

**CENTER FOR DRUG EVALUATION AND  
RESEARCH**

*APPLICATION NUMBER:*

**204958Orig1s000**

**PHARMACOLOGY REVIEW(S)**

## Tertiary Pharmacology Review

**By:** Paul C. Brown, Ph.D., ODE Associate Director for Pharmacology and Toxicology, OND IO

**NDA:** 204958

**Submission date:** 4/30/2013

**Drug:** cangrelor

**Applicant:** The Medicines Company

**Indication:** reduction of thrombotic cardiovascular events (including stent thrombosis) in patients with coronary artery disease undergoing percutaneous coronary intervention (PCI); to maintain P2Y12 inhibition in patients with acute coronary syndromes (ACS) or patients with stents who are at increased risk for thrombotic events (such as stent thrombosis) when oral P2Y12 inhibitor therapy is interrupted due to surgery

**Reviewing Division:** Division of Cardiovascular and Renal Products

### **Discussion:**

The primary pharm/tox reviewer concluded that the information submitted was adequate to assess cangrelor for the indications listed above. He raised some concern about renal, urinary tract and liver toxicity observed in the dog at exposures relevant to the PCI setting. In this setting, the C<sub>ss</sub> values of cangrelor in humans could be higher than the plasma concentration measured at the NOAEL in the dog. The reviewer recommended enhanced renal and liver function monitoring in the clinic. The cross-discipline team leader has noted and considered these recommendations in his review.

Cangrelor did not produce malformations in either the rat or rabbit reproductive studies, although some adverse effects were seen including fetal growth retardation in rats and increased incidences of abortion and intrauterine losses, as well as fetal growth retardation in rabbits. Some of these findings occurred at plasma concentrations similar to or lower than those that occurred in humans.

No carcinogenicity studies were conducted with cangrelor. This is acceptable because the indications are not considered chronic.

Cangrelor is a P2Y12 platelet inhibitor which is an existing Established Pharmacologic Class.

### **Conclusions:**

The pharmacology/toxicology reviewer conducted a thorough evaluation of the nonclinical information submitted in support of this NDA. I agree that this NDA may be approved for the above indications from a pharm/tox perspective. I will provide comments on labeling a later time.

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/s/  
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PAUL C BROWN  
04/25/2014

DEPARTMENT OF HEALTH AND HUMAN SERVICES  
PUBLIC HEALTH SERVICE  
FOOD AND DRUG ADMINISTRATION  
CENTER FOR DRUG EVALUATION AND RESEARCH

**PHARMACOLOGY/TOXICOLOGY NDA REVIEW AND EVALUATION**

Application NDA number: 204958  
Supporting document/s: Original e-submission, eCTD sequence # 0002  
CDER stamp date: 05/06/2013  
Review completion date: 12/13/2013  
Product: Cangrelor ( (b) (4) Intravenous injection  
Indication: Antithrombotic, P2Y<sub>12</sub> receptor platelet inhibitor  
Applicant: The Medicines Company, Parsippany, NJ  
Review Division: Cardiovascular and Renal Products  
(DCRP/ODE1/OND/CDER)  
Reviewer: Belay Tesfamariam, PhD  
Team Leader: Albert DeFelice, PhD  
Division Director: Norman Stockbridge, MD, PhD  
Project Manager: Alison Blaus, RAC

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# 1 Executive Summary

## 1.1 Introduction

Oral P2Y<sub>12</sub> platelet receptor inhibitors have been shown to reduce the risk of thrombotic events in acute coronary syndrome and percutaneous coronary intervention (PCI). However, oral P2Y<sub>12</sub> inhibitors have limitations of prolonged effects which creates a dilemma in patients who need to undergo surgery on whether to stop the treatment and risk thrombotic complications, or continue the treatment and risk bleeding. The sponsor has developed an analog of adenosine triphosphate (ATP), cangrelor, which is intravenously administered, and selectively blocks P2Y<sub>12</sub> receptor mediated ADP-induced platelet aggregation. Cangrelor is considered to exhibit rapid-onset that occurs within minutes, reversible binding and short half-life of 3 to 6 minutes with substantial recovery of platelet reactivity after cessation of drug infusion. This favorable pharmacodynamic profile of rapid onset and reversible P2Y<sub>12</sub> receptor inhibition may be useful in cases of urgent need of effective platelet inhibition during PCI because of high risks of ischemia and stent thrombosis following the procedure. In addition, the pharmacokinetic profile of cangrelor may be beneficial for 'bridging' between the discontinuation of oral P2Y<sub>12</sub> receptor inhibitors and major cardiac surgery, and thus may reduce thrombotic risks that may occur during the transition period.

## 1.2 Brief Discussion of Nonclinical Findings

The most notable toxicity findings of cangrelor in the rat and dog were renal cortical tubule and urinary tract lesions manifested as inflammation, ulceration and necrosis, with loss of kidney function (i.e., elevated BUN, creatinine,). These toxicities were caused by the parent molecule, whereas the metabolites did not induce any evidence of toxicity, indicating that the adverse effects were relatively immediate. This is also supported by the observation that a single bolus injection in the rat revealed the adverse effects within 6 hrs, which is consistent with the short half-life of the parent drug. The changes in the kidney tubules and ureter tend to be reversible upon cessation of treatment. Another finding was elevated liver function enzymes, but this was not corroborated by histopathological evidence of overt liver toxicity. Cangrelor caused impairment of male and female fertility, and fetal growth retardation at doses of 3 µg/kg/min (plasma concentration, C<sub>ss</sub> 428 - 494 ng/ml), which is about the same plasma concentration achieved in the PCI setting (488 ng/ml). The no observed adverse effect level (NOAEL) in the rat and dog was an infusion of 3 and 3.75 µg/kg/min for 4 weeks, which provided C<sub>ss</sub> values of 113 and 61.5 ng/ml, and AUC<sub>0-28d</sub> 46565 and 53957 ng\*h/ml, respectively. In the PCI setting, a bolus injection of cangrelor 30 µg/kg followed by 4 µg/kg/min infusion for 2 h yields a C<sub>ss</sub> of 488 ng/ml, which is estimated to be about 8-fold higher than the C<sub>ss</sub> at the NOAEL in the dog (C<sub>ss</sub> 61.5 ng/ml). The Bridge dosing of cangrelor 0.75 µg/kg/min administered for 7 days achieved AUC<sub>0-7d</sub> 11189 ng\*h/ml, which is approximately 4.5-fold lower than the exposures at the NOAEL in the dog (AUC<sub>0-28d</sub> 53957 ng\*h/ml).

## 1.3 Recommendations

**1.3.1 Approvability:** The extent and scope of the pharmacological and toxicological documentation provided are appropriate to assess the safety of cangrelor for the proposed clinical dosing regimens – i) In the Bridge setting, human exposures achieved from cangrelor

0.75 µg/kg/min infusion for 7 days ( $AUC_{0-7d}$  11189 ng\*h/ml) is about 4.5-fold lower than that at the 28-day NOAEL in the dog ( $AUC_{0-28d}$  53957 ng\*h/ml). ii) However, in the PCI setting, the  $C_{ss}$  values of 488 ng/ml achieved with a bolus injection of cangrelor 30 µg/kg followed by 4 µg/kg/min infusion for 2 hr is ~8-fold higher than the plasma concentration measured at the NOAEL in the dog ( $C_{ss}$  61.5 ng/ml), and therefore in the absence of any margin of safety over the NOAEL, enhanced renal, urinary tract and liver function monitoring are recommended.

### 1.3.2 Additional Nonclinical Recommendations

- Cangrelor is structurally similar to ATP, and thus it may aggravate conditions in patients with respiratory distress (dyspnea, COPD, asthma), which may be mediated by inhibition of P2Y<sub>12</sub> receptors in the sensory neurons thereby increasing the dyspnea sensation.
- Cangrelor at high doses may cause slowing of heart rate (negative chronotropic effect), depression of the SA node and AV nodal conduction (negative dromotropic effect) and asynchronous premature ventricular contractions ('ectopics').
- Cangrelor 30 µg/kg in humans achieves plasma concentration of 488 ng/ml, and considering 98% protein plasma binding, the unbound is ~9.8 ng/ml, which is ~22-fold higher than the half-maximal inhibitory concentration of cangrelor in human platelets ( $IC_{50}$  0.5 nM, 0.43 ng/ml), indicating excess loading dose. In the Bridge setting of 0.75 µg/kg/min ( $C_{ss}$  67 ng/ml), the unbound cangrelor (1.34 ng/ml) is ~3-fold higher than the  $IC_{50}$  of cangrelor.
- In conscious beagle dogs infused continuously with cangrelor (40 µg/kg/min) for 7 days, ADP-induced aggregation was restored at about 60 min after cessation of cangrelor infusion. This indicate that transitioning from cangrelor (direct and reversible inhibitor) to orally active and irreversible thienopyridine P2Y<sub>12</sub> receptor inhibitors may delay onset of action if administered at earlier time point because of cangrelor's high affinity for P2Y<sub>12</sub> receptors which may prevent access to the thiol active metabolite to form disulfide bridge with cysteine residues in the receptor, thereby causing a delay in the onset of platelet inhibition. Thus, careful consideration should be given to the timing of administration of an oral thienopyridine, which is estimated to be ~60 minutes after the termination of cangrelor infusion to assure complete on-off receptor kinetics.

### 1.3.3 Labeling

The proposed prescribing information includes an appropriate description of data and recommendations in the genotoxicity, impairment of fertility, and pregnancy sections.

## 2 Drug Information

### 2.1 Drug

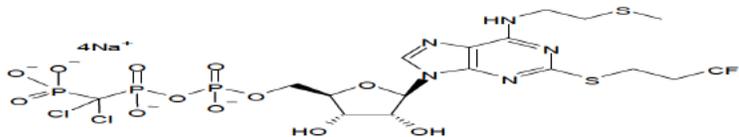
**Chemical Abstract Service (CAS) Number:** (b) (4)  
**Trade Name:** (b) (4)  
**Code Name:** AR-C69931MX, ARL 69931MX; FPL 69931MX  
**Generic Name:** Cangrelor tetrasodium

**Chemical Name:** N6-[2-(methylthio)ethyl]-2-[(3,3,3-trifluoropropyl)thio]-5'-adenylic acid

**Empirical formula:** C<sub>17</sub>H<sub>21</sub>N<sub>5</sub>Cl<sub>2</sub>F<sub>3</sub>Na<sub>4</sub>O<sub>12</sub>P<sub>3</sub>S<sub>2</sub>

**Molecular Weight:** 864.3 g/mol

**Structure:**



**Pharmacologic Class:** Platelet aggregation inhibitor, P2Y<sub>12</sub> receptor antagonist

**Route of administration:** Intravenous infusion

## 2.2 Dug Formulation

Cangrelor was formulated as tetrasodium salt with mannitol and Sorbitol USP as excipients, and reconstituted in NaCl 0.9% w/v solution.

### Impurities/Degradants

In the toxicology studies of cangrelor, the degradation products reported as percent impurities in the drug substance in various batches were [REDACTED] (b) (4)

Thus, levels above the reported percentage of impurities are considered to be toxicologically untested.

## 2.3 Clinical Indication

Cangrelor is indicated for the reduction of thrombotic cardiovascular events including stent thrombosis in patients with coronary artery disease undergoing PCI, as well as for bridging patients who need to discontinue oral P2Y<sub>12</sub> inhibitors prior to major cardiac surgery.

### 2.3.1 Proposed Dosing Regimens:

- i) For the PCI dosing with a bolus injection of 30 µg/kg followed by infusion of 4 µg/kg/min for 2 h,
- ii) For the Bridging dosing with 0.75 µg/kg/min IV infusion for up to 7 days after discontinuation of oral P2Y<sub>12</sub> inhibition prior to surgery.

### 2.3.2 Regulatory Background:

The original IND 56,812 was approved to proceed for clinical studies of cangrelor at a dose of 4 µg/kg/min infused for 24 hrs in patients undergoing PCTA with or without coronary stent implant based on a 3- and 4-fold safety margin over the one-month study in the rat and dog, respectively, using body weight analysis. In this NDA, a higher dose of cangrelor 30 µg/kg was used in the CHAMPION PHOENIX trial in a PCI setting.

## 3 Studies Submitted

**3.1 Studies Reviewed:** Sponsor's data on the pharmacology, pharmacokinetics, toxicokinetics, toxicology, genotoxicity and reproductive and developmental toxicology

studies on cangrelor tetrasodium salt (AR-C69931MX). In the original IND 56,812, the studies were performed using the triammonium salt (ARC69931MY), and later pharmacological testing and all ancillary and safety pharmacology studies were performed using the tetrasodium salt (AR-C69931MX).

### **3.2 Studies Not Reviewed:** None

**3.3 Previous Reviews Referenced:** Review of pharmacology and toxicology of the original IND 56,812 in 2/10/1999. In this NDA 204958, the preclinical studies relied on the previous data submitted in the IND 56,812 on August 1998 by Astra USA, Inc.

## **4 Pharmacology**

### **4.1 Primary Pharmacology**

Cangrelor is an intravenously administered short-acting, P2Y<sub>12</sub> receptor antagonist that blocks ADP-induced platelet aggregation intended for use in patients with coronary artery disease undergoing PCI, and for bridging patients who need to discontinue oral P2Y<sub>12</sub> inhibitors prior to surgery. Cangrelor, a synthetically derived agent structurally analogous to ATP, is as a selective P2Y<sub>12</sub> receptor antagonist.

### **4.2 Secondary Pharmacology**

Cangrelor had no effect on smooth muscle contraction induced by serotonin or angiotensin II in isolated perfused rabbit aortic rings, and did not affect histaminergic (H<sub>2</sub>) release from antigen-stimulated macaque monkey bronchiolar mast cells.

#### **4.2.1 Receptor screening assays**

Cangrelor did not exhibit agonist or antagonist activity toward other purinergic receptor subtypes (P2X<sub>1</sub>, P2X<sub>2</sub>, P2X<sub>7</sub>, P2Y<sub>1</sub> and P2Y<sub>2</sub>) at concentrations up to 30 μM. Cangrelor (100 μM) had no effect on aggregation of human washed platelets induced by a thromboxane A<sub>2</sub> mimetic U46619, or on the uptake of [<sup>3</sup>H]-adenosine by human erythrocytes. Cangrelor did not inhibit human neutrophil activation as measured by superoxide production in response to antigen stimulation, and did not interfere with binding of ligands to bovine pituitary dopaminergic (D2) receptors or inhibit adenosine uptake into erythrocytes.

#### **Pharmacological activities related to proposed indication of cangrelor**

#### **4.2.2 Inhibition of platelet aggregation**

(study # PR30144, PR30147)

In washed platelets from humans, cangrelor produced concentration-dependent inhibition of ADP-induced platelet aggregation with an IC<sub>50</sub> value of 0.45 nM in the presence or absence of the non-selective P1-purinoceptor antagonist, 8-p-sulphophenyl theophylline. The putative monophosphate metabolite of cangrelor (AR-C88558KP) showed very weak platelet inhibitor activity (IC<sub>50</sub> 360 nM). The base (AR-C71301XX) and S-oxidized base (ARC90441XX) metabolites had no inhibitory effects on platelets at concentrations up to 10 μM. In whole blood from the rat, dog and human, cangrelor exhibited concentration-related inhibition of platelet aggregation with IC<sub>50</sub> 5.1, 0.72, and 0.71 nM, respectively, measured as maximum change in impedance (ohms) within 6 minutes of addition of ADP.

**4.2.3 Ex vivo platelet aggregation in blood from rats and dogs** (study # PR30148)

In anesthetized rats, three 20 minute IV infusion of cangrelor (b) (4) salt (b) (4) produced dose-related inhibition of ADP (3 μM) induced platelet aggregation with an ID<sub>50</sub> (dose that inhibits 50%) of 256 ng/kg/min, which is approximately 11.7-fold lower than the NOAEL (3 μg/kg/min) in the 28-day toxicity study in rats (Figure 1). ADP-induced aggregation returned substantially toward pre-infusion level (79%) within 20 minutes of cessation of infusion, but still remained significantly inhibited in the high dose of 7700 ng/kg/min (Figure 1). In whole blood isolated from dogs pretreated with cangrelor, dose-related inhibition of platelet aggregation was observed with an ID<sub>50</sub> of 15 ng/kg/min, which is 250-fold lower than the NOAEL (3.75 μg/kg/min, C<sub>ss</sub> 61 ng/ml) in the 28-day toxicity study in dogs using body weight analysis. However, inhibition of platelet aggregation is anticipated to occur at lower dosages than at the ID<sub>50</sub> or NOAEL because the half-maximal inhibitory concentration of cangrelor for platelet aggregation in the dog is 0.72 nM (0.62 ng/ml).

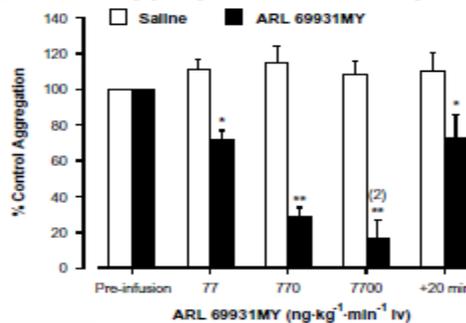


Figure 1: Effects of cangrelor (b) (4) salt on ADP-induced platelet aggregation in whole blood from rats

**4.2.4 In vivo inhibition of thrombosis** (study # PR30147)

In anaesthetized mongrel dog model of stenosed femoral artery induced by clamping for 5 minutes, cangrelor exhibited dose-related inhibition of thrombosis as measured by inhibition of cyclic blood flow, with increase in prolongation of bleeding time (Figure 2). Cangrelor inhibited thrombosis by ~90% at a dosage of 200 ng/kg/min, a dosage associated with complete inhibition of platelet aggregation. The antithrombotic activity of cangrelor quickly diminished upon cessation of infusion with restoration of platelet aggregation becoming apparent within 10 - 15 minutes post-infusion, indicating the short action of cangrelor.

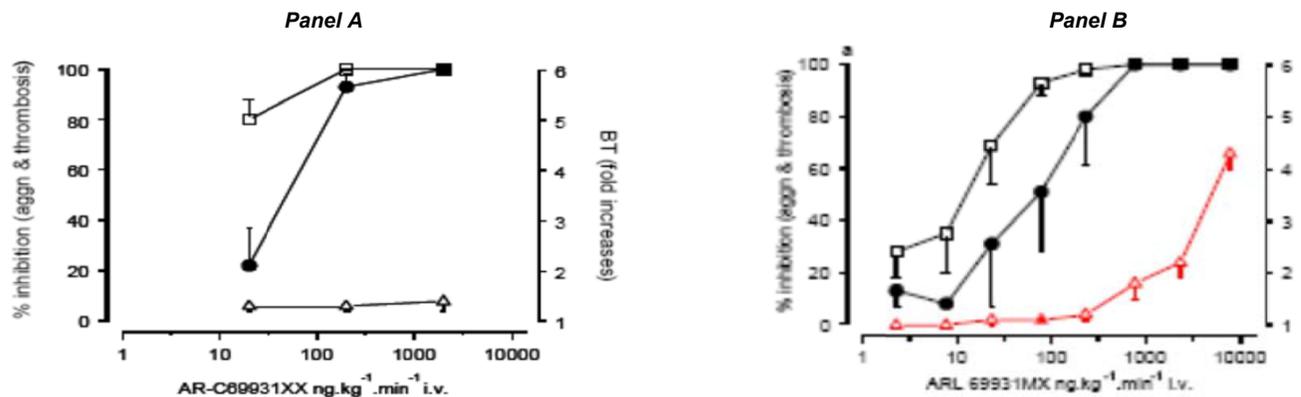


Figure 2: Effects of IV infusion of cangrelor on ADP-induced platelet aggregation (□), thrombosis (●) and bleeding time (Δ) in dogs

#### 4.2.5 Platelet recovery following cessation of cangrelor in dogs (study # SE 9861)

In conscious beagle dogs infused continuously with cangrelor (40 and 60  $\mu\text{g}/\text{kg}/\text{min}$ ) for 7 days, platelet aggregation in response to ADP was abolished at both doses, with corresponding plasma concentration of 2 - 5  $\mu\text{g}/\text{ml}$  (Figure 3). Following cessation of infusion at 40  $\mu\text{g}/\text{kg}/\text{min}$ , aggregation response recovered by at least 50% after 10 minutes, with full recovery evident within 1 - 2 hours, with no evidence of a “rebound” increase in platelet responsiveness to ADP over the 2 hour post-infusion period. In contrast, platelet inhibition was sustained for up to 120 min at the high dose (60  $\mu\text{g}/\text{kg}/\text{min}$ ). This indicates that the off-rate receptor kinetics for cangrelor is dose-dependent. Thus, following infusion of low dosages of cangrelor (reversible inhibitor, short  $t_{1/2}$ ), transitioning to orally active and irreversible thienopyridine P2Y<sub>12</sub> receptor inhibitors within 60 minutes after cessation of cangrelor may assure completion of on-off receptor kinetics.

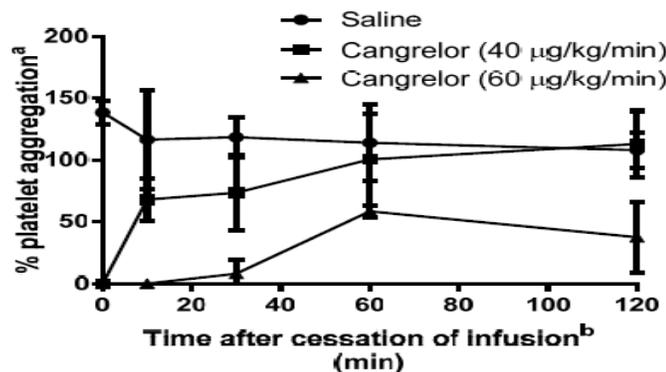


Figure 3: *Ex vivo* platelet aggregation (Ohms) in whole blood following infusion ARL 69931MX (40 or 60  $\mu\text{g}/\text{kg}/\text{min}$ ) for 7 days in conscious dog (n=2/group).

#### 4.2.6 Effects of cangrelor on thrombolysis therapy

In the dog model of occlusive thrombus and stenosis in the left circumflex coronary artery induced by electrical stimulation, no acceleration in t-PA-induced thrombolysis or increased reperfusion rate was observed in the cangrelor group (4  $\mu\text{g}/\text{kg}/\text{min}$  IV for 2 hours) compared with the placebo group. However, there were differences in the incidences of re-occlusion and cyclic flow variation between the groups. When administered as an adjuvant following administration of thrombolytic agent (t-PA, 1 mg/kg over 20 min), cangrelor reduced re-occlusion and cyclic blood flow in the coronary artery and reduced myocardial infarct size by approximately 50% compared to t-PA alone. In the cangrelor-treated dogs, there was an increase in coagulation parameters (aPTT and PT) relative to vehicle that was evident 2 hours post-reperfusion and was associated with an increase in gum bleeding time.

### 4.3 Safety Pharmacology

#### 4.3.1 Neurological effects

Behavioral study in conscious mice to infusion of cangrelor 200 mg/kg over 10 minutes (HED 16 mg/kg) showed transient increase in motor activity appearing less than 1 hour after the infusion and resolving  $\sim$ 1 hour after infusion (n=3-5). At high dosage of 400 mg/kg infusion over 10 minutes, marked features of CNS depression were noted consisting of depressed and poorly coordinated motor activities, respiratory depression, wire maneuver failure and hypothermia, lasting for up to 1 hour after the infusion. The dosages of cangrelor used in this

study were several orders of magnitude higher than the dosages that maximally inhibited platelet aggregation based on body weight analysis.

#### 4.3.2 Thermoregulation

In conscious male CD-1 mice, IV infusion of tri-ammonium salt of cangrelor (AR-C69931MY) 58 and 580 mg/kg/min (HED 4.7 and 47 mg/kg) for 10 minutes, produced dose-related hypothermia of  $-2.7^{\circ}\text{C}$  and  $-3.6^{\circ}\text{C}$ , respectively, which reached maximal at 0.5 hours post-infusion (n=2-4). The maximum fall in rectal temperature observed was  $-5.5^{\circ}\text{C}$  in the high dose group. Near complete recovery of rectal temperature was observed at 1 hour in the 58 mg/kg group, and 2 hours in the 580 mg/kg group. High doses of cangrelor could cause hypothermia, an effect mediated by its metabolite adenosine via adenosine A<sub>1</sub> receptor subtype in the CNS.

#### 4.3.3 Cardiovascular effects

There were no treatment related effects of cangrelor (30  $\mu\text{g}/\text{kg}/\text{min}$  for 6 h) on the cardiovascular system parameters including phasic and mean blood pressure, heart rate, and electrocardiography (ECG) in anesthetized rats and dogs. Telemetry studies did not show any adverse effects on hemodynamic and cardiovascular parameters including QRS and QT intervals. There were no treatment-related effects on the contraction of the nictitating membrane induced by cervico-sympathetic preganglionic nerve stimulation; heart rate as a result of vagal stimulation induced bradycardia; and blood pressure resulting from a baroreceptor-induced rise during bilateral carotid occlusion. In instrumented anesthetized male beagle dogs, cangrelor doses up to 6  $\mu\text{g}/\text{kg}/\text{min}$  (HED 3.2  $\mu\text{g}/\text{kg}/\text{min}$ ) for 30 min had no effects on blood pressure, heart rate, stroke volume, and total peripheral resistance.

#### 4.3.4 Pulmonary effects

There were no effects of cangrelor (200  $\mu\text{g}/\text{kg}/\text{min}$  for 30 min) on respiratory system parameters including respiratory flow, rate and tidal volume in anesthetized Sprague-Dawley rats. In a single dose study in the CD-1 mice, changes in the rate and depth of respiration were observed at dosage of 50 mg/kg of cangrelor (HED 4 mg/kg).

## 5 Pharmacokinetics/ADME/Toxicokinetics

### 5.1 PK/ADME

**5.1.1 Plasma protein binding:** *In vitro* binding of [<sup>3</sup>H]-cangrelor to rat, dog and human plasma proteins and blood cells was 97%, 93%, and 98%, respectively (Table 1). At plasma concentrations of 20 - 400 ng/mL, cangrelor was highly bound to plasma proteins and was not distributed inside red blood cells. Binding of [<sup>3</sup>H]-cangrelor to human plasma proteins was not affected by co-incubation with various drugs that are known to displace anionic compounds from serum albumin. The metabolite, [<sup>3</sup>H]-AR-C69712XX exhibited less plasma protein binding than the parent compound, with 85% bound in the dog and 89% bound in the rat and human.

Table 1: Binding to plasma protein blood cells in rats, dogs and humans

Species	Concentrations Tested (ng/mL)	% Bound			
		$^3\text{H}$ Cangrelor		$^3\text{H}$ AR-C69712XX	
		Plasma Protein <sup>a</sup>	Blood Cell <sup>b</sup>	Plasma Protein <sup>a</sup>	Blood Cell <sup>a</sup>
Rat (Sprague-Dawley)	20, 100, 400	97.4 (96.7 to 97.9)	0 to 7.7	88.7 (87.8 to 89.4)	35.8 (33.4 to 37.5)
Dog (Beagle)	20, 100, 400	92.6 (92.1 to 93.2)	0 to 1.3	85.4 (84.8 to 86.0)	27.4 (26.7 to 28.7)
Human	20, 100, 400	97.8 (97.3 to 98.1)	0 to 5.0	88.8 (88.5 to 89.2)	31.3 (28.0 to 33.8)

AR-C69712XX = cangrelor metabolite

### 5.1.2 Absorption

Steady state plasma levels of cangrelor were attained at the time the first blood sample was taken (10 - 30 minutes in rats, 10 - 60 minutes in dogs), and remained stable until the infusion was stopped. Cangrelor infused over a long period (72 hrs) at a high enough concentration (20  $\mu\text{g}/\text{kg}/\text{min}$ ), the drop in plasma concentration after the infusion was stopped contained two distinct phases, a rapid decline followed by a much slower decline.

### 5.1.3 Distribution

Tissue distribution studies of radiolabeled  $^3\text{H}$ -cangrelor and metabolites in rats indicated localization to highly perfused organs such as liver, kidney, heart, lung, and spleen. Radioactivity levels decreased with time and by 6 hours post-infusion, levels were at or close to background. Qualitative assessment of whole body autoradiograms indicated that in the kidney, high levels of radioactivity were observed in the cortico-medullary junction at the end of infusion (Table 2). At 10 minutes post-infusion, higher levels of radioactivity were observed in the cortex and pyramids compared to the medulla. By 6 hours post-infusion, low levels of radioactivity were uniformly distributed within the kidney, lower levels remaining in the cortex up to 24 hours post-infusion. High concentrations of radioactivity were also observed in the liver, stomach, and gastrointestinal tract (Table 2). The presence of high levels of radiolabel in the small intestine at early times post infusion suggest biliary elimination as a route of excretion. There is no evidence of retention of compound-related material from administration of  $^3\text{H}$ -cangrelor as the majority of radiolabel is excreted within 24 hours after infusion.

Table 2: Tissue distribution of single IV infusion of  $^3\text{H}$ -cangrelor 12  $\mu\text{g}/\text{kg}$  in rats

Tissue	Radiolabel Concentration (ng-equiv/g)					
	0 min	10 min	1 hr	6 hr	24 hr	168 hr
Kidney	1168.52	894.23	426.40	249.11	33.99	2.47
Liver	1234.93	727.06	247.47	108.07	12.72	1.98
Stomach	182.15	126.27	62.36	19.26	6.35	0.23
Small Intestine	2105.94	4109.14	5357.76	185.41	7.05	0.23
Large Intestine	1365.98	126.00	71.44	17.56	11.21	0.70

### 5.1.4 Metabolism

Cangrelor is rapidly metabolized similarly in rats, dogs and humans by de-phosphorylation to an inactive nucleoside metabolite, AR-C69712XX (Figure 4). The initial inactivation step of de-phosphorylation is mediated through ectonucleotidases in the vascular endothelium, followed by metabolism to various products, mainly sulfoxides. The main metabolites detected in feces were the purine base (AR-C71301XX) and its S-oxide (AR-C90441XX) and mass spectrometric analysis indicated a component which may be either the di-S-oxide or hydroxylated S-oxide of the base (AR-C71301XX). A major component in urine was AR-C90439XX, an S-oxide of AR-C69712XX, the nucleoside metabolite of cangrelor (Figure 4). The metabolic profile of cangrelor in animals is similar to that in humans, and there are no metabolites specific to humans that needed to be qualified in separate toxicity studies. There was no evidence of inhibition or induction of cytochrome P450 isoenzyme systems by cangrelor or its metabolites, indicating that cangrelor may not interfere with the hepatic metabolism of other concomitantly administered drugs.

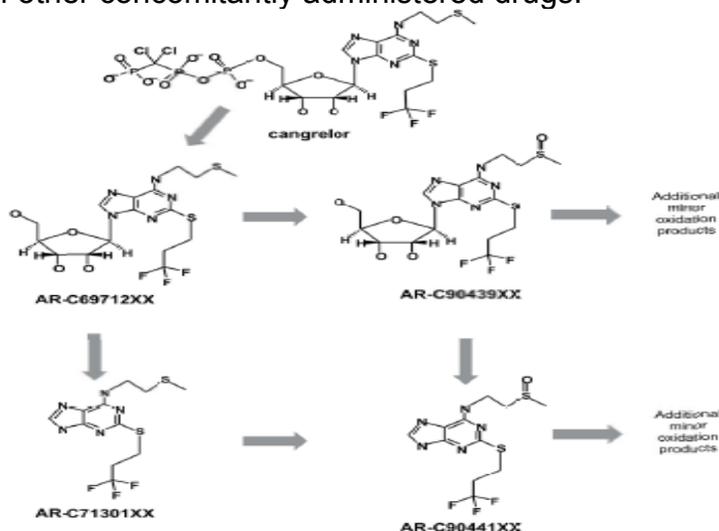


Figure 4. Postulated major routes of metabolism of cangrelor in rats, dogs and humans.

### 5.1.5 Excretion

Biliary elimination is the primary route of excretion as evident by the recovery of the majority of drug in the feces. Approximately 65% of the dose of [<sup>3</sup>H]-cangrelor is recovered in feces by 24 hours and 80% by 48 hours. The kidneys are responsible for removal of the remaining drug with 15% of the total dose recovered in the urine within 24 - 48 hours post-dose. Elimination was biphasic with an initial half-life of 1 - 2 minutes and a less defined terminal half-life ranging from 1 - 4 hours in rats and dogs. In rabbits, the initial elimination half-life was ~10-fold longer (26 minutes) than rats and dogs. However, the terminal half-life was similar (3 - 4 hours) (Table 3). Approximately 90% of a total dose of cangrelor was cleared from the plasma during the initial elimination phase. This was followed by a more prolonged terminal elimination half-life for the remaining 10% of infused drug, and the metabolite AR-C69712XX is apparent but contributes little to systemic exposures. No unchanged drug was detected in excreta. A major component in urine was AR-C90439XX, the S-oxide derivative of AR-C69712XX, the nucleoside metabolite of cangrelor.

Table 3: Pharmacokinetics of IV infused cangrelor in the rat and dog

Species	Rat (Sprague-Dawley)			Dog (Beagle)			
	Gender (# animals)	Male (n=6)	Male (n=38)	Female (n=22)	Male (n=3)	Male (n=3)	Female (n=3)
Dose ( $\mu\text{g}/\text{kg}/\text{min}$ )		4.8	48	48	6	60	60
Duration of Infusion (h)		1.0	1.0	0.5	0.5	4.0	1.0
C <sub>ss</sub> (ng/mL)		277	2013	2080	110	1021	845
CL <sub>p</sub> (mL/min/kg)		—	25.4	22.3	—	59	74
Initial t <sub>1/2</sub> (min)		—	0.75	<2	—	<1	0.6
Terminal t <sub>1/2</sub> (min)		—	180	45.5	—	354	61
Vd <sub>ss</sub> (L/kg)		—	6.03	1.47	—	2.8	0.5
MRT (min)		—	29.2	12.3	—	47	6.5
AUC <sub>0-∞</sub> (ng·hr/mL)		—	1892	1075	—	4316	819

C<sub>ss</sub> = steady-state plasma concentration; CL<sub>p</sub> = plasma clearance; t<sub>1/2</sub> = half-life;

Vd<sub>ss</sub> = steady-state volume of distribution; MRT = mean residence time;

AUC<sub>0-∞</sub> = area under the concentration curve from administration to last measured concentration extrapolated to infinity.

## 5.2 Toxicokinetics

The AUC was computed as AUC<sub>1h-t</sub> by using the appropriate plasma concentration (C<sub>ss</sub>) value and GraphPad Prism 5.00 to calculate exposure for the total duration of drug administration. The C<sub>ss</sub> values appear to represent the dosing regimen in the PCI setting which is injected as a bolus. In the PCI settings (CHAMPION PHOENIX trial), bolus injection of cangrelor 30  $\mu\text{g}/\text{kg}$  followed by 4  $\mu\text{g}/\text{kg}/\text{min}$  infusion for 2 h achieved C<sub>ss</sub> of 488 ng/ml which is estimated to be about 8-fold higher than the plasma concentration at the NOAEL in the dog infused for 4 weeks (C<sub>ss</sub> 61.5 ng/ml). In the Bridge dosing of cangrelor 0.75  $\mu\text{g}/\text{kg}/\text{min}$  infusion for 7 days achieved AUC<sub>0-7d</sub> 11189 ng·h/ml, which is approximately 4.5-fold lower than the exposures at the NOAEL in the dog infused with cangrelor 3.5  $\mu\text{g}/\text{kg}/\text{min}$  for 28 days (AUC<sub>0-28d</sub> 53957 ng·h/mL (Table 4).

Table 4. Exposures following IV administration of cangrelor for 28 days in rat and dog.

Rat						Dog					
Cangrelor			AR-C69712XX			Cangrelor			AR-C69712XX		
Dose	C <sub>ss</sub>	AUC <sub>0-28d</sub>									
$\mu\text{g}/\text{kg}/\text{min}$	ng/ml	ng·h/ml									
3	113.3	46565	3	25.6	17027	3.75	61.5	53957	3.75	31	228805
12	314	371825	12	102.4	89307	15	242	165627	15	121	87424
48	1939	1204732	48	402.6	301630	60	992	884405	60	586	407409

C<sub>ss</sub> = steady-state plasma concentration, Human Bridge dosing of cangrelor 0.75  $\mu\text{g}/\text{kg}/\text{min}$  for 7 days achieved C<sub>ss</sub> 67 ng/ml and AUC<sub>0-7d</sub> 11189 ng·h/ml, and for the metabolite AR-C69712XX C<sub>ss</sub> of 38 ng/ml and AUC<sub>0-7d</sub> 5471 ng·h/ml Human PCI dosing of cangrelor 30  $\mu\text{g}/\text{kg}$  bolus + 4  $\mu\text{g}/\text{kg}/\text{min}$  for 2 hr achieved C<sub>ss</sub> of 488 ng/ml and AUC<sub>1h-2h</sub> 1008 ng·h/ml, AUC<sub>1h-t</sub> = area under the concentration curve from 1 h post-administration to last observed concentration at t.

## 6 General Toxicology

### 6.1 Single-Dose Toxicity

#### 6.1.1 Single dose toxicity study of cangrelor in mice

Study no.:	SE 9994 / 96031
Study report location:	Astra Safety Assessment, Astra Charnwood, UK
Date of study initiation / completion:	May 9, 1996 / May 23, 1996
GLP compliance:	Yes, QA statement: ✓
Drug:	Cangrelor tetrasodium salt, Batch No: 3879F, 3960G
Vehicle	0.9% saline
Dosages	64.4, 128.8, 257.6, 515.2 $\mu\text{mol/kg}$ , Equivalent to 50, 100, 200, 280, 400 mg/kg
Route	IV bolus, volume 4 mL/kg, rate 0.4 mL/min
Species	CD-1 mice, Age: 3 - 5 weeks old
Weight:	Males 16 – 23 g, Females 17 – 22 g
Number/sex/group:	2-3/sex/dose group

**Mortality:** Deaths occurred immediately after dosing in all males and females in the 400 mg/kg dosage group and in one male and one female in the 280 mg/kg dosage group. The deaths were preceded by clonic convulsions and dyspnea.

**Clinical Signs:** Treatment related clinical observations included decreased motor activity, changes in the rate and depth of respiration, piloerection, and partially closed eyes at dosages >50 mg/kg. In most animals a bluish discoloration occurred after dosing at the site of injection, which disappeared gradually except in the surviving high dose animal where the lesion became necrotic.

**Body Weights:** A decrease in body weight gain was observed in all groups. The treated males recovered during the observation period but not as fast as control animals and treated females gained weight slower than their control counterparts, which resulted in lower mean body weights at termination.

**Gross Pathology:** At necropsy on Day 15, there were no treatment related findings except slight irritation at the injection site (tail vein).

**Histopathology:** There were no histopathological findings observed in any tissues, including the kidney. Minimal focal vacuolation of hepatocytes, loss of nuclear structure degenerative changes in the centrilobular region were observed at doses  $\geq 200$  mg/kg.

**Toxicokinetics:** Not performed.

**Summary:** Treatment-related clinical observations included decreased locomotion, irregular respiration, half-closed eyes, hunched posture, and piloerection at dosages >50 mg/kg. Typically, signs were transient and developed within 30 minutes after the start of dosing with recovery occurring within 1 - 2 hours after dosing. The lowest observed effect level in the single dose study in mice is considered to be 50 mg/kg (HED 4 mg/kg).

### 6.1.2 Single dose toxicity study of cangrelor in rats

Study no.:	SE9993 / 97093		
Study report location:	Astra Safety Assessment, Astra Charnwood, UK		
Date of study initiation / completion:	Sept 26, 1995 and May 8, 1996 / Oct 10, 1995 and May 22, 1996		
GLP compliance:	Yes,	QA statement:	✓
Drug:	Cangrelor tetrasodium salt,	Batch No:	3879F
Vehicle	0.9% NaCl		
Dosages	50, 100, 200, 400 or 800 mg/kg		
Route	IV, volume 4 mL/kg, rate 0.4 mL/min		
Species	Sprague-Dawley rat, Age: 5 - 7 weeks old		
Weight:	Males 170 – 210 g, Females 152 – 173 g		
Number of mice:	2/sex/dose group		

**Mortality:** At  $\geq 400$  mg/kg, death occurred during or immediately after dosing. At necropsy, pale spleens and congested lungs were in one of the animals, but the cause of death was not investigated.

**Body weights:** There was a 20% reduction in body weight gain in animals in the 200 mg/kg dosage group compared to controls.

**Clinical signs:** Clinical observations included decreased locomotion, increased depth of respiration, reduction in body temperature, hunched posture, dropping of eye lids and piloerection at doses  $\geq 100$  mg/kg. Typically, signs were transient and developed within 30 minutes after the start of dosing with recovery occurring within by 2 hours after dosing.

**Gross Pathology:** At necropsy (Day 15), there were no treatment-related macroscopic observations.

**Histopathology:** Microscopic changes in kidneys including basophilic tubules, cortical tubular degeneration, interstitial mononuclear cell infiltrates, tubules distended with colloid at  $\geq 100$  mg/kg. The presence of the tubular basophilia may be indicative of a regenerative response to an earlier injury.

**Toxicokinetics:** Not performed

**Key Study Findings:** Clinical observations included decreased locomotion, irregular respiration, decreased body temperature, hunched posture, dropping of eye lids and piloerection at dosages  $\geq 100$  mg/kg. Typically, signs were transient and developed within 30 minutes after the start of dosing with recovery occurring within 1 - 2 hours after dosing. The lowest observed effect level in the single dose study in the rat is considered to be 50 mg/kg (HED 8.1 mg/kg).

## 6.2 Repeat-Dose Toxicity

### 6.2.1. 28-Day repeat dose study of cangrelor in rats

Study no.:	SE9948 / SE10176
Study report location:	Astra Safety Assessment, Astra Charnwood, UK
Date of study initiation / completion:	7/27/1995 – 9/27/1995, 4/30/1996 - 5/31/1996
GLP compliance:	Yes, QA statement: $\checkmark$
Drug:	Cangrelor, Batch #: 3879F, 3960G, Purity: 98.8% w/w
Formulation/Vehicle:	0.9% NaCl
Route	IV, continuous infusion via femoral vein cannula
Dosages	3, 12, or 48 $\mu\text{g}/\text{kg}/\text{min}$ (equivalent to 4.32, 17.3, 69.1 mg/kg/d)
Species	Sprague-Dawley rat, Age: 10 – 12 weeks
Weight:	Males 180 – 229 g, Females 158 - 181 g
Number of rats:	9/sex/dose group, Satellite: 3/sex/dose group for TK

**Mortality:** In the high dosage group, mortality at Day 13 was associated with high levels of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) levels.

**Body Weights:** There was a treatment-related decrease (~4%) in body weight in the high dosage group on Day 29, but were comparable during the recovery period in all groups.

**Food Consumption:** There was a reduction of ~2% in food consumption in the high dose group during weeks 3 and 4, but were comparable during recovery period.

**Clinical Signs:** Observations including blood in urine, depressed activity, hunched posture, half shut eyes, cold extremities were more apparent in the high dose group.

**Clinical Chemistry:** Plasma clinical chemistry changes included increases in AST and ALT by 2- to 3-fold, increases in urea, and reduced triglyceride levels in the high dose group. One rat in the 3  $\mu\text{g}/\text{kg}/\text{min}$  and four rats in the 48  $\mu\text{g}/\text{kg}/\text{min}$  group showed altered protein electrophoresis patterns, including increased albumin and additional protein bands, and proteinuria indicating altered renal function. In the high dosage group, increased celluria was observed that may be related to urinary tract inflammation and bladder hemorrhage.

**Hematology:** There were increased incidences of red blood cells, white blood cells and epithelial cells in urine sediment at 48  $\mu\text{g}/\text{kg}/\text{min}$ , indicating inflammation of the urinary tract. Neither drug-related changes in hematology and coagulation parameters nor increased bleeding due to cangrelor's pharmacology were observed.

**Organ Weights:** Decrease in liver weight, increase adrenal and reduced thymus weights, and thymic involution were observed in the high dose group.

**Histopathology:** Adequate battery: yes ( $\checkmark$ ), no ( )

The urinary tract was the predominant target characterized by inflammatory and necrotic changes, associated urothelial hyperplasia in the renal pelvis and ureter in the high dose group. Injury to the urinary tract was reversible following cessation of infusion. Inflammatory changes in various organs along with local inflammation, abscess formation, and/or thrombi at the infusion site occurred at a high incidence in all dose groups and control, indicating that the kidney and lower urinary tract and liver were primary target organs. In the high dosage group, microscopic changes include marked pyelonephritis and ureteritis in two rats, hemorrhage and congestion or distention of the bladder, liver necrosis

in three rats (focal in two rats and multilobular in one rat). No histological changes to the liver were observed in rats in the 3 or 12 µg/kg/min dosage groups (Appendix Tabulated Summary 1, 2).

**Toxicokinetics:** Cangrelor plasma levels exhibited very wide variations within the dosage groups on the same day and from the same animal over time (Table 5). Systemic exposure was dosage proportional and increased linearly with increasing dose. A 16-fold increase from the low to high dosage group was associated with a 13- to 18-fold increase in cangrelor exposures. AR-C69712XX was detected in all plasma samples from animals dosed with test article and appeared to be at steady state throughout the sampling times.

Table 5. Steady-state plasma concentrations (C<sub>ss</sub>, *Panel A*) and AUC (*Panel B*) of cangrelor and metabolite AR-C69712XX in rats infused for 29 days

**Panel A: Plasma concentration C<sub>ss</sub> in rats**

Cangrelor C <sub>ss</sub> ng/ml, Male rats				Cangrelor C <sub>ss</sub> ng/ml, Female rats				Mean M + F
Dose µg/kg/min	3	8	29	Dose µg/kg/min	3	8	29	
3	206	230	57.2	3	89.2	21.2	76.7	113.3
12	341	304	339	12	74.8	95	731	314
48	2600	4300	887	48	815	455	2580	1939

AR-C69712XX C <sub>ss</sub> ng/ml, Male rats				AR-C69712XX C <sub>ss</sub> ng/ml, Female rats				Mean M + F
Dose µg/kg/min	3	8	29	Dose µg/kg/min	3	8	29	
3	22.6	21.8	19.6	3	30.7	30.2	28.8	25.6
12	91.9	112	126	12	79.2	74.6	131	102.4
48	330	480	347	48	314	424	521	402.6

**Panel B: AUC<sub>0-28d</sub> in rats**

Dose µg/kg/min	Cangrelor			Dose µg/kg/min	AR-C69712XX		
	Male AUC <sub>0-28d</sub> ng.h/ml	Female AUC <sub>0-28d</sub> ng.h/ml	Mean M + F		Male AUC <sub>0-28d</sub> ng.h/ml	Female AUC <sub>0-28d</sub> ng.h/ml	Mean M + F
3	39615	53515	46565	3	13900	20155	17027
12	235605	508045	371825	12	87570	91045	89307
48	616465	1793000	1204732	48	241165	362095	301630

**Key Study Findings:** The primary adverse effects from continuous IV infusion of cangrelor in both male and female rats occurred in the kidney, urinary tract and liver (Appendix Tabulated Summary 1 and 2). Severe pyelonephritis and ureteritis was observed in the high dosage group along with several animals exhibiting congestion or distension and hemorrhage of the bladder and distension of the ureters. The high dose group exhibited marked hepatic necrosis accompanied by increases in AST and ALT that were accompanied by increases in urea, triglyceride and cholesterol levels. Renal functional changes of elevated plasma urea levels, increased proteinuria, and abnormal molecular weight proteins in the urine were detected at dosages ≥12 µg/kg/min.

**Margin of Safety Analysis:** The NOAEL in rats infused for 28 days is considered to be 3 µg/kg/min (C<sub>ss</sub> 113 ng/ml, AUC<sub>0-29d</sub> 46565 ng\*h/ml), which is estimated to be 1.7- and 4-fold higher than the plasma concentration of C<sub>ss</sub> 67 ng/ml and AUC<sub>0-7d</sub> 11189 ng\*h/ml, respectively, obtained in human Bridge settings from infusion of cangrelor 0.75 µg/kg/min for 7 days. The NOAEL identified in the rat (C<sub>ss</sub> 113 ng/ml) is estimated to be about 4.5-fold lower than the human PCI dosing of cangrelor 30 µg/kg IV bolus followed by 4 µg/kg/min for 2 hr infusion (C<sub>ss</sub> 488 ng/ml).

### 6.2.2. 28-Day repeat dose toxicity study of cangrelor in beagle dogs

Study no.: SE9862 / SE10177  
 Study report location: Astra Safety Assessment, Astra Charnwood, UK  
 Date of study initiation / completion: 8/22/1995 – 9/21/1995, 4/23/1996 – 5/22/1996  
 GLP compliance: Yes      QA statement: ✓  
 Drug: Cangrelor, Batch #: 3879F, 3960G, Purity: 92.91%, 98.8% w/w  
 Dosages: 3.75, 15, 60 µg/kg/min (equivalent to 5.4, 21.6, 86.4 mg/kg/d)  
 Route of administration: IV infusion, continuous  
 Formulation/Vehicle: 0.9% NaCl  
 Species/Strain: Beagle dogs,      Age: 4 – 5 months  
 Weight: Males 7.6 - 10.4 kg, Females 6.5 – 9.3 kg  
 Number of dogs: 3/sex/dose group

**Mortality:** There were no unscheduled deaths.

**Body Weights:** There was no consistent dose-related effect on body weight gain, except a weight loss of 0.6 kg was recorded for one dog in the high dosage group.

**Food Consumption:** There were no effects of treatment on food consumption, except in one dog in the high dose which was reduced on day 24 and had blood in vomit.

**Clinical Signs:** Asynchronous premature ventricular contractions on Days 15 and 22, but not on Day 29, were noted in the high dosage group. Ophthalmological examination revealed slight changes in pigmentation and coloration of the fundus of both eyes in two animals in the 60 µg/kg/min dosage group and one animal in the 15 µg/kg/min dosage group. Retinal blood vessels engorged and undulating tortuous blood vessels were noted at doses ≥ 15 µg/kg/min. However, no functional or behavioral perturbations related to ocular function and no histological changes in the eye were observed.

**Clinical Chemistry:** Increased levels of aminotransferases (AST, ALT) were observed at 15 and 60 µg/kg/min dosage levels and increases in creatine kinase by 2-fold were observed at the 60 µg/kg/min dosage level. However, there were no associated histopathological changes in the liver or muscle that were cangrelor related. Renal functional changes at dosages ≥ 15 µg/kg/min were indicated by elevated plasma urea (1.5-fold) and creatinine levels (1.5-fold) and urinary N-acetyl-β-D-glucosaminidase (β-NAG, 3-fold) and proteinuria (10-fold), but the effect at 15 µg/kg/min was not associated with any histological evidence of renal toxicity (Appendix Tabulated Summary 3 and 4). One animal in the high dosage group had elevated plasma urea, indicating loss of renal function.

**Hematology:** Neutrophilia with a consequent increase in white cell count, increase in fibrinogen level (an acute phase protein), increase in platelet count and decreased hemoglobin levels were observed at dosages > 15 µg/kg/min.

**Organ Weights:** The 60 µg/kg/min dosage group exhibited decreases in absolute and relative mean spleen and kidney weights by up to 47% and 10%, respectively. No other effects on organ weights were observed.

**Histopathology:** Adequate battery: yes (✓), no ( )

In the high dosage group (60 µg/kg/min), cangrelor produced histological evidence of renal toxicity characterized by pelvic inflammation, urothelial necrosis, tubular necrosis, tubular

regeneration, basophilic tubules, interstitial nephritis, urothelial hyperplasia, and tubular dilatation. Tubular dysfunction was also indicated by the presence of glucosuria and proteinuria. Various changes were observed in the urinary bladder, including necrosis, ulceration, abscesses, transitional cell hyperplasia, inflammatory cell foci and cystitis, which may be linked to urine sampling catheter damage to the wall of the bladder. In the high dose group, gastritis was observed in the stomach with minimal erosion/ulceration and multifocal necrosis of the gastric mucosa. One dog in the 60 µg/kg/min dosage group also exhibited inflammatory changes in the large bowel (cecum, colon, rectum) that may be suggestive of an infection rather than a test article-related effect, but also reminiscent of urinary bladder and gastric mucosal inflammation. There were no histological findings in the liver that support a toxicological association with the increase in plasma enzymes (Appendix Tabulated Summary 3, 4).

**Toxicokinetics:** Systemic exposures increased with increase in dose of cangrelor but were variable (Table 6). The plasma concentrations of AR-C69712XX were less variable than cangrelor within each dosage group and were apparently at steady state throughout the times of sampling. The metabolite increased linearly with dosage and did not show changes within dosage groups with time, and represents ~60% of the concentration of parent drug.

Table 6. Steady state plasma concentrations (C<sub>ss</sub>, *Panel A*) and AUC (*Panel B*) of cangrelor and metabolite AR-C69712XX in dogs infused for 28 days

**Panel A: Plasma concentration C<sub>ss</sub> in Dogs**

Cangrelor C <sub>ss</sub> ng/ml, Male dogs				Cangrelor C <sub>ss</sub> ng/ml, Female dogs				Mean
Dose	Day			Dose	Day			
µg/kg/min	2	15	28	µg/kg/min	2	15	28	M + F
3.75	42.2	63.9	81.6	3.75	65	43.5	76.7	61.5
15	284	240	273	15	228	215	212	242
60	694	1150	1520	60	816	699	1070	992

AR-C69712XX C <sub>ss</sub> ng/ml, Male dogs				AR-C69712XX C <sub>ss</sub> ng/ml, Female dogs				Mean
Dose	Day			Dose	Day			
µg/kg/min	2	15	28	µg/kg/min	2	15	28	M + F
3.75	36.5	44.8	43.3	3.75	21	17	24	31
15	135	143	157	15	96	96	99	121
60	709	692	656	60	442	482	537	586

**Panel B: AUC<sub>0-28d</sub> in Dogs**

Dose	Cangrelor			Dose	AR-C69712XX		
	Male	Female	ng.h/ml		Male	Female	ng.h/ml
µg/kg/min	AUC <sub>0-28d</sub>	AUC <sub>0-28d</sub>	Mean	µg/kg/min	AUC <sub>0-28d</sub>	AUC <sub>0-28d</sub>	Mean
3.75	56006	51908	53957	3.75	29369	16392	228805
15	186459	144796	165627	15	107231	67617	87424
60	1038000	730810	884405	60	448048	366771	407409

**Key Study Findings:** Continuous administration of cangrelor by IV infusion for one month to the male and female beagle dogs showed histological evidence of renal toxicity characterized by pelvic inflammation, urothelial necrosis, tubular necrosis, tubular regeneration, basophilic tubules, interstitial nephritis, urothelial hyperplasia and tubular dilatation at the high dosage (Appendix Tabulated Summary 3 and 4). Renal functional disturbances were also evident by increase in plasma creatinine levels, urinary β-NAG and presence of proteinuria at doses ≥15 µg/kg/min. Tubular dysfunction was also indicated by

the presence of glucosuria and proteinuria. In the high dose group, an increase in severity and frequency of asynchronous premature ventricular contractions were also observed.

**Margin of Safety Analysis:** The NOAEL dosage in dogs infused for 28 days is considered to be 3.75 µg/kg/min (C<sub>ss</sub> 61.5 ng/ml, AUC<sub>0-28d</sub> 53957 ng\*h/ml), which is approximately the same as plasma concentration of C<sub>ss</sub> 67 ng/ml and 4.8-fold higher than the AUC<sub>0-7d</sub> 11189 ng\*h/ml obtained in the human Bridge dosing of cangrelor 0.75 µg/kg/min infused for 7 days. The C<sub>ss</sub> of 61.5 ng/ml identified in the dog infused for 28 days is estimated to be about 8-fold lower than the C<sub>ss</sub> achieved in the PCI dosing of cangrelor 30 µg/kg IV bolus followed by 4 µg/kg/min for 2 h infusion (C<sub>ss</sub> 488 ng/ml).

## 7 Genetic Toxicology

### 7.1 *In vitro* reverse mutation assay in bacterial cells

**Study no.:** SE 9940

**Conducting laboratory:** Astra Safety Assessment, Astra Charnwood, UK

**Date of study initiation:** Sept 23, 1994, June 12, 1995 **Report date:** Oct 7, 1994, Jul 13, 1995

**GLP compliance:** Yes,

**QA statement:** yes (√), no ( )

**Drug:** Cangrelor,

**Batch #:** 3879F

**Positive control:** 1,8-dihydroxyanthraquinone

**Negative control:** Vehicle 0.9% NaCl

**Strains/cell line:** *Salmonella typhimurium* (TA1535, TA1537, TA98, TA100) and TA102.

The potential of cangrelor to induce gene mutation was assessed in bacterial reverse mutation assay. No cytotoxic effects were observed at 5000 µg/plate. There was no increase in the incidence of histidine-independent revertant mutant colonies in any of the bacterial strains in the absence of metabolic activation (-S9) or presence of metabolic activation (+S9) using rat liver fraction (Table 7). Positive controls showed increase in revertant colony incidence (Table 7). In summary, cangrelor was non-mutagenic in an *in vitro* bacterial reverse mutation assay.

Table 7: Bacterial mutation assay with (+S9) and without (-S9) metabolic activation.

Cangrelor (µg/plate)	- S9 mix					+ S9 mix				
	TA1535	TA1537	TA98	TA100	TA102	TA1535	TA1537	TA98	TA100	TA102
15	15	7	16	145	216	12	7	19	103	222
50	23	8	15	119	240	11	8	25	111	185
150	24	6	20	133	204	9	5	23	121	189
500	24	7	15	127	184	15	5	20	98	184
1500	24	6	12	117	196	5	6	17	108	206
5000	27	8	12	126	212	10	10	26	100	218
Vehicle Control	19	7	18	115	230	13	12	22	100	206
Positive Control	236*	128*	325*	665*	301*	114*	300*	266*	294*	582*

Revertant cells: \* = p<0.05, Positive controls: sodium azide (1 µg/plate), 9-aminoacridine (50 µg/plate), 1,8-dihydroxyanthraquinone (10 µg/plate), 4-nitro-1,2-phenylenediamine (5 µg/plate), 4-nitroquinoline-N-oxide (0.2 µg/plate), 2-aminoanthracene (1 µg/plate), 2-aminofluorene (1 µg/plate).

## 7.2 *In vitro* gene mutation assay in mouse lymphoma

**Study no.:** 96102

**Conducting laboratory:** Astra Safety assessment, Sweden, **Report date:** Oct 21, 1996

**GLP compliance:** Yes, **QA statement:** yes (√), no ( )

**Drug:** Cangrelor, **Batch #:** 3960G, **Purity:** 99.3%

**Positive control:** 10-Dimethyl-1,2-benzanthracene and 4-Nitroquinoline-N-oxide 1,8-dihydroxyanthraquinone

**Negative control:** Vehicle 0.9% NaCl

**Strains/cell line:** Mouse lymphoma L5178Y cells TK<sup>+/-</sup>

The ability of cangrelor to induce forward mutation in the mouse lymphoma L5178Y cells at the thymidine kinase locus was determined using concentrations of test article selected on the basis of cytotoxicity study (Table 8). In the absence of metabolic activation, the relative total growth levels were decreased to 5% at 3.56 mmol/L, 11% at 4.45 mmol/L and 22% at 5.34 mmol/L, and no significant increase in the mutant frequency were observed. In the presence of metabolic activation, the relative growth was decreased in a dose-related manner up to 14% at the highest concentration (Table 8). While increases in mutant frequency were seen, they were either at decreased cell growth levels that were below 10% or not dose-related (Table 8). In summary, cangrelor is not a mutagen in the mouse lymphoma cell assay.

Table 8: Percent increase in mutant frequency in mouse Lymphoma L5178 cell line

+ S9 mix (4 hr activation)			- S9 mix (24 hr activation)		
Cangrelor mM (mg/L)	Cell growth (% control)	Mutant Frequency/10 <sup>6</sup> cells	Cangrelor mM (mg/L)	Cell growth (% control)	Mutant Frequency/10 <sup>6</sup> cells
0	-	91	0	-	57
0.89 (769)	95	75	0.89 (769)	103	51
2.68 (2310)	57	107	1.78 (1540)	30	88
6.25 (5400)	32	106	2.68 (2310)	8	105
7.12 (6160)	25	127	3.56 (3080)	5	129
8.01 (6930)	14	98	4.45 (3850)	11	82
BA (1.5 mg/L)	44	772*	5.34 (4620)	22	76.9
			NQ (0.25 mg/L)	23	996*

Positive control: BA = 10-Dimethyl-1, 2-benzanthracene; NQ = 4-Nitroquinoline-N-oxide

## 7.3 *In vitro* chromosomal aberration assay

**Study no.:** SE9958

**Conducting laboratory:** Astra Charnwood, U.K.

**Date of study initiation:** 6/26/1995

**Report date:** 9/15/1995

**GLP compliance:** Yes,

**QA statement:** yes (√), no ( )

**Drug:** Cangrelor

**Batch #:** 3879F

**Positive control:** Cyclophosphamide, 4-Nitroquinoline-N-oxide

**Cell culture:** Human peripheral blood lymphocytes

The ability of cangrelor to induce chromosomal aberrations was investigated in freshly isolated human peripheral blood lymphocytes exposed to cangrelor in the presence of S9 mix for 4 hrs and harvested at 22 or 46 hours, and in the absence of S9 mix for 22 or 46 hours (Table 9). In the absence of S9 mix, mitotic activity was suppressed by at least 50% at concentrations > 313 µg/mL, and therefore highest concentration of test article was set at 313 µg/mL. There were no increase in the incidence of cells with structural chromosomal

changes and/or polyploidy in the presence or absence of metabolic activation at concentrations up to 5000 µg/ml and 313 µg/mL cangrelor, respectively (Table 9). The positive control groups treated with cyclophosphamide (CP, 10 µg/ml) or 4-Nitroquinoline-N-oxide (NQO, 1.25 µg/ml) showed significant increase in aberrant cells (Table 9). In summary, cangrelor was non-clastogenic in a validated *in vitro* human peripheral blood lymphocyte chromosomal aberration assay.

Table 9: Percent increase in chromosomal aberration in lymphocytes treated with cangrelor

+ S9 mix				- S9 mix			
Cangrelor (µg/ml)	Mitotic index %	Polyploid cell %	Aberrant cells (%)	Cangrelor (µg/ml)	Mitotic index %	Polyploid cell %	Aberrant cells (%)
0	5.6	0.26	1.31	0	5.0	0.00	0.28
1250	4.1	1.96	0.50	39	4.9	0.00	0.00
2500	5.6	0.50	1.50	156	4.2	0.00	0.00
5000	4.9	1.48	1.50	313	2.1	0.00	0.00
CP (10 µg/ml)	-	0.00	12.63*	NQO (1.25 µg/ml)	-	0.00	11.93*

\* = p<0.05, vs. control, CP = Cyclophosphamide, NQO = 4-Nitroquinoline-N-oxide

#### 7.4 *In vivo* micronucleus assay in mice

**Study no.:** 96112

**Conducting laboratory:** (b) (4)

**Date of study initiation:** Jul 4, 1996

**Report date:** Sept 1996

**GLP compliance:** Yes,

**QA statement:** yes (√), no ( )

**Drug:** Cangrelor (ARL 69931MX)

**Batch #:** 3960G

**Doses:** 125, 250, 500 mg/kg (0.16, 0.32, 0.64 mmol/kg), IV infusion over 5 min

**Positive control:** Cyclophosphamide

**Negative control:** 0.9% NaCl

**Strains/species:** CD-1 mice male,

**Number:** 9/dose **Age:** 37- 44 days

**Basis of dose selection:** In the 500 mg/kg dose, convulsions, lethargy, tremors, irregular breathing, and eye closure were observed, and two deaths occurred, and thus a dose of 500 mg/kg was selected as the high dose level (MTD).

Cangrelor was assayed *in vivo* in a mouse bone marrow micronucleus test at dosages up to systemically toxic dose. In the blood collected from the mouse tail 48 hours after IV infusion, there was no significant increase in the incidence of micronucleated polychromatic erythrocytes in comparison with that of vehicle treated mice (Table 10). The positive control, cyclophosphamide, caused significant increase in the incidence of micronucleated polychromatic erythrocytes. In summary, cangrelor did not induce micronuclei in the polychromatic erythrocytes of the bone marrow of mice treated with dosages up to 500 mg/kg, a dose at which clinical signs and mortalities occurred.

Table 10: Micronucleated reticulocytes in blood after 48 hrs of IV infusion of cangrelor to mice

Treatment	Dose (mg/kg)	PCE/NCE	MNPCE/1000PCE	MNNCE/1000 NCE
Vehicle (saline)	-	0.91	0.72	0.33
cangrelor	125	1.11	0.67	0.44
"	250	1.05	0.67	0.22
"	500	1.07	0.43	0.00
Cyclophosphamide	40	0.83	10.10*	0.78

\* = p<0.05, vs. control, n=9/group, PCE = polychromatic erythrocytes; NCE = normochromatic erythrocytes; MNPCE = micronucleated polychromatic erythrocytes; MNNCE = micronucleated normochromatic erythrocytes;

## 8 Carcinogenicity

Cangrelor is intended for short-term administration, and thus carcinogenicity studies are not applicable because cangrelor will not be administered chronically.

## 9 Reproductive and Developmental Toxicology

### 9.1 Fertility and Early Embryonic Development

**Study no.:** SR 98073-01 / SR 98074-01  
**Conducting laboratory:** Astra Safety Assessment, Astra Charnwood, UK  
**Study report date:** Apr 7, 1999  
**GLP compliance:** Yes, **QA statement:** yes (✓), no ( )  
**Drug:** Cangrelor, Batch #: 4201J, 4035G, Purity: 97.4%, 99.49% w/w  
**Dosages:** 3, 12, 48 µg/kg/min, (4, 15, 62 µmol/kg/min), IV infusion via right femoral vein cannula  
**Vehicle:** 0.9% NaCl  
**Species/strain:** Sprague-Dawley rat, **Initial age:** 10-13 weeks  
**Number of rats:** 20/sex/dose group

**Study design:** Males were treated with cangrelor by continuous IV infusion for 28 days prior to pairing and continuing for 57 to 64 days. Female rats were treated for 14 days prior to mating, throughout pairing and up to Day 7 post-coitum (between 22 to 37 days treatment). C-section was performed on Day 14 of presumed pregnancy.

**Mortality:** Two animals in the 48 µg/kg/min dosage group were euthanized pre-term on Days 19 and 23 due to heavy blood loss in urine during the dosing period. One of these animals exhibited bilateral ureter distension at necropsy.

**Body weight:** No effect due to treatment on body weight gain.

**Food consumption:** There was no effect of cangrelor on food consumption

**Clinical signs:** Dose-related incidence of blood in urine during the treatment period involving 1, 4 and 7 males in all dose groups was observed, but not in the urine of control rats. Although mean body temperatures were comparable among all groups, the number of rats with body temperatures below 35° C during the week prior to the end of treatment was greater in the drug-treated groups.

**Organ weights:** Reduced epididymal and increased testes weights were observed in the high dose group (Appendix Tabulated Summary 5).

**Necropsy:** Two males at 48 µg/kg/min showed bilateral ureter distension at necropsy. At the end of the 8 week treatment period, abnormal sperm morphology and reduced sperm motility was seen in all males at 48 µg/kg/min. Reduced epididymal weights and sperm counts were also observed, and three rats showed testicular tubular epithelial atrophy and tubular dilatation.

**Histopathology:** Marked tubular epithelial atrophy and tubular dilatation along with epididymal oligospermia and spermatocele formation were observed at the end of the

treatment period in males at a dosage of 48 µg/kg/min. Spermatocyte degeneration was observed in the testes of one of the animals euthanized during the pre-mating period. Following the 8 week treatment-free period, marked tubular epithelial atrophy was observed in the testes of the high dosage group (3/8 rats), one of which also had Leydig cell hyperplasia.

**Fertility parameters:** The high dose group failed to produce pregnancy with female partners, exhibited high implantation loss, reduced sperm motility and count, reduced epididymal spermatid counts, high incidence of detached sperm heads, marked tubular epithelial atrophy and tubular dilatation along with epididymal oligospermia. In female rats infused for 14 days prior to mating through Day 7 post-coitum, cangrelor did not affect estrous cycle, mating performance, and pre-implantation losses. In the high dosage group (48 µg/kg/min), reduced post-implantation survival of embryos, which could have been secondary to maternal toxicity, although direct embryo-toxicity could not be excluded.

**Toxicokinetics:** Plasma concentrations of cangrelor and AR-C69712XX were variable (Table 11). However, mean steady-state concentrations for both compounds generally increased in a dose-proportional manner with similar exposures on Day 7 and Day 17 pc.

Table 11: Plasma concentrations of cangrelor and metabolite AR-C69712XX on Day 7 and 17 pc in rats (n=3)

Dose Level (µg/kg/min)	Mean Cangrelor Plasma Concentrations (ng/mL)		Mean AR-C69712XX Plasma Concentrations (ng/mL)	
	Day 7 pc	Day 17 pc	Day 7 pc	Day 17 pc
3	81	109	15.2	21
12	494	428	85.9	88.5
48	2350	1820	433	462

**Key Study Findings:** Reduced male fertility and mating performance, increased pre-implantation loss, abnormal sperm morphology, and spermatocele formation were observed in males at the high dosage (48 µg/kg/min) (Appendix Tabulated Summary 5). The high dosage group was associated with failure to produce pregnancy, high implantation loss, reduction in sperm motility, and sperm count, reduced epididymal spermatid counts, incidence of detached sperm heads, marked tubular epithelial atrophy and tubular dilatation along with epididymal oligospermia. Treatment with 48 µg/kg/min resulted in reduced post-implantation survival of embryos (Appendix Tabulated Summary 6). There was no significant effect of cangrelor on male or female rats fertility, or on early embryonic development at doses ≤12 µg/kg/min (HED 1.94 µg/kg, C<sub>ss</sub> 428 - 494 ng/ml), in which plasma concentration achieved is similar to that expected in the PCI setting (488 ng/ml), and ~6-fold higher than the Bridge setting (C<sub>ss</sub> 67 ng/ml).

## 9.2 Embryo-Fetal Development

### 9.2.1 Embryo-fetal development study in rats

<b>Study no.:</b>	SR 98002-01	
<b>Conducting laboratory:</b>	Astra Safety Assessment, Astra Charnwood, UK	
<b>Study report date:</b>	Feb 1, 1999	
<b>GLP compliance:</b>	Yes,	<b>QA statement:</b> yes (√), no ( )
<b>Drug:</b>	Cangrelor,	<b>Batch #:</b> 4035G, Purity: 99.49%

**Dosages:** 3, 12, 48 µg/kg/min, (0.004, 0.015, 0.062 µmol/kg/min)  
**Vehicle:** 0.9% NaCl  
**Route:** Continuous IV infusion via right femoral vein cannula  
**Species/strain:** Sprague-Dawley rat **Initial Age:** 13 to 17 weeks;  
**Number of rats:** 24/dose group,  
**Satellite groups:** 3/dose group for TK on days 7 and 17 pc

**Study design:** Cangrelor was administered to time-mated female rats from Day 6 to 17 post-coitum. C-section was performed on Day 21 of presumed pregnancy.

**Mortality:** There were no unscheduled deaths during the course of the study.

**Body weight:** Three animals in the 48 µg/kg/min dosage group exhibited reductions in body weight from Day 18 pc. There was a slight dose-related decrease in fetal weights.

**Food consumption:** The high dose group exhibited marked reductions in food consumption.

**Clinical signs:** One animal in the 12 µg/kg/min dosage group, euthanized on Day 12 post-coitum due to poor condition, showed piloerection, staining around nostrils, red discharge in the tray/cage, and pale kidneys at gross necropsy.

**Necropsy:** In the 48 µg/kg/min dosage group, gross necropsy findings included pale kidneys, pale liver, renal dilation; enlarged spleen, dark spleen, impacted large intestine, and dark pyloric region of the stomach. The rat with the gastrointestinal findings had a total intrauterine loss of 16 fetuses which were considered likely to be secondary to the gastrointestinal pathology.

**Offspring (malformations, variations):** There was slight retardation in development of some fetuses at all dose levels indicated by slight reductions in fetal weights and increased incidence of unossified hind limb metatarsals (Table 12). These effects were in the absence of any overt maternal toxicity. The developmental retardation was more pronounced in the litters from the dams in the 48 µg/kg/min dosage group where there was also an increased incidence of incomplete ossification of skull and sternebrae bones. In all dose groups, there was increased incidence of unossified hindlimb metatarsals (Appendix Tabulated Summary 7).

Table 12: Summary of treatment-related fetal skeletal findings

Finding	Dose (µg/kg/min)			
	Vehicle (Control)	3	12	48
Total # of Fetuses Examined	151	154	150	151
Skull-squamosal-Incomplete ossification	1%	5%	4%	8%*
Sternum-sternebrae-Incomplete ossification	15%	23%	22%	32%**
Hindlimb-metatarsal-not ossified	6%	20%**	16%*	23%**

**Toxicokinetics:** Plasma concentrations of cangrelor and AR-C69712XX were variable, however, mean steady-state concentrations increased in a dose-proportional manner with similar exposures on Day 7 and Day 17 post-coitum (Table 13). The mean plasma concentrations of AR-C69712XX generally reflected dose proportionality.

Table 13: Plasma concentrations of cangrelor and AR-C69712XX

Dose Level ( $\mu\text{g}/\text{kg}/\text{min}$ )	Mean Cangrelor Plasma Concentrations (ng/mL)		Mean AR-C69712XX Plasma Concentrations (ng/mL)	
	Day 7 pc	Day 17 pc	Day 7 pc	Day 17 pc
3	81	109	15.2	21
12	494	428	85.9	88.5
48	2350	1820	433	462

pc = post-coitum

**Key Study Findings:** Treatment with cangrelor resulted in retardation of development of fetuses at all dosage levels as shown by reductions in fetal weights and increased incidence of unossified hind limb metatarsals, but there were no malformations or overt maternal toxicity. The developmental retardation was more pronounced in the litters from the dams in the high dosage group (48  $\mu\text{g}/\text{kg}/\text{min}$ ) where there was also an increased incidence of incomplete ossification of skull and sternbrae bones, indicating retarded development in fetuses and viability of fetuses caused by direct fetotoxic effect of cangrelor at 48  $\mu\text{g}/\text{kg}/\text{min}$  (Appendix Tabulated Summary 7). At dosage levels of  $\geq 3$   $\mu\text{g}/\text{kg}/\text{min}$  (Css 81 ng/ml,  $\text{AUC}_{0-7\text{d}}$  20331 ng.h/ml), retarded development in fetuses were observed, and thus a fetal NOAEL was not identified in the rat embryo-fetal development study. Thus, the embryo-fetal toxicity was observed at about 2-fold higher exposure levels of cangrelor achieved in the Bridge dosing ( $\text{AUC}_{0-7\text{d}}$  11189 ng.h/ml), and 5-fold lower than the plasma concentration achieved in the PCI dosing (Css 488 ng/ml).

### 9.2.2 Embryo-fetal development study in rabbits

<b>Study no.:</b>	SR 97279-01	
<b>Conducting laboratory:</b>	Astra Safety Assessment, Astra Charnwood, UK	
<b>Study report date:</b>	Nov 3, 1998	
<b>GLP compliance:</b>	Yes,	<b>QA statement:</b> yes ( $\surd$ ), no ( )
<b>Drug:</b>	Cangrelor,	Batch #: 4035G, Purity: 99.49% w/w
<b>Dosages:</b>	4, 12, 36 $\mu\text{g}/\text{kg}/\text{min}$ ,	(5.2, 15.5, 46.5 nmol/kg/min)
<b>Vehicle:</b>	0.9% NaCl	
<b>Route:</b>	IV infusion via right jugular vein cannula,	
<b>Species/strain:</b>	Dutch Rabbit.	<b>Initial Age:</b> 20 to 44 weeks;
<b>Number of rabbits:</b>	22 female/dose group,	<b>Satellite group:</b> 3 - 6/group
<b>Study design:</b>	Cangrelor was administered to pregnant rabbits, from Day 6 to 19 of post-coitus. C-Section was performed at Day 29 post-coitus.	

**Mortality:** The number of abortions on Days 17 to 23 pc was slightly increased in animals in the 12 and 36  $\mu\text{g}/\text{kg}/\text{min}$  dosage groups (3 abortions in each group) compared to the 4  $\mu\text{g}/\text{kg}/\text{min}$  and control groups (1 abortion in each group). The number of viable litters on Day 29 pc in all dosage groups was comparable to those in the control. The number of viable litters in the control group was 18 and there were 17, 15, and 17 viable litters in the 4, 12 and 36  $\mu\text{g}/\text{kg}/\text{min}$  dosage groups, respectively. However, total intrauterine losses were higher in the 12 and 36  $\mu\text{g}/\text{kg}/\text{min}$  dosage groups (n= 4 and 3, respectively) compared to control and 4  $\mu\text{g}/\text{kg}/\text{min}$  dosage groups (n=0 and 1, respectively) (Appendix Tabulated Summary 8).

**Body weight:** Rabbits in the 36 µg/kg/min dosage group showed a slight body weight loss during the dosing period.

**Food consumption:** The dams exhibited dose-related reductions in food and water consumption and fecal production, which were reversible following cessation of dosing

**Clinical signs:** Treated groups were given soft food and oral rehydration therapy to encourage them to eat before they became too ill and lost their litter.

**Necropsies and fetal examination:** Fetal weights were slightly decreased in the 36 µg/kg/min dosage group with female fetuses exhibiting 9% decrease compared to control. Seven fetuses among 6 litters (5.6% of fetuses) exhibited slight increase in additional blood vessels on the descending aorta in the fetuses of the 36 µg/kg/min dosage group. Slight increase in additional blood vessels were also observed 1.9% and 1.1% in the mid (12 µg/kg) and low (4 µg/kg) dose groups, respectively, but none in the control group. There was a slight reduction in ossification and a slight increase in skeletal variants observed in all dose groups compared to control animals.

**Toxicokinetics:** Mean plasma levels of cangrelor and metabolite AR-C69712XX, were proportional to the infusion dose and respective concentrations of both compounds were similar on Day 7 and Day 18 post-coitum (Table 14).

Table 14: Plasma concentrations of cangrelor and AR-C69712XX in pregnant Dutch rabbits

Dose (µg/kg/min)	Cangrelor (ng/mL)		AR-C69712XX (ng/mL)	
	Day 7 pc	Day 18 pc	Day 7 pc	Day 18 pc
4	2120	1830	99	90
12	6210	6060	355	330
36	18900	21600	1150	772

pc = post-coitum

**Key Study Findings:** In the rabbit embryo-fetal developmental study, cangrelor produced maternal toxicity at all dosage levels as indicated by decreased food and water consumption as well as body weight. Maternal body weight change showed a dose-related suppression up to 20% in the high dosage group (36 µg/kg/min). In the high dosage group, the fetal skeletal observations are not considered to be indicators of teratogenic potential as they are typically seen in the Dutch rabbits when the dams are stressed or at maternally toxic dosages. There were increased incidences of abortion and intrauterine losses and fetal retardation at dosages ≥12 µg/kg/min (Appendix Tabulated Summary 8). Fetal growth retardation occurred at the high dosage group, and was characterized by decreased fetal weights, slight reduction in ossification, and a slight increase in skeletal variants. There were also indications of maternal toxicity at the low dosage, but no fetal effects occurred. Cangrelor is not considered to be a teratogen in the rabbit. The steady state plasma concentration at the NOAEL of 4 µg/kg/min (C<sub>ss</sub> 2120 ng/ml) is ~4-fold higher than the plasma concentration achieved in the PCI setting (488 ng/ml). The total exposure at the NOAEL of 4 µg/kg/min (AUC<sub>0-7d</sub> 532120 ng.h/ml) is 47-fold higher than the exposures achieved in the Bridge setting (AUC<sub>0-7d</sub> 11189 ng.h/ml).

### 9.3 Prenatal and Postnatal Development

<b>Study no.:</b>	901045	
<b>Conducting laboratory:</b>	(b) (4)	
<b>Date of study initiation:</b>	Nov 15, 2006	<b>Report date:</b> Apr 2, 2008
<b>GLP compliance:</b>	Yes	<b>QA statement:</b> yes (✓) no ( )
<b>Drug:</b>	Cangrelor (tetrasodium salt)	Batch no.: 902714, Purity: 102.6%
<b>Route:</b>	IV infusion,	<b>Vehicle:</b> 0.9% NaCl
<b>Dosages:</b>	3, 9, 30 µg/kg/min, (dose equivalent 4.32, 12.96, 43.2 mg/kg/day)	
<b>Species/strain:</b>	Sprague-Dawely rat,	<b>Initial Age:</b> 77 - 84 days of age
<b>Number/sex/group:</b>	F <sub>0</sub> : 24 female/dose group; F <sub>1</sub> : 20 M, 20 F/dose group	

**Study design:** Rats were dosed during gestation Day 6 to post-partum Day 21 interval, and were allowed to litter and rear their offspring to weaning (PP Day 21), and killed on Day 21 of lactation. F<sub>1</sub> females were killed on Day 14 after mating, and F<sub>1</sub> males were killed soon afterwards.

#### F<sub>0</sub>-Generation:

**Mortality:** In the F<sub>0</sub>-generation, there was a slight increase in the incidence of mortality in the high dosage group (43.2 mg/kg/d). One female in the mid-dose group (12.96 mg/kg/d) was found dead on Day 3 post-partum with no clinical observations prior to death and no gross pathology findings to implicate the cause of death. Three dams from the high dosage group (43.2 mg/kg/d) were euthanized. One was euthanized on gestation Day 22 due to prolonged parturition. Necropsy findings included a large mass at the infusion site as the likely cause of the prolonged parturition. The remaining 2 dams were in poor condition on Days 19 and 21 post-partum, and were euthanized. One female from the control, low- and high-dose groups were euthanized on Day 0 post-partum following litter cannibalization.

**Bodyweight:** Body weights and body weight gains were generally similar for all groups. A statistically significant reduction in body weight gain in the high dosage dams (43.2 mg/kg/d) from gestation Day 7 to post-partum Day 14 was attributed to biological variation.

**Food consumption:** A statistically significant reduction in food consumption in the high dosage dams (43.2 mg/kg/d) was observed during gestation Days 15 - 18 and did not impact the gestation Day 18 or Day 20 body weight, and was attributed to biological variation.

**Maternal Performance (Gestation length):** Pregnancy rates, gestation index, length of gestation, numbers of live, dead and malformed pups, sex ratio, and the live birth index were unaffected by drug treatment.

**Clinical Signs:** There were no clinical observations during the gestation and lactation periods that were attributed to the administration of cangrelor. Occasional findings of thin fur cover, fur staining, skin scabs/lesions/redness were seen in a few animals in all groups and were considered to be incidental in origin.

**Gross Pathology:** There were no gross macroscopic findings considered to be the result of administration of cangrelor at any dose level.

**Macropathology:** At necropsy, there were lesions in the kidney that included swelling, enlargement, green material at the cut surface, adhesions, capsular thickening and firmness

that were considered to be the causes of deteriorating conditions in the dams, and consistent with the elevated BUN and renal lesions observed in the 28 day study performed over a similar dosage range.

#### **F<sub>1</sub> and F<sub>2</sub>-Generation pups:**

**Mortality:** In F<sub>1</sub>-generation pups, viability, survival and lactation indices were not affected by cangrelor administration to the F<sub>0</sub>-generation dams. For the F<sub>1</sub> pups found dead at parturition or euthanized between Day 0 and Day 7 post-partum, the few findings in various organs and tissues were considered to be incidental in origin. No pups died between Days 8 and 21 post-partum.

**Bodyweight:** In F<sub>2</sub>-Generation pups, clinical condition, litter size, and body weights showed no effects of administration of cangrelor to the F<sub>0</sub>-generation. Because pup body weights were similar for all groups at the time of development of the tooth eruption parameter, the slight increase in the mean day of development is considered to be incidental and unrelated to treatment. The mean day of development for righting reflex, negative geotaxis, and auricular startle response was similar for all groups.

**Physical Development:** There were no significant differences in the mean day of development for pinna unfolding and eye opening for the males and females for any group. For tooth eruption, a statistically significant increase in the mean day of development was noted for the high dose group dams (43.2 mg/kg/d) and female pups in low dose group dams (43.2 mg/kg/d). Because pup body weights were similar for all groups at the time of development for this parameter (tooth eruption) and because there was no dose dependency, this slight increase in the mean day of development was considered to be incidental and unrelated to treatment.

**Clinical signs:** In F<sub>1</sub>-generation pups, viability, survival and lactation indices, litter size, pup weights, and developmental parameters were not affected by cangrelor administration to the F<sub>0</sub>-generation dams. In F<sub>1</sub>-generation adults, there were no unusual clinical signs or significant differences among groups in body weight, day of vaginal opening or preputial separation, pupillary closure, visual placing, motor activity, and auditory startle habituation (Appendix Tabulated Summary 9). Errors and time to complete the water maze were comparable among all groups. Estrous cycles were comparable for all groups. The mean day to mating, mating index, fertility index, and conception rates of the drug-treated rats were not affected. Maternal performance parameters of length of gestation and parturition, sex ratio, and numbers of live, dead and malformed pups, implantation scars, and the live birth index were comparable for all groups. In F<sub>2</sub>-generation pups, viability, clinical condition, pup body weights and terminal examinations showed no effect of administration of cangrelor to the F<sub>0</sub>-generation dams.

**Estrous Cycles:** Estrous cycles were comparable for all groups.

**Gross Pathology:** There were no gross pathological findings noted for the F<sub>1</sub>-generation adults that were attributed to the administration of cangrelor to the F<sub>0</sub>-generation dams. For the few pups examined, there were no external or visceral findings.

**Key Study Findings:** In the F<sub>0</sub>-generation dams, there was a slight increase in the incidence of mortality in the dams of the high dosage group of 43.2 mg/kg/d, with no clear cause (Appendix Tabulated Summary 9). The NOAEL for the F<sub>0</sub> females was considered to

be 12.96 mg/kg/d (9 µg/kg/min = HED 1.5 µg/kg/min). For the offspring, there were no effects on survival, physical development, behavior, or reproductive performance, and the NOAEL for the F<sub>1</sub>- and F<sub>2</sub>-Generation pups was considered to be 43.2 mg/kg/d (30 µg/kg/min = HED 7 µg/kg/min).

## 10 Special Toxicology Studies

### 10.1 Investigative toxicity on kidney, urinary tract and liver

#### 10.1.1 Effects of cangrelor infusion on the kidney, urinary tract, liver

Study no.:	SR97246, GLP compliance, QA statement ✓
Study report location:	Astra Safety Assessment, Astra Charnwood, UK
Date of study report:	May 20, 1998
Drug:	Cangrelor tetrasodium salt, Batch No: 40449, Purity 99.6%
Dosages	25, 75 µg/kg/min (32.2, 96.7 nmol/kg/min), 1-month infusion
Vehicle;	0.9% NaCl
Species	Sprague-Dawley Rat, 6-8 weeks old, Weight: 155 – 216 g
Number of rats:	30 Males (10/dose group for 1-week, 1-month followed by 1-month treatment-free period)

**Key Study Findings:** To obtain further information on cangrelor-related urinary tract lesions, a follow-up study was conducted to assess time of onset and reversibility in Sprague-Dawley rats treated with cangrelor infused at a rate of 25 or 75 µg/kg/min. After 1-week of dosing, histopathological analysis showed changes in the kidney and ureter at both dosages of cangrelor. There was submucosal inflammation in the pelvic region that centered on the hilus. The cellular infiltrate was largely mononuclear with hyperplasia of the overlying urothelium that also showed areas of ulceration and accompanying neutrophil infiltration. Inflammation of the ureter was focal and restricted to the upper part close to the kidney. Clinical laboratory evaluations revealed increases in plasma urea and creatinine (2.6x than control) and glutamate dehydrogenase (GLDH, 2.8-3.6x than control), AST, ALT (2x than control), and GGT (6x than control) indicating changes in kidney and liver function. After one-month, one animal administered 25 µg/kg/min and two animals administered 75 µg/kg/min showed similar treatment-related pathology changes in the kidney and ureter including vacuolation and necrosis. Two animals in the 75 µg/kg/min recovery group exhibited minimal or slight tubular degeneration that was thought to be indicative of previous injury which would be consistent with the reversal of urinalysis parameters following the recovery period. In summary, the incidences of histopathologic changes in the kidney and ureter, increases of plasma enzymes and urinalysis parameters develop within 7 days of treatment at the tested of doses ≥ 25 µg/kg/min = 4 µg/kg/min), and tend to be reversible after one-month of treatment-free period.

### 10.1.2 Time course of renal lesions induced by single dose of cangrelor

Study no.: 98098, non-GLP  
 Study report location: Astra Safety Assessment, Astra Charnwood, UK  
 Date of study report: Dec18, 2000  
 Drug: Cangrelor tetrasodium salt, Batch No: 40449, Purity 99.6%  
 Doses 200 mg/kg (258 µmol/kg), Vehicle: 0.9% NaCl  
 Species Sprague-Dawley Rat, 6-8 weeks old, Weight: 155 – 216 g  
 Number of rats: 53 Females

**Key Study Findings:** At 6 hrs post administration of cangrelor (200 mg/kg), ultrastructural examination of kidney proximal tubular cells showed increased vacuolation in 5/7 animals, and mitochondrial vesicles in one animal. Renal tubular necrosis, mainly of the inner cortex, first detected by light microscopy at 16 hours post-dosing, increased in incidence and degree with time. Minimal renal tubular degeneration, seen in 2 animals at 24 hours post-dosing, was considered to be a transitional stage leading to tubular necrosis. In plasma, increased aminotransferase levels (GLDH, AST, ALT) were detected at 6, 24, 48 and 72 hours, indicating liver dysfunctional effects. Inflammation of the bladder of 3 animals and ureter of 1 animal indicate that acute high exposure to cangrelor can induce similar effects to those seen after continual infusion at lower doses. Thus, a single intravenous dose of cangrelor 258 µmol/kg induced changes to kidney proximal tubules within 6 hours of administration which progressed to tubular necrosis with associated inflammation and loss of kidney function that showed no recovery up to 72 hours post-dose.

### 10.2 Toxicity study of metabolite AR-C90439XX

14-day continuous infusion of cangrelor and major metabolite AR-C90439XX in rats

Study no.: 99288, GLP compliance, QA statement ✓  
 Study report location: Astra Safety Assessment, Astra Charnwood, UK  
 Date of study report: Feb 6, 2002  
 Drugs: Cangrelor, Dose:100 µg/kg/min (3.86 µmol/ml)  
 AR-C90439XX, Dose: 15 µg/kg/min (0.93 µmol/ml)  
 Dose selection: Based on equivalent plasma concentrations achieved  
 Species Sprague-Dawley rat, 9-11 weeks old, Weight: 184 – 243 g  
 Number of rats: 12 Females/dose group

**Key Study Findings:** The metabolite, AR-C90439XX (15 µg/kg/min), administered at a dose similar to the plasma concentration achieved from administration of cangrelor for 14 days did not induce any evidence of toxicity. Cangrelor (AR-C69931MX) administered at 100 µg/kg/min for 14 days induced nephrotoxic effects, indicating that toxicities are mediated by the parent drug, but not by the metabolite.

### 10.2.1 *In vitro* bacterial mutagenicity assay of metabolites

Study no.: SE9563, SE9564, GLP compliance: ✓  
 Study report location: (b) (4)  
 Study dates: 9/23/1994 - 10/07/1994  
 Metabolites: ARL 69712XX (12.5 – 800 µg/plate)  
 AR-C71301XX (0.25 – 250 µg/plate)  
 Incubation: 3 days, +/- S9 metabolic activation  
 Bacterial strains: Salmonella typhimurium TA 98, TA 100, TA 102, TA 1537  
 Positive control: 4-nitroquinoline-N-oxide, 2-aminofluorene 1,8-dihydroxyanthraquinone

Bacterial mutagenicity assays (Ames Test) were conducted on the nucleoside and base metabolites of cangrelor, AR-C69712XX (major metabolite) and AR-C71301XX (minor metabolite), respectively. There were no increases in the frequency of histidine-independent (his+) revertant colony numbers in any of the strains indicating that the metabolites are not considered to be mutagenic.

### 10.3 Antigenicity

#### Antigenicity study of cangrelor in Guinea pigs

Study no.: 69812, GLP compliance, QA statement ✓  
 Study report location: (b) (4)  
 Date of study report: July 9, 1999  
 Drugs: Cangrelor, Batch No.: 4121H, Purity: 99.54%  
 Dosages: 6, 18 mg/kg, Vehicle: 0.9% NaCl  
 Route: Intravenous or subcutaneous  
 Species: Hartley strain Guinea pig, Initial age: 4-5 weeks old  
 Number of rats: 10 Females/dose group

**Key Study Findings:** In sensitized Hartley strain guinea pigs using subcutaneous or intravenous administration of cangrelor (6, 18 mg/kg) in combination with Freund's complete adjuvant (FCA) to enhance immune response, did not show any active systemic anaphylaxis (ASA) or passive cutaneous anaphylaxis (PCA) reactions indicating that cangrelor is not antigenic in the guinea pig. Guinea pigs sensitized with a mixture of ovalbumin and FCA administered SC as a positive control elicited high titers of PCA.

### 10.4 Local Tolerance

Separate local tolerance studies were not conducted. However, histopathological findings at the injection sites of rats and dogs administered cangrelor by continuous infusion for 1-month at dosages up to 48 and 60 µg/kg/min, showed comparable incidence and severity of injection-site lesions between cangrelor and vehicle treated groups. The frequently noted findings in both groups included inflammation, thrombus formation and vasculitis. At high dosages of cangrelor (≥200 mg/kg) infusion in mice or rats, there was clear evidence of irritation and development of bluish discoloration after dosing at the site of injection.

## 11 Integrated Summary and Safety Evaluation

Cangrelor is a short-acting P2Y<sub>12</sub> purinergic receptor antagonist that blocks ADP-induced platelet aggregation at IC<sub>50</sub> values of 0.5 nM. Cangrelor has characteristics of reversible binding, rapid onset of action that occurs within minutes, and short-lived upon cessation of infusion, with substantial recovery occurring within 60 minutes post-infusion. In rats and dogs, steady-state plasma concentrations of cangrelor were quickly attained after the start of infusion. Following termination of drug infusion, most of the parent compound rapidly disappeared from plasma, although a small proportion (< 10%) was eliminated more slowly, and a more prolonged terminal elimination phase was apparent. Cangrelor is metabolized by dephosphorylation to inactive form AR-C69712XX, and further metabolized by sulphoxidation, purine base oxidation, deribosylation and glucuronidation, producing multiple metabolites, and the main one is the sulphoxide, AR-C90439XX. The plasma pharmacokinetic properties of cangrelor and the initial step major plasma metabolite, AR-C69712XX, are similar between rats, dogs, and humans. Upon cessation of infusion, plasma cangrelor levels decline with an initial plasma elimination t<sub>1/2</sub> of 1 to 2 minutes. Cangrelor is eliminated in the feces, with a smaller proportion in the urine.

The most notable toxicity findings in the rat and dog were observed in the renal cortical tubule and urinary tract including the renal cortical tubules, transitional epithelium of the pelvis and ureter, ranging from mild inflammation to necrosis, and tend to appear within 6 hours of administration. The injury to the kidney is also indicated by elevated BUN, creatinine, GGT and urinary protein markers. Urinary bladder and gastric mucosa are also targets of toxicity. The primary insult appeared to be to the transitional epithelium, manifested by epithelial ulceration and necrosis, associated reactive epithelial hyperplasia and inflammation, which extended into the renal parenchyma, connective tissue of the renal hilum and into the peri-ureteral connective tissue, and these changes tend to be reversible upon cessation of treatment. These toxicities were mediated by the parent drug cangrelor which has an ultra-short half-life of 5 minutes, but the metabolite AR-C90439XX had no adverse effect. Another finding in the rat and dog was elevated liver function enzymes, as shown by increase in plasma aspartate aminotransferase (AST), alanine aminotransferase (ALT) and gamma – glutamyl-transpeptidase (GGT), but there was no associated histopathological evidence of liver toxicity.

In the 28-day repeat-dose studies in the dog, cangrelor infused at 60 µg/kg/min achieved steady state plasma concentration of 992 ng/ml, and considering protein binding of 93%, the unbound (free) drug is estimated to be 70 ng/ml. The free drug is approximately two orders magnitude higher than the half-maximal inhibitory concentration of cangrelor to inhibit platelet aggregation in the dog (IC<sub>50</sub> 0.72 nM, 0.62 ng/ml), but there were no reports of bleeding in the repeat-dose studies in the dog. The absence of bleeding incidences in this study is unexpected, because the dog has been reported to exhibit bleeding from the toenails, perioral region and ear in response to potent P2Y<sub>12</sub> receptor inhibitors.

Cangrelor caused respiratory distress, an effect likely related to its structural similarity to ATP which may be mediated by mast cell degranulation and histamine release. Alternatively, it may be mediated by inhibition of P2Y<sub>12</sub> receptors in the sensory neurons thereby increasing the dyspnea sensation. Cangrelor may cause slowing of heart rate

(negative chronotropic effect), depression of the SA node and AV nodal conduction (negative dromotropic effect) and asynchronous premature ventricular contractions ('ectopics' PVCs), which may be mediated by activation of adenosine A<sub>1</sub> receptors.

Cangrelor and its metabolites were non-genotoxic and non-mutagenic *in vitro* and *in vivo* genetic toxicology studies that included *in vitro* bacterial mutagenicity assays, the mouse lymphoma tyrosine kinase assay, chromosome aberration assay in human peripheral lymphocytes, and *in vivo* bone marrow micronucleus assays in mice. Reproductive and developmental studies indicate that cangrelor impairs male and female fertility, and these were reversible following cessation of dosing. At dosages  $\geq 48$   $\mu\text{g}/\text{kg}/\text{min}$ , impairment of male fertility were associated with abnormal sperm motility and morphology, decreased sperm counts, and microscopic lesions in testes and epididymides. In the female fertility study, cangrelor did not affect estrous cycle, mating performance, and pre-implantation losses at doses up to 48  $\mu\text{g}/\text{kg}/\text{min}$ . At doses associated with maternal toxicity there was reduced post-implantation survival of embryos and increased pre-implantation loss in pregnant females. In the rat embryo-fetal development (EFD) study, cangrelor produced fetal growth retardation, in the absence of maternal toxicity, which was most evident at 48  $\mu\text{g}/\text{kg}/\text{min}$ , characterized by increased incidences of incomplete ossification and unossified hind limb metatarsals, and the latter may be non-specific. In the rabbit EFD study, cangrelor produced maternal toxicity at  $\geq 4$   $\mu\text{g}/\text{kg}/\text{min}$  with increased incidences of abortion and intrauterine losses at  $\geq 12$   $\mu\text{g}/\text{kg}/\text{min}$ . Fetal growth retardation occurred at 36  $\mu\text{g}/\text{kg}/\text{min}$ , and was characterized by decreased fetal weights, reduction in ossification, and a slight increase in skeletal variants. Since cangrelor did not produce malformations in either the rat or rabbit EFD studies, and it is not teratogenic, but there were effects on fetal growth in rats and rabbits at high dosages. Cangrelor did not exhibit any effects on pre- and post-natal development.

In the rat and dog toxicology studies performed as a continuous infusion for 4 weeks, the NOAEL is identified to be 3 and 3.75  $\mu\text{g}/\text{kg}/\text{min}$ , which achieved C<sub>ss</sub> of 113 and 61.5 ng/ml, and AUC<sub>0-28d</sub> 46565 and 53957 ng\*h/mL, respectively. For a rapid acting and short-duration drug with no accumulation, the steady state plasma concentration appears to represent the pharmacokinetic profile in the PCI dosing injected as a bolus of cangrelor 30  $\mu\text{g}/\text{kg}$  followed by 4  $\mu\text{g}/\text{kg}/\text{min}$  infusion for 2 h. The dosing regimen in the PCI setting achieved C<sub>ss</sub> of 488 ng/ml, which is estimated to be 4.3- and 7.9-fold higher than the steady state plasma concentration in the rat and dog, respectively (Table 16). Accordingly, the preclinical data does not support the safety of cangrelor in the PCI setting where 30  $\mu\text{g}/\text{kg}$  achieves plasma concentrations appreciably higher than the estimated threshold for potential toxicity in the kidney and ureter, and thus safety assurances should be ascertained in the clinical trials. In the Bridge scenario, dosing of cangrelor at 0.75  $\mu\text{g}/\text{kg}/\text{min}$  for 7 days achieved AUC<sub>0-7d</sub> 11189 ng\*h/ml, and this is estimated to be ~4-fold lower than the NOAEL (Table 15). The metabolite AR-C69712XX also shows a similar safety margin multiples as the parent drug.

Table 15: Margin of safety analysis for the Bridge and PCI dosing over the NOAEL in the rat and dog

Rat		Bridge dosing	Safety	PCI dosing	Safety	Rat NOAEL	Metabolite	Bridge dosing	Safety
NOAEL	3 µg/kg/min	0.7 µg/kg/min	multiple	30+4 µg/kg/min	multiple	3 µg/kg/min	AR-C69712	0.7 µg/kg/min	multiple
C <sub>ss</sub>	113 ng/ml			488 ng/ml	0.23x	AUC	17027 ng.h/ml	5471 ng.h/ml	3x
AUC	46565 ng.h/ml	11189 ng.h/ml	4x						
Dog		Bridge dosing	Safety	PCI dosing	Safety	Dog NOAEL	Metabolite	Bridge dosing	Safety
NOAEL	3.75 µg/kg/min	0.7 µg/kg/min	multiple	30+4 µg/kg/min	multiple	3.75 µg/kg/min	AR-C69712	0.7 µg/kg/min	multiple
C <sub>ss</sub>	61.5 ng/ml			488 ng/ml	0.12x	AUC	22805 ng.h/ml	5471 ng.h/ml	4x
AUC	53957 ng.h/ml	11189 ng.h/ml	4.8x						

**OVERALL CONCLUSIONS AND RECOMMENDATIONS**

The extent and scope of the pharmacological and toxicological documentation provided are appropriate to assess the safety of cangrelor for the proposed clinical dosing regimens – i) In the Bridge setting, human exposures based on AUC<sub>0-7d</sub> of cangrelor achieved by a 0.75 µg/kg/min infusion for 7 days is approximately 4.5-fold less than that at the 28-day NOAEL in the dog, predicting tolerance of the human exposure. ii) However, in the PCI setting the C<sub>ss</sub> values achieved from bolus injection of cangrelor 30 µg/kg followed by 4 µg/kg/min infusion for 2 hr is ~8-fold higher than that at the 28-day NOAEL, and there is no margin of safety over the NOAEL identified in the preclinical studies, and thus enhanced renal and urinary tract function monitoring are recommended in the clinic.

**Suggested labeling:** Mutagenesis, Carcinogenesis, Impairment of fertility –

**Mutagenesis:** Cangrelor was non-mutagenic and non-clastogenic in genetic toxicology studies that included *in vitro* bacterial gene mutation assay, <sup>(b) (4)</sup> mouse lymphoma thymidine kinase assay, chromosome aberration assay in human peripheral lymphocytes, and *in vivo* bone marrow micronucleus assay in mice.

**Carcinogenesis:** No carcinogenicity studies were conducted.

**Impairment of Fertility:** <sup>(b) (4)</sup> no significant effect <sup>(b) (4)</sup> on male or female rats fertility treated for 28 days, or on early embryonic development at steady state plasma concentration (C<sub>ss</sub>) of approximately the same as that achieved in the PCI setting at the <sup>(b) (4)</sup> (MRHD), <sup>(b) (4)</sup>



**Pregnancy:** In embryo-fetal developmental toxicology studies in rats, cangrelor produced dose-related fetal growth retardation characterized by increased incidences of incomplete ossification and unossified hind limb metatarsals at plasma concentration of approximately 5 times lower than (b) (4) (b) (4). In rabbits, cangrelor was associated with increased incidences of abortion and intrauterine losses, as well as fetal growth retardation at plasma concentrations of approximately (b) (4). Cangrelor did not produce malformations in either the rat or rabbit reproductive studies, and is not considered to be a teratogen.

(b) (4)

**Pregnancy Category C:** (b) (4)

**Nursing Mothers:** It is not known whether cangrelor is excreted in human milk. (b) (4)

## 12 Appendix/Attachments

Tabulated Summary 1. Repeat-dose toxicity studies in male rats,		Study No.: SE9948		
Daily Dose (µg/kg/min)	0 (Control)	3	12	48
<b>Number of Animals Main Group</b>	9M	9M	10M <sup>a</sup>	9M
<b>Hematology<sup>h</sup></b>				
Hb (g/dL)	13.74	13.26	13.49	11.61*
RBC (10 <sup>12</sup> /L)	7.653	7.357	7.660	6.494*
HCT (1/1)	0.4214	0.3993	0.4080	0.3464*
<b>Coagulation</b>	-	-	-	-
<b>Clinical Chemistry<sup>h</sup></b>				
Urea (mmol/L)	6.41	6.22	6.13	9.57**
AST (IU/L)	118.9	133.9	164.3	292.0***
ALT (IU/L)	55.4	60.4	81.4**	114.7**
Phosphate (mmol/L)	2.197	2.003	1.813*	1.726*
Triglyceride	0.679	0.468*	0.446*	0.459*
Total Protein (g/l)	67.9	69.0	74.3*	68.9
<b>Organ Weights</b>				
Left adrenal gland (mg)	40.19	42.36	44.95	50.72***
<b>Gross Pathology</b>				
Pyelonephritis & Ureteritis	0	0	0	2
Hepatic Necrosis	0	0	0	2
Bladder Hemorrhage/Congestion/Necrosis	0	0	0	2
<b>Kidney</b>				
Pyelonephritis				
Moderate	0	0	0	1
Marked	0	0	0	1
Intraepithelial neutrophils pelvis				
Minimal	0	1	0	1
Slight	0	1	0	0
Pelvic area plasma cell infiltration (Marked)	0	0	0	1
Pelvic region mixed inflammatory cell infiltration				
Slight	0	1	0	1
Moderate	0	0	0	1
Erythrocytes/inflammatory cells pelvic space				
Minimal	0	0	0	1
Pelvic epithelium, hyperplastic (Slight)	0	1	0	1
Pelvic epithelium, neutrophil infiltrate (Slight)	0	0	0	1
<b>Liver</b>				
Centrilobular vacuolation (minimal)	0	0	0	1
Focal necrosis (Slight)	0	0	0	1
Lobar necrosis	0	0	0	1
Congestion (Slight)	1	0	0	0
Hemopoiesis				
Minimal	0	0	0	1
Slight	0	0	0	1
Inflammatory cell foci				
Minimal	5	6	4	1
Slight	0	0	1	3
<b>Ureter</b>				
Intraepithelial inflammatory cells (Slight)	0	0	0	1
Periurethral inflammation				
Minimal	1	2	2	0
Slight	1	4	4	2
<b>Urinary Bladder</b>				
Colloid plug	0	0	1	0
Hemorrhage (Marked)	0	0	0	1
Submucosal neutrophils (Slight)	0	0	0	1

<sup>a</sup> One additional animal was added, <sup>h</sup> Group means.

## Tabulated Summary 2. Repeat-dose toxicity studies in female rats, Study No.: SE10176

Daily Dose (µg/kg/min)	0 (Control)	3	12	48
<b>Number of Animals Main Group</b>	9F	9F	9F	9F
<b>Clinical Chemistry (Terminal)<sup>e</sup></b>				
Urea (mmol/L)	6.71	6.66	6.13	8.18*
Phosphate (mmol/L)	1.995	1.902	1.748**	1.717**
AST (IU/L)	98.6	146.7	167.2***	185.2***
ALT (IU/L)	55.0	94.3	86.3*	102.0**
<b>Urinalysis<sup>f</sup></b>				
Presence of RBC and/or WBC <sup>g</sup>	1	2	1	4
Presence of Albumin <sup>h</sup>	0	-	0	2
Qualitative proteinuria	0	0	1	2
<b>Organ Weights<sup>i</sup></b>				
Liver	3.31	3.24	3.17	2.85**
Pituitary	6.8	6.8	6.5	5.85*
Adrenal (both)	31.11	30.01	28.52	25.13*
<b>Histopathology<sup>f</sup></b>				
Kidney				
Pelvic/hilar inflammation				
slight	0	0	0	4
Pelvic necrosis				
minimal	0	0	0	1
slight	0	0	0	1
Pyelonephritis				
slight	0	0	0	1
Urothelial hyperplasia				
slight	0	0	0	4
Pyelitis				
slight	0	0	0	1
Interstitial nephritis				
minimal	0	0	0	1
Ureter				
Urothelial hyperplasia				
moderate	0	0	0	1
Necrosis				
minimal	0	0	0	1
moderate	0	0	0	1
Ureteritis				
slight	0	0	0	1
marked	0	0	0	1
Urothelial erosion				
slight	0	0	0	1

<sup>e</sup> Group means, <sup>f</sup> Reported as number observed, <sup>g</sup> Incidence of RBC and WBC in urine sediment microscopically, <sup>h</sup> Incidence of albumin in urine by electrophoresis on Day 15 and Day 28, <sup>i</sup> Mean values.

## Tabulated Summary 3. Repeat-dose toxicity studies in male dogs,

Study No.: SE9862

Daily Dose (µg/kg/min)	0 (Control)	3.75	15	60
<b>Number of Animals</b>	3M	3M	3M	3M
<b>Hematology<sup>e</sup></b>				
Fibrinogen (g/l)	3.1	3.2	3.3	4.7
<b>Coagulation</b>	-	-	-	-
<b>Clinical Chemistry<sup>e</sup></b>				
Creatinine (µmol/L)	73.3	67.7	85.0	121.0
Urea (mmol/L)	4.90	3.80	6.40	9.37
AST (IU/L)	35.3	35.3	37.3	80.3
ALT (IU/L)	37.3	26.7	40.0	48.0
CK (IU/L)	173.7	168.0	238.0	275.0
<b>Urinalysis<sup>f</sup></b>	-	-	-	-
Blood	-	+	-	+
Urinary protein	-	+	-	+
WBC and RBC	-	-	+	+
<b>Organ Weights<sup>g</sup></b>	-	-	-	-
Thymus (g)	21.75	-3	-11	-58
Thymus relative to body weight (%)	0.204	-2	-11	-55
<b>Gross Pathology</b>	-	-	-	-
<b>Number of Animals</b>				
<b>Histopathology<sup>h</sup></b>				
<b>Kidney</b>				
Tubular dilatation				
slight	0	0	0	2
moderate	0	0	0	1
Tubular necrosis				
moderate	0	0	0	1
Basophilic tubules				
slight	0	0	0	1
Tubular regeneration				
slight	0	0	0	1
Interstitial nephritis				
slight	0	0	0	1
Pelvic inflammation				
slight	0	0	0	1
Urothelial hyperplasia				
minimal	0	0	0	1
slight	0	0	0	1
Urothelial necrosis				
slight	0	0	0	1
<b>Stomach</b>				
Gastritis				
slight	0	0	0	1
moderate	0	0	0	2
Erosion/ulceration				
minimal	0	0	0	1
Necrosis				
slight	0	0	0	1
<b>Thymus</b>				
Involution				
minimal	0	0	0	2
slight	0	0	0	1
Lymphocytolysis				
minimal	0	1	0	1
slight	0	0	0	2

<sup>e</sup> Terminal group means, <sup>f</sup> "+" = present in animals in dose group, <sup>g</sup> Vehicle control values as means. Cangrelor group values expressed as percent difference from vehicle control values, <sup>h</sup> Reported as number observed.

### Tabulated Summary 4. Repeat-dose toxicity studies in female dogs, Study No.: SE10177

Daily Dose (µg/kg/min)	0 (Control)	3.75	15	60
<b>Number of Animals in Main Group</b>	3F	3F	3F	3F
<b>Body Weight<sup>b</sup></b>	0	0	0	1
<b>Food Consumption<sup>c</sup></b>	0	0	0	1
<b>Clinical Observations<sup>d</sup></b>				
Dark urine/blood in urine	0	0	0	1
Blood in vomit	0	0	0	2
<b>Clinical Chemistry<sup>e</sup></b>				
Urea (mmol/L)				
Day 15	5.23	6.00	4.13	7.73
Terminal	5.93	5.50	4.76	8.63
Creatinine (µmol/L)				
Day 15	71.6	77.6	69.6	89.0
Terminal	71.3	68.0	64.0	97.6
Total protein (g/L)				
Terminal	49.6	56.6	54.0	48.6
Albumin (g/L)				
Terminal	30.3	30.6	30.0	26.6
<b>Urinalysis</b>				
Protein/creatinine				
Day 15	6.3	5.5	8.0	68.7
Terminal	6.0	8.0	6.0	140.0
β-NAG/creatinine				
Day 15	0.840	1.200	0.773	2.700
Terminal	0.860	0.830	0.777	4.840
<b>Organ Weights<sup>f</sup></b>				
Kidney (g)	46.433	-1	3	-10
Kidney to body weight ratio (%)	0.506	8	4	-7*
<b>Gross Pathology</b>	-	-	-	-
<b>Histopathology<sup>g</sup></b>				
<b>Kidney</b>				
Basophilic tubules				
minimal	0	0	0	2
Inflammatory cell foci				
minimal	0	0	0	1
Tubules distended with colloid/cellular debris				
minimal	0	0	0	2
<b>Stomach</b>				
Edema				
minimal	0	0	0	1
Mucosal inflammation				
minimal	0	0	0	1
moderate	0	0	0	1
Diluted glands				
minimal	0	0	0	1
Mucosal atrophy				
minimal	0	0	0	1

<sup>b</sup> animals with decreased body weight, <sup>c</sup> animals with decreased food consumption, <sup>d</sup> animals affected on occasions  
<sup>e</sup> Group means, <sup>f</sup> Vehicle control values as means. Treated group values as percent difference from vehicle control values  
<sup>g</sup> Reported as number observed

Tabulated Summary 5. **Effects of cangrelor on Fertility and early embryonic development in male rats**  
Study No.: SR 98073-01

Daily Dose (µg/kg/min)	0 (Control)	3	12	48
<b>Treatment Phase (continued)</b>				
<b>Implantation Data</b>				
No. Aborted/Total Resorption of Litter	0	0	0	0
Mean No. Implants per Litter	16.2	15.5	14.9	11.8**
Mean No. Live Implants per Litter	15.2	15.0	14.2	10.7**
Mean % Per Litter Pre-implantation Loss	4.3	8.0	8.7	24.4*
Mean % Per Litter Post-implantation Loss	6.1	3.5	5.2	10.0
No. Evaluated	11	11	12	12
<b>Necropsy Observations<sup>b</sup></b>				
Ureters Distended, bilateral	0	0	0	1
Testes (large)	0	0	0	1
Epididymides (Discoloration)	0	0	0	1
No. Evaluated	9	10	9	9
<b>Organ Weights<sup>c</sup></b>				
Epididymides (g)	1.40	-6	0	-23*
Epididymides % Body Weight	0.286	-0.264	0.271	0.224*
No. Evaluated	9	10	9	9
<b>Sperm Morphology (#/200 sperm assessed)<sup>d</sup></b>				
Normal Appearance	192.3	196.3	195.6	11.1**
Detached Head	6.3	2.1	3.0	186.9**
Number of Males Affected	0	0	0	9
<b>Sperm Motility and Count</b>				
Percent Mobile (at 0 hours)	79.0	74.3	75.1	0.4**
Sperm Count per g Vas Deferens (x10 <sup>6</sup> )	47.2	32.4	47.7	10.6**
Spermatid Count per g Epididymis (x10 <sup>6</sup> )	434.2	447.8	428.3	319.1**
<b>Histopathology<sup>b</sup></b>				
<b>Testes</b>				
Tubular epithelial atrophy	0	0	0	3
Dilatation	0	0	0	3
Epididymal Oligospermia	0	0	0	3
Spermatocoele formation	0	0	0	1
<b>Reversal Phase:</b>				
No. Evaluated	9	9	9	8
<b>Male Mating Performance<sup>e</sup></b>				
No. of Males Mated & Produced a Pregnancy	9	NA	NA	8
Necropsy Observations	-	-	-	-
Organ Weights	-	-	-	-
<b>Sperm Morphology (#/200 sperm assessed)<sup>d</sup></b>				
Normal Appearance	195.9	195.3	195.8	177.1 <sup>f</sup>
Detached Head	2.2	4.1	2.6	21.6 <sup>f</sup>
Number of Males Affected	0	0	0	1 <sup>f</sup>
<b>Histopathology<sup>b</sup></b>				
Testicular Tubular Epithelial Atrophy	1	0	1	3
Leydig cell hyperplasia	0	0	0	1
Epididymides Oligospermia or Aspermia	0	0	0	3
Spermatocoele Formation	0	0	0	3

<sup>b</sup> Number of animals affected, <sup>c</sup> Cangrelor groups expressed as percent difference from vehicle control value, <sup>d</sup> Mean number of sperm affected /200 sperm assessed, <sup>e</sup> Males in the low and mid dose groups were not paired with females in the reversal period, <sup>f</sup> One male had high percentage of abnormal sperm with detached heads, other males were comparable to controls.

Tabulated Summary 6. **Effects of cangrelor on Fertility and early embryonic development in female rat**  
Study No.: SR 98074-01

Daily Dose (µg/kg/min)	0 (Control)	3	12	48
<b>F<sub>0</sub> Females</b>				
<b>Necropsy Observations<sup>b</sup></b>				
Kidney				
Bilateral renal pelvic dilatation	0	0	1	1
Ureters				
Distended	0	0	1	2
Premating Body Weight (%)	-	-	-	-
Gestation Body Weight (%)	-	-	-	-
Premating Food Consumption (%)	-	-	-	-
Gestation Food Consumption (%)	-	-	-	-
Number of Females Sperm Positive	20	20	19	19
Number of Pregnant Females	20	20	19	19
Number Aborted or with Total Resorption of Litter (Total Intrauterine Loss)	0	0	0	0
Mean Number Corpora Lutea	15.9	15.6	15.1	15.4
Mean Number Implantations	15.0	13.7	13.8	14.6
Mean % Per Litter Pre-implantation Loss	4.57	11.23	8.07	4.70
Mean Number Viable Fetuses	13.9	12.9	12.9	12.3
Mean % Per Litter Post-implantation Loss	7.58	5.96	6.53	15.97*

<sup>b</sup> Reported as number of animals affected, \* Significantly different from control mean,  $p \leq 0.01$

Tabulated Summary 7. **Effects of cangrelor on Embryo-fetal development study in rats**

Study No: SR 98002-01

Daily Dose (µg/kg/min)	0 (Control)	3	12	48
<b>F<sub>0</sub> Females</b>				
<b>Toxicokinetics: (Mean Plasma Concentration)</b>				
No. Animals	3	3	3	3
Cangrelor (ng/mL)				
Day 7 post coitum	<10	81.3	494	2350 <sup>d</sup>
Day 17 post coitum	<10	109	428	1820 <sup>d</sup>
AR-C89712XX <sup>a</sup> (ng/mL)				
Day 7 post coitum	<5	15.2	85.9	433
Day 17 post coitum	<5	21.0	88.5	462
<b>Main Study:</b>				
No. Animals	24	24	24	24
No. Pregnant	23	24	24	24
No. Aborted or with Total Resorption of Litter (Total loss of litter)	0	0	1	2 <sup>h</sup>
<b>Clinical Observations<sup>b</sup></b>				
Staining around nostrils	0	0	1	0
Fluid feces	0	0	1	0
Piloerection	0	0	0	1
Cold body surface	0	0	0	1
Red discharge in tray/cage	0	0	1	1
<b>Necropsy Observations<sup>c</sup></b>				
Kidney				
Pale kidney(s)	0	0	1	2
Spleen				
Enlarged	0	0	0	1
Dark	0	0	0	1
Large intestine				
Impacted	0	0	0	1
Stomach				
Pyloric region dark	0	0	0	1
Mean Body Weight Gain (g) (%) <sup>d,e</sup>				
Days 18-21 post coitum	58.2	59	56	46.4*
Food Consumption (g/animal/day) (%) <sup>d,e</sup>				
Days 18-21 post coitum	36.7	35.7	34.9	33.7*
Mean No. Corpora Lutea	15.52	15.71	16.00	16.39
Mean No. Implantations	15.00	14.79	15.00	14.35
Mean % Per Litter Pre-implantation Loss	3.51	5.76	5.72	9.83
<b>F<sub>1</sub> Litters</b>				
Total No. of Viable Litters	23	24	23	22
Total No. of Viable Fetuses	331	333	330	296
No. of Litters with Dead Fetuses	0	0	0	1 <sup>f</sup>
Mean % Per Litter Post-implantation Loss	4.02	5.90	4.27	9.81
Mean Fetal Body Weight (g)	5.69	5.52	5.48	5.46
Fetal Sex Ratios (% males)	52	45	44	49
<b>Fetal Anomalies:</b>				
Number of Fetuses Examined:	331	333	330	296+16DF <sup>g</sup>
Gross External	-	-	-	-
Number of Fetuses Examined:	170	169	170	161
Visceral Anomalies	-	-	-	-
Number of Fetuses Examined:	161	164	160	151
<b>Skeletal Anomalies (% examined fetuses affected)</b>				
Skull, squamosal – incomplete ossification	1	5	4	8*
Sternum, sternbra(e) – incomplete ossification	16	23	22	32**
Hindlimb, metatarsal(s) – not ossified	6	20**	16**	23**

<sup>a</sup> Metabolite of cangrelor, <sup>b</sup> Number of animals with clinical signs between Days 6 to 21 post coitum, <sup>c</sup> Number of animals affected, <sup>d</sup> Mean values, <sup>e</sup> Excludes TK animals, <sup>f</sup> Litter with 16 dead fetuses, <sup>g</sup> Includes 16 dead fetuses from one animal, <sup>h</sup> One animal cannibalized its litter, another animal with total intrauterine loss, <sup>i</sup> Mean value slightly different from issued report.

Tabulated Summary 8. **Effects of cangrelor on Embryo-fetal development study in rabbits**

Study No: SR 97297-01

Daily Dose (µg/kg/min)	0 (Control)	4	12	36
<b>F<sub>0</sub> Females</b>				
<b>Toxicokinetics: (Mean Plasma Concentration)</b>				
No. Animals <sup>a</sup>	3	3	3	3
Cangrelor (ng/mL)				
Day 7 post coitum	<50	2120	6210	18900
Day 18 post coitum	<50	1830	6060	21600
AR-C89712X <sup>b</sup> (ng/mL)				
Day 7 post coitum	<10	98.7	355	1150
Day 18 post coitum	<10	90.0	330	772
<b>Main Study:</b>				
No. Animals <sup>c</sup>	26	27	29	26
No. Pregnant <sup>e</sup>	20	18	24	24
No. Euthanized Early <sup>d</sup>	1	0	2	1
No. Aborted	1	1	3	3
Total No. Intrauterine Loss	1	0	4	3
Clinical Observations <sup>e</sup>				
Reduced fecal production	1	4	7	21
Reduced food consumption	0	3	4	17
Reduced water consumption	2	5	4	17
Necropsy Observations	-	-	-	-
Body Weight Change (kg)				
Days 6-18 post coitum	0.10	0.02*	0.04*	-0.06***
Mean No. Corpora Lutea	8.79	8.65	7.89	9.30
Mean No. Implantations	7.21	6.82	5.84	7.95
Mean % Per Litter Pre-implantation Loss	18.41	20.82	24.43	13.38
<b>F<sub>1</sub> Litters</b>				
Total No. of Viable Litters	18	17	15	17
Total No. of Viable Fetuses	123	104	92	124
No. of Litters with Dead Fetuses	0	0	0	0
Mean % Per Litter Post-implantation Loss	13.58	9.82	26.95	22.40
Mean Fetal Body Weight (g)				
Males	39.9	40.0	38.5	37.5
Females	39.5	40.4	39.0	36.1*
Fetal Sex Ratios (% males)	44.7	45.2	50.0	49.2
<b>Fetal Anomalies:</b>				
Number of Fetuses Examined:	123	104	92	124
Gross External	-	-	-	-
Number of Fetuses Examined:				
Visceral Anomalies (total No. fetuses affected)	0	2	1	7
Descending aorta, additional blood vessel				
Number of Fetuses Examined:	123	104	92	124
<b>Skeletal Anomalies (% examined fetuses affected)</b>				
Skull, additional suture line/fissure	0.8	0	3.3	4
Sternum, sternebrae, misshapen	4.1	2.9	1.1	8.1
Sternum, additional ossification center	0	3.8	3.3	4.8
Sternum, sternebrae, misaligned	3.3	7.7	6.5	7.3
Hindlimbs, heads, not ossified	0	3.8	5.4	13.7

<sup>a</sup> Number of animals, <sup>b</sup> Metabolite of cangrelor, <sup>c</sup> Number of animals mated including satellite animals, <sup>d</sup> No deaths, <sup>e</sup> Number of animals affected for >2 days, \* Significantly different from control p≤0.05, \*\* Significantly different from control p≤0.001

Tabulated Summary 9. **Effects of cangrelor on Pre and postnatal development study in rats**

Study No: 901045

Daily Dose (µg/kg/min)	0 (Control)	3	9	30
<b>F<sub>0</sub> Females</b>				
No. Pregnant	22	24	24	23
No. Died or Sacrificed Moribund	0	0	1	3
Clinical Observations	-	-	-	-
Necropsy Observations	-	-	-	-
Gestation Body Weight (%) <sup>a</sup>	404.8	0	0	0
Lactation Body Weight (%) <sup>a</sup>	346.8	0	0	0
Gestation Food Consumption (%) <sup>b</sup>	34.0	0	0	-7*
Mean Duration of Gestation (days)	21.5	21.4	21.3	21.2
Abnormal Parturition	-	-	-	-
<b>F<sub>1</sub> Litters (Prewaning)</b>				
No. Litters Evaluated				
Mean No. Pups/Litter	13.7	13.8	13.2	13.4
Mean No. Liveborn Pups/Litter	13.5	13.4	12.9	13.4
Mean No. Stillborn Pups/Litter	0.2	0.4	0.3	0
Postnatal Survival to Day 4	-	-	-	-
Postnatal Survival to Weaning	-	-	-	-
Change in Pup Body Weights (g)	-	-	-	-
Pup Sex Ratios (% males)	42.47	51.79*	53.73*	51.8*
Pup Clinical Signs	-	-	-	-
Pup Necropsy Observations	-	-	-	-
<b>F<sub>1</sub> Litters (Postweaning)</b>				
No. Placed for Mating	21	23	23	21
Mean No. Days Prior to Mating	3.6	3.1	2.8	3.1
No. Mating	19	22	23	20
No. Pregnant Females	19	22	22	19
<b>F<sub>1</sub> Males (Postweaning)</b>				
No. Evaluated Postweaning	21	23	23	21
No. Died or Sacrificed Moribund	0	0	0	0
Clinical Observations	-	-	-	-
Necropsy Observations	-	-	-	-
Body Weight Change (g)	473.2	475.5	472.1	475.5
<b>F<sub>1</sub> Females (Postweaning)</b>				
No. Evaluated Postweaning				
No. Died or Sacrificed Moribund	1	0	0	0
Clinical Observations	-	-	-	-
Necropsy Observations	-	-	-	-
Premating Body-Weight Change (g)	209.1	202.9	207.3	211.8
Gestation Body-Weight Change (g)	138.2	143.9	138.3	145.4
Mean Age of Vaginal Patency (days)	-	-	-	-
Sensory Function	-	-	-	-
Motor Activity	-	-	-	-
Learning and Memory	-	-	-	-
Mean No. Days Prior to Mating	3.6	3.1	2.8	3.1
No. of Pregnant Females	19	22	22	19
Live Birth Index (%)	93.2	91.2	92.1	94.2
Day 4 Viability Index (%)	99.3	98.8	99.7	99.6
<b>F<sub>2</sub> Litters</b>				
Group Mean Litter Size	14.2	14.5	13.7	14.3
No. of Dead Conceptuses/Litter	0.2	0.2	0.3	0.4
Mean Group Fetal Body Weights (g)	6.8	6.9	6.8	6.8
Sex Ratios (% males)	47	45	50	55
Fetal Anomalies	-	-	-	-

A, b = At end of gestation or lactation

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/s/  
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BELAY TESFAMARIAM  
12/13/2013

ALBERT F DEFELICE  
01/10/2014

## PHARMACOLOGY/TOXICOLOGY FILING CHECKLIST FOR NDA

**NDA Number:** 204958

**Applicant:** The Medicines Co

**Stamp Date:** 05/06/2013

**Drug Name:** Cangrelor (b) (4) **NDA Type:** 505(b)(1)

On **initial** overview of the NDA/BLA application for filing:

	<b>Content Parameter</b>	<b>Yes</b>	<b>No</b>	<b>Comment</b>
1	Is the pharmacology/toxicology section organized in accord with current regulations and guidelines for format and content in a manner to allow substantive review to begin?	√		e-submission, eCTD format
2	Is the pharmacology/toxicology section indexed and paginated in a manner allowing substantive review to begin?	√		Reviewable submission
3	Is the pharmacology/toxicology section legible so that substantive review can begin?	√		Acceptable
4	Are all required (*) and requested IND studies (in accord with 505 b1 and b2 including referenced literature) completed and submitted (carcinogenicity, mutagenicity, teratogenicity, effects on fertility, juvenile studies, acute and repeat dose adult animal studies, animal ADME studies, safety pharmacology, etc)?	√		Carcinogenicity studies are not applicable because cangrelor is intended for short-term administration. Juvenile studies were not performed.
5	If the formulation to be marketed is different from the formulation used in the toxicology studies, have studies by the appropriate route been conducted with appropriate formulations? (For other than the oral route, some studies may be by routes different from the clinical route intentionally and by desire of the FDA).	√		Cangrelor for injection reconstituted in NaCl 0.9%
6	Does the route of administration used in the animal studies appear to be the same as the intended human exposure route? If not, has the applicant <u>submitted</u> a rationale to justify the alternative route?	√		Intravenous infusion
7	Has the applicant <u>submitted</u> a statement(s) that all of the pivotal pharm/tox studies have been performed in accordance with the GLP regulations (21 CFR 58) <u>or</u> an explanation for any significant deviations?	√		GLP and QA statements are included.
8	Has the applicant submitted all special studies/data requested by the Division during pre-submission discussions?	√		Investigative kidney and urinary tract toxicity studies included.

File name: 5\_Pharmacology\_Toxicology Filing Checklist for NDA\_BLA or Supplement 010908

## PHARMACOLOGY/TOXICOLOGY FILING CHECKLIST FOR NDA

	Content Parameter	Yes	No	Comment
9	Are the proposed labeling sections relative to pharmacology/toxicology appropriate (including human dose multiples expressed in either mg/m <sup>2</sup> or comparative serum/plasma levels) and in accordance with 201.57?		√	Preclinical studies were performed as continuous IV infusion. Dosing regimen in PCI trial is bolus injection, and thus C <sub>max</sub> may be a better comparator of exposures than AUC.  BRIDGE trial is continuous infusion, and AUC shows adequate safety multiples.
10	Have any impurity – etc. issues been addressed? (New toxicity studies may not be needed.)	√		No issues of impurities. Genotoxicity studies were also conducted on base metabolites of cangrelor. Metabolic profile of cangrelor in animals is similar to humans, and no metabolites specific to humans need to be qualified.
11	Has the applicant addressed any abuse potential issues in the submission?		√	Not applicable
12	If this NDA/BLA is to support a Rx to OTC switch, have all relevant studies been submitted?		√	Not applicable

**IS THE PHARMACOLOGY/TOXICOLOGY SECTION OF THE APPLICATION FILEABLE? Yes**

**If the NDA/BLA is not fileable from the pharmacology/toxicology perspective, state the reasons and provide comments to be sent to the Applicant.**

This NDA is fileable.

**Please identify and list any potential review issues to be forwarded to the Applicant for the 74-day letter.**

The preclinical studies were performed as continuous IV infusion for 4 weeks, whereas the dosing regimen in the PCI trial is as a bolus injection followed by low dose infusion for 2 hrs. Thus, for a drug characterized by rapid acting, short-duration and rapid clearance, the C<sub>max</sub> (steady state plasma concentration) appears to be a better comparator of exposures than the AUC. A request will be made to the Sponsor to provide a plot of semi-log concentration vs time profile of cangrelor and ARL 69712XX metabolite to assess exposure comparability of the preclinical studies to the dosing regimen in the PCI as a bolus injection.

Belay Tesfamariam	6/28/2013
Reviewing Pharmacologist	Date
Albert DeFelice	6/28/2013
Team Leader/Supervisor	Date

File name: 5\_Pharmacology\_Toxicology Filing Checklist for NDA\_BLA or Supplement 010908

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/s/  
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BELAY TESFAMARIAM  
07/01/2013

ALBERT F DEFELICE  
07/02/2013