number, blinded IP name (PZ-128/Placebo), IP dosage form (for intravenous infusion), final weight-adjusted dose (mg) and total volume (mL), single-use only, method of administration/infusion rate (IV infusion over 120 min; infuse entire contents for full dose), preparation and expiration dates, storage conditions, study identifier, prescribing investigator name, pharmacist name, and investigational use statement. [redacted]

6.4.7.7 Administration

The study drug will be delivered once as a continuous IV infusion using an intravenous infusion pump given over a 2-hour period [redacted] This will be a dedicated line for the study drug administration compatible with the institution's IV pump module and will be supplied by the clinical site. PZ-128/placebo will be administered by a qualified member of the study staff or institution's clinical staff following successful enrollment/ randomization prior to cardiac catheterization. [redacted]
Intravenous PZ-128/placebo will be administered peripherally through an intravenous catheter which should not be smaller than 20-gauge. It is recommended that PK/PD/exploratory

biomarkers/pharmacogenomics research samples are obtained from a distal venous site, ideally from the opposite arm or alternatively from a site distal to the site of drug infusion. If a subject has a central line in place, PZ-128/placebo may be administered via that route. If PZ-128/placebo is administered via central venous catheter, it is strongly recommended that PK/PD/exploratory biomarkers/ pharmacogenomics research samples are obtained by peripheral venipuncture if at all possible. Due to the potential overfill volume in the commercial D5W IV bag, the entire contents of the IV bag should be infused for complete dosing. [redacted]

The investigator or designated medical staff must be present during the administration of the investigational product to assess and treat adverse events that may arise during dosing. Anaphylaxis precautions must be observed during the PZ-128/placebo administration and for at least 1 hour following the completion of the infusion. Subjects will be monitored in the cardiac catheterization lab until discharge or transfer to another hospital unit per the protocol and local standards of care.

There will be no dose modifications or alterations to the rate of infusion. Subjects should receive the entire study drug infusion regardless of whether a PCI is performed. Potential drug allergic reactions that occur during the administration of PZ-128/placebo may necessitate stopping and restarting the infusion (interruption/temporary cessation of the infusion is allowed for up to 1 hour only) or discontinuing (permanent cessation) the study drug infusion, regardless of whether the reaction is deemed related or not-related to PZ-128/placebo (See **Management of Drug Allergic Reactions Section 6.7.2.3**).

Based on the judgment of the investigator or qualified designee (i.e., enrolling/treating sub-investigator), dosing with the study drug may be withheld, interrupted and/or discontinued for any clinical situation in order to preserve the protection, safety and well-being of the subject.

[redacted]

6.4.7.8 Drug Accountability

An accurate and current accounting of the supplies of investigational products that were received from the sponsor/designee, dispensed and administered to subjects and returned/destroyed/disposed of as dictated by the sponsor, will be maintained on an ongoing basis by the designated research/investigational pharmacist on the Investigational Product Accountability Record at each clinical site and will be verified by the sponsor's study monitor throughout the conduct of the study. The PZ-128/placebo Dose Preparation Worksheet will be completed for each subject and the subject's receipt of study drug administration will be recorded in their medical records and on the eCRF.

The sponsor's designated study monitor will perform routine site monitoring activities for pharmacy, drug accountability and reconciliation of the investigational products. At the completion of the trial, after the study monitor has conducted a final inventory and drug accountability, all unused drug will be returned to the sponsor, Tufts Medical Center (or designee) or destroyed at the clinical site as authorized by the sponsor and in accordance with the institution's SOP. In the event that study drug needs to be returned for any other reason, the site will receive a written request from the sponsor/designee with the identity of the drug product listing the vial number(s)/box number(s) to be returned and the reason for the return request.

Inventory records must be readily available for inspection by the study monitor and/or auditor, and open to government inspection at any time.

6.4.8 Non-Study Treatments

6.4.8.1 Prior and Concomitant Medications

All prior medications (prescription and over-the-counter, including dietary supplements) within 30 days prior to randomization, and all concomitant therapy taken by the subject for the duration of the study are to be recorded in source documentation and on the eCRF. The identity of the therapy, the dose, route, and regimen, the dates started and stopped (or notation of "continuing" if that is the case), and the reason for use must be recorded. The use of any concomitant medication must relate to an adverse event or the subject's medical history.

6.4.8.1.1 Medications/Therapies Prohibited Prior to Randomized Study Treatment Assignment and During the Study

The medications/therapies prohibited prior to randomization treatment assignment and during the study after randomized treatment assignment are listed under **Exclusionary Prior/ Concomitant/Anticipated Medication/Therapy** (exclusion criteria 25 through 31) in **Section 6.3.2**. A categorical summary is provided below:

IV Antiplatelets
A subject is not eligible for the study if any other IV antiplatelet drug has been administered within 30 days prior to randomization and they should not be used after randomization up until 30 days following the study drug administration.

Other Oral Antiplatelets
Aside from aspirin and fish oil supplements, clinical experience of other drugs with any antiplatelet effect in combination with PZ-128 is not available at this time. Treatment with these types of drugs (including NSAIDs) are allowed pre-study and/or during the study, at the investigator's discretion. Treatment with cycloxygenase-2 inhibitors is permitted. However, as per the exclusion criteria (**Section 6.3.2**), treatment with another PAR1 inhibitor within 60 days prior to randomization or the concurrent/anticipated use of one after randomization up until 30 days following the study drug administration is not allowed.

Oral Anticoagulants
A subject is not eligible for the study if warfarin treatment has been administered within 5 days prior to randomization and it should not be used after randomization up until 2 days following the study drug administration. A subject is not eligible for the study if oral Factor Xa or direct thrombin inhibitor treatment has been administered within 2 days prior to randomization and they should not be used after randomization up until 2 days following the study drug administration.

If treatment with oral anticoagulant drugs is considered essential during this study period, the sponsor should be contacted.

Thrombolytic/Fibrinolytic Therapy
A subject is not eligible for the study if thrombolytic/fibrinolytic therapy has been administered within 7 days prior to randomization and they should not be used after randomization up until 30 days following the study drug administration. Clinical experience of thrombolytics/ fibrinolytics in combination with PZ-128 is not available at this time and caution should be used.

Other Medications
All previous prescription and non-prescription concomitant medications administered up to 30 days prior to randomization, on an ongoing basis, as well as changes in such concomitant medication and any new concomitant medication taken while the subject is on study, should be recorded in the subject's source documents and on the appropriate eCRF through 90 days after the completion of the study drug administration. The administration of all medication (including investigational products) must be recorded in the subject's source documents/ medical records and in the appropriate sections of the eCRF.

6.4.8.1.2 Medications/Therapies Allowed During the Study

Medications or supplements that are prohibited prior to randomization and during the study period are identified in **Section 6.3.2** and **Section 6.4.8.1.1.** Other medication(s), which are considered necessary for the subject's safety and well-being, may be given at the discretion of the investigator(s).

6.4.8.2 Other Restrictions

Due to the potentially acute nature of randomization of the subject prior to the administration of the study IP and cardiac catheterization/PCI procedure, there are no protocol restrictions on the food and drink intake prior to the randomization or procedure, although fatty meals should be avoided if possible.

Subjects should not donate blood at any time during the study period.

6.4.9 Procedures for Monitoring Compliance

This study will be conducted as described in this protocol, except for any emergency situation

in which the protection, safety and well-being of the subject requires immediate intervention, based on the judgment of the investigator or qualified designee. In the event of a significant deviation from the protocol due to an emergency, accident, or mistake, the investigator or designee should contact the sponsor at the earliest possible time by telephone. This will allow an early joint decision regarding the subject's continuation in the study. The investigator will be responsible for complying with the local IRB established procedures for executing and reporting all protocol deviations. All protocol deviations will be tracked by the sponsor and used in the evaluation of the data sets for the protocol analysis.

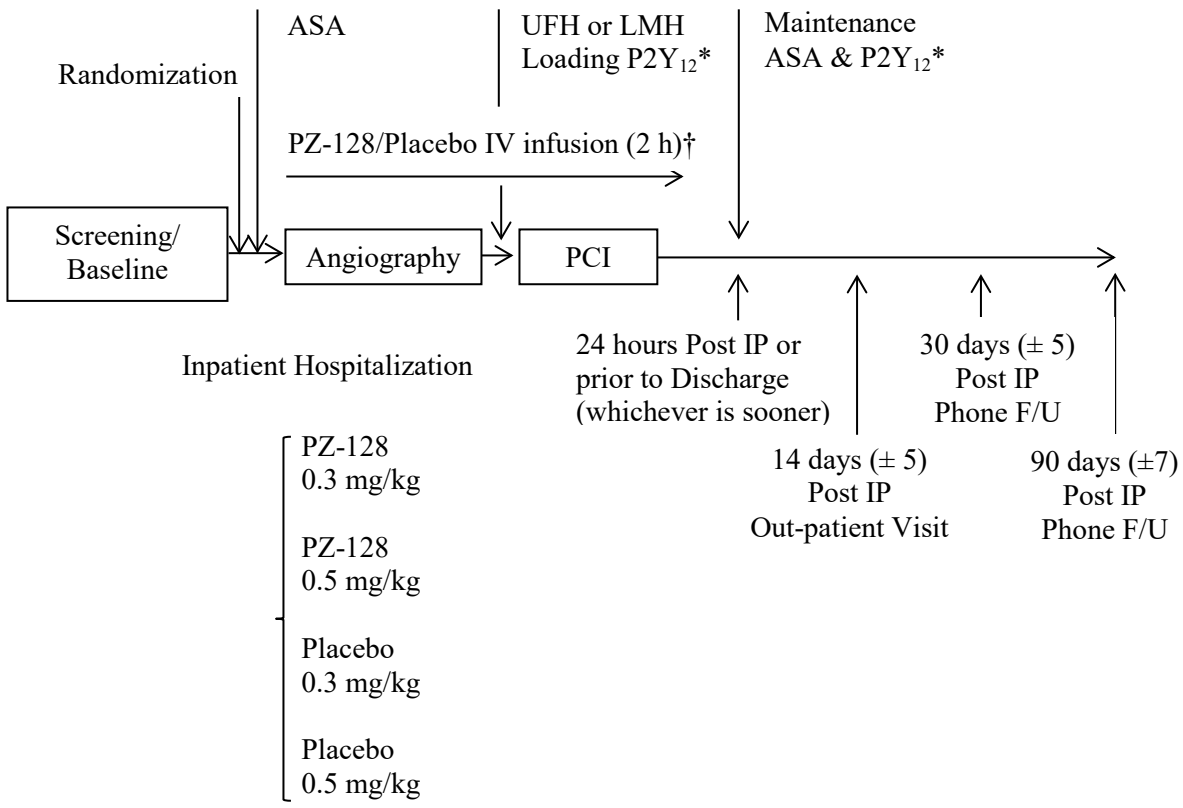
The investigator or qualified designee is to note in source documentation and in the appropriate section of the eCRF whether study drug infusion had been administered per protocol. If not, the reason for deviation must be recorded. In addition, the designated pharmacy staff will maintain drug accountability logs, and investigational product (IP) dose preparation worksheets on an ongoing basis and the supply of the IP will be cross-referenced with the infusion records at routine interim monitoring visits (IMV) conducted by the sponsor's monitor.

6.5 Study Schedule

An overview of the study is provided in the Study Design Diagram in **Section 1.1** and the schedule of assessments/events is shown in the Study Flow Chart in **Section 1.2**. See **Section 6.6** for specific information on processes. Figure 1 provides a treatment and follow-up schema.

The sponsor has specified a schedule of intervals relative to the day of randomized treatment for the return visit to the clinic and the telephone contacts, with a "window" of days around each visit/contact. The sponsor strongly requests that the subjects return for clinical visits or undergo telephone contacts as close to the specified interval as possible, and preferable within the defined window. If it is not possible for the subject to return within the visit "window", then the visit should be scheduled as close to the interval as is convenient for the subject and clinical site. Note that these are regularly scheduled visits/time points; if an unscheduled visit is required by the investigator or sponsor because of clinical circumstances or other unforeseen event, this will be allowed. All other aspects of care beyond those specified in the protocol will be based on the judgment and discretion of the investigator. All data collected will be used in the conduct of the study.

Figure 1. Study and Treatment Schema



*Specific $P2Y_{12}$ inhibitor drugs and doses will be administered as per the investigator's discretion/standard of care (e.g., clopidogrel, prasugrel or ticagrelor)

[redacted]

ASA=aspirin; IP=investigational product; LMH=low molecular weight heparin; UFH=unfractionated heparin

Each subject randomized in this trial will participate for approximately 15 weeks, including a screening period of up to 14 days prior to the administration of the study drug and cardiac catheterization, the hospitalization phase, and a 90-day follow-up phase after the study drug administration as follows:

- Screening/Enrollment Period: up to 14 days prior to cardiac catheterization/PCI and study drug administration
- Hospitalization Phase: inpatient or outpatient period of hospitalization for care associated with the study drug administration and cardiac catheterization ± PCI through discharge
- Outpatient follow-up visit: 14 days (± 5 d) post study drug administration
- Telephone follow-up contact: 30 days (± 5 d) post study drug administration
- Telephone follow-up contact: 90 days (± 7 d) post study drug administration

6.5.1 Screening (within 14 days prior to cardiac catheterization/study drug administration)

Screening should occur as soon as a prospective study participant is identified. The investigator or qualified designee should ascertain as much as possible, and as allowed, that a subject would be eligible to participate before obtaining informed written consent. All screening parameters for enrollment must be performed prior to randomization and must be within 14 days before the cardiac catheterization/PCI and study drug administration. Subjects may or may not already be hospitalized due to their acute condition. Any data collected and any procedure performed as part of standard of care preadmission testing or other testing before written consent has been obtained may be used as part of the assessment for eligibility as long as it would have been performed regardless of subject participation in the study and it is within the specified time frame for inclusion/exclusion criteria (this includes data/results collected from another facility). All samples, tests, and evaluations collected/ performed/ recorded as part of the procedures specified in this protocol that are not part of standard of care may occur only after written informed consent has been obtained.

A subject who signs the informed consent to participate in the study but does not progress to a randomization assignment (by decision of either the subject or investigator) will be considered as a screen failure. Screen failure subjects will not be considered enrolled in the trial by the sponsor and will not be followed beyond the point it is determined that the subject will not be randomized- no further data will be collected, and the subject will not be included in any study safety and efficacy analyses. However, the investigator and/or designee will submit a log record of those subjects as per **Section 8.2**.

The clinical laboratory at each study site will be used to perform all standard clinical labs according to its internal SOP's unless otherwise stated.

The following will be performed or obtained during the baseline screening period prior to randomization:

1. Explain the study to the subject and obtain appropriate written informed consents (i.e., main study and optional PK/PD sub-study, exploratory biomarkers research and pharmacogenomics). Review acceptable methods of contraception (birth control) while taking part in this study with both female subjects of child-bearing potential and male subjects.
2. Obtain a complete medical history (medical, surgical and cardiovascular) and demographics.
3. Review and record all previous and concomitant medications taken within the past 30 days with particular attention to anti-platelet, anticoagulant and thrombolytic/ fibrinolytic therapy.
4. Perform a complete physical exam (review of systems).
5. Obtain vital signs, including heart rate, blood pressure, and height and weight.
6. Record a standard 12-lead electrocardiogram (ECG).

7. Obtain blood samples from the subject for the following standard clinical laboratory evaluations to be performed locally:
    a. Hematology- complete blood count (CBC), differential white blood count and platelet count;
    b. Serum chemistry- sodium, potassium, chloride, carbon dioxide (CO_2), glucose, blood urea nitrogen (BUN), creatinine, alkaline phosphatase, total bilirubin, alanine transaminase (ALT), and aspartate transaminase (AST);
    c. PT/INR and aPTT;
    d. Lipid panel- total cholesterol, high-density lipoprotein cholesterol, calculated low-density lipoprotein cholesterol and triglycerides (does not have to be fasting); and
    e. Glycosylated Hemoglobin (HbA1c).
8. Perform and document urine pregnancy test (urine beta-HCG point-of-care test) for all women of child-bearing potential (this must be completed during screening and also within the 24 hours prior to study drug administration). Perform eligibility assessment (review of inclusion & exclusion criteria, See **Section 6.3.1** and **Section 6.3.2**).

Subjects may be randomized prior to the return of the baseline local clinical laboratory test results unless the test is specifically listed in the inclusion/exclusion criteria (i.e., hemoglobin, hematocrit, platelet count). However, all baseline laboratory blood draws must be done prior to randomization and within the specified time frame (i.e., 14 days prior to study drug administration/cardiac catheterization ± PCI).

6.5.2 Enrollment and Randomization

Randomization must occur far enough in advance of the cardiac catheterization/PCI to allow the institution's pharmacy staff to prepare and deliver the study drug to the catheterization lab and to ensure that the single dose, 2-hour IV infusion is started within one hour prior to the beginning of the coronary angiography following [redacted]

1. When a subject qualifies for enrollment/randomization, the clinical site staff will access the web-based, randomization program (available 24 hours a day, 7 days a week) and enter confirmation that the subject meets all of the inclusion criteria and none of the exclusion criteria. The clinical site will provide information on the urgency of the procedure to allow for proper stratification of study treatment assignment (i.e., elective versus urgent, See **Section 6.3.1**). Following a successful randomization process, the subject will be assigned a unique subject randomization number.

2. The clinical site staff will notify the investigational pharmacy staff of the randomization and will coordinate the delivery of ready-to-infuse, blinded study drug per the appropriate time/date of the anticipated procedure.

Appropriate eCRFs must be completed for all randomized subjects regardless of whether they

receive study drug. Randomized subjects cannot be considered screen failures.

6.5.3 Pre-Cardiac Catheterization Procedure

1. Prepare study drug for administration in investigational/research pharmacy (see *Investigational Product Instruction Manual-IPIM*).
2. Record any changes to concomitant medication(s).
3. Confirm subject still meets eligibility criteria to receive study drug.
4. Perform and document urine pregnancy test (urine beta-HCG point-of-care test) for women of child-bearing potential if screening assessment was not within the 24 hours prior to study drug administration. The result must be known and negative prior to administration of the study drug.
5. Administer aspirin.
6. [redacted]
7. Obtain vital signs, including heart rate and blood pressure <u>just prior</u> to study drug administration (i.e., Time=0).
8. Collect blood sample (serum) for centralized research analysis of Cardiac Troponin I (cTnI) just prior to study drug administration (i.e., Time=0).
9. Collect blood samples for optional research according to site participation and subject's individual consent (see **Section 6.6.3.2**) just prior to study drug administration (i.e., Time=0):
    a. Samples for pharmacokinetics (plasma) and pharmacodynamics-platelet aggregation (whole blood) (PK/PD sub-study; participating sites only);
    b. Plasma sample for exploratory biochemical, inflammatory and metabolic markers; and
    c. Whole blood sample for pharmacogenomics.
10. **Start blinded PZ-128/placebo** IV administration 0-60 min prior to start of coronary angiography and infuse continuously over 2 hours (infuse entire contents for complete dosing). Observe anaphylaxis precautions during the infusion and for at least 1 hour following cessation of study drug.
11. Perform any additional standard of care assessments in the pre-cardiac catheterization/PCI setting.
12. Assess and record adverse events, including bleeding and clinical efficacy endpoint events (MACE, stent thrombosis).

6.5.4 Cardiac Catheterization ± Percutaneous Coronary Intervention

1. Perform diagnostic angiogram per institutional practices and determine if percutaneous coronary intervention (PCI) is indicated. Study drug infusion will be run through completion regardless of the outcome from the diagnostic angiography.
2. Perform PCI per institutional guidelines and standards of care including parenteral anti-coagulant (i.e., UFH or LMWH) and oral anti-platelet (i.e., $P2Y_{12}$) if indicated (See **Section 6.4.3** and **Section 6.4.8** for standard of care treatment(s) and study protocol directives). PCI is deemed to have commenced when the guide wire crosses the lesion. If the randomized subject does not undergo PCI, the reason for non-intervention must be documented in the subject's medical records by the operator (i.e., interventional cardiologist) making the decision to not perform PCI and entered into the eCRF.
3. Following administration of the initial bolus of anti-coagulant (UFH or LMWH), ACT will be measured and documented (point-of-care testing) locally at "peak dose" and just prior to sheath removal, as well as according to the standard of care.
4. Record all concomitant medications administered to the subject.

   Note, during PCI, a GPI for thrombotic bailout may be used (per the package insert) in subjects with new ischemia on telemetry or ECG, worsening of angiographic coronary flow during procedure, or thrombotic complication related to stenting during PCI, at the investigator's discretion. The GPI used must be recorded on the case report form along with the indication and dose. However, subjects in whom the administration of a GPI is planned must not be enrolled in the study.
5. Assess and record adverse events, including bleeding and clinical efficacy endpoint events (MACE, stent thrombosis).

6.5.5 Post-PZ-128/Placebo Infusion

1. Obtain vital signs including heart rate and blood pressure immediately after the completion of the study drug administration (i.e., Time=2 h).
2. Assess and record adverse events, including bleeding and efficacy endpoint events (MACE, stent thrombosis).
3. Collect blood samples for optional research according to site participation and subject's individual consent (see **Section 6.6.3.2**):
   a. Samples for pharmacokinetics (plasma) and pharmacodynamics- platelet aggregation (whole blood) (PK/PD sub-study; participating sites only) will be drawn at the following time points: 30 min ± 5 min, 1 h ± 5 min, 2 h + 15 min (immediately after completion of infusion), and 6 h – 2 h (4 to 6 h) after the start of the PZ-128 infusion; and
   b. Plasma sample for exploratory biochemical, inflammatory and metabolic markers will be drawn at the following time points: 2 h + 15 min (immediately after completion of infusion) and 6 h – 2 h (4 to 6 h) after the start of the PZ-128 infusion.

6.5.6 Post-Cardiac Catheterization/PCI

1. Collect blood sample (serum) for centralized research analysis of Cardiac Troponin I (cTnI) at 4 to 8 h and 12 to 24 h after the completion of PCI. In addition, if applicable, collect samples immediately after occurrence of symptoms suggestive of acute coronary syndrome, and again 4 to 8 h and 12 to 24 h after symptoms if the subject has not yet been discharged from the hospital.
2. Assess and record adverse events, including bleeding and clinical efficacy endpoint events (MACE, stent thrombosis).
3. Record concomitant medications.
4. Perform vital signs including heart rate and blood pressure (timing as per standard of care).
5. Record a standard 12-lead ECG (timing as per standard of care).
6. Obtain blood sample for the following standard clinical laboratory evaluations (timing as per standard of care):
    a. Hematology- complete blood count (CBC), differential white blood count and platelet count;
7. Perform other post-procedural tests and evaluations in accordance with institutional guidelines and standards (e.g., arterial access sheath removal).

6.5.7 24 hours (± 2 h) Post Start of Study Drug Infusion or At Discharge from Hospital (whichever occurs sooner)

Perform the following at 24 hours ± 2 h post start of the IP infusion, or Prior to Discharge, whichever occurs sooner.

1. Review and record all concomitant medications taken during hospitalization.
2. Perform a physical examination (note, the clinical site's standard of care, cardiac catheterization nursing assessment may be performed to evaluate the subject's clinical status in lieu of a complete physical exam at this time point).
3. Obtain vital signs, including heart rate and blood pressure.
4. Obtain blood samples from the subject for the following standard clinical laboratory evaluations:
    a. Hematology- complete blood count (CBC), differential white blood count and platelet count;
    b. Serum chemistry- sodium, potassium, chloride, carbon dioxide (CO_2), glucose, blood urea nitrogen (BUN), creatinine, alkaline phosphatase, total bilirubin, alanine transaminase (ALT), and aspartate transaminase (AST); and
    c. PT/INR and aPTT.

5. Assess and record adverse events, including bleeding and clinical efficacy endpoint events (MACE, stent thrombosis).
6. Collect blood sample (serum) for centralized research analysis of Cardiac Troponin I (cTnI) if a sample was not already collected at 12 to 24 h after the completion of PCI.
7. Collect blood samples for optional research according to site participation and subject's individual consent (see **Section 6.6.3.2**):
    a. Samples for pharmacokinetics (plasma) and pharmacodynamics- platelet aggregation (whole blood) (PK/PD sub-study; participating sites only); and
    b. Plasma sample for exploratory biochemical, inflammatory and metabolic markers.
8. Perform other tests and evaluations in accordance with institutional guidelines and standards (e.g., start maintenance anti-platelet regimen, aspirin and $P2Y_{12}$, if indicated).
9. Provide subject with discharge instructions, wallet medication card for study drug and schedule follow-up study office visit appointment for 14 days ± 5 d post study drug administration.

6.5.8 Hospitalization lasting >24 Hours

If the subject remains in the hospital >24 hours, perform the following daily until discharge:

1. Obtain routine clinical tests and evaluations in accordance with institutional guidelines and standard of care (e.g., vital signs, blood samples for standard clinical laboratory evaluations, ECG, etc.).
2. Assess and record adverse events, including bleeding and clinical efficacy endpoint events (MACE, stent thrombosis).
3. Review and record all concomitant medications taken during hospitalization.

If the subject remains in the hospital for ≥48 hours, collect blood samples for optional research according to site participation and subject's individual consent (see **Section 6.6.3.2**) just prior to discharge:

a. Samples for pharmacokinetics (plasma) and pharmacodynamics- platelet aggregation (whole blood) (PK/PD sub-study; participating sites only); and
b. Plasma sample for exploratory biochemical, inflammatory and metabolic markers.

6.5.9 14 days (± 5 d) Post Study Drug Infusion Follow-Up Outpatient Visit

The subject will return to the investigational site approximately 14 days following their dosing with the study drug.

1. Review and record interval history since discharge including any concomitant illnesses, procedures, therapies (e.g., transfusions), adverse events, including bleeding and clinical

efficacy endpoint events (MACE, stent thrombosis). The subject should be questioned regarding any hospitalization and/or medical visits relating to episodes of ischemia or bleeding or adverse events that may have occurred since discharge. Original source documents must be collected for any events. Should a subject be re-admitted or undergo care at an outside facility, all possible efforts should be made to obtain the original source documents.

2. Review and record concomitant medications (including maintenance anti-platelet treatment) taken since discharge.
3. Perform a complete physical examination.
4. Obtain vital signs, including heart rate and blood pressure.
5. Record a standard 12-lead ECG.
6. Obtain blood sample from the subject for the following standard clinical laboratory evaluations:
    a. Hematology- complete blood count (CBC), differential white blood count and platelet count.
7. Perform and document urine pregnancy test (urine beta-HCG point-of-care test) for all female subjects of child-bearing potential.
8. Additional clinical laboratory tests and/or other evaluations may be performed for standard of care.
9. Collect blood samples for optional research according to site participation and subject's individual consent (see **Section 6.6.3.2**):
    a. Samples for pharmacokinetics (plasma) and pharmacodynamics- platelet aggregation (whole blood) (PK/PD sub-study; participating sites only); and
    b. Plasma sample for exploratory biochemical, inflammatory and metabolic markers.
10. Schedule follow-up telephone contact 30 days (± 5 d) post study drug administration.

6.5.10 30 days (± 5 d) Post Study Drug Infusion Follow-Up Telephone Contact

The subject will be contacted approximately 30 days following dosing with the study drug.

1. Review and record interval history since the 14-day study visit including any concomitant illnesses, procedures, therapies (e.g., transfusions), adverse events, including bleeding and clinical efficacy endpoint events (MACE, stent thrombosis). The subject should be questioned regarding any hospitalization and/or medical visits relating to episodes of ischemia or bleeding or adverse events that may have occurred since discharge. Original source documents must be collected for any events. Should a subject be re-admitted or undergo care at an outside facility, all possible efforts should be made to obtain original source documents.
2. Review and record concomitant medications (including maintenance anti-platelet treatment) taken since 14-day study visit.

3. Schedule follow-up phone contact for 90 days (± 7 d) post study drug administration.

6.5.11 90 days (± 7 d) Post Study Drug Infusion Follow-up Telephone Contact

The subject should be contacted approximately 90 days following dosing with the study drug.

1. Review and record interval history since the 30-day study telephone call including any concomitant illnesses, procedures, therapies (e.g., transfusions), adverse events, including bleeding and clinical efficacy endpoint events (MACE, stent thrombosis). The subject should be questioned regarding any hospitalization and/or medical visits relating to episodes of ischemia or bleeding or adverse events that may have occurred since discharge. Original source documents must be collected for any events. Should a subject be re-admitted or undergo care at an outside facility, all possible efforts should be made to obtain original source documents. Note that non-serious adverse events (bleeding and non-bleeding) will not be collected in the eCRF beyond the 30-day contact. All serious adverse events and clinical efficacy endpoint events will be collected through the 90-day contact.
2. Review and record concomitant medications (including maintenance anti-platelet treatment) taken since 30-day study telephone call.

Appropriate treatment for each subject after completing the study shall be at the investigator's discretion.

6.5.12 Non-PCI Subjects: Catheterization to Study Completion

Non-PCI subjects will receive medical management or CABG. The following provisions apply to all subjects when it is determined that PCI will not commence:

- Subjects should receive all subsequent care according to local standards.
- All post cardiac catheterization protocol-specified tests, evaluations and collections should be performed as per **Section 1.2** and **Section 6.5** and **Section 6.6** as usual.

Additional information/documentation for **medical management**:

- During the subject's hospitalization, the investigator or qualified designee should be sure to record the methods of medical management in source documentation/medical records and in the eCRF.

Additional information/documentation for **CABG:**

Every effort must be taken to obtain and record in source documentation and in the eCRF the following information for subjects who undergo CABG:

- Postoperative bleeding volume, measured by
  - chest tube drainage for the first 24 hours after completion of the procedure, and
  - total chest tube drainage;

- type and number of units of transfused blood products within a 48 hour period;
- reoperation following closure of the sternotomy incision for the purpose of controlling bleeding;
- abnormalities in laboratory tests for cardiac enzymes and CBC; and
- ECG abnormalities.

6.6 Study Procedures

The Study Flow Chart in **Section 1.2** summarizes the study procedures to be performed at each protocol time point. Individual study procedures are described below. For details of the procedures for assessment and reporting of adverse events, see **Section 6.7.2.2** (Assessment and Reporting of Adverse Events).

In order to minimize variability of evaluations, it is recommended that the same individuals perform the same types of evaluations for all subjects at each clinical site.

If a subject's participation in the study is discontinued for any reason, every attempt should be made to obtain final information for each test and evaluation as shown in the Study Flow Chart in **Section 1.2** pending the subject's consent to undergo the study-directed procedures and permission to disclose the corresponding information/data.

6.6.1 Standard Procedures and Clinical Evaluations

- **Explain Study and Obtain Written Informed Consent for the Main Study: Screening**
  The investigator or qualified designee will explain the study to the prospective subject, answer all of his/her question (verbally or in writing), and obtain written informed consent before performing any study-related procedure. The investigator or qualified designee will also review acceptable methods of contraception (birth control) while taking part in this study with both female subjects of child-bearing potential and male subjects. The execution of the informed consent will be documented in the subject's medical records and a copy(s) of the informed consent(s) will be given to the subject.

- **Explain Optional Sampling for Pharmacokinetics/Pharmacodynamics, Exploratory Biomarkers and Pharmacogenomics: Screening**
  The investigator or qualified designee will explain the optional sampling to the prospective subject, answer all of his/her question (verbally or in writing), and obtain written informed consent before performing any procedure related to the optional sampling. Written informed consent for the optional sampling for PK/PD sub-study (select sites only), exploratory biomarkers and pharmacogenomics (DNA and RNA) **(See Sections 6.6.3.2, 6.7.3 and Appendix E)** research may be contained in the same instrument as written informed consent for the rest of the study, or may be separate documents, at the discretion of the local Institutional Review Board (IRB). Regardless, separate signatures of informed consent are required to collect the optional samples for

PK/PD, exploratory biomarkers and pharmacogenomics research. Subjects who decline to sign the consent for optional research may still continue in the study. The execution of the informed consent will be documented in the subject's medical records and a copy(s) of the informed consent(s) will be given to the subject.

- **Review Inclusion/Exclusion Criteria (Including Prior and Current Medications and Therapies): Screening**
  The inclusion and exclusion criteria will be reviewed by the investigator or qualified designee to ensure that the subject qualifies for the study. All medications and therapies during the 30 days prior to enrollment/randomization will be recorded in the source documentation and transcribed to the eCRF.

- **Medical History: Screening**
  A medical history will be obtained by the investigator or qualified designee. Subject history should include information on family history and personal history. Subject history should include information concerning family history of chest pain and/or heart attack, general cardiac history, alcohol intake, smoking, as well as other coronary risk factors such as diabetes, hypertension, dyslipidemia, and obesity. Particular attention and documentation should be given to the subject's cardiac history, the clinical evaluation leading to the cardiac catheterization procedure (i.e., CAD presentation, Canadian Cardiovascular Society Classification for angina, Thrombolysis in Myocardial Infarction Risk Score for UA/NSTEMI, and New York Heart Association Classification of Functional Status for heart failure), bleeding history and allergy history (drugs, IV contrast media, food, stings and atopic conditions and any required treatment/ medical intervention).

- **Physical Examination: Screening, 24-hour/Discharge*, 14-day Visit**
  The investigator or qualified designee will perform an examination to include skin, head, neck, pulses, lungs, heart, abdomen, extremities, and gross neurologic system. Abnormal findings not present at screening or abnormal findings that were present at screening but have become worse in severity/grading over baseline will be considered adverse events and recorded as per the AE procedures **in Section 6.7.2.2**.

  *Note, the clinical site's standard of care, cardiac catheterization nursing assessment may be performed to evaluate the subject's clinical status in lieu of a complete physical exam at the 24 hour/discharge time point. This exam should be completed by qualified nursing staff with particular attention being paid to signs/symptoms related to the known side effects of PZ-128 (i.e., allergic reaction, injection site reaction and paresthesia) and other possible side effects from the procedure and/or concomitant medications (i.e., bleeding).

- **Vital Signs Assessment: Screening, Just Before Study Drug Infusion, Just After Completion of Study Drug, After Catheterization/PCI, 24-hour/Discharge, 14-day Visit**
  At screening, the investigator or qualified designee will measure blood pressure and

pulse (after the subject has been supine for at least 5 minutes), and measure body weight and height without shoes and heavy clothing. Thereafter, only blood pressure and pulse will be measured. Measurements will be made with the subject lying down just before study drug infusion, just after study drug completion, after catheterization/PCI and 24-hour/discharge. Measurements will be made with the subject seated at the 14-day visit.

- **Assay- Pregnancy Test (Urine beta-HCG) for All Female Subjects of Child-Bearing Potential: Screening (must be within 24 hour of study drug administration), 14-day Visit**
  The investigator or qualified designee will record the point-of-care urine beta-HCG test. If more frequent pregnancy testing is required by local standards of care, perform the testing as required and report pregnancy as specified in **Section 6.7.2.2.7.**

- **Electrocardiogram: Screening, After Catheterization/PCI, 14-day Visit**
  A standard 12-lead ECG will be recorded by a qualified ECG technician or other qualified individual after the subject has been supine for at least 5 minutes. All ECG reports in the study must include heart rate (HR) and QRS, QT, QT_c (with method of correction) and PR intervals. Timing after catheterization/PCI will be dictated by standard practice and clinical signs/symptoms of cardiac ischemia.

- **Record Previous and Concomitant Medications and Therapies: Screening, 24-hour/Discharge, 14-day Visit, 30-day Phone Call, 90-day Phone Call**
  Concomitant medications will be recorded in the subject's source documents and the appropriate section of the eCRF (See **Section 6.4.3**, Other Required Treatments and **Section 6.4.8**, Non-Study Treatments).

  The following medications/therapies are exempt from being reported to the sponsor, unless they are deemed the cause of or contributor to an adverse event or used in the treatment of an adverse event:

  - Electrolyte or nutritional replacement
  - IV fluids
  - Contrast media
  - Anesthetic medications that are inhaled or topical
  - Medications for surgical or nonsurgical procedures (e.g., sedatives, anesthestic)
  - Mediations/therapies that are not directly administered to the subject, such as cardioplegic solution or antithrombin used in conjunction with the cardiopulmonary bypass pump
  - Nonsystemic bowel prep (e.g., enema)
  - Vaccines

- **Adverse Events Assessment (Bleeding and Non-Bleeding): Continually During Hospitalization Following Enrollment/Randomization, 14-day Visit, 30-day Phone Call**

- **Serious Adverse Events Assessment (Bleeding and Non-Bleeding): Continually During Hospitalization Following Enrollment/Randomization, 14-day Visit, 30-day Phone Call, 90-day Phone Call**

- **Suspected Clinical Efficacy Endpoint Events (MACE & Stent Thrombosis): Continually During Hospitalization Following Enrollment/Randomization, 14-day Visit, 30-day Phone Call, 90-day Phone Call**

  All adverse events (AEs) (bleeding and non-bleeding) will be reported to the sponsor in the eCRF from the time of randomization through 30 days after study drug administration. Only AEs which are classified as serious adverse events (SAEs) (bleeding and non-bleeding) will be reported to the sponsor in the eCRF after 30 days up until 90 days after study drug administration. All clinical efficacy endpoint events (MACE, stent thrombosis) will be reported to the sponsor from the time of randomization through 90 days after study drug administration.

  Recording of adverse events, including reports by subjects, observations by study staff, and results of laboratory tests or other procedures, is described in **Section 6.7.2.2** (Assessment and Reporting of Adverse Events). Endpoint events (and associated symptoms) are identified as part of **Section 6.7.1** (Safety/Activity/Efficacy Endpoints) and are defined specifically in **Appendix B**, and will be detailed in the separate CEC charter for purposes of adjudication.

  Note that clinical efficacy endpoint events- major adverse cardiac events **(MACE)** (any of cardiovascular death, non-fatal myocardial infarction (MI), non-fatal stroke, recurrent ischemia requiring hospitalization or urgent coronary revascularization) and **definite or probable stent thrombosis-** as well as **bleeding events** (safety) are considered "Clinical Endpoint Events" and will be adjudicated periodically during the conduct of the trial. The clinical endpoint events will be monitored in aggregate by the DSMB at appropriate intervals (i.e., quarterly). As such, these clinical endpoint events will <u>not</u> be reported separately in an expedited manner as "serious adverse events" to the sponsor's SAE contact (see **Section 6.7.2.2.10**- Protocol-Specific Exceptions to SAE Reporting).

6.6.2 Standard Laboratory Tests

The investigator or qualified designee will collect all appropriate samples for analysis which will be then be performed at the local clinical laboratory on site. The values from Screening (Enrollment/Randomization) will be considered Baseline values for all standard laboratory tests. Results of testing should be reviewed by the investigator/qualified designee in a timely manner for pertinent clinically significant values. If any abnormal test result is considered to be clinically significant, the test should be repeated at appropriate time intervals until it returns to Baseline or becomes a clinically insignificant finding or the investigator/qualified designee decides it is not appropriate to continue.

- **Hematology: Screening, After Catheterization/PCI, 24-hour/Discharge, 14-day Visit**
  Blood samples will be collected to quantify red blood cell (RBC) count, total and differential white blood cell (WBC) count, hemoglobin concentration, hematocrit, and platelet count. Timing after catheterization/PCI will be dictated by standard of care.

- **Serum Chemistry: Screening, 24-hour/Discharge**
  Blood samples will be collected to quantify the following variables:
  Sodium, potassium, chloride, carbon dioxide, glucose, blood urea nitrogen (BUN), creatinine, alkaline phosphatase, total bilirubin, alanine aminotransferase (ALT) and aspartate aminotransferase (AST).

- **PT/INR and aPTT: Screening, 24-hour/Discharge**

- **Activated Clotting Time: During and After PCI**
  Following administration of the initial bolus of anti-coagulant, point-of-care testing of the activated clotting time (ACT) should be measured at "peak dose" and just prior to sheath removal, as well as according to the standard of care and local routine practice.

- **Lipid Panel: Screening**
  Blood samples (does not need to be fasting) will be collected to quantify total cholesterol, high-density-lipoprotein cholesterol (HDL-C), calculated low-density-lipoprotein cholesterol (LDL-C) and triglycerides.

- **Glycosylated Hemoglobin: Screening**
  Blood sample (does not require fasting) will be collected to quantify glycosylated hemoglobin (HgA1c) levels.

6.6.3 Research Laboratory Tests and Collections

Detailed procedures for collection, handling/processing, packaging, and shipment of samples associated with the various tests mentioned in this section will be included in a separate *Research Laboratory Manual* and will be provided to each clinical site. The investigator or qualified designee will collect all appropriate samples for analysis. Results from the analyses will be transmitted electronically from the sponsor/sponsor's designated laboratory into a research laboratory study database and will not be individually reported back to the clinical site. Therefore the results will not become part of the subject's medical records or be used by the investigator for medical management/clinical decision-making.

6.6.3.1 Mandatory

Samples will be collected from all subjects who sign the consent to participate in the main research study. The time points provided for the collection of blood samples for the Cardiac Troponin I (cTnI) will be indexed from the completion of the PCI.

- **Cardiac Troponin I (Serum): Just Prior to Study Drug Infusion, After PCI**
  Blood samples will be collected to quantify the Cardiac Troponin I (cTnI) marker just before the administration of the study drug, 4 to 8 hours and 12 to 24 hours after completion of the **PCI** (Note, PCI subjects only). In addition, samples will be collected immediately after the occurrence of symptoms suggestive of acute coronary syndrome (ACS), and again 4 to 8 hours and 12 to 24 hours after symptoms if the subject has not yet been discharged from the hospital.

  Note, the evaluation of cardiac biomarker(s) (i.e., Cardiac Troponin I, Cardiac Troponin T, Creatine Kinase or Creatine Kinase-Myocardial Band) done locally for the clinical management of the subject will be performed as per standard of care and local routine practice. The results from those standard clinical laboratory tests should be recorded in the appropriate section of the eCRF.

6.6.3.2 Optional

Samples will be collected only from subjects who sign the appropriate section in the consent form(s) for these optional collections and analysis. The time points provided for the collection of blood samples for exploratory biomarkers research, pharmacogenomics and pharmacokinetics/pharmacodynamics-platelet aggregation will be indexed from the start of the PZ-128/placebo infusion (i.e., Time=0).

These blood samples and any other components may be retained by the sponsor for analysis until the study-specific scientific objectives have been completed. The subject retains the right to have the sample material destroyed at any time by contacting the investigator. The sponsor will be the exclusive owner of any data, discoveries, or derivative materials from the sample materials and is responsible for the destruction of the sample(s) at the request of the research subject through the investigator or at the end of the storage period. Following the request from a research subject, the investigator will provide the sponsor with the required study and subject number so that any remaining blood samples and any other components can be located and destroyed. If a commercial product is developed from this research, the sponsor will own the commercial product. The subject will have no commercial rights to such product and will have no commercial rights to the data, information, discoveries, or derivative materials gained or produced from the samples. See **Section 8.2** and **Appendix E** for subject confidentiality.

Subjects at All Centers

- **Exploratory Biomarkers Research (Plasma): Just Prior to Study Drug Infusion, Select Times During Hospitalization, 24-hour/Discharge, 14-day Visit**
  Blood samples for exploratory research of systemic markers of thrombosis and platelet reactivity, inflammation, and metabolomics will be collected just before the administration of the study drug, then 2 hours + 15 min (immediately after completion of infusion), 6 hours – 2 h (4 to 6 h) and 24 hours ± 2 h or discharge (whichever occurs sooner) after the start of the study drug infusion and at the 14-day visit. After the 24-hour collection, if a subject remains in the hospital for ≥48 hours, a blood sample will

be collected just prior to discharge. The samples will be analyzed by the sponsor under non-GLP conditions.

- **Pharmacogenomics (DNA and RNA) (Whole Blood): Just Prior to Study Drug Infusion**
  One blood sample for DNA, RNA and microRNA analysis will be collected just before the administration of the study drug. Pharmacogenomics analysis will be undertaken to investigate whether DNA polymorphisms, RNA levels and microRNAs correlate with the expression of genes involved in thrombosis, inflammation, and cardiometabolic diseases and whether these genetic components correlate with PZ-128 outcomes. The samples will be analyzed by the sponsor and/or at one of the participating clinical sites under non-GLP conditions.

Subjects at Select Clinical Site(s) Participating in the PK/PD Sub-study

- **Pharmacodynamics- Platelet Aggregation (Whole Blood): Just Prior to Study Drug Infusion, Select Times During Hospitalization, 24-hour/Discharge, 14-day Visit**
  Blood samples will be collected just before the administration of the study drug, then 30 minutes ± 5 min, 1 hour ± 5 min, 2 hours + 15 min (immediately after completion of infusion), 6 hours – 2 h (4 to 6 h) and 24 hours ± 2 h or discharge (whichever occurs sooner) after the start of the study drug infusion and at the 14-day visit. After the 24-hour collection, if a subject remains in the hospital for ≥48 hours, a blood sample will be collected just prior to discharge. The samples will be analyzed using light transmission aggregometry (LTA) on-site under non-GLP conditions.

- **Pharmacokinetics (Plasma): Just Prior to Study Drug Infusion, Select Times During Hospitalization, 24-hour/Discharge, 14-day Visit**
  Blood samples will be collected just before the administration of the study drug, then 30 minutes ± 5 min, 1 hour ± 5 min, 2 hours + 15 min (immediately after completion of infusion), 6 hours – 2 h (4 to 6 h) and 24 hours ± 2 h or discharge (whichever occurs sooner) after the start of the study drug infusion and at the 14-day visit. After the 24-hour collection, if a subject remains in the hospital for ≥48 hours, a blood sample will be collected just prior to discharge. The samples will be analyzed by the sponsor/sponsor's designee under non-GLP conditions.

The PK/PD sub-study will also include the collection of plasma for exploratory biomarkers research, meaning that subjects at select clinical site(s) participating in the PK/PD sub-study will be asked in one question whether they will allow additional blood to be collected for the PK/PD samples and exploratory biomarkers samples. Subjects at clinical site(s) not participating in the PK/PD sub-study will be asked in one question whether they will allow additional blood to be collected just for the exploratory biomarkers samples.

Approximately 54-64 mL of blood will be collected from each subject during their participation in the main portion of the study.

Subjects in whom informed consent has been obtained will have approximately 25-30 mL of additional blood collected during their participation in the exploratory biomarkers research component of the study.

Subjects in whom informed consent has been obtained and are participating at a clinical site that has the capability to perform LTA will have approximately 200-230 mL of additional blood collected during their participation in this PK/PD sub-study (includes the PK/PD and exploratory biomarkers samples).

Subjects in whom informed consent has been obtained will have approximately 5 mL of additional blood collected one time during their participation in the pharmacogenomics component of the study.

Table 1 summarizes the collection of blood samples for the protocol-directed tests and optional research evaluations.

Table 1. Summary of Total Blood Sampling

	Number of Samples per Visit						**Volume (mL)**	
	Screening Enrollment	**Pre-Study Drug**	**Post-Cath ± PCI**	**24 hours / Discharge**	**14 Days**	**Total Draws**	**Blood Volume per Test**	**Sub-total**
Hematology	1		1	1	1	4	4.5	18
Serum Chemistry	1			1		2	8.5	17
Lipid Panel	1					1	1	1
Hemoglobin A1C	1					1	3	3
PT/INR, aPTT	1			1		2	4.5	9
ACT			2			2	2	4
Cardiac Troponin I (Sample for Centralized Research Analysis)		1	0-1^	0-1^		1-6^	2	2-12^
Volume per Visit (mL)	21.5	2	10.5	19.5	4.5		**TOTAL**	54-64^
Optional								
Exploratory Biomarkers		1	2	1-2*	1	5-6*	5	25-30*
Pharmacogenomics		1				1	5	5
Sub-Study (site specific)								
Pharmacodynamics-Platelet Aggregation		1	4	1-2*	1	7-8*	20	140-160*
Pharmacokinetics		1	4	1-2*	1	7-8*	5	35-40*

^ depending upon performance of PCI, length of stay and the occurrence of symptoms suggestive of ACS
*depending upon total length of stay

6.7 Study Assessments

Study assessments that require adjudication by the CEC are defined in Appendix B and will be detailed in a separate CEC charter.

6.7.1 Safety/Activity/Efficacy Endpoints

6.7.1.1 Primary Endpoint (Safety)

The primary endpoint of the study will be the incidence of TIMI major plus minor bleeding not related to CABG through 30 days after treatment with the study drug. Definitions are as follows:

- TIMI Major bleeding is defined as
  - intracranial hemorrhage, or
  - clinically significant overt signs of bleeding associated with a decrease in hemoglobin concentration of ≥5 g/dL (or hematocrit ≥15%), or
  - fatal bleeding within 7 days.

- TIMI Minor bleeding is defined as clinically overt signs of bleeding (including imaging) associated with a decrease in hemoglobin concentration of 3 to <5 g/dL (or hematocrit of 9 to <15%) that does not otherwise meet criteria for major bleeding.

Hemoglobin concentration and hematocrit will be adjusted for any transfusion of packed red blood cells or whole blood given between enrollment/randomization and the post-transfusion measurements by Landefeld and coworkers as follows:

Δ Hemoglobin (Hgb) = [Baseline Hgb – post-transfusion Hgb] + [number of transfused units]
Δ Hematocrit (Hct) = [Baseline Hct – post-transfusion Hct] + [number of transfused units x 3]

6.7.1.2 Key Secondary Endpoint (Efficacy)

The key secondary endpoint is the incidence of any component of the MACE composite through 30 days and 90 days after treatment with the study drug.

6.7.1.3 Other Secondary Endpoints Related to Safety

Other secondary safety endpoints include the incidences of the following:

1. TIMI minimal bleeding not related to CABG (defined as clinically significant overt signs of bleeding associated with a drop in hemoglobin concentration of <3 g/dL (or hematocrit of <9%) that does not otherwise meet criteria for major or minor bleeding) through 30 days after treatment;
2. "clinically significant" bleeding (defined as TIMI major or minor bleeding or bleeding requiring medical attention) through 30 days after treatment; and

3. TIMI bleeding related to CABG through 30 days after treatment (subjects who undergo CABG will specifically have assessment of (a) surgical wound bleeding using quantitative total chest tube drainage in the first 24 hours; (b) need for transfusion of blood products in the first 48 hours; and (c) need for surgical re-exploration).

6.7.1.4 Other Secondary Endpoints Related to Efficacy

Other secondary efficacy endpoints will include the following:

1. incidence of the individual components of the MACE composite through 30 days and 90 days after treatment;
2. incidence of definite or probable stent thrombosis through 30 days and 90 days after treatment;
3. inhibition of platelet aggregation induced by SFLLRN, thrombin, ADP, collagen and AYPGKF relative to baseline at several time points following treatment at select sites;
4. population pharmacokinetics at baseline and at several time points following treatment; and
5. effect of the investigator choice of P2Y12 inhibitor (i.e., clopidogrel versus prasugrel/ticagrelor) on the individual components of the MACE composite.

6.7.1.5 Exploratory Endpoints

These endpoints are not part of the formal objectives and are included as additional ways to evaluate bleeding and to gather information for future research.

1. incidence of bleeding events according to the BARC (Bleeding Academic Research Consortium) classification through 30 days after treatment;
2. change from baseline in select systemic markers of thrombosis and platelet reactivity, inflammation, and metabolomics at select time points; and
3. population pharmacogenomics at baseline.

6.7.1.6 Appropriateness of Measurements

The study design is appropriate for the indication studied. Validated methods of data collection, analysis, and evaluation will be used for the study.

6.7.2 Additional Safety Assessment

6.7.2.1 Specification of Safety Variables

In addition to bleeding, safety variables to be assessed include all other adverse events, and results of physical examination, ECG, and standard clinical laboratory tests.

6.7.2.2 Assessment and Reporting of Adverse Events

An adverse event (AE) is defined as any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug related. An AE can be any unfavorable and unintended sign (e.g., an abnormal laboratory finding), symptom or disease temporally associated with the use of a drug, and does not imply any judgment about causality. An adverse event can arise with any use of the drug (e.g., use in combination with another drug) and with any route of administration, formulation, or dose, including an overdose.

AEs may include the onset of new illness and the exacerbation of pre-existing medical conditions. Worsening indicates the pre-existing medical condition (e.g., diabetes, headaches) has increased in severity, frequency, and/or duration, and/or has an association with a significantly worse outcome. A pre-existing condition that has not worsened during the study, and involves an intervention such as elective cosmetic surgery or a medical procedure while on study is not considered an adverse event.

All AEs whether volunteered by the subject, discovered by study personnel during questioning, or detected through physical examination, laboratory test, or other means, and the treatment provided must be assessed by the investigator or qualified designee and recorded in the subject's source documents/medical records.

All adverse events (AEs) (bleeding and non-bleeding) will be reported to the sponsor in the eCRF from the time of randomization through 30 days after study drug administration. Only AEs which are classified as serious adverse events (SAEs) (bleeding and non-bleeding) will be reported to the sponsor in the eCRF after 30 days up until 90 days after study drug administration. All clinical efficacy endpoint events (MACE, stent thrombosis) will be reported to the sponsor from the time of randomization through 90 days after study drug administration.

The investigator must assign the following adverse event attributes:

- Adverse event diagnosis or syndrome(s), if known (if not known, signs or symptoms),
- Dates of onset and resolution,
- Severity,
- Assessment of relatedness to PZ-128 or PZ-128 placebo, and
- Action taken and/or outcome (e.g., discontinuation of study drug administration, hospitalization, etc.).

Subjects will be questioned and/or examined by the investigator or a qualified designee for evidence of AEs. Overall, the questioning of subjects with regard to the possible occurrence of adverse events will be generalized such as, "How have you been feeling since your last visit/contact?" The presence or absence of specific AEs should not be elicited from subjects. However, subjects will be questioned about the occurrence of any study-specific safety (bleeding) and efficacy (MACE, stent thrombosis) endpoint events (e.g., overt signs of

bleeding, chest pain, need for interim medical care, procedure(s) or hospital admission).

The investigator is responsible for reviewing laboratory and other test (e.g., ECG) results and determining whether an abnormal value in an individual subject represents a change from the subject's baseline values. In general, only abnormal laboratory findings that represent a change from the subject's baseline values that have clinical significance (based on the investigator's judgment) should be recorded as adverse events. A clinically significant lab value is one that indicates a new disease process, an exacerbation or worsening of an existing condition or requires further action(s) to be taken. Where applicable, clinical sequelae (not the laboratory value) should be recorded as the adverse event.

In particular, the following laboratory results must be captured as AEs:

- decrease from Baseline in platelet count to a value <75,000/mm^3 (note, <50,000 is a serious adverse event- see **Section 6.7.2.2.5**);
- increase from Baseline in ALT or AST activity to a value greater than three times the upper limit of the reference range (i.e., >3 x ULN);
- increase from Baseline in total bilirubin concentration to a value greater than one and a half times the upper limit of the reference range (i.e., >1.5 x ULN); and
- increase from Baseline in serum creatinine to a value greater than one and a half times the upper limit of the reference range (i.e., >1.5 x ULN) or greater than one and a half times the baseline value (i.e., >1.5 x baseline).

6.7.2.2.1 Assessment of Adverse Event Severity and Relationship to Treatment

Where the determination of adverse event severity rests on medical judgment, the determination of severity must be made with the appropriate involvement of a medically-qualified investigator.

The descriptions and grading scales found in National Cancer Institute Common Terminology Criteria for Adverse Events (NCI CTCAE) version 4.0 will be used for all events with an assigned CTCAE grading. For events without assigned CTCAE grades, the recommendation is that the CTCAE criteria that convert the severity description into CTCAE grades should be used (see guideline below). For each episode on an adverse event, the highest attained CTC grade should be reported. All appropriate treating areas should have access to a copy of the CTCAE version 4.0. A copy of the CTCAE version 4.0 can be downloaded:
http://evs.nci.nih.gov/ftp1/CTCAE/CTCAE_4.03_2010-06-14_QuickReference_8.5x11.pdf

This version of CTCAE is MedDRA v12.0 (Medical Dictionary for Regulatory Activities Terminology) compatible at the AE (Adverse Event) term level where each CTCAE term is a MedDRA LLT (Lowest Level Term). CTCAE v4.0 includes 764 AE terms and 26 'Other, specify' options for reporting text terms not listed in CTCAE. Each AE term is associated with a 5-point severity scale. For events without corresponding CTCAE terms, the MedDRA online resource should be consulted for the LLT:
http://bioportal.bioontology.org/ontologies/MEDDRA?p=classes&conceptid=10063933

The CTCAE displays Grade 1 through 5 with unique clinical descriptions for severity for each AE based on this general guideline:

Grade 0	No AE (or within normal limits).
Grade 1	Mild; asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
Grade 2	Moderate; minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental activities of daily living (ADL).
Grade 3	Severe or medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL.
Grade 4	Life-threatening consequences; immediate risk of death; urgent intervention indicated.
Grade 5	Death related to AE.

After naming and grading the event, a medically qualified investigator must assess the relationship of any AE to the use of study drug (i.e., PZ-128/ placebo), based on available information, using the following guidelines:

Unlikely related:	no temporal association, or the cause of the event has been identified, or the investigational drug cannot be implicated;
Possibly related:	temporal association, but other etiologies are likely to be the cause; however, involvement of the investigational drug cannot be excluded;
Probably related:	temporal association, other etiologies are possible, but unlikely.

For purposes of regulatory reporting, AEs listed as "possibly" and "probably" related to the investigational drug will be considered to have a suspected "reasonable causal relationship" to the investigational drug. For serious events that are unexpected, the sponsor will consider the investigator's causality assessment when reporting to regulatory authorities, but ultimately the sponsor will determine if there is a reasonable possibility that the drug caused the event. For SAEs, causal relationship will also be assessed for other medication and study procedures. Further guidance to the interpretation of the causality question is found in **Appendix C**.

The expectedness of an adverse event shall be determined according to the reference document. The reference document for this current study is to be the current version of the sponsor's investigator brochure (IB). An adverse event is considered "unexpected" if it is not listed in the IB or is not listed at the specificity or severity that is observed. Adverse events listed in the IB as occurring with members of the same class of drugs, or as anticipated from the pharmacological properties of the drug, are considered unexpected until they have been observed with the drug under investigation.

6.7.2.2.2 Monitoring Adverse Events

Subjects experiencing AEs should be followed clinically until their health has returned to baseline status or until all parameters have returned to normal or have otherwise been explained. It is expected that the investigator will provide or arrange appropriate supporting care for the subject if necessary. The investigator or qualified designee is expected to report ongoing AEs at the subject's completion of the clinical study to the subject's primary care physician or referring cardiologist who will determine the need for and provide standard medical care.

Any actions taken and follow-up results must be recorded either on the appropriate page of the eCRF or in a follow-up letter to the sponsor, as well as in the subject's source documentation/medical records.

For all AEs that require the subject to be discontinued from the study drug or the study procedures, relevant clinical assessments and laboratory tests will be repeated as clinically appropriate until final resolution or stabilization of the event(s).

The sponsor, Tufts Medical Center, retains the right to request additional information for any subject with ongoing AEs/SAEs at the end of the study, if judged necessary.

6.7.2.2.3 Known Adverse Events Relating to the Underlying Clinical Condition

The underlying clinical condition in the subjects in the current trial is coronary artery disease and non-ST-segment-elevation ACS (NSTE-ACS). Therefore, any signs and symptoms associated atherosclerosis, NSTE-ACS and/or subjects undergoing cardiac catheterization/PCI may be expected/ anticipated. These may include (but are not limited to) coronary, cerebral or peripheral ischemia, ACS, stroke/transient ischemic attack (TIA), MI, and life-threatening ischemic events and any signs and symptoms associated with these conditions, such as (but not limited to) chest/jaw/arm pain, dyspnea, diaphoresis, hypotension, nausea, vomiting, dizziness, changes in ECG pattern, hemiparesis, and claudication.

[redacted]

No bleeding risk (i.e., bleeding or any other coagulation abnormalities) has been observed in animal or human investigations with PZ-128 to date and therefore bleeding should be considered unexpected for PZ-128. However, bleeding is anticipated to occur in this trial with some frequency, regardless of PZ-128 exposure, due to it being a known consequence of the underlying standard of care procedures for the study population (cardiac catheterization/PCI) and background drug regimen (i.e., anti-coagulant and anti-platelet drugs). Bleeding is a known risk of other PAR1 inhibitors [Zontivity® Full prescribing information]. The AEs mentioned under this section still need to be recorded in the subject's source documents/ medical records and on the eCRF, regardless of causality.

The current Investigator's Brochure (IB) for PZ-128 is the reference document containing safety information for the investigational product (IP) in this study, PZ-128. Adverse events expected for the IP are those described as such in the most recent IB.

6.7.2.2.5 Definition of Serious Adverse Events

A **serious adverse event (SAE)** is any adverse drug experience occurring at any dose and any study phase (e.g., treatment, follow-up) that results in any of the following outcomes (as determined by either the investigator or sponsor):

- death;
- life-threatening AE (i.e., one that places the subject, in the view of the investigator or sponsor, at immediate risk of death from the AE as it occurs; it does not include an adverse event that, had it occurred in a more severe form, might have caused death);
- requires in-patient hospitalization (i.e., admission) or prolongs existing hospitalization (for ≥24 hours);
- persistent or significant disability/incapacity (i.e., substantial disruption of the ability to conduct normal life functions);
- congenital anomaly or birth defect;
- an important medical event that may not result in death, be life-threatening,

or require hospitalization may be considered a serious adverse event when, based upon appropriate medical judgment, they may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias, or convulsions that do not result in in-patient hospitalization, or the development of drug dependency or drug abuse.

In addition to the general definitions above for SAEs, **the following specific events will be considered as SAEs** in the context of this study:

- Anaphylaxis: defined as ≥CTCAE Grade 3 or meeting Sampson criteria (**Appendix C**);
- Decrease from Baseline in platelet count to a value <50,000/mm^3 (=CTC Grade 3)

Clarification should be made between the terms "serious" and "severe" since they are not synonymous. The term "severe" is often used to describe the intensity (and defined by the grading/severity scale) of a specific event (as in mild, moderate, or severe pain); the event itself, however, may be of relatively minor medical significance. This is NOT the same as "serious," which is based on the strict definition listed above and is based on subject or event outcome or action criteria usually associated with events that pose a threat to a subject's life or functioning. A severe AE does not necessarily need to be considered serious. Investigators should assess adverse event severity and seriousness independently.

Since the criteria for CTCAE severity differs from the regulatory criteria for serious adverse events, if adverse events correspond to grade 4 "life-threatening" CTCAE severity criteria (e.g., laboratory abnormality reported as grade 4 without manifestation of life-threatening status), it will be left to the investigator's judgment to also report these abnormalities as serious adverse events. For any adverse event that applies to this situation, comprehensive documentation of the event's severity status must be recorded in the subject's medical record.

All SAEs, whether or not deemed study drug-related or expected, **must be reported** by the investigator or qualified designee to the sponsor **within 24 hours of first becoming aware of the event.** The investigator/qualified designee will enter the required information about the SAE into the appropriate module of the eCRF, which will automatically result in distribution of the information to the appropriate sponsor contact(s). If the eCRF system is temporarily unavailable, then a full description of the event and any sequelae should be provided as a telefacsimile (FAX). If both the eCRF and FAX are not available, the event should be reported by telephone.

SAE Reporting Contact Information

If the report is initially given to the sponsor via telephone, a full description of the event and any sequelae, including the investigator-determined causality to study drug must be provided in writing (by FAX or eCRF) as soon as possible, so that the appropriate written report can be completed by the sponsor. If the report is initially given by FAX or telephone, then the required information about the SAE will be entered into the appropriate module of the eCRF immediately after the eCRF system is available.

The investigator is responsible for ensuring that all SAEs that occur at any time after the subject has been randomized up through 90 days following the study drug administration are reported to the sponsor. New information relating to a previously reported SAE must be submitted to the sponsor within 24 hours of receipt. The investigator may be asked to provide additional follow-up information, which may include a discharge summary or extracts from the medical record. Information provided about the SAE must be consistent with that recorded on the applicable eCRF.

Any SAE (including those which are ongoing when a subject completes his/her participation in the trial) must be followed by the site investigator (and will be followed by the sponsor) until one of the following occurs:

- the event resolves or stabilizes; or
- the event returns to baseline condition or value (if a baseline value is available).

Events which are ongoing at the time the clinical database is closed will be recorded as unresolved.

Reports of all SAEs must be communicated as soon as possible by the investigator to the appropriate IRB and/or reported in accordance with local law and regulations.

Federal regulations require that the sponsor, Tufts Medical Center, to report any suspected adverse reactions ("reasonable possibility" that the drug caused the event) that are both serious and unexpected (21 CFR 312.32). Such events must be reported to the Food and Drug Administration (FDA), the NHLBI/NIH and all participating investigators (i.e., all investigators to whom the sponsor is providing drug under its IND) within 15 calendar days after the sponsor receives the information and determines that it qualifies for reporting. Such notification must occur within 7 calendar days if the serious unexpected suspected adverse reaction (SUSAR) was fatal or life-threatening. The sponsor is also required to report other serious adverse events and qualifying adverse events in an expedited manner to its IRB in

compliance with local institutional and IRB policies. Therefore, once the investigator has determined that an SAE has occurred, it is important to adhere to the reporting timeframes outlined above. After receiving the investigator's SAE report, including the investigator's assessment of causality, the sponsor will also assess the event for seriousness and the relationship to PZ-128/Placebo, and will determine if the event was unexpected. Investigational New Drug Safety Reports (INDSRs) will be reported to the FDA and NHLBI/NIH by Tufts Medical Center and copies of the report(s) will be distributed to all participating clinical investigators. The investigator is responsible for notifying the relevant IRB of all INDSRs in accordance with institutional and IRB policies in addition to any other safety information which has been provided by the sponsor throughout the duration of the trial.

To comply with reporting regulations for serious adverse events, the treatment assignment of subjects who develop serious, unexpected, and related adverse events may be unblinded by the sponsor before submission to regulatory authorities.

6.7.2.2.6 Reporting of Subject Death

The **death of any subject** after enrollment/randomization up through 90 days following the study drug administration, regardless of the cause, **must be reported** by the investigator or qualified designee to the sponsor **within 24 hours of first becoming aware of the death**. If the report is given to the sponsor via telephone rather than in writing on the form designated for SAE reporting, a full description of the event and any sequelae, including the investigator-determined causality to study drug must be provided, so that the appropriate written report can be completed by the sponsor. If an autopsy is performed, the report must be provided to the sponsor. Reports of all deaths must be communicated as soon as possible to the appropriate IRB and/or reported in accordance with local law and regulations.

6.7.2.2.7 Reporting of Pregnancies

Pre-clinical genotoxicity studies with PZ-128 were negative and the reproductive and developmental risk of PZ-128 is likely low. However, reproductive and teratogenicity studies have not been performed and therefore the risks to the human embryo or fetus or sperm are unknown at this time. Women of child-bearing potential recruited into the study must have a negative urine pregnancy test (urine beta-HCG) prior to randomization and within 24 hours of treatment with the study drug. Women of child-bearing potential must agree to use a medically accepted method of contraception during the study from the time of providing written informed consent through 90 days after dosing with the study drug and will undergo a post study drug infusion pregnancy test (urine beta-HCG) at the 14-day Follow-Up Visit. If a female subject becomes pregnant or suspects she is pregnant while participating in this study, she must inform her treating obstetrician and principal investigator immediately. Pregnancy information on female clinical study subjects will be collected by the sponsor. If a female subject becomes pregnant during the course of the study, the study drug must be discontinued (if not already administered) and the investigator or qualified designee must contact the sponsor within 5 working days of the investigator or qualified designee first becoming aware

of the pregnancy. If a serious adverse event occurs in conjunction with the pregnancy, then the reporting time frame for an SAE (1 working day) and method of reporting must be met. The sponsor's representative will provide instructions on how to collect pregnancy information (i.e., Pregnancy Notification Form) including follow-up data on the outcome of the pregnancy.

Male participants will be advised to use a medically accepted method of contraception during the study from the time of study drug administration through 90 days after dosing with the study drug. Written information will be provided to the participating male subjects at the time of enrollment in the study's informed consent document which may be shared with their female partner(s) as to the unknown risks to a future pregnancy. If a female partner conceives and becomes pregnant while the male subject is participating in this study, the sponsor should be notified as per the procedures described above. However, information about the pregnancy may only be collected by the investigator and released to the sponsor if the female partner of the male subject provides written consent to release such information via an IRB-approved authorization/ consent form. A female partner of a male subject should only be presented with the authorization/consent form if a pregnancy or suspected pregnancy occurs. The sponsor will provide clinical sites with templates for these related documents to use as a model and the local IRB guidelines and approval process will be adhered to.

6.7.2.2.8 Preplanned Hospitalizations or Procedures

Hospitalization for SAE reporting purposes is defined as an inpatient hospital stay equal to or greater than 24 hours. Emergency room visits that do not result in admission to the hospital should be evaluated for one of the other "serious" outcomes.

Hospitalization admissions and/or surgical procedures/operations scheduled to occur during the entire study period, but planned prior to study enrollment/randomization are not considered AEs if the illness or disease existed before the subject was enrolled/randomized in the trial, provided that it did not deteriorate in an unexpected manner during the trial (e.g., surgery performed earlier than planned). If the event/condition worsens during the study, however, it must be reported as an AE (or SAE, if the event/condition results in a "serious" outcome).

As per the exclusion criteria (**Section 6.3.2**), major surgical procedures should not be planned to take place within 30 days following the study drug administration/catheterization/PCI.

6.7.2.2.9 Reports of Overdose

The doses of 0.3 mg/kg and 0.5 mg/kg are expected to be well-tolerated. The half-life of PZ-128 ranged from 1.3 to 1.8 hours in volunteers with CAD risk factors, so the blood levels of PZ-128 will be substantially lower in 8 hours (i.e., 3 to 4 half-lives).

Overdose is defined as any single dose above the maximum dose allowed by the clinical protocol.

The effects of overdose of PZ-128 in humans are not known. Potential adverse effects include bleeding, anaphylaxis and thrombophlebitis/injection site irritation based upon the experience in the previous phase 1 study which involved the administration of PZ-128 at dose levels up to and including 2 mg/kg. Although there are no known antidotes to overdose, in case of overdose, the subject should be closely monitored, and supportive treatment should be administered to treat the presenting clinical manifestations. Bleeding events should be treated according to standard clinical practice, depending upon the location and severity of the bleed. In severe cases, the administration of platelets may be considered, as it may counteract the pharmacologic effect of PZ-128.

Information on overdoses in clinical subjects will be collected by the sponsor. Should a subject experience an overdose during the course of the study (whether symptomatic or not), the investigator or qualified designee must contact the sponsor within 5 working days of the investigator or qualified designee first becoming aware of the overdose. Follow-up information on the outcome of the overdose should be forwarded to the sponsor.

Any event associated with, or observed in conjunction with, a product overdose (whether accidental or intentional) or a product abuse and/or withdrawal is considered an AE should be reported as such (**Section 6.7.2.2**). If a serious adverse event occurs in conjunction with the overdose, then the reporting time frame for an SAE (24 hours) must be met. The sponsor's representative will provide instructions on how to collect this information.

6.7.2.2.10 Protocol-Specific Exceptions to SAE Reporting to Sponsor

Suspected clinical endpoint events that are also serious adverse events are to be recorded in the subject's eCRF (bleeding or clinical efficacy endpoint modules), but will **not** be reported in an expedited manner to the sponsor's SAE contact as serious adverse events. The suspected endpoint events recorded in the eCRF will be monitored in aggregate by the DSMB at appropriate intervals in order to detect any potential imbalance between the study arms which would suggest that the events are occurring more frequently or at higher severity in the study drug treatment group than the control group.

Specifically, the following events will **not** be reported in an expedited manner to the sponsor's SAE contact:

- activity/efficacy endpoints- MACE- any of cardiovascular death, non-fatal MI, non-fatal stroke, recurrent ischemia requiring hospitalization, or urgent coronary revascularization with PCI or CABG; and stent thrombosis
- safety endpoints- any bleeding that does **not** result in death, regardless of severity

Note that death unrelated to cardiovascular disease and death attributable to bleeding **will** be reported in an expedited manner to the sponsor's SAE contact.

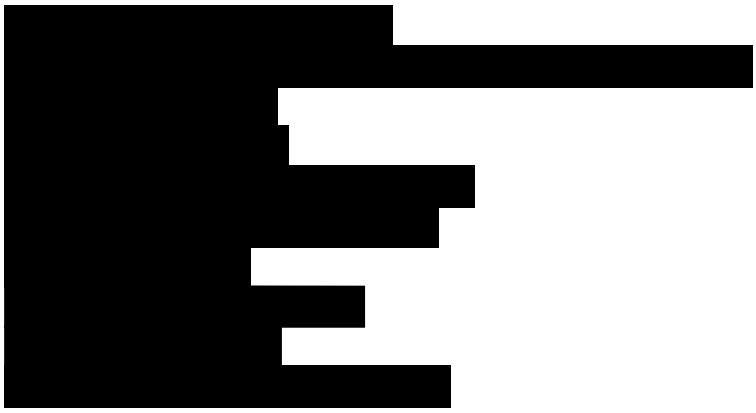
6.7.2.2.11 Reporting of Investigational Medicinal Product Quality Complaints

Any defect or possible defect in an investigational drug product (defined as a pharmaceutical form of an active substance or placebo being tested or used as a reference in a clinical trial) must be reported by the investigator or qualified designee to the sponsor **within 24 hours** of first becoming aware of the possible defect. This report to the sponsor may be made by telephone to the designated sponsor representative. The product and packaging components in question, if available must be stored in a secure area under specified storage conditions until it is determined whether the product is required to be returned for investigation of the defect. If the product complaint is associated with an SAE, the SAE must be reported separately in accordance with the protocol, and the SAE report should mention the product quality complaint.

6.7.2.3 Guidelines and Management of Special Situations

The principal investigator(s) is responsible for ensuring that procedures and expertise are available to handle medical situations including emergencies during the study. A medical emergency usually constitutes an SAE and should be reported as such (**Section 6.7.2.2**).

The Sponsor Representative/Medical Director may be contacted with any questions/concerns:



Management of Coronary Artery Bypass Surgery (CABG)

If the investigator determines that a subject requires CABG, the subject should be treated as if they had received PZ-128. Pre-surgical prep and treatment (e.g., administration of blood products) and the timing of the CABG should be based on a sufficient washout of 48 hours. The need for CABG should not require unblinding of study drug. However, in the event that it becomes medically necessary to unblind the subject, the procedures are outlined in **Section 6.4.6**.

Management of Bleeding Events

Bleeding complications should be managed according to usual clinical practice and may include the administration of blood products. While high levels of platelet inhibition are achieved, especially after the IV infusion of PZ-128, the effects of the drug are readily reversible and decline with declining blood concentrations. After a single dose with 0.3 or 0.5

mg/kg IV administration, the effects are ≥50% recovered within about 24 hours. If a clinically significant bleeding event occurs, the following data should be obtained immediately:

- Complete blood count
- Platelet count
- Prothrombin time (PT) / INR
- Activated partial thromboplastin time (aPTT) for bleeding not associated with PCI, or peak ACT (if available) for bleeding associated with PCI

Transfusions of blood products (packed red blood cells, whole blood, platelets, fresh frozen plasma, cryoprecipitate, etc.) may be necessary for subjects with clinically significant bleeding events and these decisions are left to the discretion of the investigator. The type of blood, number of units, site of bleeding (if any), and hemoglobin value at the time of transfusion will be recorded on the case report form.

Management of Drug Allergic Reactions

[redacted]

Anaphylaxis precautions must be observed during PZ-128/placebo administration and for at least 1 hour following the completion of the infusion. Medical equipment and supplies needed for resuscitation (pharmacologic agents including epinephrine, anti-histamines, corticosteroids, intravenous fluids, and aerosolized bronchodilators, oxygen, devices for intubation and mechanical ventilation, tracheostomy equipment, and a defibrillator) will be readily available in the treatment area where the study drug is administered. An anesthesiologist will be available 24 hours/day at the facility for assistance with intubation, mechanical ventilation and advanced life support.

Subjects will be observed carefully for possible infusion reactions and managed appropriately if indicated. The investigator may use his/her discretion in determining the regimen to be used according to standard clinical practice and tailored to the specific clinical situation (i.e., symptoms and severity of the reaction and responsiveness). Management may include, but is not limited to the following:

- Interruption and/or discontinuation of the study drug infusion
- Antihistamine(s) (e.g., Diphenhydramine (H1), Ranitidine (H2))
- Corticosteroid (s) (e.g., Methylprednisolone)
- Brochodilator(s) (e.g., Albuterol)
- Adrenergic Agonist(s) (e.g., Epinephrine)
- Inotropic Agent(s) (e.g., Glucagon)
- Vasopressor(s) (e.g., Dopamine)
- Intubation

- IV fluids
- Oxygen

Mild to moderate symptoms include localized skin reactions (pruritus, flushing, rash) and usually can be controlled with symptom management (e.g. administration of antihistamines and/or corticosteroids) and if necessary, temporary interruption of the study drug infusion. The study drug infusion may be interrupted and/or restarted at the discretion of the investigator depending upon the type and severity of the reaction and the responsiveness to the symptom management and/or the brief (no longer than 1 hour) interruption of the infusion. Any recurrence of symptoms following an initial improvement and/or following drug re-challenge is cause for discontinuation of the study drug.

Severe symptoms, such as clinically significant hypertension or hypotension, dyspnea, bronchospasm, angioedema or generalized urticaria require immediate discontinuation of the study drug and aggressive symptom management. The criteria for the diagnosis of Anaphylaxis [48] is attached in **Appendix C**. Anaphylaxis is a medical emergency that requires prompt recognition and immediate discontinuation of the study drug and intervention/treatment. A subject who experiences anaphylaxis will not be re-challenged with the study drug.

Prompt and accurate documentation of any allergic reaction should include the following:

- Initial symptoms and course of progression
- The timing of symptom onset
- Intervention, timing and subject response
- Time of symptom resolution
- Discharge instructions (e.g., follow-up plans, prescribed medications, appropriate allergy referral, etc.) or transfer to emergency services.

The disposition and follow-up of subjects with an allergic reaction depends on the severity of the initial reaction, the response to treatment and the clinical judgment of the investigator and medical staff. Subjects with non-life-threatening symptoms should be observed for 4-6 hours after successful treatment (resolution of symptoms) if discharge is being considered. Subjects who have severe reactions/anaphylaxis with cardiovascular and/or respiratory symptoms or refractory symptoms should be admitted or treated and observed for a longer prior of time especially if the condition is not promptly resolved by therapy. Other high-risk groups include those on beta-blocker therapy who have significant reactions and those with severe late-phase or prolonged reaction. Some subjects who have problematic social situations (e.g., no phone) or live far from medical care may also require admission or prolonged observation.

Subjects experiencing any allergic reaction should be provided with education regarding risk with future exposure to agents that might initiate an allergic response and precautions at the time of discharge including instructions for seeking emergency medical care upon recurrence

of signs/symptoms.

Need for Administration of Prohibited Medications
There are no anticipated prohibitive medications in the case of a medical emergency.

6.7.3 Other Assessments

6.7.3.1 Optional Sampling for Pharmacogenomics (DNA and RNA) Analysis

Platelets, endothelium and atherosclerotic plaques express and secrete a large number of diverse pro-thrombotic and anti-thrombotic components. Prothrombotic and hemorrhagic risk may be influenced in large part by the individual's genetic make-up and accordingly, the study will explore whether DNA polymorphisms (specific DNA sequences that differ among individuals), RNA levels and microRNAs correlate with the expression of genes involved in thrombosis, inflammation, and cardiometabolic diseases and whether these genetic components correlate with PZ-128 clinical outcomes in subjects providing informed consent for the whole blood collection. A number of CAD-related genetic loci have been recently identified (n=198), but it is essentially unknown how these genetic variants are related to cardiovascular outcomes in PCI/ACS patients or whether the variants influence platelet reactivity and thrombotic risk. In particular, the newly identified PAR1 agonist, MMP-1 is significantly elevated in plasma from patients following acute myocardial infarction. A commonly occurring MMP-1 promoter polymorphism will be assessed for increased risk of myocardial infarction in subjects with high promoter activity haplotypes versus significantly decreased risk in subjects with low promoter activity haplotypes. It has also been suggested that polymorphisms in the PAR4 gene region may be associated with platelet reactivity and cardiovascular outcomes. Therefore, in an effort to determine if DNA sequence variability in PAR4 is associated with disease status and response to PZ-128, genotype for single nucleotide polymorphisms (SNPs) in PAR4 will be determined. In addition, genome-wide association studies may be performed to identify genetic factors predisposing subjects to cardiometabolic diseases which are associated with the population under study in this trial (i.e., obesity, lipid metabolism, CVD, diabetes).

In all cases of genetic analysis of DNA and gene expression analysis from the whole blood samples provided by the subjects participating in the TRIP-PCI study, strict confidentiality will be maintained. No subject names or initials will be associated with the whole blood, the derived samples (DNA, RNA, or microRNA) or the corresponding laboratory research data. More information appears in **Appendix E.**

6.7.3.2 Optional Sampling for Pharmacokinetics, Pharmacodynamics (Platelet Aggregation), Exploratory Biomarkers

Thrombin is the major protease in the coagulation cascade. Its pleiotropic activity can ultimately lead to thrombosis and tissue injury. Thrombin also has numerous direct effects on vascular cells and is the most potent activator of platelets identified to date. The purpose of this optional sampling is to gather data to further investigate the effects of PAR1 blockade on

platelet and other vascular reactivities and to continue to characterize the pharmacokinetic profile.

6.7.4 Criteria for Termination of the Trial

The DSMB (**see Section 7.8 and Section 9.3.1**) will retain responsibility for recommending early termination of the study to the sponsor, who will have ultimate authority/responsibility for making the decision. The criteria that the DSMB will follow to determine when to terminate the study will be described in the separate DSMB charter.

7.0 STATISTICAL AND ANALYTICAL PLAN

7.1 Study Design

This is a multi-center, prospective, randomized, double-blind, placebo-controlled, balanced-parallel-groups study to evaluate the safety of PZ-128 compared with placebo, given in addition to standard of care, in subjects undergoing non-emergent PCI or non-emergent cardiac catheterization with the intention to perform PCI.

Prior to coronary angiography, approximately 600 subjects are expected to be randomized in a 2:2:1:1 parallel, double-blind fashion to a single, 2-hour continuous intravenous infusion of either: PZ-128 0.3 mg/kg, PZ-128 0.5 mg/kg, or Placebo 0.3 mg/kg, or Placebo 0.5 mg/kg (the two placebo groups will be merged for the purposes of data analysis). The study drug infusion will be initiated within one hour prior to the start of the cardiac catheterization procedure (i.e., angiography).

The planned administration of GPI for a subject excludes that subject from participation in this study. During PCI, bail-out GPI may be used, at the investigator's discretion, in subjects with new ischemia on telemetry or ECG, worsening of angiographic coronary flow during procedure, or thrombotic complication related to stenting during PCI.

Subjects meeting the inclusion/exclusion criteria (**Section 6.3.1 and Section 6.3.2**) will follow the randomization procedures (**Section 6.4.4 and Section 6.5.2)**. The randomization code and starting seed will be generated and maintained by RTI. It will not be provided to the sponsor, or sites until the database has been locked with the exceptions of parties listed in **Section 6.4.6**. The randomization will be generated using random permuted blocks, stratified by clinical site according to the urgency of the procedure at the time the operator decides to perform the cardiac catheterization (i.e., elective versus urgent).

7.2 General Aspects of Statistical Evaluation

Statistical analyses will be performed using SAS version 9.23 and, where appropriate (e.g., specific pharmacokinetic analyses) other validated software. Statistical analyses will be performed in accordance with the study protocol, and a comprehensive Statistical Analysis Plan (SAP) which will be prepared before unmasking of the data. The SAP will provide further details of the analyses and presentation of the data and will contain ground rules and data handling conventions used to perform the analyses. Both the data and the proposed methods of analyses will be reviewed continuously during the study by the study team, which includes input from statistical, clinical and data management personnel. Before database lock, a masked review will take place during which all important protocol deviations will be discussed and agreed by the team and any ambiguities in study outcomes will be adjudicated in a masked fashion by the protocol team.

The primary and secondary outcome measures will be treated as binary outcomes with any individual experiencing the event within the specified time frame after treatment classified as

positive for the outcome and other individuals classified as negative. Details of the measures will be included in the SAP.

7.3 Data Sets

Safety analysis set
Safety evaluations will include subjects who have been randomized and who have received any amount of study drug treatment. Subjects will be evaluated according to treatment actually received. The safety analysis set will be used as the primary analysis dataset for the reporting of both the primary and secondary safety analyses as described below.

Intention-to-treat analysis set
Efficacy analyses will be carried out on an intent-to-treat basis, and all evaluations will include all subjects who receive randomization assignment, irrespective of initiation of treatment, who provide at least one outcome assessment. Subjects will be analyzed according to the assigned study treatment irrespective of any treatment allocation errors.

Per-protocol analysis sets
Per-protocol analysis sets will also be defined during the masked review for total bleeding events, and key efficacy outcome measures. Subjects will be excluded from the per protocol analysis sets as a result of protocol deviations likely to influence the outcome variables (exact details of possible protocol deviations will be detailed in the SAP). Similar conclusions from both the safety analysis set and per protocol analysis set are required for a robust interpretation of the bleeding event results.

7.4 Demographic and Other Baseline Characteristics

Demographic variables (e.g., sex, race, age, weight) and baseline clinical characteristics will be summarized by treatment group to assess comparability of treatment groups. No formal statistical analyses of these data comparing treatment arms are planned.

7.5 Safety Analyses

All safety endpoints for bleeding are defined in **Appendix B**. Only bleeding events adjudicated by the Clinical Events Committee (CEC) will be used in the primary and secondary analyses.

7.5.1 Primary Endpoint

The overall objective of this study is to evaluate the safety of two dose levels of PZ-128 compared to placebo. The outcome used to assess the safety of PZ-128 will be the incidence of major and minor bleeding events, as assessed by the TIMI system of classification, when PZ-128 or placebo is added to the standard of care (dual anti-platelet therapy with aspirin and $P2Y_{12}$ inhibitor) in subjects undergoing non-emergent PCI or non-emergent cardiac catheterization with the intent to perform PCI.

The primary safety outcome variable is the incidence of TIMI major plus minor bleeding not related to CABG through 30 days after treatment.

7.5.2 Secondary Safety Endpoints

The secondary safety outcome variables will be the following:

- incidence of bleeding that does not meet the TIMI criteria for major or minor (i.e., minimal bleeding) through 30 days after treatment;
- incidence of "clinically significant" bleeding through 30 days after treatment (TIMI major or minor or bleeding requiring medical attention);
- incidence of TIMI bleeding related to CABG through 30 days after treatment;

7.5.3 Exploratory Safety Endpoints

The following exploratory safety endpoint is not part of the objectives of the trial and is included as an additional way to evaluate bleeding for future consideration:

- incidence of bleeding events according to the BARC (Bleeding Academic Research Consortium) classification through 30 days after treatment.

7.5.4 Methodology

The primary objective of this study is to evaluate the safety of two dose levels of PZ-128 compared to placebo as measured by the incidence of TIMI major plus minor bleeding not related to CABG through 30 days after treatment. Specifically, the study will is designed to test the hypothesis that from a safety perspective, PZ-128 is non-inferior to placebo, where non-inferiority is demonstrating that the difference in risk of bleeding incidence between the two treatments is less than a specified clinically important margin; the acceptable safety margin will depend in part on the efficacy benefit found for PZ-128. Specifically, the analysis of the bleeding outcome variables will assess the risks of bleeding on the two treatment regimens by generating point and one-sided 95% confidence interval estimates of the risk difference using the following formula:

$$\text{Risk Difference} = \pi\text{PZ-128} - \pi\text{Pbo}$$

where πPZ-128 is the bleeding incidence across the two PZ-128 arms and πPbo is the bleeding incidence for the placebo arm. The point and interval estimates for this risk difference will be obtained using a generalized linear model with the binomial distribution assumption and an identity link. To address the potential underlying risk differences associated subpopulations defined by underlying subject risk and whether or not the subject received CABG, those factors will be included in the model. The robust variance estimator as defined by Liang and Zeger [49] will be used to generate the interval estimates. Comparable models will be used to generate estimates for both the primary outcome and the secondary outcomes defined above.

Adverse Events
National Cancer Institute Common Terminology Criteria for Adverse Events (NCI CTCAE) version 4.0 will be used for AE/SAE reporting in the trial database. This version of CTCAE is MedDRA v12.0 (Medical Dictionary for Regulatory Activities Terminology) compatible at the AE (Adverse Event) term level where each CTCAE term is a MedDRA LLT (Lowest Level Term). All adverse events will be captured from the time of randomization up until 30 days after study drug administration and all serious adverse events will be captured from the time of randomization up until 90 days after study drug administration. AEs will be summarized by system organ class and preferred term using MedDRA, with summaries reported separately for the 3 treatment arms aggregated across all participants in those treatment arms, and stratified by the four risk groups defined by the cross-classification of whether subjects received CABG or not and the urgency of the procedure at the time the operator decides to perform the cardiac catheterization (i.e., elective versus urgent). All adverse event data will be listed for all subjects. Separate listings of all SAEs, deaths and discontinuations due to AEs will be presented. Adverse event data will be summarized by relationship and severity. All AEs relating to bleeding will be summarized separately.

Laboratory data
All laboratory data will be listed and absolute values outside the normal range will be flagged if applicable. Changes from pre-dose baseline at Screening may also be summarized. Numerical laboratory data will be summarized using standard summary statistics and will be presented by treatment group.

Other safety data
Physical examination, ECG, and vital signs data will be listed and summarized using standard summary statistics by treatment group.

7.6 Efficacy Analyses

Efficacy endpoints are defined in **Appendix B**. Only efficacy endpoint events adjudicated by the CEC will be used in the secondary efficacy analyses.

7.6.1 Secondary Efficacy Endpoints

While the primary objective of this study is to determine the safety of PZ-128, secondary analyses will be used to evaluate potential efficacy signals that will allow an assessment of the comparative risk/benefit calculus of the treatment in determination of a future Phase 3 study.

Specific secondary efficacy outcome measures include:

- incidence of the composite endpoint of major adverse cardiac events (MACE)- any of cardiovascular death, non-fatal myocardial infarction, non-fatal stroke, recurrent ischemia requiring hospitalization or urgent coronary revascularization with either CABG or subsequent PCI evaluated through 30 days and 90 days after treatment;

- incidence of the individual components of the MACE composite as measures of potential clinical benefit evaluated through 30 days and 90 days after treatment:
    - cardiovascular death
    - non-fatal MI
    - non-fatal stroke
    - recurrent ischemia requiring hospitalization
    - urgent coronary revascularization
- incidence of definite or probable stent thrombosis evaluated through 30 days and 90 days after treatment;
- effect of the investigator choice of $P2Y_{12}$ inhibitor (i.e., clopidogrel versus prasugrel/ticagrelor) on the individual components of the MACE composite measure through 30 days and 90 days post treatment.

7.6.2 Methodology

The proposed phase 2 study is not of sufficient size or of sufficient follow-up duration to allow analysis of clinical event rates.

Two alternative approaches will be used to evaluate the differences in risk of the various secondary outcome measures. The first will utilize generalized linear models analogous to those used for the safety outcomes. In addition because each of these outcome measures can be evaluated using a time to event approach, survival analyses will be used to examine those outcomes. Kaplan-Meier estimates of the cumulative risk of each outcome will be calculated to investigate differences between the treatment groups in the time to occurrence, and a Cox Proportional hazard model that accounts for the risk stratification variables described above will be used to assess risk differences in the 3 treatment groups. All of these analyses will be interpreted descriptively, with no adjustment for multiple comparisons. We anticipate that some analyses will be conducted for specific subgroups (e.g., influence of baseline risk factors, concomitant therapies), but these subgroup analyses will again be considered exploratory and treated as descriptive analyses. Details will be provided in the SAP. Generalized linear models comparable to those used for the safety outcomes will also be used to evaluate the effect of investigator choice of $P2Y_{12}$ inhibitor on each of the MACE components.

7.7 Determination of Sample Size/Power

Power calculations were used to evaluate the size of non-inferiority margins that were able to be detected from a study of 600 participants, 400 of whom are randomized to PZ-128 and 200 of whom are randomized to placebo. Available data on bleeding rates from previous studies [29, 32, 33] suggest that the incidence of bleeding in the placebo arm is likely to be in the range of 2% to 6%. Furthermore, the sponsor assumes that the true difference in the incidence

of the participants on the PZ-128 arms and those in the placebo arms are likely to be in the range of 0% to 2%. Based on these assumptions, the table below shows the minimum non-inferiority margin that could be demonstrated with at least 80% power, where that non-inferiority margin is based on the upper limit of the 95% confidence interval identified above. This sample size is adequate to demonstrate acceptable upper safety bounds.

True Incidence or Risk of Bleeding for placebo	Minimum Demonstrable Non-inferiority margin	
	True Incidence Difference of 0%	True Incidence Difference of 2%
2%	2.6%	5.0%
3%	3.2%	5.5%
4%	3.7%	5.9 %
5%	4.1%	6.3%
6%	4.4%	6.6%

7.8 Interim Analysis

Although no formal stopping boundaries will be utilized, one interim safety analysis will be performed at approximately 1 year following the enrollment of the first subject, or after enrollment of 50% of the study population, whichever occurs first. The Data and Safety Monitoring Board (DSMB) will be provided point and interval estimates of PZ-128 benefit at the time of that analysis based on survival estimates, and will evaluate evidence of bleeding risk in light of the potential efficacy benefits in making recommendations about stopping the trial.

An independent DSMB, as described in **Section 9.3.1**, will monitor safety data on an ongoing basis to ensure that subject safety is not compromised. The DSMB-supporting statistician will have access to the randomization code to allow grouping of the safety data. The DSMB will receive blinded data but has the option to unblind the data if deemed warranted. The DSMB will make safety recommendations for the alteration or termination of the study to the study sponsor.

7.9 Other Analyses

7.9.1 Pharmacokinetics

The plasma portion of the blood samples from each consenting participant will be assayed at a centralized lab for PZ-128 under non-GLP conditions at baseline and select time points following the start of the study drug administration as specified in **Section 6.6.3.** PK parameters will be calculated over time (C_{max}, AUC, $t_{1/2}$, Clearance and Distribution) and will be summarized by dose level. Data will be presented using graphical summaries and descriptive statistics.

7.9.2 Pharmacodynamics

At select sites, blood samples from each consenting participant will be analyzed onsite using Light Transmission Aggregometry (LTA) under non-GLP conditions at baseline and select time points following the start of the study drug administration as specified in **Section 6.6.3**.

The following will be measured:

- Inhibition of platelet aggregation (maximum and final) induced by SFLLRN 5-20 μM (or SFLLRN of equivalent potency) using blood collected in 0.4% sodium citrate.
- Inhibition of platelet aggregation (maximum and final) induced by AYPGKF 160 μM using blood collected in 0.4% sodium citrate.
- Inhibition of platelet aggregation (maximum and final) induced by collagen 4-20 μg/mL using blood collected in 0.4% sodium citrate.
- Inhibition of platelet aggregation (maximum and final) induced by 5-20 μM ADP using blood collected in 0.4% sodium citrate.
- Inhibition of platelet aggregation (maximum and final) induced by thrombin 1 nM-1 μM using blood collected in 0.4% sodium citrate (with GPRP added for 1mM final concentration).

Maximal and final extent inhibition of platelet aggregation (IPA) from pre-dose baseline at Screening will be calculated at all subsequent time points using

$$\text{Percentage of Inhibition} = 100\% \times \frac{(PAs - PA)}{(PAs)}$$

Where PA is the mean response at the given time point, and PAs is the mean response at pre-dose baseline (Screening). Percentage inhibition will be restricted to the closed interval [0,100]; any data falling outside this range will be truncated to the appropriate limit. Individual peak inhibition of platelet aggregation (IPA_{max}) will be estimated as the highest IPA within the collection time period and $TIPA_{max}$ will be the time to IPA_{max} for final extent and maximum extent of IPA.

The percentage inhibition of the specific agonist-induced platelet aggregation (final and maximum extent) will be summarized at the scheduled protocol time points using standard summary statistics for each treatment group. Mean plots showing changes within treatment groups across time will be presented. The derived PK and PD parameters will be summarized using standard summary statistics and graphical presentations.

7.9.3 Exploratory Biomarkers

The plasma portion of the blood samples from each consenting participant will be assayed at a centralized lab under non-GLP conditions for systemic biomarkers of platelet reactivity, inflammation and metabolomics at baseline and at select time points following the start of the study drug administration as specified in **Section 6.6.3**. This will include two PAR1 agonists,

thrombin (in complex with anti-thrombin III: TAT) and MMP-1. Changes from the pre-dose baseline (Screening) to each of the subsequent time points will be compared between treatment groups using descriptive statistics and correlated with outcomes.

8.0 ADHERENCE TO ETHICAL, REGULATORY, AND ADMINISTRATIVE CONSIDERATIONS

The ethical and regulatory requirements that must be observed to comply with Principles of Good Clinical Practice (GCP) for the conduct and monitoring of clinical investigations are presented in this section and **Appendix D**, General Requirements for Clinical Trials.

The study must be conducted in accordance with GCP as outlined in the International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) Guidelines, E6 Good Clinical Practice: Consolidated Guidance and other applicable laws and regulations. In addition, the study must be conducted in accordance with the USA Code of Federal Regulations (CFR) since the study is conducted under a USA IND (Investigational New Drug Application).

By signing this protocol (**Appendix A**), the investigator agrees to adhere to these requirements.

8.1 Ethical Conduct of the Study

8.1.1 Institutional Review Board

Prior to initiation of the study at any site, the study, including the protocol, informed consent form, other written subject information, and any proposed advertising material must be approved by an appropriate Institutional Review Board (IRB). The IRB must be constituted according to applicable regulatory requirements. The IRB approval must be obtained in writing, clearly identifying the trial, the documents reviewed (e.g., IB, informed consent, advertisement), the Principal Investigator's name and the date of the review/approval. In the event that the IRB requires changes in the protocol, the sponsor shall be advised and must approve the changes prior to implementation. The investigator shall not modify the study described in the protocol once finalized and after approval by the IRB without the prior written approval of the sponsor. A copy of the written approval of the protocol and informed consent form must be received by Tufts Medical Center before recruitment of subjects into the study and shipment of investigational product to the site. The investigator or qualified designee will forward all subsequent IRB approvals to the sponsor.

Protocol amendments, except where necessary to eliminate an immediate hazard to subjects, must be made only with the prior approval of the sponsor. Agreement from the investigator must be obtained for all amendments relating to the protocol, Investigator's Brochure and informed consent document(s) issued by the sponsor. The IRB must be informed of all amendments and give written approval before implementation at the site.

The investigator will be responsible for obtaining annual IRB approval/renewal throughout the duration of the study. Copies of the IRB's continuance of approval must be sent to Tufts Medical Center. The investigator will submit a report to the IRB (with a copy to the sponsor) upon notification by the sponsor of the completion of study enrollment (study closure) and the

overall termination of the study.

All investigators will be responsible for notifying their IRB of deviations from the protocol and AEs/SAEs occurring at their site according to local procedures. All other AE/SAE reports received by the investigator from Tufts Medical Center should be processed by the site in accordance with ICH GCP Guidelines, CFR and local procedures.

8.1.2 Subject Information and Consent

The investigator is responsible for obtaining written informed consent from the subject after adequate explanation of the aims, methods, anticipated benefits, and potential risks of the study and before any protocol-specific screening procedures or any investigational products are administered. In obtaining informed consent, the information must be provided in language and terms understandable to the subject. The execution of informed consent (acquisition of informed consent and the subject's agreement or refusal) must be documented in the subject's medical records, and the informed consent form must be signed and personally dated by the subject and by the person who conducted the informed consent discussion. The original signed and dated consent form must be retained by the investigator as part of the trial records in accordance with institutional policy and a copy of the signed and dated consent form must be given to the subject.

The consent form must include all of the required elements of informed consent in accordance with ICH Guidelines E6 and the USA FDA as set forth in Title 21 CFR, Part 50. In addition, the sponsor specifically requests that the consent form identify it as the sponsor and state that use of the drug is investigational/ experimental and the side effects of the drug are not completely known. The sponsor will provide a suggested model generic informed consent document for the investigator to prepare the informed consent form to be used at his/her site. The consent form must be approved by the appropriate IRB and sponsor before study initiation at a clinical site. Updates to the template will be communicated in writing from the sponsor to the investigator. Any subsequent changes to the approved informed consent form must be reviewed and approved by the appropriate IRB and sponsor before implementation.

8.1.3 Protocol-Related Regulatory Issues

The investigator is responsible for forwarding the following documents to the sponsor for review before study initiation can occur:

- Signed and dated protocol signature page (Investigator's Agreement, **Appendix A**)
- Signed and dated Investigator's Brochure signature page
- Copy of approved/validated IRB informed consent form (and HIPAA Form if not included in the ICF), other written subject information, and any proposed advertising material
- Copy of the IRB approval letter of the protocol and informed consent form

- The IRB composition or written statement and assurance number (FWA) that IRB is in compliance with regulations
- Completed Form FDA 1572
- Up-to-date curriculum vitae (signed and dated within one year) and medical license number of Principal Investigator and all Sub-Investigators listed on the Form FDA 1572, if applicable
- Laboratory normal ranges and documentation of laboratory certification (or equivalent license) for clinical labs
- Completed Financial Disclosure statements for the Principal Investigator and all Sub-Investigators listed on Form FDA 1572
- Delegation of Authority (signed study staff log)
- Signed study contract

8.2 Reporting to Sponsor and Subject Confidentiality

Contact with the investigator will be maintained by the sponsor and sponsor's monitor who will visit the investigator at the initiation and closure of the study and at periodic intervals during the study to assess the conduct of the study.

The investigator must ensure that the subject's confidentiality is maintained:

- On the eCRFs or other documents submitted to the sponsor/sponsor's designee, subjects should be identified by their initials and a subject identification number only, with a complete and accurate date of birth on the demographics eCRF.
- Documents that are not for submission to the sponsor (e.g., signed informed consent forms) should be kept in strict confidence by the investigator.
- All optional pharmacokinetic, pharmacodynamics, exploratory biomarkers research and pharmacogenomics blood samples (for subjects who sign separate consent) submitted to the sponsor should be identified by the subject's unique study number only and the collection date/time. All results will be coded by the sponsor/sponsor's designee with the subject's unique study number (or a random genetics sample code in the case of the pharmacogenomics) and stored in a secure database to ensure confidentiality while enabling destruction of the samples upon completion of the study's objectives or the subject's request (i.e., withdrawal of consent for further analysis). Since the research laboratory evaluations are not expected to benefit the subject directly or to impact medical management, the results will not be individually reported back to the investigator, will not be placed in the subject's medical record and will not be made available to the subject or third parties (e.g., medical providers). See **Appendix E** for information relating to pharmacogenomics.

In compliance with Federal regulations/ICH GCP Guidelines, it is required that the investigator and institution permit authorized representatives of the sponsor, of the regulatory agency(s), and the IRB direct access to review the subject's original medical records for

verification of study-related procedures and data. Direct access includes examining, analyzing, verifying and reproducing any records and reports that are important to the evaluation of the study. The investigator is obligated to inform and obtain the consent of the subject to permit named representatives to have access to his/her study-related records, including personal health information, without violating the confidentiality of the subject.

An entire eCRF will be completed for all subjects who give informed consent and undergo successful randomization (i.e., considered "enrolled" by the sponsor), whereas subjects who give informed consent but do not subsequently undergo successful randomization will be registered in the interactive web-based system as screen failures and will have the following information completed: 1) demographics (date of birth, sex/gender, race, ethnicity) and 2) subject status including the reason for failure to be randomized. Any other institutional screening/enrollment logs used by the clinical site staff should be securely maintained and any identifying fields should be removed if the log is accessible to the sponsor/sponsor's monitor.

All eCRF screens should be completed soon after the evaluation has occurred.

It is essential that all dates appearing on the sponsor's subject data collection forms for laboratory tests, evaluations, etc. be the dates on which the samples/specimens were obtained, or the procedures performed. The investigator will acknowledge in writing that he/she has verified the accuracy of the recorded data.

Further details are provided in **Appendices D and E.**

8.3 Publications and Other Rights

A. The investigator has the right to publish or publicly present the results of the study in accordance with this **Section 8.3** of the protocol. Since this protocol is a part of a multicenter study, it is understood that it is the intent of the sponsor and the investigator to initially only publish or present the study results together with the other centers, unless specific written permission is obtained in advance from the sponsor to publish separate results. The sponsor shall advise as to the implications of timing of any publication in the event clinical trials are still in progress at centers other than the investigator's center.

   The investigator agrees not to publish or publicly present any interim results of the study without the prior written consent of the sponsor. The investigator further agrees to provide to the sponsor 60 days prior to submission for publication or presentation, review copies of abstracts or manuscripts for publication (including, without limitation, slides and texts of oral or other public presentations and texts of any transmission through any electronic media (e.g., any computer access system such as the Internet) that report any results of the study. The sponsor shall have the right to review and comment with respect to publications, abstracts, slides, and manuscripts and the right to review and comment on the data analysis and presentation with regard to (1) proprietary information that is protected by the provisions contained in paragraph B below, (2) the accuracy of the information contained in the publication, and (3) to ensure that the presentation is fairly balanced and

in compliance with FDA regulations.

If the parties disagree concerning the appropriateness of the data analysis and presentation, and/or confidentiality of the sponsor's confidential information, the investigator agrees to meet with the sponsor's representatives at the clinical site or as otherwise agreed, prior to submission for publication, for the purpose of making good faith efforts to discuss and resolve any such issues or disagreement.

B. No publication or manuscript shall contain any trade secret information of the sponsor or any proprietary or confidential information of the sponsor and shall be confined to new discoveries and interpretations of scientific fact. If the sponsor believes there is patentable subject matter contained in any publication or manuscript submitted for review, the sponsor shall promptly identify such subject matter to the investigator. If the sponsor requests and at the sponsor's expense, the investigator shall use its best efforts to assist the sponsor to file a patent application covering such subject matter with the USA Patent and Trademark Office or through the Patent Cooperation Treaty prior to any publication.

C. The investigator is granted the right, subject to the provisions of this protocol, to use the results of all work provided by the investigator under this protocol, including but not limited to, the results of tests and any raw data and statistical data generated for the investigator's own teaching, research, and publication purposes only. The investigator/institution agrees, on behalf of itself and its employees, officers, trustees, and agents, not to cause said results to be knowingly used for any commercial purpose whatsoever except as authorized by the sponsor in writing.

9.0 INVESTIGATORS AND STUDY ADMINISTRATIVE STRUCTURE

9.1 Sponsor

The sponsor of this study is indicated in the Title Page.

9.2 Investigators

The investigators of this study are indicated in the Title Page.

Only investigators qualified by training and experience in interventional cardiology to perform a clinical investigation with PZ-128 in subjects undergoing cardiac catheterization/PCI are selected. The sponsor will contact and select all investigators (i.e., the legally responsible party(s) at each clinical site). It is the responsibility of each study-site investigator to select appropriately qualified persons to whom he/she will delegate study duties and document this on the study-specific Delegation of Authority Form. The investigator will provide the current study protocol and other relevant study documents to all sub-investigators and other staff responsible for study conduct, as well as provide for the training of all sub-investigators or other staff involved in the conduct of this research.

Both the sponsor and the investigator reserve the right to terminate the investigator's participation in the study according to the study contract/agreement.

A clinical study report (CSR) will be prepared by the sponsor or its qualified designee to describe the results of the study. One of the investigators shall be selected by the sponsor to review the CSR and provide approval of the final CSR in writing. The investigator chosen to review and approve the CSR is to be called the CSR Coordinating Investigator. A second investigator shall be selected as the Alternate CSR Coordinating Investigator. The Alternate CSR Coordinating Investigator is to review and approve the CSR should the first CSR Coordinating Investigator be unable to do so. The sponsor is to select the CSR Coordinating Investigator and Alternate CSR Coordinating Investigator from the investigators using the following criteria:

- must be the Principal Investigator at a clinical study site actively enrolling subjects and participating in the study;
- must be willing and capable of completing the necessary reviews and providing approval of the CSR in writing; and
- must have participated in clinical research prior to participating in this current study.

9.3 Committees and Central Organizations

Addresses of central organizations will be identified on Forms FDA 1571.

9.3.1 Data and Safety Monitoring Board (DSMB)

An independent DSMB will be established to further the rights, safety and well-being of subjects who will be participating in this study by monitoring the progress and results. The DSMB will review in an aggregated and blinded fashion, with the option to unblind these data, if deemed warranted:

- bleeding and associated data (aggregated) through 30 days, primarily to determine whether the type and occurrence of bleeding is in accord with anticipated results (i.e., no potential meaningful additional risk), and
- MACE and stent thrombosis results.

Reviewed data will not have been adjudicated by the CEC (see **Section 9.3.2**). The DSMB will monitor SAEs and aggregated AEs and may also elect to review data on platelet aggregation, if deemed necessary. The DSMB will perform one interim safety analysis to evaluate evidence of bleeding risk in light of any potential efficacy benefit(s) and to allow the DSMB to make recommendations to the sponsor (i.e., continuing the study under the current protocol, amending the current protocol, or stopping the study). Otherwise, the DSMB will meet quarterly or as required to perform its function. The DSMB may request an unplanned review of all safety data if a concern arises. No formal statistical rule for stopping the trial will be defined.

The DSMB will comprise scientifically inclined clinicians who are not investigators in the study and not otherwise associated with the sponsor and an independent statistician (i.e., not from the sponsor's statistical designee, RTI and not otherwise associated with the sponsor or clinical sites). The DSMB will be described in detail in the separate DSMB Charter, which will include a description of the interim analysis to be performed, including how the interim results will be analyzed and how the interim analysis will affect the overall study. All activities of the DSMB will be documented. This documentation will include data summaries and analyses provided to the committee as well as minutes of the meetings. The unmasked documentation will remain confidential within the DSMB until the study is unblinded.

9.3.2 Clinical Events Committee (CEC)

An independent CEC will be established to review and adjudicate each suspected bleeding event, MACE (CV death, non-fatal MI, non-fatal stroke, recurrent ischemia with hospitalization, urgent coronary revascularization) and probable/definite stent thrombosis identified by investigators or triggered by predefined criteria with the appropriate supporting documentation for the event. The group will use the definitions of the endpoints of the protocol (**Appendix B**) and additional instructions for its interpretation so that the adjudication proceeds in consistent fashion overtime. The CEC will comprise physician personnel (interventional cardiologists) who are not investigators in the study and not otherwise directly associated with the sponsor. The CEC judges will remain blinded to treatment throughout the adjudication process and the study. The CEC-adjudicated data will

be used in the final analyses of certain endpoints. The CEC will be described in detail in a separate CEC Charter.

9.3.3 Central Randomization Service (Medidata Rave®)

Central randomization will be performed via an interactive web-based system within the Medidata Rave® platform which is available 24 hours a day, 7 days a week to facilitate real-time enrollment/ randomization. Investigators or designee will login to iMedidata under the specific study to enroll/randomize the subject and obtain the following: unique subject study number and randomization number. The designated unblinded pharmacist at the clinical site will login to iMedidata via a restricted access mechanism to obtain the randomized treatment assignment (active PZ-128 or placebo and dose level designation) in order to provide blinded IV solution for infusion. The system will allow the study sponsor and designated parties to track the progress of the study closely.

9.3.4 Research Triangle Institute, International (RTI)

RTI will provide intellectual services with respect to the design and operation (regulatory consulting, biostatistics, computer programming and data management) of the clinical trial.

9.3.5 Clinical Supplies Management (CSM)

CSM is the sponsor's designated partner for the labeling, packaging, storing, management of inventory of the investigational product(s) (IP) and the distribution of the IP to the approved clinical sites.

9.3.6 Central Research Lab- Tufts Center for Hemostasis and Thrombosis Research

Tufts Center for Hemostasis and Thrombosis Research at Tufts Medical Center (study sponsor) will serve as the recipient and processing/distribution center for samples for the following research tests and will perform the analysis on the samples unless otherwise indicated:

- Cardiac Troponin I (sponsor will send samples to a designated laboratory for analysis and provide results to the CEC members for adjudication purposes related to suspected MI cases)
- Exploratory Biomarkers- thrombin in complex with anti-thrombin III: TAT and MMP-1
- Pharmacokinetics (sponsor will send samples to a designated laboratory for analysis)
- Pharmacodynamics (sponsor will perform LTA only for the Tufts Medical Center clinical site)
- Pharmacogenomics- DNA, RNA and microRNA analysis (sponsor may send samples to one of the participating clinical sites to perform analysis)

All of the above-listed tests are being conducted for research purposes only. The subject's individual results will not be returned to the corresponding investigator but will be transmitted electronically to the study's research laboratory database by the sponsor.

10.0 REFERENCES

1. Gurbel PA, Bliden KP, Saucedo JF, Suarez TA, DiChiara J, Antonino MJ, Mahla E, Singla A, Herzog WR, Bassi AK, Hennebry TA, Gesheff TB, Tantry US. Bivalirudin and clopidogrel with and without eptifibatide for elective stenting: effects on platelet function, thrombelastographic indexes, and their relation to periprocedural infarction results of the CLEAR PLATELETS-2 (Clopidogrel with Eptifibatide to Arrest the Reactivity of Platelets) study. J Am Coll Cardiol 2009;53:648-57.

2. Gurbel PA, Bliden KP, Zaman KA, Yoho JA, Hayes KM, Tantry US. Clopidogrel loading with eptifibatide to arrest the reactivity of platelets: results of the Clopidogrel Loading With Eptifibatide to Arrest the Reactivity of Platelets (CLEAR PLATELETS) study. Circulation 2005;111:1153-9.

3. Gurbel PA, Bliden KP, Hayes KM, Tantry U. Platelet activation in myocardial ischemic syndromes. Expert Rev Cardiovasc Ther 2004;2:535-45.

4. Davi G, Patrono C. Platelet activation and atherothrombosis. N Engl J Med 2007;357:2482-94.

5. Smyth SS, Woulfe DS, Weitz JI, Gachet C, Conley PB, Goodman SG, Roe MT, Kuliopulos A, Moliterno DJ, French PA, Steinhubl SR, Becker RC. G-protein-coupled receptors as signaling targets for antiplatelet therapy. Arterioscler Thromb Vasc Biol 2009;29:449-57.

6. Tantry US, Etherington A, Bliden KP, Gurbel PA. Antiplatelet therapy: current strategies and future trends. Future Cardiol 2006;2:343-66.

7. Yusuf S, Zhao F, Mehta SR, Chrolavicius S, Tognoni G, Fox KK. Effects of clopidogrel in addition to aspirin in patients with acute coronary syndromes without ST-segment elevation. N Engl J Med 2001;345:494-502.

8. Mehta SR, Yusuf S, Peters RJ, Bertrand ME, Lewis BS, Natarajan MK. Effects of pretreatment with clopidogrel and aspirin followed by long-term therapy in patients undergoing percutaneous coronary intervention: the PCI-CURE study. Lancet 2001;358:527-33.

9. Bonello L, Tantry US, Marcucci R, Blindt R, Angiolillo DJ, Becker R, Bhatt DL, Cattaneo M, Collet JP, Cuisset T, et al. Consensus and future directions on the definition of high on-treatment platelet reactivity to adenosine diphosphate. J Am Coll Cardiol 2010;56:919-33.

10. Wallentin L, Becker RC, Budaj A, Cannon CP, Emanuelsson H, Held C, Horrow J, Husted S, James S, Katus H, et al. Ticagrelor versus clopidogrel in patients with acute coronary syndromes. N Engl J Med 2009;361:1045-57.

11. Wiviott SD, Braunwald E, McCabe CH, Montalescot G, Ruzyllo W, Gottlieb S, Neumann FJ, Ardissino D, De Servi S, Murphy SA, et al. Prasugrel versus clopidogrel in patients with acute coronary syndromes. N Engl J Med 2007;357:2001-15.

12. Storey RF, Bliden KP, Patil SB, Karunakaran A, Ecob R, Butler K, Teng R, Wei C, Tantry US, Gurbel PA. Incidence of dyspnea and assessment of cardiac and pulmonary function in patients with stable coronary artery disease receiving ticagrelor, clopidogrel, or placebo in the ONSET/OFFSET study. J Am Coll Cardiol 2010;56:185-93.

13. Amsterdam EA, Wenger NK, Brindis RG, Casey DE, Jr., Ganiats TG, Holmes DR, Jr., Jaffe AS, Jneid H, Kelly RF, Kontos MC, Levine GN, Liebson PR, Mukherjee D, Peterson ED, Sabatine MS, Smalling RW, Zieman SJ. 2014 AHA/ACC Guideline for the Management of Patients with Non-ST-Elevation Acute Coronary Syndromes: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines. J Am Coll Cardiol. 2014;64:e139-228.

14. Gurbel PA, Tantry US. Combination antithrombotic therapies. Circulation 2010;121:569-83.

15. Leger AJ, Covic L, Kuliopulos A. Protease-Activated Receptors and Cardiovascular Diseases. Circulation 2006;113:1070-7.

16. Jacques S, LeMasurier M, Sheridan PJ, Seeley SK, Kuliopulos A. Substrate-Assisted Catalysis of the PAR1 Thrombin Receptor: Enhancement of Macromolecular Association and Cleavage. J Biol Chem 2000;275:40671-8.

17. Seeley S, Covic L, Jacques SL, Sudmeier J, Baleja JD, Kuliopulos A. Structural Basis for Thrombin Activation of a Protease-Activated Receptor: Inhibition of Intramolecular Liganding. Chemistry & Biology 2003;10:1033-41.

18. Ossovskaya VS, Bunnett NW. Protease-activated receptors: contribution to physiology and disease. Physiol Rev 2004;84:579-621.

19. Kimmelstiel C, Zhang P, Kapur NK, Weintraub A, Krishnamurthy B, Castaneda V, Covic L, Kuliopulos A. Bivalirudin is a Dual Inhibitor of Thrombin and Collagen-Dependent Platelet Activation in Patients Undergoing Percutaneous Coronary Intervention. Circ Cardiovasc Interv 2011;4:171-9.

20. Patterson C, Stouffer GA, Madamanchi N, Runge MS. New tricks for old dogs: nonthrombotic effects of thrombin in vessel wall biology. Circ Res 2001;88:987-97.

21. Smyth SS, McEver RP, Weyrich AS, Morrell CN, Hoffman MR, Arepally GM, French PA, Dauerman HL, Becker RC. Platelet functions beyond hemostasis. J Thromb Haemost 2009;7:1759-66.

22. Derian CK, Damiano BP, Addo MF, Darrow AL, D'Andrea MR, Nedelman M, Zhang HC, Maryanoff BE, Andrade-Gordon P. Blockade of the thrombin receptor protease-activated receptor-1 with a small-molecule antagonist prevents thrombus formation and vascular occlusion in nonhuman primates. J Pharmacol Exp Ther 2003;304:855-61.

23. Kato Y, Kita Y, Hirasawa-Taniyama Y, Nishio M, Mihara K, Ito K, Yamanaka T, Seki J, Miyata S, Mutoh S. Inhibition of arterial thrombosis by a protease-activated receptor 1 antagonist, FR171113, in the guinea pig. Eur J Pharmacol 2003;473:163-9.

24. Jennings LK. Mechanisms of platelet activation: need for new strategies to protect against platelet-mediated atherothrombosis. Thromb Haemost 2009;102:248-57.

25. Chackalamannil, S, Wang, Y, Greenlee, WJ, Hu Z, Xia Y, Ahn HS, Boykow G, Hsieh Y, Palamanda J, Agans-Fantuzzi J, Kurowski S, Graziano M, Chintala M. Discovery of a novel, orally active himbacine-based thrombin receptor antagonist (SCH 530348) with potent antiplatelet activity. J Med Chem 2008;51:3061-64.

26. TRA*CER EaSC. The Thrombin Receptor Antagonist for Clinical Event Reduction in Acute Coronary Syndrome (TRA*CER) trial: study design and rationale. Am Heart J 2009;158:327-34 e4.

27. Zhang C, Srinivasan Y, Arlow DH, Fung JJ, Palmer D, Zheng Y, Green HF, Pandey A, Dror RO, Shaw DE, Weis WI, Coughlin SR, Kobilka BK. High-resolution crystal structure of human protease-activated receptor 1. Nature 2012;492:387-92.

28. Kosoglou T, Reyderman L, Tiessen RG, van Vliet AA, Fales RF, Keller R, Yang B, Cutler DL. Pharmacodynamics and pharmacokinetics of the novel PAR-1 antagonist vorapaxar (formerly SCH 530348) in healthy subjects. Eur J Clin Pharmacol 2012;68:249-58.

29. Becker RC, Moliterno DJ, Jennings LK, Pieper KS, Pei J, Niederman A, Ziada KM, Berman G, Strony J, Joseph D, et al. Safety and tolerability of SCH 530348 in patients undergoing non-urgent percutaneous coronary intervention: a randomised, double-blind, placebo-controlled phase II study. Lancet 2009;373:919-28.

30. Goto S, Yamaguchi T, Ikeda Y, Kato K, Yamaguchi H, Jensen P. Safety and exploratory efficacy of the novel thrombin receptor (PAR-1) antagonist SCH 530348 for non-ST-segment elevation acute coronary syndrome. J Atheroscler Thromb 2010;17:156-64.

31. Shinohara Y, Goto S, Doi M, Jensen P. Safety of the novel protease-activated receptor-1 antagonist vorapaxar in Japanese patients with a history of ischemic stroke. J Stroke Cerebrovas Dis 2012;21:318-24.

32. Morrow, DA, Braunwald E, Bonaca MP, et al. Vorapaxar in the secondary prevention of atherothrombotic events. N Engl J Med 2012;366:1404-13.

33. Tricoci P, Huang Z, Held C, Moliterno DJ, Armstrong PW, Van de Werf F, White HD, Aylward PE, Wallentin L, Chen E, Lokhnygina Y, Pei J, Leonardi S, Rorick TL, Kilian AM, Jennings LH, Ambrosio G, Bode C, Cequier A, Cornel JH, Diaz R, Erkan A, Huber K, Hudson MP, Jiang L, Jukema JW, Lewis BS, Lincoff AM, Montalescot G, Nicolau JC, Ogawa H, Pfisterer M, Prieto JC, Ruzyllo W, Sinnaeve PR, Storey RF, Valgimigli M, Whellan DJ, Widimsky P, Strony J, Harrington RA, Mahaffey KW. Thrombin-receptor antagonist vorapaxar in acute coronary syndromes. N Engl J Med 2012;366:20-33.

34. Gurbel PA, Bliden KP, Guyer K, Cho PW, Zaman KA, Kreutz RP, Bassi AK, Tantry US. Platelet reactivity in patients and recurrent events post-stenting: results of the PREPARE POST-STENTING Study. J Am Coll Cardiol 2005;46:1820-6.

35. Covic L, Tchernychev B, Jacques S, Kuliopulos A. Pharmacology and in vivo efficacy of pepducins in hemostasis and arterial thrombosis. In: Langel U, ed. Handbook of Cell-Penetrating Peptides. 2nd ed. New York: Taylor & Francis; 2007:245-57.

36. Tressel SL, Koukos G, Tchernychev B, Jacques SL, Covic L, Kuliopulos A. 2010. Pharmacology, biodistribution, and efficacy of GPCR-based pepducins in disease models. Methods Mol Biol 2011;683: 259-75.

37. Covic L, Gresser AL, Talavera J, Swift S, Kuliopulos A. Activation and inhibition of G protein-coupled receptors by cell-penetrating membrane-tethered peptides. Proc Natl Acad Sci (USA) 2002;99:643-8.

38. Covic L, Misra M, Badar J, Singh C, Kuliopulos A. Pepducin-based intervention of thrombin receptor signaling and systemic platelet activation. Nature Med 2002;8:1161-5.

39. Kaneider NC, Agarwal A, Leger AJ, Kuliopulos A. Reversing Systemic Inflammatory Response Syndrome with Chemokine Receptor Pepducins. Nature Med 2005;11:661-5.

40. Leger A, Jacques SL, Badar J, Kaneider NC, Derian CK, Andrade-Gordon P, Covic L, Kuliopulos A. Blocking the Protease-Activated Receptor 1-4 Heterodimer in Platelet-Mediated Thrombosis. Circulation 2006;113:1244-54.

41. Kaneider NC, Leger AJ, Agarwal A, Nguyen N, Perides G, Derian C, Covic L, Kuliopulos A. Role reversal for the receptor PAR1 in sepsis-induced vascular damage. Nature Imm 2007;8:1303-12.

42. Trivedi V, Boire A, Tchernychev B, Kaneider NC, Leger AJ, O'Callaghan K, Covic L, Kuliopulos A. Platelet Matrix Metalloprotease-1 Mediates Thrombogenesis By Activating PAR1 at a Cryptic Ligand Site. Cell 2009;137:332-43.

43. Zhang P, Gruber A, Kasuda S, Kimmelstiel C, O'Callaghan K, Cox DH, Bohm A, Baleja JD, Covic L, Kuliopulos A. Suppression of arterial thrombosis without affecting hemostatic parameters with a cell-penetrating PAR1 pepducin. Circulation 2012;126:83-91.

44. O'Callaghan K, Kuliopulos A, Covic L. Turning receptors on and off with intracellular pepducins: new insights into G-protein-coupled receptor drug development. J Biol Chem 2012;287:12787-96.

45. Fontanini KB, Janz J, Looby R, Hamilton JA. Rapid binding and transmembrane diffusion of pepducins in phospholipid bilayers. Biophysical J 2010;98:278a.

46. Van Geffen, JP, Kleinegris MC, Verdoold R, Baaten CCFMJ, Cosemans JMEM, Clemetson KJ, ten Cate H, Roest M, de Laat B, Heemskerk JWM. Normal platelet activation profile in patients with peripheral arterial disease on aspirin. Thromb Res 2015;135:513-520.

47. Levine GN, Bates ER, Blankenship JC Levine GN, Bates ER, Blankenship JC, Bailey SR, Bittl JA, Cercek B, Chambers CE, Ellis SG, Guyton RA, Hollenberg SM, Khot UN, Lange RA, Mauri L, Mehran R, Moussa ID, Mukherjee D, Nallamothu BK, Ting HH. 2011 ACCF/AHA/SCAI guideline for percutaneous coronary intervention: a report of the American College of Cardiology Foundation/American Heart Association Task Force on Practice Guidelines and the Society for Cardiovascular Angiography and Interventions. Circulation 2011;124:e574–e651.

48. Sampson HA, Muñoz-Furlong A, Campbell RL, et al. Second symposium on the definition and management of anaphylaxis: summary report- Second National Institute of Allergy and Infectious Disease/Food Allergy and Anaphylaxis Network symposium. J Allergy Clin Immunol 2006;117:391-7.

49. Liang, KY, Zeger SL. Longitudinal data analysis using generalized linear models. Biometrika 1986;73:13-22.

50. Wiviott SD, Antman EM, Gibson CM, et al. Evaluation of prasugrel compared with clopidogrel in patients with acute coronary syndromes: design and rationale for the Trial to assess Improvement in Therapeutic Outcomes by optimizing platelet InhibitioN with prasugrel Thrombolysis in Myocardial Infarction 38 (TRITON-TIMI 38). Am Heart J 2006;152:627-35.

51. Morrow DA, Scirica BM, Fox KA, Berman G, Strony J, Veltri E, Bonaca MP, Fish P, McCabe CH, Braunwald E. Evaluation of a novel antiplatelet agent for secondary prevention in patients with a history of atherosclerotic disease: design and rationale for the Thrombin-Receptor Antagonist in Secondary Prevention of Atherothrombotic Ischemic Events (TRA 2 degrees P)-TIMI 50 trial. Am Heart J 2009;158:335-41 e3.

52. Mehran R, Rao SV, Bhatt DL., et al. Standardized bleeding definitions for cardiovascular clinical trials: a consensus report from the Bleeding Academic Research Consortium. Circulation 2011;123:2736-47.

53. Cannon CP, Brindis RG, Chaitman BR, et al. 2013 ACCF/AHA key data elements and definitions for measuring the clinical management and outcomes of patients with acute coronary syndromes and coronary artery disease: a report of the American College of Cardiiology Foundation/American Heart Association Task Force on Clinical Data Standards (Writing Committee to Develop Acute Coronary Syndromes and Coronary Artery Disease Clinical Data Standards). Circulation 2013;127:1052-89.

54. Hicks KA, Tcheng JE, Bozkurt B, Chaitman BR, Cutlip DE, FArb A, Fonarow GC, Jacobs JP, Jaff MR, Lichtman JH, Limacher MC, Mahaffey KW, Mehran R, Nissen SE, Smith EE, Targum SL. 2014 ACC/AHA key data elements and definitions for cardiovascular endpoint events in clinical trials: a report of the American College of Cardiology/American Heart Association Task Force on Clinical Data Standards (Writing Committee to Develop Cardiovascular Endpoints Data Standards). Circulation 2015;131:000-000.

55. Thygesen K, Alpert JS, Jaffe AS, on behalf of the Joint ESC/ACCF/AHA/WHF Task Force for the Universal Definition of Myocardial Infarction. Third universal definition of myocardial infarction. Circulation 2012;126:2020-35.

56. Cutlip DE, Windecker S, Mehran R, Boam A, Cohen DJ, van Es GA, Steg PG, Morel MA, Mauri L, Vranckx P, et al. Clinical end points in coronary stent trials: a case for standardized definitions. Circulation 2007;115:2344-51.

APPENDIX A: SIGNATURES

INVESTIGATOR'S AGREEMENT

I have read the accompanying protocol entitled, "A Multicenter, Randomized, Double-Blind, Placebo-Controlled Study to Evaluate the Safety of PZ-128 in Subjects Undergoing Non-Emergent Percutaneous Coronary Intervention- **T**hrombin **R**eceptor **I**nhibitory **P**epducin in **PCI** (TRIP-PCI) (Protocol No. TMC-PZ128-02)" and agree to abide by all provisions set forth therein.

I agree to comply with the International Conference on Harmonisation Tripartite Guideline on Good Clinical Practice and applicable FDA regulations/guidelines set forth in 21 CFR Parts 11, 50, 54, 56, and 312.

I agree to ensure that Financial Disclosure Statements will be completed by:

- me (including, if applicable, my spouse [or legal partner] and dependent children)
- my sub-investigators (including, if applicable, their spouses [or legal partners] and dependent children)

before study initiation, during the study if there are changes that affect our financial disclosure status, and for one year following the completion of the study (i.e., one year after the last subject has completed the study as specified in the protocol).

I agree to ensure that the confidential information contained in this document will not be used for any purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of Tufts Medical Center, Inc.

_____
Signature

_____	_____
Name of Principal Investigator	Date (DD Month YYYY)
_____	_____
Name of Investigational Site	Investigational Site #

SPONSOR'S AGREEMENT

I have read the accompanying protocol entitled, "A Multicenter, Randomized, Double-Blind, Placebo-Controlled Study to Evaluate the Safety of PZ-128 in Subjects Undergoing Non-Emergent Percutaneous Coronary Intervention- **T**hrombin **R**eceptor **I**nhibitory **P**epducin in **PCI** (TRIP-PCI) (Protocol No. TMC-PZ128-02)" and agree to abide by all provisions set forth therein.

I agree to comply with the International Conference on Harmonisation Tripartite Guideline on Good Clinical Practice and applicable FDA regulations/guidelines set forth in 21 CFR Parts 11, 50, 54, 56, and 312.

APPENDIX B: DEFINITIONS OF CLINICAL ENDPOINTS

An Independent Clinical Events Committee (CEC) will adjudicate each suspected bleeding and efficacy endpoint event (MACE- any of CV death, non-fatal MI, non-fatal stroke, recurrent ischemia with hospitalization, or urgent coronary revascularization; and stent thrombosis), while blinded to treatment.

SAFETY (BLEEDING)

Bleeding will be classified according to 2 separate classifications.

1. **Thrombolysis in Myocardial Infarction (TIMI)** [26, 50, 51]
   Primary and secondary endpoints

The following definitions apply to all settings outside of peri-CABG:

Major:
1. Any intracranial (ICH)*, or
2. Clinically significant overt signs of hemorrhage associated with a drop in hemoglobin (Hgb) of ≥5 g/dL (or, when Hgb is not available, an absolute drop in hematocrit (Hct) of ≥15%), or
3. Fatal bleeding (bleeding that directly results in death within 7 d)

Minor:
Any clinically significant overt sign of hemorrhage (including imaging) that is associated with a fall in Hgb of 3 to <5 g/dL (or, when Hgb is not available, a fall in Hct of 9 to <15%).

Minimal:
Clinically significant overt signs of hemorrhage associated with a drop in Hgb of <3 g/dL (or, when Hgb is not available, a fall in Hct of <9%) that did not otherwise meet criteria for minor or major bleeding.

To account for transfusions, Hgb and Hct measurements will be adjusted for any packed red blood cells (PRBCs) or whole blood given between baseline (enrollment) and post-transfusion measurements by the method of Landefeld and coworkers. A transfusion of one unit of blood will be assumed to result in an increase by 1 g/dL in Hgb or by 3% in Hct. Thus, to calculate the true change in hemoglobin or hematocrit, if there has been an intervening transfusion between 2 blood measurements, the following calculations should be performed:

Δ Hemoglobin (Hgb) = [Baseline Hgb – post-transfusion Hgb] + [number of transfused units]
Δ Hematocrit (Hct) = [Baseline Hct – post-transfusion Hct] + [number of transfused units x 3]

Bleeding events will also be classified as spontaneous or induced as defined below:

- Spontaneous: any bleeding which there is no relation to any cause or reason.
- Induced: any bleeding that is precipitated by a cause or cause reason.

Bleeding requiring medical attention:
Any overt sign of hemorrhage that meets one of the following criteria and does not meet criteria for a major or minor bleeding event, as defined above:

- Requiring intervention (medical practitioner-guided medical or surgical treatment to stop or treat bleeding, including temporarily or permanently discontinuing or changing the dose of a medication or study drug)
- Leading to or prolonging hospitalization
- Prompting evaluation (leading to an unscheduled visit to a healthcare professional and diagnostic testing, either laboratory or imaging)

TIMI Clinically significant Bleeding:
The presence of either TIMI major or TIMI minor bleeding, or bleeding requiring medical attention.

For subjects experiencing a hemorrhage that occurs as a result of CABG, the following criteria will be used:

Major: Any hemorrhage that meets any of the following criteria:

a. Fatal bleeding (i.e., bleeding that directly results in death), or
b. Perioperative intracranial bleeding*, or
c. Reoperation following closure of the sternotomy incision for the purpose of controlling bleeding, or
d. Transfusion** of ≥5 U of whole blood or PRBCs within a 48 hour period, or
e. Chest tube output >2 L within a 24-hour period.

None: Not qualifying as a major bleed in setting of CABG.

*In light of the increased sensitivity of brain imaging for microhemorrhages of uncertain clinical significance, brain imaging with an incidental finding of microhemorrage (<10 mm evident only on gradient-echo MRI) in the absence of associated clinical symptoms/findings will not be considered to meet the protocol definition of intracranial hemorrhage.

**Cell saver transfusion will not be counted in calculations of blood products.

2. Bleeding Academic Research Consortium [52]
Exploratory endpoint

Type 0: no bleeding

Type 1: bleeding that is not actionable and does not cause the subject to seek unscheduled performance of studies, hospitalization, or treatment by a healthcare professional; may include episodes leading to self-discontinuation of medical therapy by the subject without consulting a healthcare professional

Type 2: any overt, actionable sign of hemorrhage (e.g., more bleeding than would be expected for a clinical circumstance, including bleeding found by imaging alone) that does not fit the criteria for type 3, 4, or 5 but does meet at least one of the following criteria: (1) requiring nonsurgical, medical intervention by a healthcare professional, (2) leading to hospitalization or increased level of care, or (3) prompting evaluation

Type 3
Type 3a
Overt bleeding plus hemoglobin drop of 3 to <5 g/dL* (provided hemoglobin drop is related to bleed)
Any transfusion with overt bleeding

Type 3b
Overt bleeding plus hemoglobin drop ≥5 g/dL* (provided hemoglobin drop is related to bleed)
Cardiac tamponade
Bleeding requiring surgical intervention for control (excluding dental/nasal/skin/hemorrhoid)
Bleeding requiring intravenous vasoactive agents

Type 3c
Intracranial hemorrhage (does not include microbleeds or hemorrhagic transformation, does include intraspinal)
Subcategories confirmed by autopsy or imaging or lumbar puncture
Intraocular bleed compromising vision

Type 4: CABG-related bleeding
Perioperative intracranial bleeding within 48 h
Reoperation after closure of sternotomy for the purpose of controlling bleeding
Transfusion of ≥5 U whole blood or packed red blood cells within a 48-h period†
Chest tube output ≥2L within a 24-h period

Type 5: fatal bleeding
Type 5a
Probable fatal bleeding; no autopsy or imaging confirmation but clinically suspicious

Type 5b
Definite fatal bleeding; overt bleeding or autopsy or imaging confirmation

Platelet transfusions should be recorded and reported but are not included in these definitions until further information is obtained about the relationship to outcomes. If a CABG-related bleed is not adjudicated as at least a type 3 severity event, it will be classified as not a bleeding event. If a bleeding event occurs with a clear temporal relationship to CABG (i.e., within a 48-h time frame) but does not meet type 4 severity criteria, it will be classified as not a bleeding event.

*Corrected for transfusion (1 U packed red blood cells or 1 U whole blood=1 g/dL hemoglobin).
†Cell saver products are not counted.

EFFICACY

Major Adverse Cardiac Event (MACE)
Defined as the composite endpoint of cardiovascular death, non-fatal MI, non-fatal stroke, recurrent ischemia with hospitalization, or urgent coronary revascularization.

Death [53, 54]
Cardiovascular death is the endpoint defined in this trial (as opposed to all-cause death) and only suspected CV death cases will be submitted for adjudication to the CEC. Death will be classified as cardiovascular, non-cardiovascular, or unknown/undetermined. All deaths will be assumed to be cardiovascular in nature unless a non-cardiovascular cause can be clearly shown, with the exception of death without any additional information, which will be classified as unknown/undetermined.

The primary cause of death is determined by the principal condition that caused the death, not the immediate mode of death, as per the following:

Cardiovascular death:

- Acute MI
  Death by any cardiovascular mechanism (arrhythmia, sudden death, HF, stroke, pulmonary embolus, PAD) within 30 days after an acute MI, related to the immediate consequences of the MI, such as progressive HF or recalcitrant arrhythmia. There may be assessable (attributable) mechanisms of cardiovascular death during this time period, but for simplicity, if the cardiovascular death occurs within 30 d of an acute MI, it will be considered a death due to MI.
  Note: Acute MI should be verified to the extent possible by the diagnostic criteria outlined for acute MI or by autopsy findings showing recent MI or recent coronary thrombosis. Death resulting from a procedure to treat an MI (PCI or CABG), or to treat a complication resulting from an elective coronary procedure to treat myocardial ischemia (i.e., chronic stable angina) or death due to an MI that occurs as a direct consequence of a cardiovascular investigation/procedure/operation should be considered as a death due to a cardiovascular procedure.

- Sudden cardiac death
  Death that occurs unexpectedly and not within 30 d of an acute MI.
  Note: Sudden cardiac death includes the following scenarios:
  -Death witnessed and occurring without new or worsening symptoms;
  -Death witnessed within 60 min of the onset of new or worsening cardiac symptoms unless the symptoms suggest acute MI;
  -Death witnessed and attributed to an identified arrhythmia (e.g., captured on an ECG recording, witnessed on a monitor, or unwitnessed but found on ICD review);
  -Death after unsuccessful resuscitation from cardiac arrest (e.g., ICD unresponsive sudden cardiac death, pulseless electrical activity arrest);
  -Death after successful resuscitation from cardiac arrest and without identification of a specific cardiac or noncardiac etiology;

-Unwitnessed death in a subject seen alive and clinically stable ≤24 h before being found dead without any evidence supporting a specific non-cardiovascular cause of death (information about the subject's clinical status preceding death should be provided if available)
Unless additional information suggests an alternate specific cause of death (e.g., death due to Other cardiovascular causes), if a subject is seen alive ≤24 h before being found dead, sudden cardiac death should be recorded (e.g., subject found dead in bed but who had not been seen by a family member for >24 h).

- Death due to heart failure
  Death associated with clinically worsening symptoms and/or signs of HF, regardless of etiology.
  Note: Deaths due to HF can have various etiologies, including singe or recurrent MIs, ischemic or nonischemic cardiomyopathy, hypertension, or valvular disease.

- Death due to stroke
  Death after stroke that is either a direct consequence of the stroke or a complication of the stroke.
  Note: Acute stroke should be verified to the extent possible by the diagnostic criteria outlined for stroke.

- Death due to cardiovascular procedures
  Death caused by the immediate complication(s) of a cardiovascular procedure.

- Death due to cardiovascular hemorrhage
  Death related to hemorrhage such as a nonstroke intracranial hemorrhage (e.g., subdural hematoma), nonprocedural or nontraumatic vascular rupture (e.g., aortic aneurysm), or hemorrhage causing cardiac tamponade.

- Death due to other cardiovascular causes
  Cardiovascular death not included in the above categories but with specific, known cause (e.g., PE, PAD).

Non-cardiovascular death: defined as any death with a specific cause that is not thought to be cardiovascular in nature:

- Pulmonary
- Renal
- GI
- Hepatobiliary
- Pancreatic
- Infection (includes sepsis)
- Inflammatory/immune (includes SIRS, immunological, autoimmune diseases and disorders, anaphylaxis from environmental allergies)
- Hemorrhage that is neither cardiovascular bleeding nor a stroke
- Non-cardiovascular procedure or surgery

- Trauma (includes homicide)
- Suicide
- Prescription drug reaction or overdose (includes anaphylaxis)
- Nonprescription drug reaction or overdose
- Neurological (excludes cardiovascular death from ischemic stroke, hemorrhagic stroke, or undetermined cause of stroke, or cardiovascular hemorrhage of central nervous system)
- Malignancy
- Other non-cardiovascular cause of death

Unknown/Undetermined cause of death: refers to death not attributable to either cardiovascular or noncardiovascular cause

Myocardial Infarction (MI)

Third Universal Definition of Myocardial Infarction [55]

Note: cTnI or cTnT is the preferred biomarker. If a cTnI or cTnT assay is not available, the best alternative is CK-MB (measured by mass assay). In this study, the centralized cTnI results will be used to reduce intermarker and interassay variability. The locally available results will be submitted by the clinical sites and may be used in the adjudication process as a secondary source.

Acute MI

The term *acute* myocardial infarction should be used when there is evidence of myocardial necrosis in a clinical setting consistent with acute myocardial ischemia. Under these conditions, any one of the following criteria meets the diagnosis for myocardial infarction:

Type 1: Spontaneous

Spontaneous clinical syndrome related to atherosclerotic plaque rupture, ulceration, fissuring, erosion, or dissection, with resulting intraluminal thrombus, and leading to decreased myocardial blood flow or distal platelet emboli with ensuing myocyte necrosis. This classification requires:

a) Detection of a rise and/or fall of cardiac biomarker values (preferably cTn) with at least 1 value >99th percentile of the URL and

b) At least one of the following:

(i) Symptoms of myocardial ischemia
(ii) New or presumed new significant ST-segment–T wave (ST–T) changes or new left bundle branch block (LBBB) on the ECG
(iii) Development of pathological Q waves on the ECG
(iv) Imaging evidence of new loss of viable myocardium or new regional wall motion abnormality
(v) Identification of an intracoronary thrombus by angiography or autopsy.

Notes: One or more coronary arteries may be involved. The subject may have underlying severe CAD but on occasion may have non-obstructive CAD.

Type 2: Ischemic imbalance
Spontaneous clinical syndrome where a condition other than CAD contributes to an imbalance between myocardial oxygen supply and/or demand (e.g., coronary endothelial dysfunction, coronary artery spasm, coronary embolism, tachy-/bradyarrhythmias, anemia, respiratory failure, hypotension, and hypertension with or without LVH). This classification requires:
a) Detection of a rise and/or fall of cardiac biomarker values (preferably cTn) with at least 1 value >99th percentile of the URL and
b) At least one of the following:
(i) Symptoms of myocardial ischemia
(ii) New or presumed new significant ST-segment–T wave (ST–T) changes or new left bundle branch block (LBBB) on the ECG
(iii) Development of pathological Q waves on the ECG
(iv) Imaging evidence of new loss of viable myocardium or new regional wall motion abnormality

Type 3: Death, no markers
Death where symptoms suggestive of myocardial ischemia are present, and with (presumed) new ischemic changes or new LBBB on ECG, but where death occurs before cardiac biomarkers can be obtained or could rise or (in rare cases) were not collected.

Type 4a: Percutaneous coronary intervention (PCI)-related
MI associated with and occurring within 48 h of PCI, with elevation of cardiac biomarker values to >5 × 99th percentile of the URL in subjects with normal baseline values (≤99th percentile URL), or a rise of biomarker values >20% if the baseline values are elevated and are stable or falling. This classification also requires at least 1 of the following:
(i) Symptoms suggestive of myocardial ischemia (i.e., prolonged ischemia ≥20 minutes)
(ii) New ischemic changes on ECG or new LBBB
(iii) Angiographic loss of patency of a major coronary artery or a side branch or persistent slow flow or no flow or embolization
(iv) Imaging evidence of new loss of viable myocardium or new regional wall motion abnormality.

Type 4b: Stent thrombosis
MI associated with stent thrombosis as detected by coronary angiography or at autopsy, where symptoms suggestive of myocardial ischemia are present, and with a rise and/or fall of cardiac biomarker values, with at least 1 value >99th percentile of the URL.

Type 4c: Stent restenosis
MI associated with stent restenosis as detected by coronary angiography or at autopsy, occurring >48 h after PCI, without evidence of stent thrombosis but with symptoms suggestive of myocardial ischemia, and with elevation of cardiac biomarker values to >99th percentile of the URL. This classification also requires the following:
(i) Does not meet criteria for any other classification of MI
(ii) Presence of a ≥50% stenosis at the site of previous successful stent PCI or a complex lesion and no other significant obstructive CAD of greater severity following 1)

Initially successful stent deployment, or 2) Dilation of a coronary artery stenosis with balloon angioplasty to <50% stenosis

Type 5: Coronary artery bypass grafting (CABG)-related
MI associated with and occurring within 48 h of CABG surgery, with elevation of cardiac biomarker values to >10 × 99th percentile of the URL in subjects with normal baseline cardiac biomarker values (≤99th percentile URL). This classification also requires at least 1 of the following:

(i) New pathological Q waves, new LBBB on ECG
(ii) Angiographic new graft or new native coronary artery occlusion
(iii) Imaging evidence of new loss of viable myocardium or new regional wall motion abnormality.

Acute MI- Symptoms
Presence of acute symptoms of myocardial ischemia, such as chest, upper extremity, mandibular, or epigastric discomfort, or an ischemic equivalent such as dyspnea or fatigue. This is one of the noncardiac marker criteria supporting the diagnosis of acute MI Types 1, 2, 3, 4a, 4b, and 4c.

Acute MI- Acute Ischemic Changes on ECG
Presence of new or presumed new significant ST-segment-T wave (ST-T) changes or new LBBB consistent with acute myocardial ischemia. This is one of the noncardiac marker criteria supporting the diagnosis of acute MI Types 1, 2, 3, and 4.

- Ischemic changes on ECG
  In the absence of LVH and LBBB pattern (or other confounder such as a paced rhythm) on ECG, either:

  a) ST elevation
  New (or presumed new) ST elevation at the J point in 2 contiguous leads with the following cut points: ≥0.1 mV in all leads other than leads V_2 to V_3 where the following cut points apply: ≥0.2 mV in men ≥40 y of age; ≥0.25 mV in men <40 y of age, or ≥0.15 mV in women; or

  b) ST depression and T-wave changes
  New (or presumed new) horizontal or downsloping ST-segment depression ≥0.05 mV in 2 contiguous leads and/or T inversion ≥0.1 mV in 2 contiguous leads with prominent R wave or R/S ratio >1.

- LBBB
  New (or presumed new) LBBB pattern on ECG.

Acute MI- New Q Waves on ECG
Presence of new or presumed new pathological Q waves consistent with MI. This is one of the noncardiac marker criteria supporting the diagnosis of acute MI Types 1, 2, 4, and 5.

New (or presumed new):

a) Any Q wave in leads V_2 to V_3 ≥0.02 s or QS complex in leads V_2 and V_3
b) Q wave ≥0.03 s and ≥0.1 mV deep or QS complex in leads I, II, aVL, aVF, or V_4 to V_6 in any 2 leads of a contiguous lead grouping (I, aVL; V_1 to V_6; II, III, aVF)
The same criteria are used for supplemental leads V_7 to V_9.

Acute MI- Coronary Thrombus Present
Presence of thrombus in a major epicardial vessel consistent with an acute MI. This is one of the noncardiac marker criteria supporting the diagnosis of acute MI Type 1.

- Thrombus on angiography
  In the patient with a presumed acute STEMI, the angiographic appearance of thrombus (typically a filling defect) on angiography. This includes the aspiration of thrombus from the infarct vessel before coronary intervention during primary PCI for acute STEMI.

- Thrombus at autopsy
  Identification of thrombus in a major epicardial vessel at autopsy.

Acute MI- Change in Noninvasive Imaging
Demonstration of a new change in myocardial viability or function consistent with MI. This is one of the noncardiac marker criteria supporting the diagnosis of acute MI Types 1, 2, 4a, and 5.

- New loss of viable myocardium
  Noninvasive imaging evidence of a loss of viable myocardium when compared with the most recent previous noninvasive imaging study.

- New regional wall motion abnormality
  Noninvasive imaging evidence of a decrease in regional wall motion contractility compared with the most recent previous noninvasive imaging study.

Acute MI- PCI Angiographic Complication
Occurrence of an adverse angiographic finding during PCI consistent with acute myocardial ischemia. This is one of the noncardiac marker criteria supporting the diagnosis of acute MI Type 4a.

- Loss of major coronary
  Angiographic loss of patency of a major epicardial vessel.

- Loss of side branch
  Angiographic loss of patency of a side branch.

- Slow flow/no flow/embolization
  Angiographic reduction of flow into the coronary microcirculation.

Acute MI- Acute Vessel Occlusion After CABG

Angiographic documentation of a new CABG or new native coronary artery occlusion within 48 h of CABG surgery. This is one of the noncardiac marker criteria supporting the diagnosis of acute MI Type 5.

Prior MI

Presence of any 1 of the following criteria meets the diagnosis for prior MI (before study initiation):

- Pathological Q waves with or without symptoms in the absence of non-ischemic causes
    a) Any Q wave in leads V_2 to V_3 ≥0.02 s or QS complex in leads V_2 and V_3
    b) Q wave ≥0.03 s and ≥0.1 mV deep or QS complex in leads I, II, aVL, aVF, or V_4 to V_6 in any 2 leads of a contiguous lead grouping (I, aVL; V_1 to V_6; II, III, aVF)
    The same criteria are used for supplemental leads V_7 to V_9
    c) R wave ≥0.04 s in V_1 to V_2 and R/S ≥1 with a concordant positive T wave in the absence of a conduction defect
- Imaging evidence of a region of loss of viable myocardium that is thinned and fails to contract, in the absence of a non-ischemic cause
- Pathological findings of a prior MI

Silent MI

Asymptomatic patients who develop new pathologic Q wave criteria for MI detected during routine ECG follow-up, or reveal evidence of MI by cardiac imaging, that cannot be directly attributed to a coronary revascularization procedure, should be termed ‘silent MI’. The diagnosis of a new silent Q wave MI should be confirmed by a repeat ECG with correct lead placement, or by an imaging study, and by focused questioning about potential interim ischemic symptoms.

Stroke [54]
Stroke is defined as an acute episode of focal or global neurological dysfunction caused by brain, spinal cord, or retinal vascular injury as a result of hemorrhage or infarction of sudden onset,

(a) with persistence of symptoms for ≥24 hours or results in death (in <24 hours) and is not due to an identifiable non-vascular cause (i.e., brain tumor, trauma), or
(b) with symptoms of short duration (<24 hours) but evidence of infarction on cerebral imaging.

Stroke will be subclassified into 1 of 3 mutually exclusive categories:

1. Ischemic
   An acute episode of focal cerebral, spinal, or retinal dysfunction caused by infarction of central nervous system tissue.
   Note: Hemorrhage may be a consequence of ischemic stroke. In this situation, the stroke is an ischemic stroke with hemorrhagic transformation and not a hemorrhagic stroke.

2. Hemorrhagic
   An acute episode of focal or global cerebral or spinal dysfunction caused by intraparenchymal, intraventricular or subarachnoid hemorrhage.
   Note: Subdural hematomas are intracranial hemorrhagic events and not strokes.

3. Undetermined
   An acute episode of focal or global neurological dysfunction caused by presumed brain, spinal cord, or retinal vascular injury as a result of hemorrhage or infarction but with insufficient information (brain image via CT/MRI or autopsy) to allow categorization as either ischemic or hemorrhagic (without documentation or if tests are inconclusive).

As a matter of differentiation, transient ischemic attack **(TIA)** is a transient episode (resolves spontaneously without any evidence of residual deficit <24 hours) of focal neurological dysfunction caused by brain, spinal cord, or retinal ischemia, without acute infarction. TIAs will not be reported as endpoints but will be reported as an AE.

Recurrent Ischemia with Hospitalization [26]
Recurrent ischemia with hospitalization is defined as ischemic discomfort or equivalent meeting the following criteria:

(a) lasting ≥10 minutes at rest, or repeated episodes at rest lasting ≥5 minutes, or an accelerating pattern of ischemic discomfort (episodes that are more frequent, severe, longer in duration, and precipitated by minimal exertion), considered to be myocardial ischemia upon final diagnosis, and
(b) prompting hospitalization (including an overnight stay on an inpatient unit) within 48 hours of the most recent symptoms, and
(c) at least 1 of the following additional criteria for coronary artery disease and/or

ischemia:

- new and/or dynamic ST depression ≥0.05 mV, ST elevation ≥0.1 mV, or symmetric T-wave inversion ≥0.2 mV on a resting ECG; or
- definite evidence of ischemia on stress echocardiography, myocardial scintigraphy (e.g., an area of clear reversible ischemia), or ECG-only stress test (e.g., significant dynamic ST shift, horizontal or downsloping); or
- angiographic evidence of epicardial coronary artery stenosis of ≥70% diameter reduction and/or evidence for intraluminal arterial thrombus.

If subjects are admitted with suspected myocardial ischemia, and subsequent testing reveals a noncardiac or non-ischemic etiology, this will not be recorded as meeting this end point. Potential ischemic events meeting the criteria for myocardial infarction will not be adjudicated as ischemia requiring hospitalization.

Urgent Coronary Revascularization [26]
Urgent coronary revascularization is defined as ischemic discomfort or equivalent meeting the following criteria:

(a) lasting ≥10 minutes at rest, or repeated episodes at rest lasting ≥5 minutes, considered to be myocardial ischemia upon final diagnosis, and
(b) prompting coronary revascularization during an unscheduled visit to health care facility or during an unplanned (or prolonged) hospitalization for these symptoms or revascularization, which was either done emergently or not previously planned during the course of hospitalization.

Attempted revascularization procedures, even if not successful, will be counted. Potential ischemic events meeting the criteria for myocardial infarction will not be adjudicated as urgent coronary revascularization.

Coronary Stent Thrombosis [56]
All cases of reported death, MI, recurrent ischemia with hospitalization and urgent coronary revascularization will be reviewed and adjudicated with respect to stent thrombosis according to the Academic Research Consortium definition below:

Timing:

Acute stent thrombosis*	0 to 24 hours after stent implantation
Subacute stent thrombosis*	>24 hours to 30 days after stent implantation
Late stent thrombosis†	>30 days to 1 year after stent implantation
Very late stent thrombosis†	>1 year after stent implantation

Stent thrombosis should be reported as a cumulative value over time and at the various individual time points specified below. Time 0 is defined as the time point after the guiding catheter has been removed and the subject has left the catheter laboratory.

*Acute or subacute can also be replaced by the term early stent thrombosis. Early stent

thrombosis (0-30 days)
†Includes primary as well as secondary late stent thrombosis; secondary late stent thrombosis is a stent thrombosis after a target lesion revascularization.

Grading:
Three categories of evidence define the probability that coronary artery stent thrombosis has occurred:

1) Definite stent thrombosis*
   a. Angiographic confirmation of stent thrombosis†
   The presence of a thrombus‡ that originates in the stent or in the segment 5 mm proximal or distal to the stent and presence of at least 1 of the following criteria within a 48-hour time window:
   - Acute onset of ischemic symptoms at rest
   - New ischemic ECG changes that suggest acute ischemia
   - Typical rise and fall in cardiac biomarkers (refer to definition of spontaneous MI)
   - Nonocclusive thrombus
     Intracoronary thrombus is defined as a (spheric, ovoid, or irregular) noncalcified filling defect or lucency surrounded by contrast material (on 3 sides or within a coronary stenosis) seen in multiple projections, or persistence of contrast material within the lumen, or a visible embolization of intraluminal material downstream.
   - Occlusive thrombus
     TIMI 0 or TIMI 1 intrastent or proximal to a stent up to the most adjacent proximal side branch or main branch (if originates from the side branch).

   b. Pathological confirmation of stent thrombosis
   Evidence of recent thrombus within the stent determined at autopsy or via examination of tissue retrieved following thrombectomy.
2) Probable stent thrombosis
   Clinical definition of probable stent thrombosis is considered to have occurred after intracoronary stenting in the following cases:
   - Any unexplained death within the first 30 days§
   - Irrespective of the time after the index procedure, any MI that is related to documented acute ischemia in the territory of the implanted stent without angiographic confirmation of stent thrombosis and in the absence of any other obvious cause.

3) Possible stent thrombosis
   Clinical definition of possible stent thrombosis is considered to have occurred with any unexplained death >30 days after intracoronary stenting.

*Definite stent thrombosis is considered to have occurred by either angiographic or pathological confirmation.
†The incidental angiographic documentation of stent occlusion in the absence of clinical signs

or symptoms is not considered a confirmed stent thrombosis (silent occlusion).
‡Intracoronary thrombus.
§For studies with ST-elevation MI population, one may consider the exclusion of unexplained death within 30 days as evidence of probable stent thrombosis.

When available data support >1 classification, the highest level of certainty should be reported.

APPENDIX C: ADDITIONAL SAFETY INFORMATION

A Guide to Interpreting the Causality Question

The following factors should be considered when deciding if there is a "reasonable possibility" (i.e., "possibly" or "probably") that an AE may have been caused by the drug.

- Time Course. Exposure to suspect drug. Has the subject actually received the suspect drug? Did the AE occur in a reasonable temporal relationship to the administration of the suspect drug?
- Consistency with known drug profile. Was the AE consistent with the previous knowledge of the suspect drug (pharmacology and toxicology) or drugs of the same pharmacological class? OR could the AE be anticipated from its pharmacological properties?
- Dechallenge experience. Did the AE resolve or improve on stopping the suspect drug?
- No alternative cause. The AE cannot be reasonably explained by another etiology such as the underlying disease, other drugs, other host or environmental factors.
- Rechallenge experience. Did the AE reoccur if the suspected drug was reintroduced after having been stopped?
- Laboratory tests. A specific laboratory investigation (if performed) has confirmed the relationship?

A "reasonable possibility" could be considered to exist for an AE where one or more of these factors exist.

In contrast, there would not be a "reasonable possibility" of causality if none of the above criteria apply or where there is evidence of exposure and a reasonable time course but any de-challenge (if performed) is negative or ambiguous or there is another more likely cause of the AE.

In difficult cases, other factors could be considered such as:

- Is this a recognized feature of overdose of the drug?
- Is there a known mechanism?

Ambiguous cases should be considered as being a "reasonable possibility" of a causal relationship unless further evidence becomes available to refute this.

Causal relationship in cases where the disease under study has deteriorated due to lack of effect should be classified as no reasonable possibility.

National Institute of Allergy and Infectious Disease and Food Allergy and Anaphylaxis Network criteria for anaphylaxis [48]

Anaphylaxis is highly likely when any one of the following 3 criteria are fulfilled:
1. Acute onset of an illness (minutes to several hours) with involvement of the skin, mucosal tissue, or both (e.g., generalized hives, pruritus or flushing, swollen lips-tongue-uvula)
AND AT LEAST ONE OF THE FOLLOWING
a. Respiratory compromise (e.g., dyspnea, wheeze-bronchospasm, stridor, reduced PEF, hypoxemia)
b. Reduced BP or associated symptoms of end-organ dysfunction (e.g., hypotonia [collapse], syncope, incontinence)
2. Two or more of the following that occur rapidly after exposure *to a likely stimuli for that subject* (minutes to several hours):
a. Involvement of the skin-mucosal tissue (e.g., generalized hives, itch-flush, swollen lips-tongue-uvula)
b. Respiratory compromise (e.g., dyspnea, wheeze-bronchospasm, stridor, reduced PEF, hypoxemia)
c. Reduced BP or associated symptoms (e.g., hypotonia [collapse], syncope, incontinence)
d. Persistent gastrointestinal symptoms (e.g., crampy abdominal pain, vomiting)
3. Reduced BP after exposure to *known stimuli for that subject* (minutes to several hours):
a. Infants and children: low systolic BP (age specific) or greater than 30% decrease in systolic BP
b. Adults: systolic BP of less than 90 mm Hg or greater than 30% decrease from that person's baseline
PEF, Peak expiratory flow; *BP*, blood pressure.

Modified from Sampson et al., 2006

APPENDIX D: GENERAL REQUIREMENTS FOR CLINICAL TRIALS

1 General Requirements for Clinical Trials

Sponsor (which includes its affiliates/partners) and the investigator agree to conduct the trial as described in the protocol, according to applicable laws, regulations, and principles of good clinical practice. It is the intention that the trial be conducted according to practices that will satisfy worldwide registration requirements. Therefore, in addition to satisfying locally relevant requirements for the conduct of the trial, there may be a need to satisfy other requirements, as specified in this protocol. These additional requirements may follow laws, regulations, and practices of health authorities and/or sponsor policies. While health authorities may not directly oversee the conduct of the trial, they may be recipients of marketing applications that contain the report of the trial. Sponsor will take into account the appropriateness of the trial site and facilities and be assured of the investigator's qualifications and availability for the entire duration of the trial. The investigator will ensure that there are sufficient time, staff and facilities available for the duration of the trial.

Both sponsor (represented by the "sponsor representative") and the investigator must sign the protocol (and subsequent versions of the protocol if amended) as an agreement of the details of the clinical trial. The agreement will include the acceptance of procedures for source document verification, data collection, monitoring, audit, inspection, and quality assurance. Any amendments to the protocol or changes to any trial documents must have the agreement of sponsor, the investigator, and when appropriate, the Institutional Review Board (IRB) before the amendment or change is implemented; any such agreement must be documented in writing.

It is the responsibility of the investigator to ensure that all staff personnel who will be handling, packaging, and/or shipping clinical specimens or any other hazardous or dangerous goods act in conformance with International Air Transport Association (IATA) regulations relating to the handling and shipping of hazardous or dangerous goods.

2 Monitoring and Inspections

The sponsor representative and regulatory authority inspectors are responsible for contacting and visiting the investigator for the purpose of inspecting the facilities and, upon request, inspecting the various records of the clinical study (e.g., eCRFs and other pertinent data) provided that subject confidentially is respected.

The sponsor-appointed, study monitor is responsible for verifying the eCRFs at regular intervals throughout the study to verify adherence to the protocol-completeness, accuracy and consistency of the data- and adherence to local regulations on the conduct of clinical research. The monitor should have access to subject medical records, source documents and other study-related records needed to verify the entries on the eCRFs.

The investigator agrees to cooperate with the monitor to ensure that any problems detected in the course of these monitoring visits, including delays in completing eCRFs, are resolved. It is essential that the investigator and designated staff set aside a sufficient amount of time for these study monitoring visits to permit an adequate review of the trial's progress and study data.

This study may be selected for audit by the sponsor or inspection by regulatory authorities (e.g., FDA, DHHS, NHLBI/NIH). Inspection of site facilities (e.g., pharmacy, drug storage areas, treatment areas, laboratories) and review of original study-related records will occur to evaluate the study conduct and compliance with the protocol and applicable regulatory requirements.

3 Recording Subjects' Data

The investigator must conduct the trial and maintain records and data during and after the term or early termination of the trial in compliance with all applicable legal and regulatory requirements, including any applicable requirements of the USA Code of Federal Regulations and specific local regulations, and in accordance with ICH.

The investigator is to provide such subject data on completed Case Report Forms (CRFs) or via computer entry on an Electronic Data Capture (EDC) system. The investigator must maintain a separate subject list containing the name and address for each participant so that all can quickly be contacted by the investigator, if necessary.

Case Report Forms are an integral part of the trial and subsequent reports. They need not be completed by the investigator, but all entries are the responsibility of the investigator. The investigator is expected to review subject data before attesting in writing to the accuracy and completeness (and legibility when paper CRFs are used) of the information contained therein.

Any records or documents used as the source of information in the CRF are to be maintained and readily available for review by authorized representatives of the sponsor or regulatory agency. These records or documents are called the subject source data. These include but are not limited to hospital records, clinical and office charts, laboratory and pharmacy records, diaries, microfiches, and radiographs and correspondence.

Completed paper CRFs are to be collected by the study monitor after source document verification has been completed. Paper and electronic CRFs and the electronic database shall be the exclusive property of sponsor.

3.A Paper Case Report Forms

All paper CRFs are to be filled out using a black ball-point pen. Errors should be lined out but not obliterated and the correction inserted, initialed and dated.

3.B Electronic Data Capture (EDC)

Data collection and data management will be performed using the Medidata Rave® EDC System, developed by Medidata Solutions, Inc. Medidata Rave® is an internet-based system that enables subject randomization, data collection through electronic CRFs, review of data through automated and manual data edit checks, and online source document verification. Electronic CRFs (eCRFs) are used in place of paper CRFs, for the transcription of source document data.

3.Bi Third Party Housing

The Medidata Rave® application and the eCRF data for the clinical trial is installed and stored on a server in a central, secure facility where systems administration and backups can be performed regularly. The central facility where the server is located is neither owned nor operated by Tufts Medical Center. The data-hosting facility utilizes card-key access and video surveillance. Redundant power supply, raised floors, disaster proofing, bulletproof glass/walls, motion detection, and servers inside locked cages are all security features. Data stored at the hosting facility is backed-up and stored at an offsite location on a frequent basis.

3.Bii Authentication

Each user of the system accesses the central application through a standard web browser (e.g., Internet Explorer). At the beginning of each session, user authentication is performed through a unique and confidential combination of User identification (user ID) and password. Activities in the Medidata Rave® System are time, date, and user stamped in the database, along with duration of the sessions. After login, the system establishes a user session, so that all data recorded by the user is stored in the database along with the user identification.

3.Biii Data Collection

Each user in Medidata Rave® is granted specific rights within the system, limiting the data that they can view or edit and the particular functions that they can perform. Data consistency checks may be performed prior to data submission to ensure that the proper type of data is submitted in the field (e.g., alphabetic vs. numeric, number of significant digits, etc.). Once data are submitted, an audit trail independently records the user ID, date and time of the entry and reason for change. Any actions that create, modify, or delete an electronic record are recorded. The audit trail is incremental and cannot be modified by any user.

The Medidata Rave® System performs edit checks on submitted data and notifies users with the results of these checks. In both cases, the user will be presented with a description of the discrepancy, and will be expected to address the problem. As with paper CRFs, the investigator is expected to review eCRF data on an ongoing basis.

To ensure the quality of clinical data across all subjects and sites, a clinical data management review will be performed on subject data received at the sponsor/designee. During this review, subject data will be checked for consistency, omissions, and any apparent discrepancies. In addition, the data will be reviewed for adherence to the protocol and regulatory standards. To resolve any questions arising from the clinical data management review process, data queries and/or site notifications will be created in the EDC system database for site resolution and closed by the sponsor/designee reviewer.

3.B.iv Data Access

During the conduct of the study, each investigative site can access all of its entered data, including complete audit information, through the Medidata Rave® System using a standard web browser. At the completion of the study, each investigator will be required to sign an EDC memo attesting to the completeness and accuracy of the data for each subject at his/her site. Also, each site will receive an archival copy on compact disc (CD) of the eCRFs,

representing all data entered for its subjects, in Adobe Acrobat (PDF) format. The CDs can be accessed, search, and copied.

4 Trial Documents and Records Retention

The investigator and designated staff are responsible for maintaining a comprehensive and centralized filing system of all study-related (essential) documentation, suitable for inspection at any time by representatives from the sponsor and/or applicable regulatory authorities. Elements include but are not limited to: the protocol with all amendments; Investigator's Brochure with all amendments; subject files containing completed eCRFs, original source documentation and informed consents; correspondence to and from the sponsor, health authorities and IRB; investigator's curricula vitae, license and financial disclosure; study monitor logs and reports; laboratory reference ranges and laboratory certifications; and pharmacy information pertaining to the accountability, handling and storage of the investigational product(s).

Subject files and other source data must be kept for the maximum period of time permitted by the hospital, institution or private practice, or as specified below. The study monitor must be consulted if the investigator wishes to assign the files to someone else, remove them to another location or is unable to retain them for the specified period.

The investigator must retain trial records for the amount of time specified by applicable laws and regulations. At a minimum, trial records must be retained for the amount of time specified by the Code of Federal Regulations, Title 21, Volume 5, which state that an investigator shall retain records required to be maintained under this part for a period of 2 years following the date a marketing application is approved for the drug for the indication for which it is being investigated; or, if no application is to be filed or if the application is not approved for such indication, until 2 years after the investigation is discontinued and FDA is notified. All trial documents shall be made available if required by relevant health authorities. The investigator should consult with the study monitor prior to discarding trial and/or subject files.

Sponsor will retain all sponsor-required documentation pertaining to the trial for the lifetime of the product. Archived data may be held on microfiche or electronic record, provided that a back-up exists and that a paper copy can be obtained from it, if required.

The sponsor will post trial information, including the results, on public registries in which the sponsor participates (e.g., clinicaltrials.gov, a service of the United States National Institutes of Health). The information on public registries may be transferred to other countries, as appropriate, for the uses described above. No subjects will be identified in this information.

5 FDA Financial Disclosure Requirement

In connection with the clinical study described in the protocol, the investigator certifies that, if asked, the investigator will read and answer the Clinical Investigator Financial Disclosure Form truthfully and to the best of investigator's ability. Investigator also certifies that, if asked, the investigator will have any other applicable party(s) (e.g., sub-investigators) read and answer the Clinical Investigator Financial Disclosure Form as a condition of their participation in the

study.

If the financial interests reported on the Clinical Investigator Financial Disclosure Form change during the course of the study or within one year after the last subject has completed the study as specified in the protocol, the investigator and the other applicable party(s) are obligated to inform the sponsor of such financial change.

6 Investigator Information

The investigator agrees that the sponsor may store, in an electronic database, for sponsor's ongoing and future processing and use, including for purposes of monitoring the conduct of this study and considering potential investigators for future studies, both administrative and qualitative information, including personal data (e.g., name, address, curriculum vitae, etc.), relating to the investigator's conduct of the study. The investigator further agrees that such information may be provided to FDA and/or other appropriate regulatory authority, consistent with sponsor's obligations to same. The investigator also agrees that the sponsor may post certain information concerning the investigator's participation in the study (e.g., study site phone contact information and study site location) on public registries in which the sponsor participates (e.g., clinicaltrials.gov, a service of the United States National Institutes of Health). The information on public registries may be used to contact the investigator by prospective study subjects or referring physicians. The information on public registries may be transferred to other countries, as appropriate, for the uses described above.

APPENDIX E: CONFIDENTIALITY AND SECURITY OF PHARMACOGENOMICS RESEARCH

Confidential Subject Information for Pharmacogenomics Study

All information in this trial will be treated as confidential. The whole blood sample from randomized research subjects who have provided separate informed consent will be collected and sent to the sponsor (Tufts Center for Hemostasis and Thrombosis Research) for pharmacogenomics analysis. The sample will be labeled by the clinical site with the subject's unique study identification number, the sample type code (i.e., PG), study time point and the date/time of collection. A corresponding eCRF will be entered by the clinical site and will contain certain standard subject identifiers that are normally collected in the course of this clinical trial, such as subject initials, subject study identification number, and date of birth. This will enable authorized study sponsor personnel to accurately track the collection, storage and processing of the blood sample.

The sponsor, Tufts Center for Hemostasis and Thrombosis Research, will perform the DNA and RNA purification/extraction procedures and the subsequent genetic analysis utilizing the whole blood sample and derivative specimens (i.e., DNA and RNA). Upon receipt of the whole blood sample, the sponsor's designated staff will enter the subject study identification number into a coded data base and assign a new identifier (genetic sample code) to each sample. This new identifier will be linked to the subject study identification number assigned to the subject. From that point on, the whole blood sample, its derivative specimens and any results from the genetic analysis will be labeled with the same genetic sample code for each subject. The genetic sample code or "key" will be stored separately from the subject's eCRF and from the data resulting from the genetic analysis. This "key" will be maintained by the sponsor's designated staff. The underlying clinical trial database for the study will be stored by the unique subject study identification number within the electronic data capture (EDC) system (i.e., Medidata Rave®) maintained by the sponsor and sponsor's designee, RTI, International. The genetic research laboratory database will be stored according to the genetic sample code in a separate file (i.e., Microsoft Excel® spreadsheet) maintained by the sponsor. The sponsor will request certain clinical variables and outcomes collected during the entire course of the subject's participation in the underlying trial (i.e., up to 90 days following study drug administration) from RTI at the time of data analysis. The secure "key" will be used to match the genetic sample code to the original subject identification number to allow the sponsor to correlate the clinical variables and outcomes information collected during the course of the study (identified by unique subject identification number) with the information obtained from the genetic analysis (identified only by genetic sample code). The genetic research laboratory data will not be merged into the main clinical trial database and the personal identifying information from the clinical trial database will not be sent to the sponsor by RTI. The "key" could potentially be utilized to reconstruct the link between genetic information and identifiable clinical information, under other specific circumstances- verification of data accuracy, request for sample/specimen(s) destruction by the investigator on behalf of the subject or as mandated by regulatory authorities.

The "key" will be destroyed at the conclusion of the pharmacogenomics study, after the clinical information has been associated with the subjects' genetic sample code. Once this "key" is destroyed, the sample/specimen(s) and associated data derived from the genetic analyses of these sample/specimen(s) will be rendered anonymous and it will be impossible to identify any

specific individual subject's sample/specimen(s) and/or associated data.

Genetic analysis utilizing the DNA and RNA sample/specimen(s) will be performed by the sponsor and may be performed at one of the participating clinical trial sites designated by the sponsor if expertise in a particular genetic/genetic expression test is indicated. The pharmacogenomics sample/specimen(s) will not be used by investigators other than those conducting the original research protocol (main study), and will not be subjected to testing outside the stated purpose and objectives of the original research protocol (See **Section 6.7.3.1**), and will not be disseminated to other researchers who are not part of the original protocol. If applicable, sample/specimen(s) shipped by the sponsor to a clinical trial site will be identified only by the genetic sample code. If required for analysis, the sponsor may provide the clinical trial site with the clinical trial data set identified only by the genetic sample code. The clinical trial site will not have access to the "key" to link the genetic sample code to the individual's identifiers. Any sample/specimen(s) remaining with the clinical trial site after the genetic analysis has been performed will be returned to the sponsor or destroyed.

The informed consent form will be kept under secure storage at the clinical site for regulatory purposes and cannot be traced to any samples, test results, or medical information once the specimens have been rendered anonymous. Laboratory personnel performing the genetic testing will not have access to the informed consent document or the clinical trial database, nor will they be able to identify subjects from the coded sample/specimen(s). Sample/Specimen(s) will be identified to the laboratory staff only by the genetic sample code. Subjects who decline to sign the informed consent for the optional pharmacogenomics study will not have the whole blood sample collected, nor will they be excluded from participating in the underlying clinical trial.

Withdrawal from Pharmacogenomics Study

Subjects may withdraw their consent for the pharmacogenomics sample/specimen(s) to be archived for subsequent testing at any time. Subjects may request from the local principal investigator (in writing or by telephone) that their sample/specimen(s) be destroyed and the investigator will notify the sponsor. Sample/Specimen(s) which have not already been analyzed will be destroyed in a manner documented and prescribed by the sponsor. If applicable, the sponsor will contact the clinical site performing any genetic analysis and request that those sample/specimen(s) be destroyed if they have not already been used. Any genetic or genetic expression data and clinical data that have been rendered from the sample/specimen(s) up until the time of the subject's documented informed consent withdrawal will be utilized in the study's analysis.

Retention of Samples and Data

The archived pharmacogenomics sample/specimen(s) (whole blood sample, DNA and RNA derivatives) will be analyzed within 15 years from the end of the underlying clinical trial (i.e., when the final subject completes the main study protocol), and then destroyed in a manner documented and prescribed by the sponsor. It is possible that the sample/specimen(s) may be exhausted prior to that point in time.

It is anticipated that the data generated from the pharmacogenomics study will be retained by the sponsor for an indefinite period of time.

Data Security

The pharmacogenomics research database will be accessible only to authorized sponsor research personnel and/or designated collaborators, and will be stored and accessible by the genetic sample code. Until the "key" is destroyed, there exists a means to decipher the genetic code which would enable linkage of the data to the identity and private information of the subject. However, this could only be achieved if an individual possessed both the "key" and the access to the underlying clinical trial database. This is limited to two designated staff members at the sponsor site.

These data are collected for pharmacogenomics research purposes only as specified in **Section 6.7.3.1**, and will not be used for any other purpose without explicit consent form the research subject.

APPENDIX F: SUGGESTED MODEL INFORMED CONSENT DOCUMENT