

EFFECT OF DIPYRIDAMOLE ON ATP METABOLISM IN THE RED BLOOD
CELLS AND CARDIOVASCULAR PROTECTION

by

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*This dissertation is dedicated to my mom, dad, brother and friends. Thank you for all of
your love, support, and belief in me.*

Table Of Contents

LIST OF TABLES	vi
LIST OF FIGURES	vii
ABSTRACT.....	ix
LIST OF ABBREVIATIONS AND SYMBOLS USED	x
ACKNOWLEDGEMENTS	xiv
CHAPTER: 1 INTRODUCTION.....	1
1.1 Adenosine And Cardiovascular Protection	2
1.2 Adenosine Production And Metabolism In The Cardiovascular System	3
1.3 Adenosine Receptors And Cardiovascular Protection	7
1.4 Nucleoside Transport Inhibitors (NTI) And Cardioprotection.....	8
1.5 Dipyridamole And Cardiovascular Protection	9
CHAPTER: 2 RESEARCH OBJECTIVES, SIGNIFICANCE AND SCOPE OF THE PROJECT	11
CHAPTER: 3 MATERIALS AND METHODS	12
3.1 HPLC Assay To Determine ATP And Adenine Nucleotides Concentrations In RBC Lysate In Rats.	12
3.1.1 Chemical And Reagents.....	12
3.1.2 Instrumentation	12
3.1.3 Preparation Of Stock Solutions, Spiking Solutions And Internal Standard Solutions	14

3.1.4 Solid Phase Extraction And HPLC Procedure.....	17
3.2 HPLC Assay To Determine Plasma Concentration Of Adenosine And Its Purine Metabolites	18
3.2.1 Chemical And Reagents.....	18
3.2.2 Instrumentation	18
3.2.3 Preparation Of Stock Solutions, Spiking Solutions And Internal Standard Solutions	20
3.2.4 Solid Phase Extraction And HPLC Procedure.....	21
3.3.1 Chemical, Reagents And Consumables	23
3.3.2 Instrumentation	23
3.3.3 Preparation Of Stock Solutions, Spiking Solutions And Internal Standard Solutions	25
3.3.4 Solid Phase Extraction And HPLC Procedure.....	26
3.4 Animal Study.....	27
3.5 Data Analysis.....	28
CHAPTER: 4 RESULTS	30
4.1 Mortality	30
4.2 Concentrations Of ATP, Adenosine, Dipyridamole And Their Metabolites....	31
4.2.1 RBC Concentrations Of Purine Nucleotides.....	31
4.2.2 Plasma Concentrations of Adenosine and its Purine Metabolites	41
4.2.3 Plasma Concentrations of Dipyridamole	53
4.2.4 Assay Reproducibility During Analysis	55

CHAPTER: 5 DISCUSSION	59
CHAPTER: 6 LIMITATIONS AND FUTURE DIRECTIONS.....	67
CHAPTER: 7 CONCLUSION.....	68
REFERENCES.....	69
APPENDIX 1: Rat 371	76
APPENDIX 2: Rat 372	89
APPENDIX 3: Rat 373	102
APPENDIX 4: Rat 374	115
APPENDIX 5: Rat 375	128
APPENDIX 6: Rat 376	141
APPENDIX 7: Rat 377	154
APPENDIX 8: Rat 378	167

LIST OF TABLES

<i>Table 1: Mortality of rats induced by isoproterenol in normal saline control and dipyridamole treated groups.</i>	30
<i>Table 2: Pharmacokinetic of dipyridamole in rats following the last subcutaneous (sc) injection (10 mg/kg).</i>	54
<i>Table 3: Summary of the reproducibility data of HPLC Assay for Purine nucleotides in RBC.</i>	56
<i>Table 4: Summary of the reproducibility data of HPLC Assay for adenosine and its metabolites in plasma.</i>	57
<i>Table 5: Summary of the reproducibility data of HPLC Assay for dipyridamole in plasma</i>	58
<i>Table 6: Optimization of SPE of dipyridamole and losartan from plasma samples</i>	61
<i>Table 7: Effect of dipyridamole on changes of RBC ATP and adenine nucleotides concentration after isoproterenol injection</i>	62
<i>Table 8: Effect of dipyridamole on RBC ATP and adenine nucleotide ratios before isoproterenol injection</i>	63
<i>Table 9: Effect of dipyridamole on RBC ATP and adenine nucleotide ratios after isoproterenol injection</i>	63
<i>Table 10: Effect of dipyridamole on changes of adenosine and its metabolites after isoproterenol injection.</i>	65
<i>Table 11: Effect of dipyridamole on adenosine and its purine metabolite ratios in plasma before isoproterenol injection in rats</i>	66
<i>Table 12: Effect of dipyridamole on adenosine and its purine metabolite ratios in plasma after isoproterenol injection in rats</i>	66

LIST OF FIGURES

<i>Figure 1: ATP and adenosine metabolism in the myocardium.....</i>	<i>5</i>
<i>Figure 2: ATP and adenosine metabolism in the endothelial cells and RBC's.....</i>	<i>6</i>
<i>Figure 3: Effect of dipyridamole (10 mg/kg) on RBC ATP concentration vs. normal saline control. (Data presented in mean \pm SEM).....</i>	<i>31</i>
<i>Figure 4: AUC of ATP in RBC after isoproterenol injection.</i>	<i>32</i>
<i>Figure 5: Effect of dipyridamole (10 mg/kg) on RBC ADP concentration vs. normal saline control. (Data presented in mean \pm SEM).....</i>	<i>33</i>
<i>Figure 6: AUC of ADP in RBC before isoproterenol injection.</i>	<i>33</i>
<i>Figure 7: AUC of ADP in RBC after isoproterenol injection.....</i>	<i>34</i>
<i>Figure 8: Cmax of ADP in RBC after isoproterenol injection.</i>	<i>34</i>
<i>Figure 9: Effect of dipyridamole (10 mg/kg) on RBC AMP concentration vs. normal saline control. (Data presented in mean \pm SEM).....</i>	<i>35</i>
<i>Figure 10: AUC of AMP in RBC before isoproterenol injection.....</i>	<i>36</i>
<i>Figure 11: AUC of AMP in RBC after isoproterenol injection.</i>	<i>36</i>
<i>Figure 12: Cmax of AMP in RBC after isoproterenol injection.</i>	<i>37</i>
<i>Figure 13: Effect of dipyridamole (10 mg/kg) on RBC GTP concentration vs. normal saline control. (Data presented in mean \pm SEM).....</i>	<i>38</i>
<i>Figure 14: AUC of GTP in RBC before isoproterenol injection.</i>	<i>38</i>
<i>Figure 15: Cmax of GTP in RBC after isoproterenol injection.....</i>	<i>39</i>
<i>Figure 16: AUC of GTP in RBC after isoproterenol injection.</i>	<i>39</i>
<i>Figure 17: Effect of dipyridamole on (10 mg/kg) on RBC GDP concentrations vs. normal saline control. (Data presented in mean \pm SEM).....</i>	<i>40</i>
<i>Figure 18: Cmax of GDP in RBC after isoproterenol injection.....</i>	<i>41</i>
<i>Figure 19: Effect of dipyridamole (10mg/kg) on plasma adenosine concentrations vs. normal saline control. (Data presented in mean \pm SEM).....</i>	<i>42</i>

<i>Figure 20: AUC of adenosine in plasma before isoproterenol injection.....</i>	<i>42</i>
<i>Figure 21: AUC of adenosine in plasma after isoproterenol injection.</i>	<i>43</i>
<i>Figure 22: Cmax of adenosine in plasma after isoproterenol injection.</i>	<i>43</i>
<i>Figure 23: Effect of dipyridamole (10mg/kg) on plasma inosine concentrations vs. normal saline control. (Data presented in mean ± SEM).</i>	<i>44</i>
<i>Figure 24: AUC of inosine in plasma before isoproterenol injection.</i>	<i>45</i>
<i>Figure 25: AUC of inosine in plasma after isoproterenol injection.</i>	<i>45</i>
<i>Figure 26: Effect of dipyridamole (10mg/kg) on plasma hypoxanthine concentrations vs. normal saline control. (Data presented in mean ± SEM).</i>	<i>46</i>
<i>Figure 27: AUC of hypoxanthine in plasma before isoproterenol injection.</i>	<i>47</i>
<i>Figure 28: AUC of hypoxanthine in plasma after isoproterenol injection.</i>	<i>47</i>
<i>Figure 29: Effect of dipyridamole (10mg/kg) on plasma uric acid concentrations vs. normal saline control. (Data presented in mean ± SEM).</i>	<i>48</i>
<i>Figure 30: AUC of uric acid in plasma before isoproterenol injection.</i>	<i>49</i>
<i>Figure 31: AUC of uric acid in plasma after isoproterenol injection.</i>	<i>49</i>
<i>Figure 32: Cmax of uric acid in plasma after isoproterenol injection.</i>	<i>50</i>
<i>Figure 33: Effect of dipyridamole (10mg/kg) on plasma guanosine concentrations vs. normal saline control. (Data presented in mean ± SEM).</i>	<i>51</i>
<i>Figure 34: AUC of guanosine concentrations in plasma before isoproterenol injection..</i>	<i>52</i>
<i>Figure 35: AUC of guanosine concentrations in plasma after isoproterenol injection. ...</i>	<i>52</i>
<i>Figure 36: Plasma concentration time profile of dipyridamole in rats after the 5th subcutaneous injection of dipyridamole (10 mg/kg).</i>	<i>53</i>

ABSTRACT

Dipyridamole (DYP) is an adenosine uptake inhibitor used clinically as a coronary vasodilator, although the mechanism of action is not fully understood. The objective of my thesis research was to study the effect of DYP on ATP metabolism in the red blood cell (RBC) in response to acute cardiovascular injury induced by isoproterenol (30 mg/kg). Rats were each received either 10 mg/kg of DYP or normal saline twice daily for 5 doses by subcutaneous (sc) injection. Blood samples were collected from 0 to 6 hours for measurement of circulating ATP and adenosine concentrations. Isoproterenol induced 50% mortality and increased the breakdown of ATP in the RBC to AMP and adenosine in the control group. Treatment with DYP decreased mortality to 25%, and significantly reduced the breakdown of ATP in the RBC. In conclusion, DYP is protective against cardiovascular injury induced by isoproterenol most likely by preserving ATP in the RBC.

LIST OF ABBREVIATIONS AND SYMBOLS USED

ADO	Adenosine
ADP	Adenosine diphosphate
AMP	Adenosine monophosphate
AR	Adenosine receptors
ATP	Adenosine triphosphate
AUC	Area under the curve
cAMP	Cyclic Adenosine monophosphate
cGMP	Cyclic guanosine monophosphate
CL	Clearance
C _{max}	Maximum concentration
C _{mean}	Average concentration
CN	Cyanopropylsilica
CVD	Cardiovascular disease
C18	Octadecyl carbon chain
DMUA	3-7 Dimethyluric acid

DYP	Dipyridamole
EDTA	Ethylenediaminetetraacetic acid disodium salt
EHNA	Erythro-9- (2-hydroxyl-3-nonyl) adenine
GDP	Guanosine diphosphate
GMP	Guanosine monophosphate
GTP	Guanosine triphosphate
HCL	Hydrochloric acid
HOM	Homocysteine
HPLC	High performance liquid chromatography
HYP	Hypoxanthine
IHD	Ischemic heart disease
IMP	Inosine monophosphate
INO	Inosine
ISF	Interstitial fluid
ISO	Isoproterenol
I.V	Intravenous

KH_2PO_4	Potassium phosphate monobasic
Kpsi	Kilo pound per square inch
MDC	Methylene dichloride
Meth	Methioine
MI	Myocardial Ischemia/Infarction
MTBE	Methyl tert-butyl ether
NaOH	Sodium hydroxide
NBMPR	S-(4-Nitrobenzyl)-6-thioinosine
$\text{NH}_4\text{H}_2\text{PO}_4$	Ammonium dihydrogen orthophosphate
NTI	Nucleoside transport inhibitors
PBS	Phosphate buffer saline
PEG	Polyethylene Glycol
pH	Negative log of hydrogen ion concentration
PTFE	Polytetrafluoroethylene
QC	Quality control
RBC	Red blood cell

SAH	S-adenosyl homocysteine
SAM	S-adenosyl methionine
SC	Subcutaneous
SD	Sprague Dawley
SPE	Solid phase extraction
TBAS	Tetrabutylammonium hydrogen sulfate
TCA	Trichloroacetic acid
TEA	Triethylamine
Tmax	Time to reach maximum concentration
UA	Uric acid
ug/mL	Micrograms per milliliter
ug/L.h	Micrograms per liter times hour
mm	Millimeter
uL	Microliter
mmHG	Millimeter of mercury

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CHAPTER: 1 INTRODUCTION

Cardiovascular disease (CVD) is one of the leading causes of death globally, accounting for 17.3 million deaths every year [1]. Based on the latest statistics for heart and stroke disease, 85.6 millions of American adults have more than one type of cardiovascular diseases of which 80 millions suffer from hypertension and 15.5 millions from ischemic heart diseases (IHD) such as myocardial ischemia (MI), angina pectoris and heart failure. The annual expenditure in the US for physicians, hospital service, medication, home healthcare including the loss of future productivity due to CVD and stroke was estimated for a total of \$320 billions in 2011 [1]. The total direct medical cost of CVD in the US alone is projected to reach \$918 billions by 2030 [2]. Despite current advancement in the treatment for cardiovascular ailments its estimated prevalence in the future is still unacceptably high. Thus focus on prevention by better diagnosis and effective drug treatment could significantly reduce the growing healthcare costs.

Cardiovascular biomarkers can be a useful tool for early identification of individuals at risk for CVD and to stratify patients for the most suitable treatment. A biomarker is defined as a characteristic that is objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes, or pharmacologic responses to a therapeutic intervention [3]. Although clinical use of current cardiovascular biomarkers is growing, finding an ideal biomarker with predictive value for prognostic assessment and optimum therapy remains a challenge. It has been known for many years adenosine and its catabolites such as adenosine monophosphate (AMP), adenosine diphosphate (ADP)

and adenosine triphosphate (ATP) play an important role in myocardial energy metabolism [4, 5]. More recently, it has been shown ATP was broken down to AMP in red blood cell (RBC) which was closely associated with cardiovascular mortality in an experimental rat model of acute myocardial infarction [6]. This suggests that adenosine and ATP metabolism in the RBC may be used as a biomarker for cardiovascular protection and a target for anti-ischemic drugs, which is one of the main hypotheses of my thesis project.

1.1 Adenosine And Cardiovascular Protection

Adenosine is a well-known endogenous purine nucleoside widely distributed in the body. It is also an important intermediary metabolite of adenosine 5'-triphosphate (ATP) metabolism. The role of adenosine as a signaling molecule in cardiovascular system and its cardioprotective effects has been studied for nearly half a century [7, 8]. Adenosine mediates numerous physiological effects beneficial to the cardiovascular system [9] which include negative chronotropic and dromotropic effect on the heart tissue, vasodilatation, inhibition of platelet aggregation, and ischemic preconditioning [10]. Adenosine exerts the actions by activating the adenosine receptors (AR) present in the surface membrane of most cells in the body. Production and maintaining an adequate level of adenosine in the myocardium reflects its vitality and has an important role in maintaining a balanced cardiac energy metabolism [11].

In response to stress induced by ischemia, hypoxia, trauma, seizure and inflammation there is a considerable increase in demand for energy, which triggers a rapid increase in ATP catabolism and subsequent release of adenosine resulting in elevated levels of

adenosine in both intracellular and extracellular space [12, 13]. The increased concentration of adenosine mediates the protective effects by preventing any further damage to the myocardium [14]. Several studies, both in animal models and humans have shown a wide range of cardiovascular effects of adenosine in the past few decades. These studies have established an abundant amount of knowledge and encouraged future research work to explore further the role of adenosine, adenosine agonists and adenosine re-uptake inhibitors as cardiovascular protective agents [15].

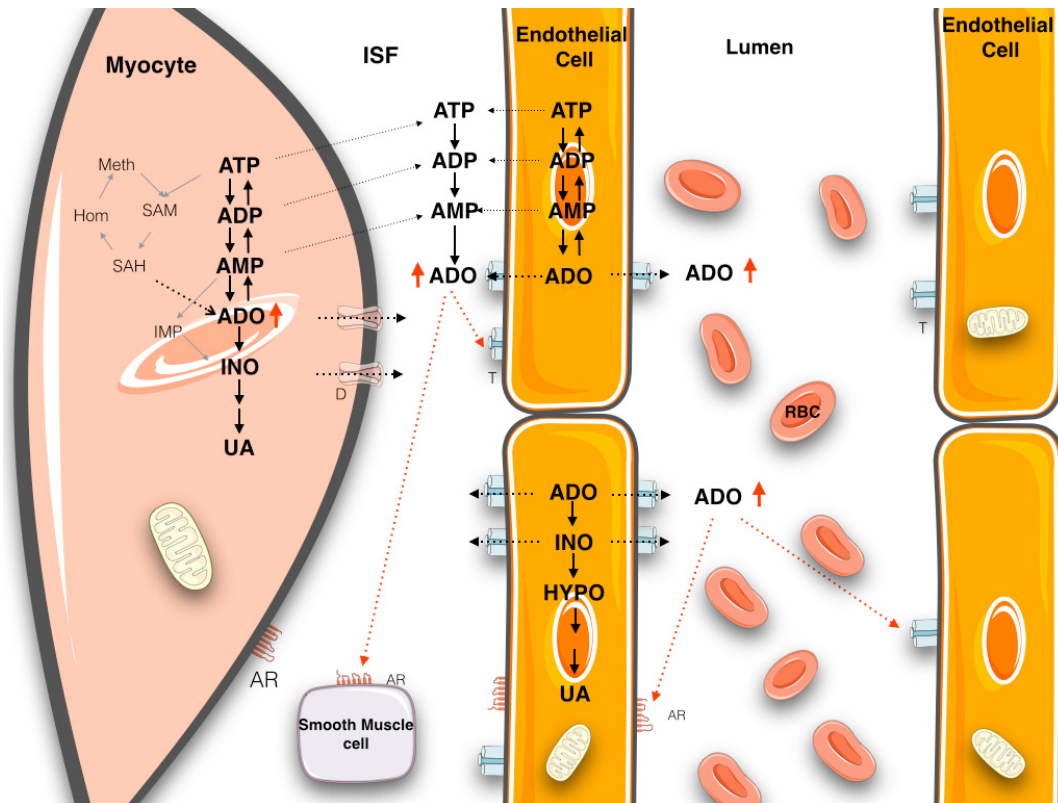
1.2 Adenosine Production And Metabolism In The Cardiovascular System

Under physiological conditions the concentration of adenosine in plasma and other extracellular fluid is low because of a constant uptake by nucleoside transporters [16]. The primary source of intracellular adenosine is from catabolism of intracellular ATP to ADP and then to AMP, which is subsequently catabolized by endo 5' nucleotidase to adenosine [17]. Another source of intracellular adenosine is via intracellular hydrolysis of S-adenosylhomocysteine derived from S-adenosylmethionine in the transmethylation pathway (Figure 1) [18]. The adenosine generated may be released to the plasma and other extracellular space or converted to inosine by adenosine deaminase [19, 20]. Both adenosine and inosine produced intracellularly may be released into extracellular space by similar nucleoside transporters [21]. Pending on the cellular demand and supply of energy, it may also undergo rephosphorylation to form AMP by adenosine kinase, and other higher nucleotides by adenylate kinase [22-24]. These metabolic reactions are known to occur in the myocardium, endothelium and the RBC as shown in Figures 1 and 2. While inosine (INO) is further metabolized to hypoxanthine (HYP) and other

oxypurine metabolites (i.e. xanthine, uric acid, etc.) in myocardium and endothelium [19, 25], it is not clear if the same also occurs in the RBC.

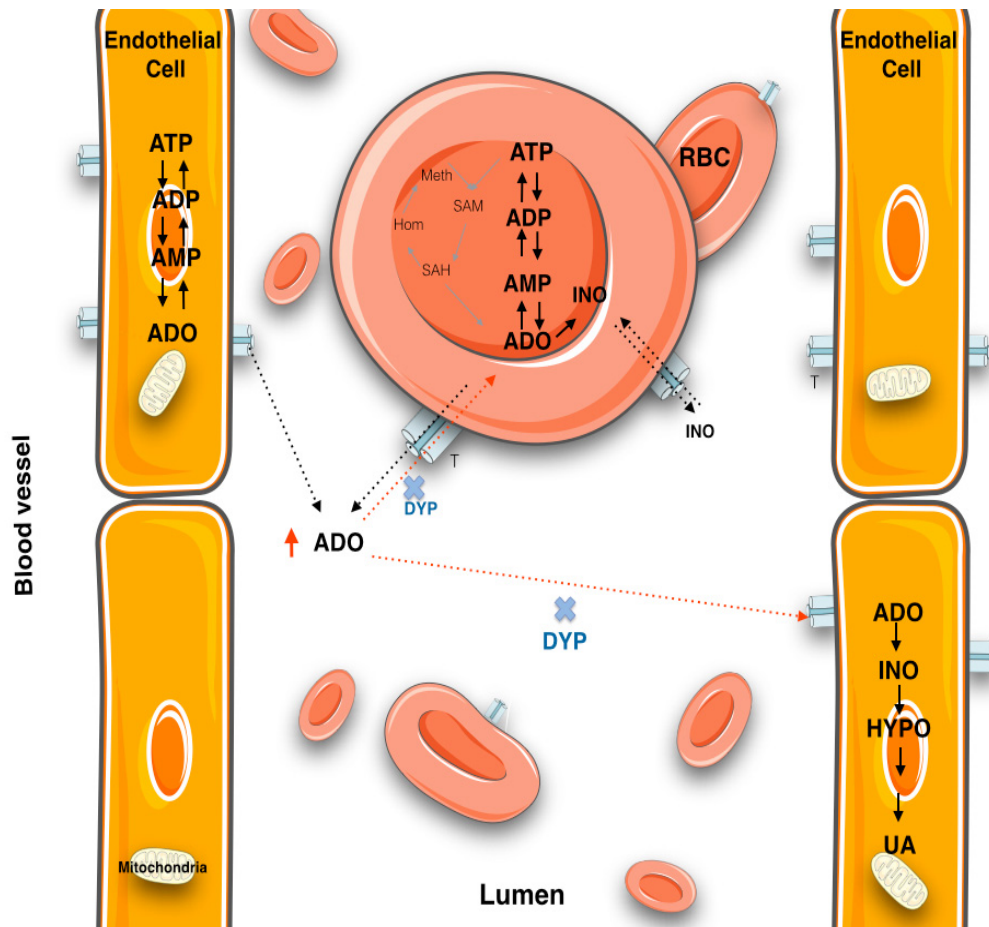
However, during ischemia/hypoxia or excessive physical work there is an increase in demand of energy causing an imbalance between energy supply and demand, which triggers the breakdown of ATP to produce other high energy phosphates (eg: ADP and AMP) and adenosine [26]. This leads to an increase of adenosine concentration in the myocytes, vascular endothelium, and RBC which is released into the interstitial fluid and other extracellular space [27] [6]. The effect of adenosine is short-lived as it is rapidly taken up back to RBC's and endothelium by the nucleoside transporters. The adenosine returning to the cells is then utilized to regenerate adenine nucleotides (AMP, ADP and ATP) in myocardium, endothelium as well as in RBC as shown in Figures 1 and 2 to maintaining adequate level of intracellular ATP by adenylate kinase [20, 28, 29]. However, RBC does not have mitochondria, the mechanism of regulating ATP metabolism in the RBC may be different than in other cell types. It has been shown that RBC's also express AMP- and guanosine mono phosphate (GMP)- dependent protein kinases which are known to be involved in energy metabolism in the RBCs [30] [31]. These enzymes regulating ATP metabolism in the RBC may play a pivotal role in energy metabolism and cardiovascular homeostasis, which is an important hypothesis yet to be tested.

Figure 1: ATP and adenosine metabolism in the myocardium.



ATP/ADP/AMP=Adenosine triphosphate/diphosphate/ monophosphate,
 ADO=Adenosine, INO=Inosine, IMP=inosinemonophosphate, Hom=Homocysteine,
 Meth=Methioine, SAM=S-adenosylethionine, SAH=S-adenosylhomocysteine,
 HYPO=Hypoxanthine, UA=Uric acid, RBC=Red blood cell, D=Diffusion,
 T=Transporter, AR=Adenosine rector, ISF=Interstitial fluid.

Figure 2: ATP and adenosine metabolism in the endothelial cells and RBC's.



ATP/ADP/AMP=Adenosine triphosphate/diphosphate/ monophosphate,
 ADO=Adenosine, INO=Inosine, IMP=inosinemonophosphate, Hom=Homocysteine,
 Meth=Methioine, SAM=S-adenosylethionine, SAH=S-adenosylhomocysteine,
 HYPO=Hypoxanthine, UA=Uric acid, RBC=Red blood cell, D=Diffusion,
 T=Transporter, AR=Adenosine receptor, ISF=Interstitial fluid.

1.3 Adenosine Receptors And Cardiovascular Protection

Adenosine exerts a range of potentially beneficial cardiovascular effects [32]. It is also a key mediator in ischemic preconditioning which is an important factor responsible for cardiovascular protection [33]. Adenosine mediates these effects via membrane bound adenosine receptors (AR's) coupled to G-proteins, which are subdivided into 4 different subtypes: A₁, A_{2a}, A_{2b} and A₃ [34-36]. Interaction between the activated adenosine receptor (AR) subtypes mediate and regulates a specific cardiovascular response. For example, vascular tone is regulated by the cross talks between A₁ receptor mediated vasoconstriction and A₂ receptor mediated vasodilatation [37]. Similarly, activation of A₂ receptor increases myocardial contractility, which is attenuated by the response mediated via A₁ subtype [38]. It was also shown that cardiovascular protection elicited by adenosine against myocardial ischemia is mediated by activation of the A₁ receptor [39]. Furthermore evidence suggests that adenosine receptors can also interact with opioid receptors which together mediate a response to limit the injury and cell death during ischemia – reperfusion [40]. Several experimental studies have demonstrated a key role of adenosine in reactive hyperemia and cardioprotection by increasing coronary blood flow and attenuating breakdown of ATP in the myocardium during ischemia and facilitating its repletion during reperfusion [41-44]. However due to the rapid cellular uptake of adenosine into RBC, myocardium and endothelium by nucleoside transporters, its pharmacological effects are extremely short lived (<1 min) [45, 46].

Several therapeutic strategies and pharmacologic tools have been explored to prolong the action of adenosine. Intra-coronary infusion of adenosine exhibited cardioprotection

in a rabbit model of ischemia and reperfusion, but clinical application of this approach is restricted and not practical in most clinical settings. Another approach to potentiate the actions of adenosine is use of adenosine agonists, which are much more stable and longer acting. However finding a suitable adenosine receptor agonist with optimum safety and efficacy profile for cardiovascular and inflammatory diseases remains a therapeutic challenge [8]. The complexity of adenosine signaling contributes to some debilitating side effects of adenosine receptor agonists [47]. An alternate therapeutic strategy is to inhibit adenosine uptake by the transporter to potentiate the beneficial effects of adenosine in the cardiovascular system [28]. Administration of exogenous adenosine in the presence of an inhibitor of the transporter such as calcium channel blockers or dipyridamole has been shown to enhance the action of adenosine in vivo in animal models and in patients [48-50]. These approaches have many potential therapeutic implications and warrant further investigation.

1.4 Nucleoside Transport Inhibitors (NTI) And Cardioprotection

Experimental studies have shown that nucleoside transport inhibitors (NTI) such as R75231 improves the functional recovery from an isolated rabbit heart model of ischemia followed by reperfusion [51]. Similarly, dipyridamole and other NTI such as dilazep have been reported to show cardioprotective effect via enhancing the activity of endogenous adenosine on adenosine A1 receptor in various animal models of myocardial ischemia [52, 53]. Further, anti-platelet agents including dipyridamole have been shown to reduce myocardial necrosis induced by epinephrine and that lidoflazine was beneficial for coronary artery surgery [54, 55]. The order of potency inhibiting adenosine uptake by

RBC is draflazine \geq dilazep $>$ R75231 $>$ S-(4-Nitrobenzyl)-6-thioinosine (NBMPR) $>$ mioflazine = dipyridamole $>$ lidoflazine as suggested by previous workers [56]. Experimental studies using mammalian cardiac myocytes, showed that adenosine attenuated the stimulatory effects of isoproterenol via blockade of calcium influx [57], which partly explains the cardiovascular protective effect of calcium channel blockers [58-61], although these agents may also have a direct effect inhibiting the adenosine transporters [62].

1.5 Dipyridamole And Cardiovascular Protection

Dipyridamole is a well-known coronary vasodilator and an antiplatelet agent, with diverse mechanism of action. It inhibits phosphodiesterase enzyme in platelets, which increases cyclic AMP (cAMP) and cyclic GMP (cGMP) levels thereby inhibiting platelet aggregation [63, 64]. It also acts as a nucleoside transport inhibitor to block the adenosine reuptake into the RBC and endothelium thereby increasing plasma adenosine concentration which attributes to its coronary vasodilatory effect [65]. Elevated adenosine level also stimulates adenylyl cyclase in platelets to increase cAMP resulting in inhibition of platelet aggregation [63, 64]. Further, dipyridamole has direct and indirect beneficial actions on the endothelium such as inhibition of proliferation, anti-oxidant and anti-inflammatory effect [66]. Apart from being used clinically as an anti-platelet agent, it is also widely used as a coronary vasodilator in pharmacologic stress myocardial perfusion imaging [67].

Following a bolus intravenous (I.V) dose in man, plasma levels of dipyridamole decreased tri-exponentially when monitored up to 60 hours with half-lives ($T_{1/2}$) of 5

minutes, 53 minutes and 10-12 hours. Its volume of distribution is 140 litres with 92-99% bound to plasma proteins [68]. As an effective anti-platelet agent used clinically with a mechanism of action inhibiting adenosine re-uptake by the RBC and endothelium, it is a suitable drug candidate for my thesis work to probe the importance of adenosine and ATP metabolism in the RBC for cardiovascular protection.

CHAPTER: 2 RESEARCH OBJECTIVES, SIGNIFICANCE AND SCOPE OF THE PROJECT

The aim of this research is to study the effect of dipyridamole on ATP metabolism and cardiovascular protection and to investigate the role of ATP and adenine nucleotides in the RBC in acute myocardial infarction induced by isoproterenol. Also it assesses the feasibility of adenosine and ATP metabolism in the RBC as biomarker targets for anti-ischemia drugs. To accomplish these objectives our research adopts HPLC assays to measure concentrations of ATP and adenine nucleotides in RBC lysate, plasma concentration of adenosine and its oxypurine metabolites and plasma concentration of dipyridamole. The data are analyzed by standard pharmacokinetic and pharmacodynamics procedures and compared between before and after isoproterenol, and also between the rats treated with and without dipyridamole. Results of this research will advance the knowledge of the action of dipyridamole on ATP metabolism and support the role of adenosine and ATP metabolism in RBC as a potential sensitive biomarker for CVD.

CHAPTER: 3 MATERIALS AND METHODS

3.1 HPLC Assay To Determine ATP And Adenine Nucleotides Concentrations In RBC Lysate In Rats.

3.1.1 Chemical And Reagents

Dipyridamole was obtained from Boehringer-Mannheim Canada Ltd (Laval, QC, Canada). Adenosine monophosphate (AMP), adenosine diphosphate (ADP), adenosine triphosphate (ATP), guanosine monophosphate (GMP), guanosine diphosphate (GDP), guanosine triphosphate (GTP) were purchased from Sigma-Aldrich Chemical Co. (PO Box 14508 ST. Louis MO 63178, USA). Ethylenediaminetetraacetic acid disodium salt (EDTA), and methyl tert-butyl ether (MTBE) were purchased from BDH Chemicals Inc. (Dartmouth, NS, Canada). 3-7 Dimethyluric acid (DMUA) was acquired from Sigma-Aldrich Chemical Co (St. Louis, MO, USA). Tetrabutylammonium hydrogen sulfate (TBAS) was purchased from Fluka Chemicals, Sigma-Aldrich Canada Ltd, (Oakville, ON, Canada). Conical and round bottom glass culture tubes (100 x 16 mm I.D.) with polytetrafluoroethylene (PTFE) - lined screw caps (Kimax Brand) and graduated glass pipettes were procured from Fisher Scientific (Ottawa, ONT, Canada). Methanol, acetonitrile and other solvents were HPLC grade and all other chemicals were reagent grade and purchased from Fisher Scientific Ltd. (Ottawa, ONT, Canada).

3.1.2 Instrumentation

3.1.2.1 High Performance Liquid Chromatography System

The high performance liquid chromatography system used in this assay consisted

of a Shimadzu LC-10AT *vp* solvent delivery module (Man-Tech Associate Inc., Guelph, ON, N1H 6J3), a 250 mm × 3.0 mm I.D. Supelcosil™ LC-18-T column bonded to a 5 µm spherical silica packing, pore size 120 Å (Supelco Inc., Bellefonte, PA, USA purchased from Sigma-Aldrich Canada, Oakville, ON), a HPLC guard column (5 µm 4.0 mm × 4.0 mm I.D. C18 reversed phase cartridge guard column (LiChrocart®3 E.M. Merck, Germany) for protection of the analytical column, a variable wavelength ultraviolet (UV) detector (Spectra 100, Spectra-Physics Inc. San Jose, CA, USA) and an integrator (Integrator-Hewlett-Packard HP3395 Integrator, Palo Alto, CA, USA) for a hard copy record of the output from the detector. The system was operated at room temperature (23–25°C) with a flow rate of 0.5 mL/min and an operating pressure of 1.7 kpsi. The wavelength was set at 254 nm for detection and quantification. The mobile phase was a mixture of 0.0005 M tetra butyl ammonium hydrogen sulfate (TBAS) solution in a 0.1 M KH₂PO₄: acetonitrile: methanol (9.6:0.3:0.1, v/v/v) with final pH adjusted to 6.2 – 6.3. Fisher Accumet pH meter 900 (Fisher Scientific, Toronto, ON, Canada) was used to determine the pH of the mobile phase and other solutions. The pH meter was standardized prior to each use by a standard buffer solution (pH 4, Colourkey™, BDH Inc., Dartmouth, NS, Canada).

3.1.2.2 Solid Phase Extraction Equipment

The Solid phase extraction (SPE) was performed using CN (cyanopropylsilica) bonded extraction column (100 mg/mL) purchased commercially (Chromosep®, Chromatographic Specialties Inc., Brockville, ON, Canada). The extraction clean up procedure was carried out on a 24-column position vacuum manifold fitted with a

stopcock at each position (Adsorbex™ SPU, E. Meerck, Darmstadt, Germany). Other equipment utilized included: a multi-tube mixer (IKA-VIBRAX-VX2®, Janke & Kunkel GMBH & Co., IKA-Labortechnik, Staufen, Germany), a refrigerated centrifuge (Model TJ-6R, Beckman Instruments San Ramon, CA, USA), and a Thermolyne Dri-bath (Fisher Scientific Co., ON, Canada).

3.1.3 Preparation Of Stock Solutions, Spiking Solutions And Internal Standard Solutions

3.1.3.1 Preparation Of Stock Solution

For the preparation of a standard stock solution of adenosine monophosphate (AMP) (4 mg/mL), 20 mg of AMP was weighed and dissolved in 5 mL of distilled water. Similarly, 100 mg of adenosine diphosphate (ADP) and adenosine triphosphate (ATP) was weighed and dissolved in 5 mL of distilled water to obtain standard stock solutions of ADP and ATP (20 mg/mL each), respectively. To prepare standard stock solutions of guanosine monophosphate (GMP), guanosine diphosphate (GDP) and guanosine triphosphate (GTP) (4 mg/mL each), authentic samples (20 mg each) were weighed out and dissolved in 5 mL of distilled water. A standard stock solution of 3,7-dimethyluric Acid (DMUA) (1mg/1mL) was prepared by dissolving 10mg of DMUA in 10 mL of 0.1% sodium hydroxide (NaOH).

3.1.3.2 Preparation Of Stopping Solution

The stopping solution contained a mixture of 6 uM erythro-9 (2-hydroxy-3-nonyl) adenine (EHNA), 100 uM of dipyridamole, 4 mM of ethylenediaminetetraacetic acid

disodium salt (EDTA), 2 ug/mL indomethacin, and 10 IU heparin/mL. The stopping solution was prepared as follow: EDTA disodium salt (148.9 mg) was dissolved in 50 mL of 0.9% sodium chloride solution and transferred to 250 mL beaker. Erythro-9-(2-hydroxyl-3-nonyl)adenine (EHNA) (10mg) was dissolved in a 10 mL of distilled water and an aliquot (0.86 mL) of this solution was then transferred to the beaker. A 5 mL volume of dipyridamole in methanol solution (10 mg/10 mL), a 0.2 mL of indomethacin methanol solution (10/10 mL) and a 1 mL volume of 1000 IU units/mL heparin were added to the beaker respectively. The pH was adjusted to 7.4 with 1% sodium hydroxide solution. This mixture of solution was transferred to a 100 mL volumetric flask and the volume was made up to 100 mL with 0.9% sodium chloride solution. The stopping solution was stored in a refrigerator at 4°C.

3.1.3.3 Preparation Of Spiking Solution

Spiking Solution 1:

It was prepared by combining standard stock solutions of 1ml of each of ATP, ADP (20 mg/ml) and 1ml of each of AMP, GTP, GDP, GMP (4 mg/ml) and making up the volume to 10 ml with phosphate buffer saline (PBS) to make a final concentration of 2000 ug/ml of ATP, ADP, and 400 ug/ml of AMP, GTP, GDP and GMP.

Spiking Solution 2:

Spiking solution 1 (1 ml) was diluted with 8mL of PBS to make a solution with a final concentration of 250 ug/mL of ATP, ADP and 50 ug/mL of AMP,

GTP, GDP, and GMP.

Spiking Solution 3:

Spiking solution 2 (4 mL) was diluted to 10 mL with PBS to make a solution with a final concentration of 100 ug/mL of ATP, ADP and 20 ug/mL of AMP, GTP, GDP, and GMP.

The spiking solutions were kept at -20oC and stable for more than 6 months.

3.1.3.4 Preparation Of Calibrating Solution For HPLC Stability Testing

A stock solution containing 1mg/ mL (1/1) of each ATP, ADP, AMP, GTP, GDP and GMP was prepared separately in HPLC water. Each of the stock nucleotide solution (0.1 mL each) was mixed with 0.1mL of a stock solution of 3,7- DMUA standard solution (in 0.1 % sodium hydroxide prepared as described in 3.1.3.1) and the volume was made up to 10 mL with HPLC water to prepare 0.01 mg/mL (1/100) solution mixture. This solution mixture was diluted with HPLC water to prepare a 0.001 mg/mL (1/1000) of calibrating solution. The calibrating solution was stored at 4°C and stable up to 3-4 months.

3.1.3.5 Preparation Of Working Internal Standard Solution

The stock internal standard solution of 1 mg/ml of 3,7 DMUA (1/1) was diluted with HPLC water to prepare the working internal standard solution of 0.05 mg/ml of 3,7 DMUA (1/20) on the day of the extraction. This solution was stable up to two weeks after it was prepared.

3.1.4 Solid Phase Extraction And HPLC Procedure

The solid phase extraction procedure was adopted for clean up of the RBC lysate samples and has been previously reported [69]. The RBC lysate samples collected from the study and the quality control (QC) samples were thawed at room temperature on the day of extraction. The QC samples were analyzed in duplicates at high spiked concentration (250 ug/mL), low spiked concentration (100 ug/ mL) and blank lysate. They were prepared by adding 0.1 mL of spiking solution 2, spiking solution 3 or PBS respectively to 0.1 mL of QC blank lysate in conical glass culture tubes. For each 0.1 mL of study lysate sample, 0.1 mL of PBS was added to make up the volume. Stopping solution (50 uL) and 100 uL of freshly prepared working internal standard (DMUA) were added into all the lysate samples. To each culture tube 1 mL of methyl tert-butyl ether (MTBE) was added and the contents were shaken on a multi-tube vortex mixer for 5 minutes to remove trichloroacetic acid (TCA) in the lysate samples. The contents of the tube were centrifuged at 4°C and 3000 rpm for 10 minutes using the Beckman centrifuge Model TJ-6R. The upper organic layer of MTBE was carefully removed and a gentle stream of high purity nitrogen was applied to the culture tubes at 55°C in the Thermolyne Dri-bath for 5 minutes to remove any residual organic layer left in the tubes. The remaining aqueous layer of each sample was passed through a 100 mg CN-bonded extraction column pre-conditioned with methanol, water and then PBS (2 mL each) on top of a vacuum manifold under 5 - 10 inches Hg of vacuum. Each column was washed with 0.2 mL of 10% methanol in HPLC water and the extracted sample was collected in a round bottom glass culture tube and stored at -20°C until analysis. Each extracted samples

were thawed and aliquots immediately injected into the HPLC system for analysis.

3.2 HPLC Assay To Determine Plasma Concentration Of Adenosine And Its Purine Metabolites

3.2.1 Chemical And Reagents

Adenosine (ADO), inosine (INO), hypoxanthine (HYP), guanosine (GUA), xanthine (XAN), and uric acid (UA) were purchased from Sigma-Aldrich Chemical Co. (PO Box 14508 ST. Louis MO 63178, USA). Trichloroacetic acid was purchased from J.T. Baker Chemical Co. (Phillisburg, NJ, USA.). Conical and round bottom glass culture tube (Kimax Brand, 100 x 16 mm I.D.) with PTFE-lined screw caps and graduated glass pipette were procured from Fisher Scientific (Ottawa, ONT, Canada). Methanol, acetonitrile and other solvents were HPLC grade and all other chemicals were reagent grade purchased from, Fisher Scientific Ltd. (Ottawa, ONT, Canada).

3.2.2 Instrumentation

3.2.2.1 High Performance Liquid Chromatography System

The high performance liquid chromatography system used in this assay consisted of a Beckman 114 M solvent delivery module (Beckman Instruments Inc., Berkeley, CA, USA), a 250 mm × 4.6 mm I.D. C18 reversed-phase column bonded to a 5 um spherical silica packing (Ultramex column from Phenomenex, Torrance, CA, USA) connected to a C18 reversed phase cartridge guard column (EM Science, Cherry Hill, NJ, USA.) and a C18 4.0 x 2.0 mm Security guard column (Phenomenex, Torrance, CA, USA.), a Shimadzu SPD-6AV UV-VIS spectrophotometric detector from Shimadzu Scientific

Instruments Inc. (Man-Tech Assoc. Inc., Guelph, ON, Canada.) and an Hewlett-Packard HP3395 Integrator (Palo Alto, CA, USA) for a hard copy of the output from the detector. The system was operated at room temperature (23–25°C) with a flow rate of 1.0 mL/ min at an operating pressure of 1.5 kpsi. The wavelength of the detector was set at 254 nm and sensitivity at 0.01 absorbance units for full-scale deflection for detection and quantification. The mobile phase was a mixture of 95% of 0.01M-ammonium dihydrogen orthophosphate ($\text{NH}_4\text{H}_2\text{PO}_4$) and 5% of methanol with the final pH adjusted to 3.9 using the Fisher Accumet pH meter 900 (Fisher Scientific, Toronto, ON, Canada). To obtain reliable results, the pH meter was standardized prior to each use with a standard buffer solution (pH 4, Colourkey™, BDH Inc., Dartmouth, NS, Canada).

3.2.2.2 Solid Phase Extraction Equipment

The Solid phase extraction (SPE) was performed using cyanopropylsilica (CN) bonded extraction column (100 mg/mL) purchased commercially (Chromosep®, Chromatographic Specialties Inc., Brockville, ON, Canada). The extraction/clean up procedure was carried out on a 24-column position vacuum manifold fitted with a stopcock at each position (Adsorbex™ SPU, E. Merck, Darmstadt, Germany). Other equipment utilized included: a multi-tube mixer (IKA-VIBRAX-VX2®, 6 Janke & Kunkel GMBH & Co., IKA-Labortechnik, Staufen, Germany), a refrigerated centrifuge (Model TJ-6R, Beckman Instruments San Ramon, CA, USA), a Thermolyne Dri-bath (Fisher Scientific Co., ON, Canada), and an Eppendorf centrifuge (purchased from Brinkmann Instrument Westbury, NY, USA.).

3.2.3 Preparation Of Stock Solutions, Spiking Solutions And Internal Standard

Solutions

3.2.3.1 Preparation Of Stock Solution

For preparation of a standard stock solution of ADO, 10 mg of the authentic standard was weighed and dissolved in 10 mL of 0.01M hydrochloric acid (HCL) to make a 1mg/mL (1/1) solution. A standard stock of INO (1 mg/mL) was similarly prepared in HPLC water. Standard stock solutions of HYP, GUA and XAN were prepared by weighing and dissolving 10mg each separately in 10mL of 0.1% sodium hydroxide (NaOH) to make 1mg/mL (1/1) solutions. To prepare the UA standard stock solution 10 mg of UA was first suspended in 1 mL of 0.01M hydrochloric acid (HCL) and to this suspension 9 mL of 0.1% of NaOH was added to make a 1mg/mL UA solution. This dissolving method prevented precipitation of UA from the solution when stored at cold temperature. A standard stock solution of 3,7-dimethyluric Acid (DMUA) (1mg/1mL) was prepared by dissolving 10mg of DMUA in 10 mL of 0.1% sodium hydroxide (NaOH).

3.2.3.2 Preparation Of Stopping Solution

The preparation followed similar procedure as described in 3.1.3.2

3.2.3.3 Preparation Of Spiking Solution

Spiking Solution 1:

Pipette 0.125 mL of each of the stock solution of ADO, INO, GUA (1 mg/mL, 1/1 solution), and 1.25 mL of each of the stock solution of UA, HYP,

XAN (1 mg/mL, 1/1 solution) to a volumetric flask and qs to 25 mL with PBS to make a final concentration of 5 ug/mL of ADO, INO, GUA and 50 μ g/mL of UA, HYP, XAN, and pH of the solution mixture should be about 7. Transfer the spiking solution to a plastic bottle with proper label and store at -20°C .

Spiking Solution 2:

Spiking solution 1 (5 mL) was diluted to 25 mL with PBS to make a final concentration of 1ug/mL of ADO, INO, GUA and 10 ug/mL of UA, HYP, XAN and pH of the solution mixture should be about 7.0. Transfer the solution to a plastic bottle with proper label and store at -20°C .

3.2.3.4 Preparation Of Calibrating Solution For HPLC Stability Testing

A calibration solution was prepared by mixing 0.1 mL of each 1/1 solution of ADO, INO, HYP, GUA, XAN, UA, and DMUA with methanol to make a 0.01 mg/mL (1/100) solution mixture. Diluting the 1/100 solution mixture ten folds with methanol yielded a 0.001 mg/mL (1/1000) calibrating solution.

3.2.3.5 Preparation Of Working Internal Standard Solution

The preparation followed same procedure as mentioned in 3.1.3.5

3.2.4 Solid Phase Extraction And HPLC Procedure

This SPE and HPLC procedure was adopted from a standardized protocol which was published in detail previously [70]. Briefly, the plasma study samples with stopping solution stored in -80°C freezer and plasma quality control (QC) samples were thawed at

room temperature on the day of extraction. They were each (0.1 mL) added to a 2 mL size polyethylene micro-centrifuge tubes. For quality control of the study sample analysis duplicates of spiked high concentration (2.5 ug/mL), spiked low concentration (0.5 ug/mL) and blank plasma were prepared by adding 50 uL of spiking solution 1, spiking solution 2 and PBS respectively into the micro-centrifuge tube for QC samples, and followed by adding 50 uL of stopping solution to each of the QC samples. To each of the plasma study samples (0.1 mL each) was added PBS (0.1 mL) to make up the volume. Ice cold 10% trichloroacetic acid (TCA) (0.1 mL) was added to each sample to precipitate the plasma proteins and the contents were centrifuged for 5 minutes at 5000 rpm at room temperature using the Eppendorf centrifuge. The plasma supernatant was separated from the precipitate and transferred to a clean conical glass culture tube. To each culture tube 0.1 mL of the working internal standard solution and 1 mL of methyl tert-butyl ether (MTBE) were added and the contents were shaken on a multi-tube vortex mixer for 5 minutes. The contents of the tube were then centrifuged at 4°C and 3000 rpm for 10 minutes using the Beckman Model TJ-6R centrifuge. The upper organic layer of MTBE was carefully removed and a gentle stream of high purity nitrogen was applied to the culture tubes at 55°C in the Thermolyne Dri-bath for 5 minutes to remove any residual MTBE. The remaining aqueous layer of each sample was passed through a 100 mg CN-bonded extraction column pre-conditioned with methanol, water and then PBS (2 mL each) on top of a vacuum manifold under 5 – 10 inches Hg of vacuum. Each column was washed with 0.2 mL 10% methanol and the sample was collected in a round bottom glass culture tube and stored at -20°C until analysis. After thawing, aliquots of the extracted

samples were injected into the HPLC system for analysis.

3.3 HPLC Assay To Measure The Plasma Concentration Of Dipyridamole

3.3.1 Chemical, Reagents And Consumables

Dipyridamole and losartan (as maleate salt) were obtained as gifts from Boehringer-Mannheim Canada Ltd (Laval, QC, Canada), and Merck Co. (Rahway, NJ, USA), respectively. Potassium phosphate monobasic (KH_2PO_4) was purchased from Fisher Scientific Co. (Ottawa, Ont., Canada), and sodium azide and triethylamine (TEA) from Sigma-Aldrich Chemical Co., (St. Louis, MO 63178, USA). Round bottom and conical glass culture tubes (100 x 16 mm I.D.) with PTFE-lined screw caps (Kimax Brand) and graduated Glass Pipettes were procured from Fisher Scientific (Ottawa, ONT, Canada). Methanol, acetonitrile, methylene dichloride (MDC) and other solvents were HPLC grade and all other chemicals were reagent grade and purchased from, Fisher Scientific Ltd., (Ottawa, ONT, Canada).

3.3.2 Instrumentation

3.3.2.1 High Performance Liquid Chromatography System

The high performance liquid chromatography system used in the assay consisted of a Beckman 114 M solvent delivery module purchased from Beckman Instruments Inc. (Berkeley, CA, USA), a Rheodyne syringe loading injector (model 9725), with a 100 μL PEEK injection loop (Scientific Products & Equipment, Concord, ON, Canada), a 5 μm 250 mm \times 4.6 mm I.D. C18 reversed-phase column (Gemini, Phenomenex, Torrance, CA 90501, USA) with a 5 μm 4.0 x 3.0 mm I.D. C18 reversed phase cartridge

guard column (Security Guard Cartridges, Phenomenex, Torrance, CA 90501, USA), a Shimadzu SPD-VIS spectrophotometric detector (Man-Tech Assoc. Inc., Guelph, ON, Canada.) and an Hewlett-Packard HP3395 Integrator (Agilent Technologies, Santa Clara CA, USA) to obtain a hard copy of the output from the detector. The system was operated at room temperature (23–25°C) with a flow rate of 0.5mL/min and an operating pressure of 1.7-2.0 Kpsi. The wavelength of the detector was set at 280 nm and sensitivity at 0.01 absorbance units for a full-scale deflection for detection and quantification. The mobile phase consisted of a mixture of 0.01M potassium phosphate monobasic (KH₂PO₄) buffer, acetonitrile and methanol (50:25:25) containing 0.04% of TEA. The final pH of the mobile phase was adjusted to 3.2 with 80% phosphoric acid using the Fisher Accumet pH meter 900 (Fisher Scientific, Toronto, ON, Canada). To ensure accuracy, the pH meter was standardized prior to each use with a standard pH 4 buffer solution (Colourkey™, BDH Inc., Dartmouth, NS, Canada).

3.3.2.2 Solid Phase Extraction System

The solid phase extraction was performed using 100 mg/mL C₁₈ columns (Chromosep®, Chromatographic Specialties, Brockville, Ont., Canada) on top of a 10-column position SPE vacuum manifold (Vac-Elute, Varian, Harbour City, CA) with a stopcock at each position.

3.3.3 Preparation Of Stock Solutions, Spiking Solutions And Internal Standard Solutions

3.3.3.1 Preparation Of Stock Solution

To prepare a 1.0mg/mL stock solution of dipyridamole and losartan, 10 mg of dipyridamole and losartan were each weighed and dissolved in 10 mL methanol separately. The stock solution was diluted 10 times with methanol to prepare a 0.1 mg/mL (1/10) standard solution of dipyridamole and losartan, respectively. These solutions were stored at -20°C.

3.3.3.2 Preparation Of Standard Spiking Solution

Standard dipyridamole solution (1/10) was diluted with 0.01 M potassium phosphate buffer (pH 6.5) to prepare a 1 ug/mL (1/1000) standard spiking solution 1. The solution 1 was further diluted ten times with 0.01 M potassium phosphate buffer (pH 6.5) to prepare 0.1 ug/mL standard spiking solution 2. These spiking solutions were used within one week after they were prepared.

3.3.3.3 Preparation Of Calibrating Solution For HPLC Stability Testing

The calibrating solution was prepared by mixing 0.1 mL of the 0.1 mg/mL (1/10) standard solution of dipyridamole and 0.3 mL of the 0.1 mg/mL (1/10) solution of losartan with 9.6 mL of methanol. The final concentration of the calibrating solution was 1 ug/mL of dipyridamole and 3 ug/mL of losartan, respectively.

3.3.3.4 Preparation Of Working Internal Standard Solution

Standard 0.1 mg/mL losartan solution (1/10) was diluted 20 folds with 0.01M

potassium phosphate buffer (pH6.5) to prepare a 1/200 working internal standard solution (5 ug/mL).

3.3.4 Solid Phase Extraction And HPLC Procedure

The HPLC assay procedure was adopted from a similar protocol for losartan which was previously published [71]. The round bottom glass culture tubes used in the extraction were first pre-rinsed with 1mL each of methylene dichloride (MDC) and then with methanol. The plasma study samples and the quality control (QC) samples were thawed at room temperature on the day of extraction. The C₁₈ columns were set up on the SPE vacuum manifold and preconditioned with methanol, water and then potassium buffer (pH 6.5) under a vacuum pressure of 5 inch of Hg. To each preconditioned SPE columns 50µL each of QC plasma and study samples was added. For quality control of the analysis duplicates of high spiked concentration (1 ug/mL), low spiked concentration (0.1 ug/mL) and blank plasma were prepared by adding 50 uL each of standard spiking solution 1 (1/1000), standard spiking solution 2 (1/10000) and 0.01M Phosphate buffer (pH 6.5) respectively into the columns with QC plasma samples. A further 100 uL of the phosphate buffer and 50 uL of working internal standard solution were added to each of the columns. The mixture of solutions on top of the column was allowed to equilibrate for 5 minutes. A vacuum of 10 inches Hg was applied in the SPE vacuum manifold and the stopcocks were slowly opened to collect the filtrate in the waste collecting glass culture tubes. The columns were each washed with 1mL of 10% methanol and the filtrate was collected in the same waste collecting glass culture tubes. The pump was turned off and the waste collecting glass culture tubes were replaced with the pre-rinsed round bottom

glass culture tubes. The columns were eluted with a mixture of acetonitrile and methanol (1:9) under a vacuum pressure of 5 inches Hg. The filtrate was collected and evaporated to dryness using a gentle stream of nitrogen at 55°C in thermolyne dri-bath. The dried sample extracts were reconstituted with 200 uL of filtered mobile phase and aliquots of the reconstituted samples were injected into the HPLC system for further analysis.

3.4 Animal Study

The animal experiment study protocol was approved by Dalhousie University Committee on Laboratory Animals (UCLA). Mr. Patrick Tillman who was a former student in Dr. Pollen Yeung's lab performed the animal experiments following a standardized protocol to induce acute myocardial infarction using isoproterenol which have been previously published [6]. Briefly, male Sprague-Dawley rats with carotid artery catheter weighing 250 to 320g were purchased for this study (Charles River laboratories, Wilmington, MA, USA). They were housed for a 48-hour acclimatization period with access to food and water at the Carleton Animal Care Center before the experiment. The treatment group (n = 8) was administered with five doses of dipyridamole (10 mg/kg) in polyethylene glycol 400 (PEG): normal saline (8:2) twice daily by (sc) injection on the dorsal area of the rat. The control group (n = 10) received normal saline (1ml/kg) injection subcutaneously (sc) also twice daily for five doses. During the experiment each rat was kept in a freely moving caging environment with free access to drinking water. Each rat in the treatment and control groups received isoproterenol hydrochloride (30mg/kg) in normal saline (30 mg/kg) subcutaneously one hour after the last injection as described previously [6, 72]. Blood samples (0.5mL each)

were collected through the catheter starting from 0 hr or T0 (before the last dose), and then at 0.05, 0.25, 1, 1.2, 1.5, 2, 3, 4, 5 and 6 hours after the last dose. The blood samples collected (0.3mL) from each rat were instantly mixed with a “Stopping Solution” which is a mixture of 26 uM EHNA, 100 mM dipyridamole, 4 mM EDTA, and a final concentration of 2 ug/mL of indomethacin in heparinized normal saline with pH adjusted to 7.4 to minimize in vitro degradation and production of adenosine during sample processing [69]. The other blood sample collected (0.2 mL) was mixed with EDTA solution for measurement of dipyridamole concentrations. The blood samples collected were immediately centrifuged (within 30 min) to separate plasma and RBC at 4°C using the Beckman TJ-6R centrifuge. The RBC samples collected were processed and lysed immediately using an ice-cold 10% trichloroacetic acid (TCA). The lysate and plasma samples were stored at -80 °C until analysis by HPLC as described earlier in the thesis (Sections 3.1, 3.2 and 3.3). The rats survived till the end of the experiment (5 hours after isoproterenol) were considered survivors from the isoproterenol injection and euthanized by cardiac puncture under general anesthesia with isoflurane.

3.5 Data Analysis

Areas under the curve (AUC's) of RBC concentrations of ATP and other purine nucleotides, and plasma concentrations of adenosine and its purine metabolites from T0 to the last blood sample time were calculated using trapezoidal method (Prism®-6, Graphpad Software Inc., La Jolla, USA). Pharmacokinetics variables including AUC, maximum plasma concentrations (C_{max}), time to maximum plasma concentrations (T_{max}), plasma half-life (T_{1/2}) and system clearance (CL) were determined using

compartmental modeling with a first order input to simulate subcutaneous injection (Rstrips, Micromath, Saint Louis, MO, USA). Data between the control and dipyridamole treated groups were analyzed by student's t-test and the differences were considered significant when $p < 0.05$ (Minitab® Inc. Release 17 State College, PA, USA).

CHAPTER: 4 RESULTS

4.1 Mortality

In control group (n=10) treated with normal saline (1 mL/kg), a sc injection of isoproterenol (30 mg/kg) induced 50 % mortality within 5 hours after the injection ($p < 0.05$) [6]. On the other hand the mortality was reduced to 25 % (2 out of 8 died) in the treatment group (n=8), which received dipyridamole (10mg/kg) twice a day for 5 doses by sc injection (Table 1). However due to small the sample size in each group the difference did not reach statistical significance.

Table 1: Mortality of rats induced by isoproterenol in normal saline control and dipyridamole treated groups.

Animal experiment		
Rats	Normal saline control (1 mL/kg)	Dipyridamole (10mg/kg)
Total	10	8
Died	5	2
Mortality	50%	25%

4.2 Concentrations Of ATP, Adenosine, Dipyridamole And Their Metabolites

4.2.1 RBC Concentrations Of Purine Nucleotides

Under the described experimental condition, in normal saline control group there was an apparent decline in RBC ATP concentration shortly after isoproterenol injection, but it was not observed in the dipyridamole treated rats (Figure 3). The area under the curve (AUC) of RBC ATP after isoproterenol injection was also higher in treatment group than in the normal saline control group (8.49 ± 4.35 vs. 7.34 ± 3.05 mM*T), however the difference was not statistically significant (Figure 4).

Figure 3: Effect of dipyridamole (10 mg/kg) on RBC ATP concentration vs. normal saline control. (Data presented in mean \pm SEM).

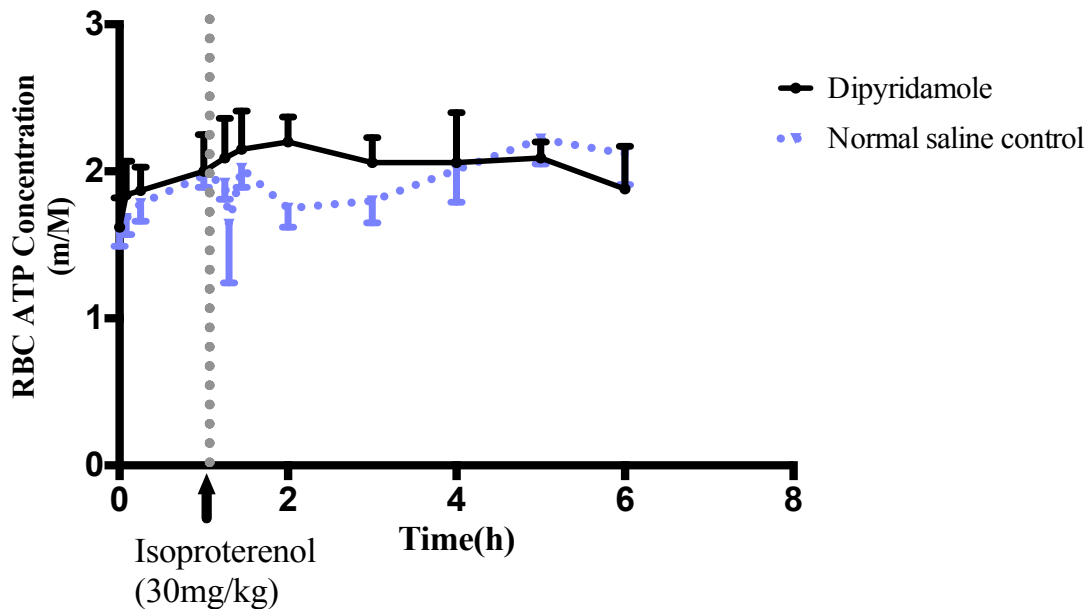
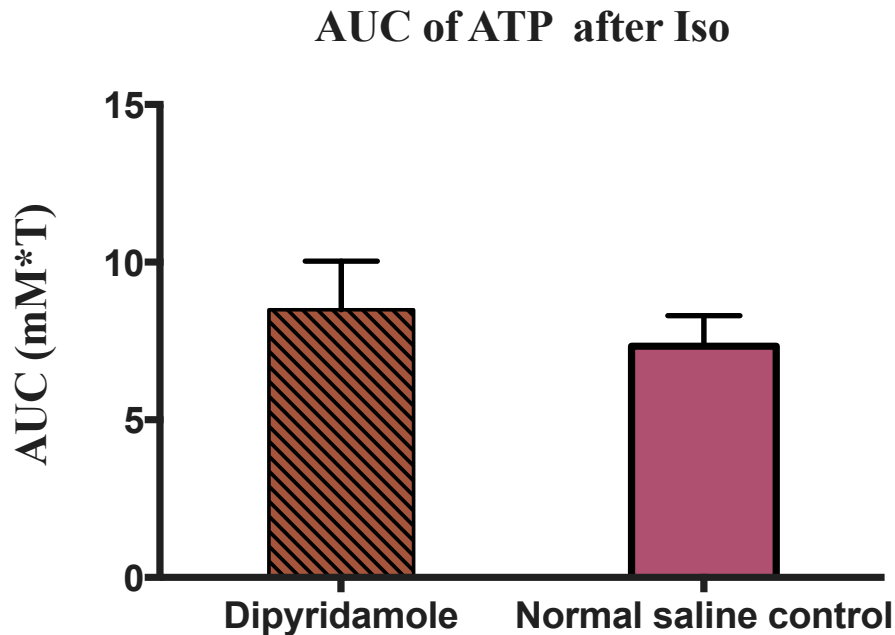


Figure 4: AUC of ATP in RBC after isoproterenol injection.



The RBC concentrations of ADP were higher in the control group throughout the experiment (Figure 5). There was an increase of ADP concentrations shortly after the isoproterenol injection in the control group, but not in the dipyrindamole treated group (Figure 5). In the dipyrindamole treatment group the AUC of ADP in the RBC before isoproterenol was significantly lower than in normal saline control group (0.32 ± 0.11 vs. 0.46 ± 0.10 mM*T, $p < 0.05$) (Figure 6). Similarly, AUC and maximum concentration (C_{max}) of ADP in RBC after isoproterenol injection in dipyrindamole treated rats also were significantly lower compared to control (AUC 1.19 ± 0.74 vs. 2.21 ± 0.75 mM*T; C_{max} 0.40 ± 0.13 vs. 0.81 ± 0.29 mM; $p < 0.05$ for both) (Figures 7 & 8).

Figure 5: Effect of dipyridamole (10 mg/kg) on RBC ADP concentration vs. normal saline control. (Data presented in mean \pm SEM).

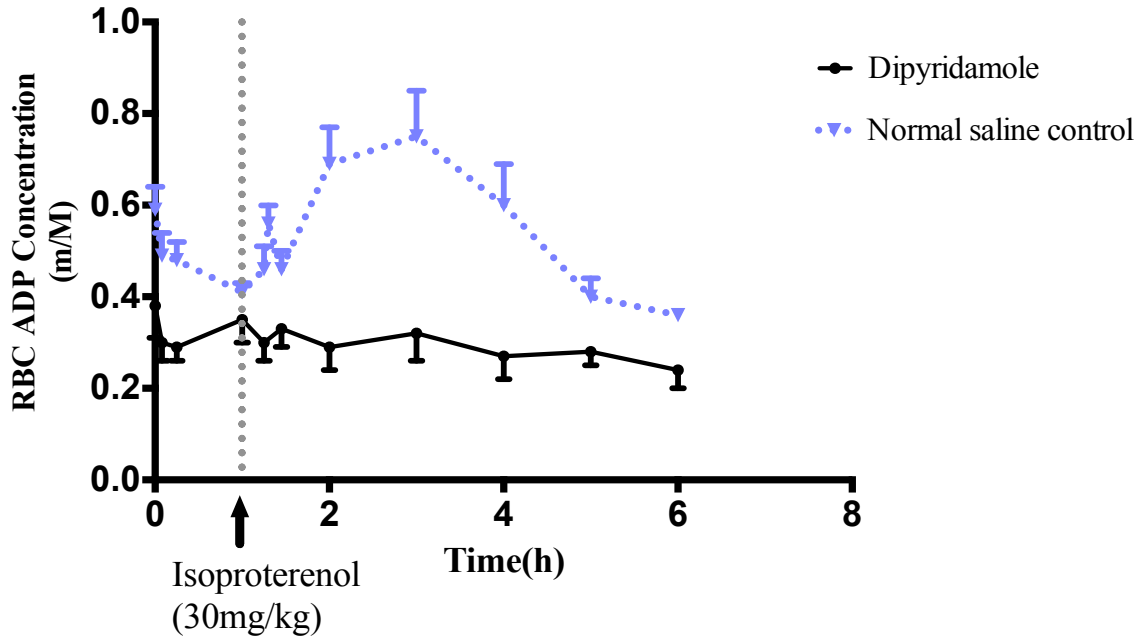


Figure 6: AUC of ADP in RBC before isoproterenol injection.

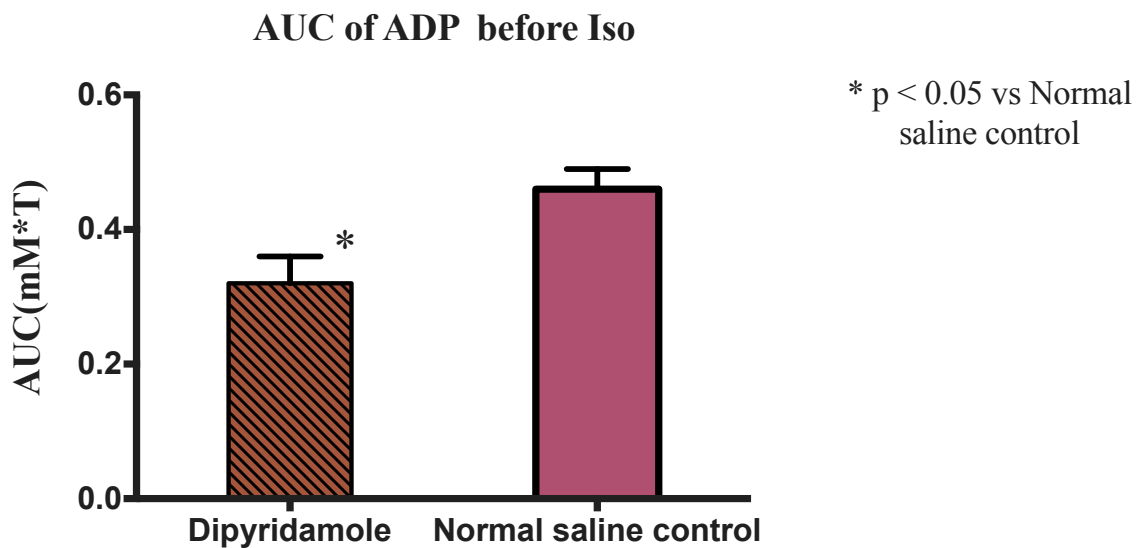


Figure 7: AUC of ADP in RBC after isoproterenol injection.

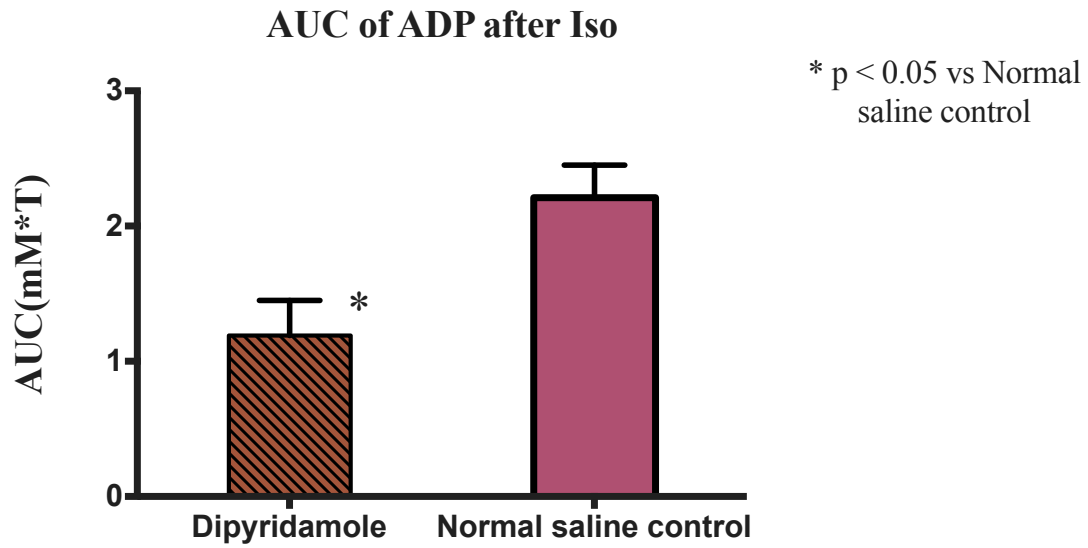
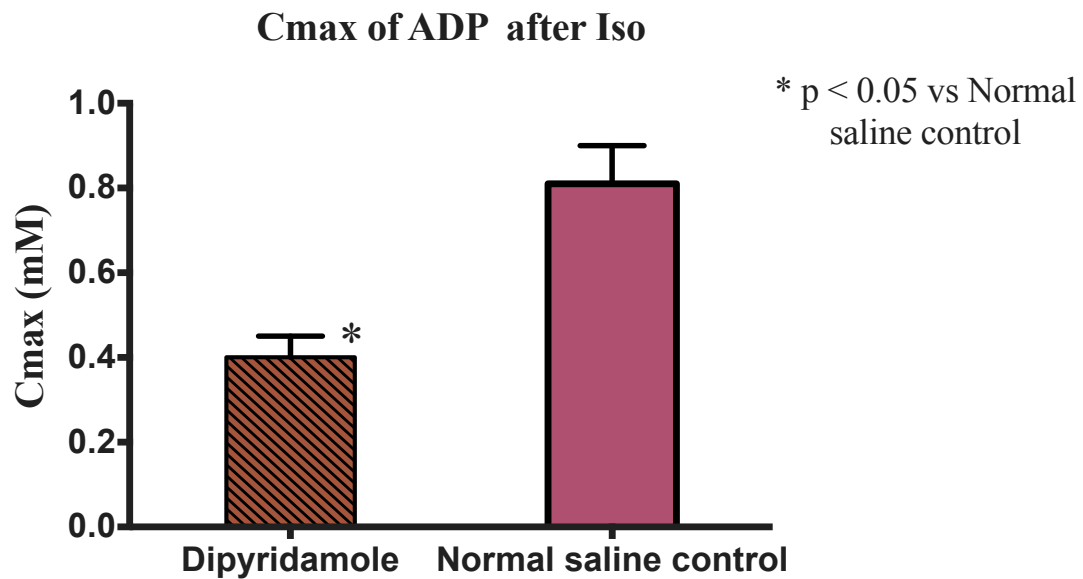


Figure 8: Cmax of ADP in RBC after isoproterenol injection.



Similarly, the RBC concentrations of AMP were also higher in the normal saline control group (Figure 9). Shortly after isoproterenol injection there was increase in AMP concentrations in control group but it was not observed in the dipyridamole treated group (Figure 9). The AUC of AMP in RBC before isoproterenol injection was significantly higher in the control than in dipyridamole treated group (0.05 ± 0.01 vs. 0.03 ± 0.02 mM*T, $p < 0.05$) (Figure 10). The AUC and Cmax of AMP in RBC after isoproterenol were also found to be significantly lower in the dipyridamole treated group (AUC 0.12 ± 0.10 vs. 0.52 ± 0.39 mM*T; Cmax 0.08 ± 0.09 vs. 0.27 ± 0.22 mM; $p < 0.05$ for both) (Figures 11 & 12).

Figure 9: Effect of dipyridamole (10 mg/kg) on RBC AMP concentration vs. normal saline control. (Data presented in mean \pm SEM).

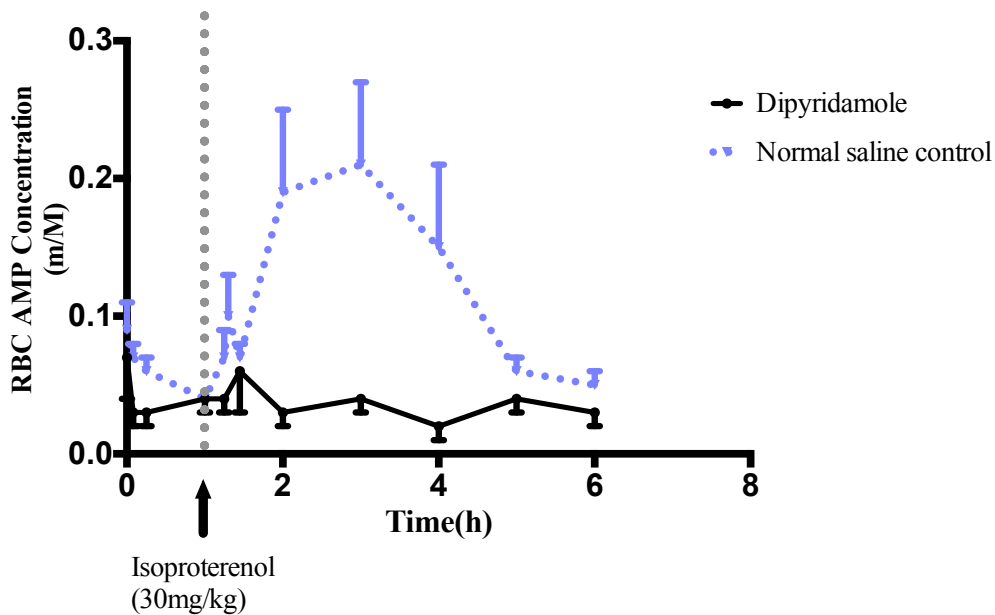


Figure 10: AUC of AMP in RBC before isoproterenol injection.

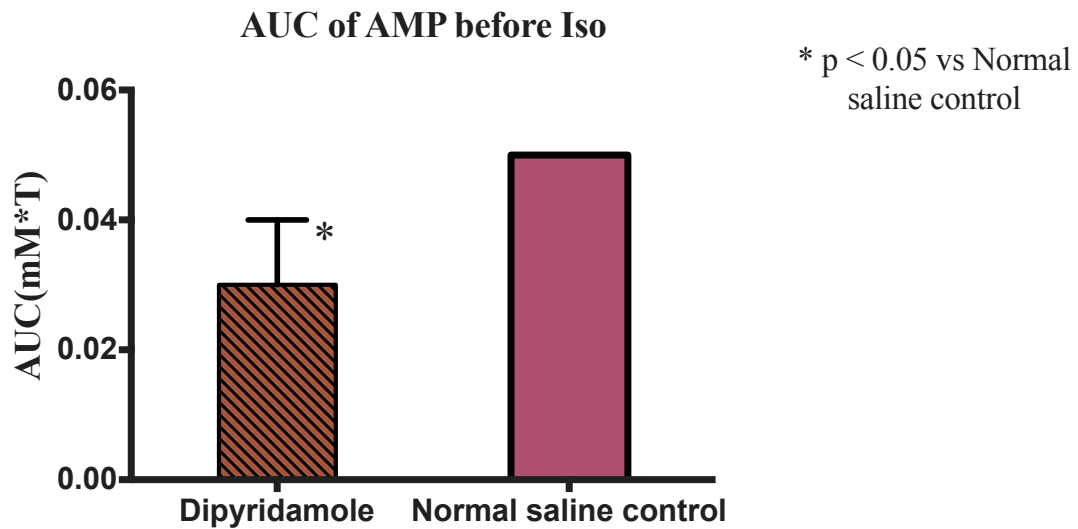


Figure 11: AUC of AMP in RBC after isoproterenol injection.

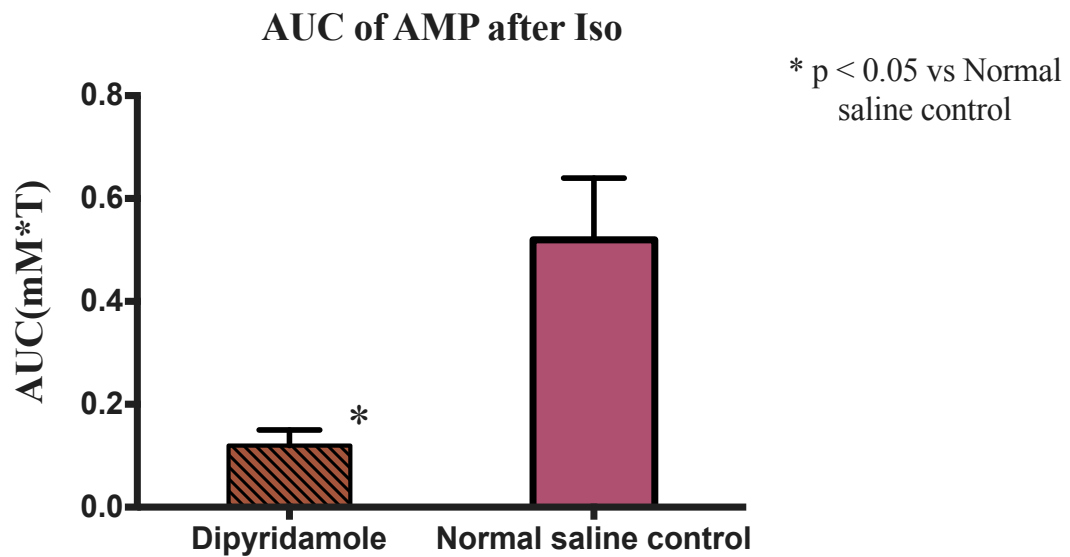
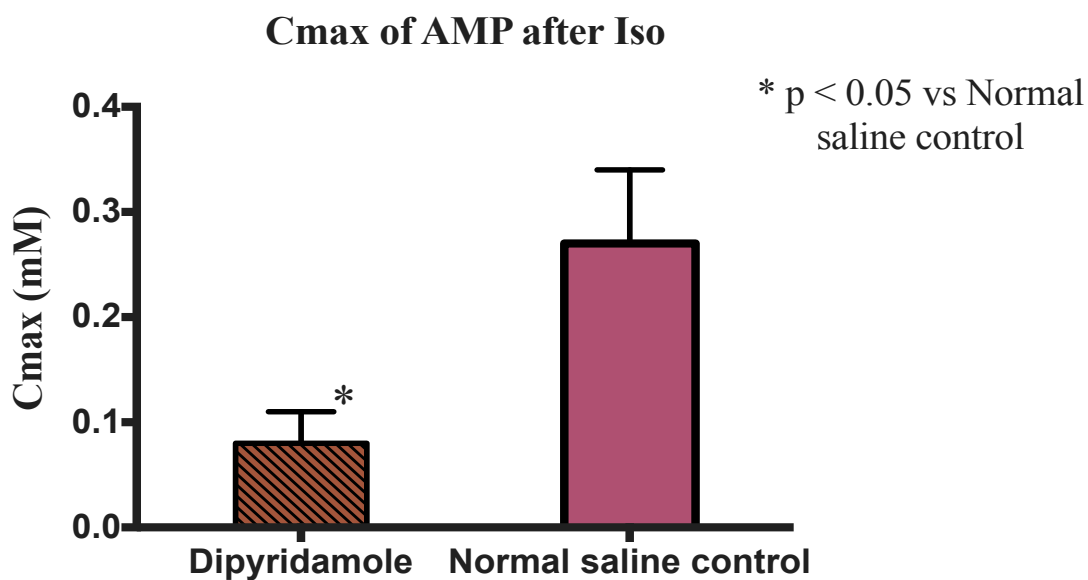


Figure 12: Cmax of AMP in RBC after isoproterenol injection.



The RBC concentrations of GTP were higher in the dipyrindamole treated group (Figure 13). In the control group the GTP concentration increased gradually shortly throughout the experiment (Figure 13). The AUC of GTP in the RBC before the isoproterenol injection was significantly higher in the dipyrindamole treatment group (AUC 0.22 ± 0.04 vs. control 0.14 ± 0.03 mM*T, $p < 0.05$) (Figure 14). The Cmax of GTP in RBC after isoproterenol injection was also significantly higher in the dipyrindamole treated group (0.31 ± 0.06 vs. control 0.25 ± 0.04 mM, $p < 0.05$) (Figure 15). The AUC of GTP in the RBC after isoproterenol was also higher in the dipyrindamole treatment group but the difference was not statistically significant (AUC 0.22 ± 0.04 vs. control 0.14 ± 0.03 mM*T) and (0.31 ± 0.06 vs. control 0.25 ± 0.04 mM) (Figure 14&15) (Figure 16)

Figure 13: Effect of dipyridamole (10 mg/kg) on RBC GTP concentration vs. normal saline control. (Data presented in mean \pm SEM).

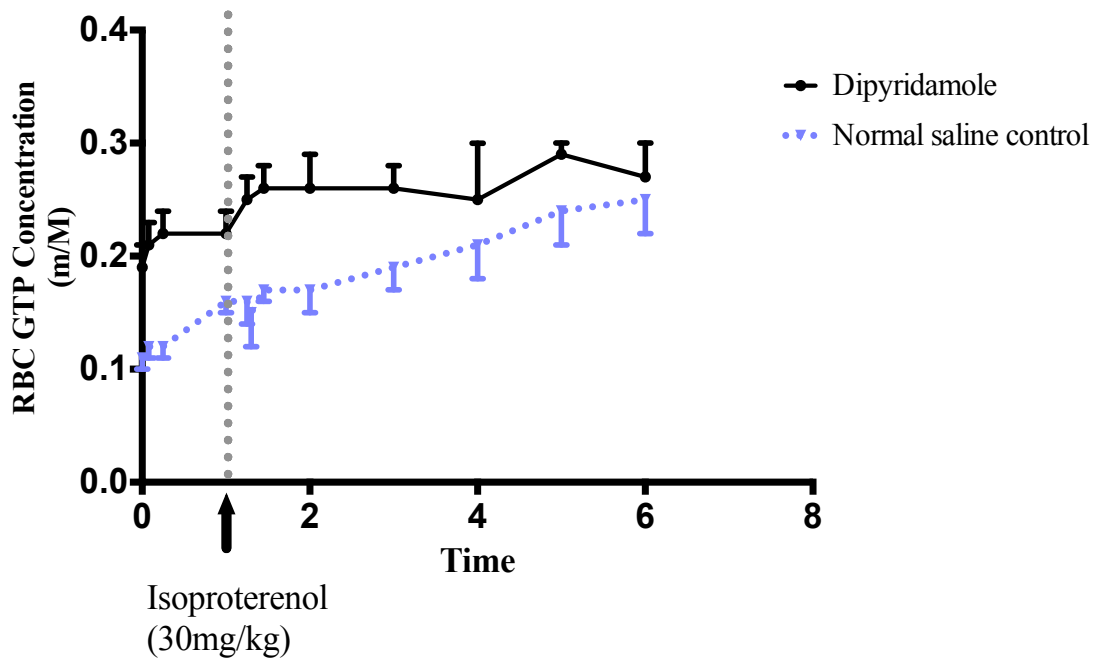


Figure 14: AUC of GTP in RBC before isoproterenol injection.

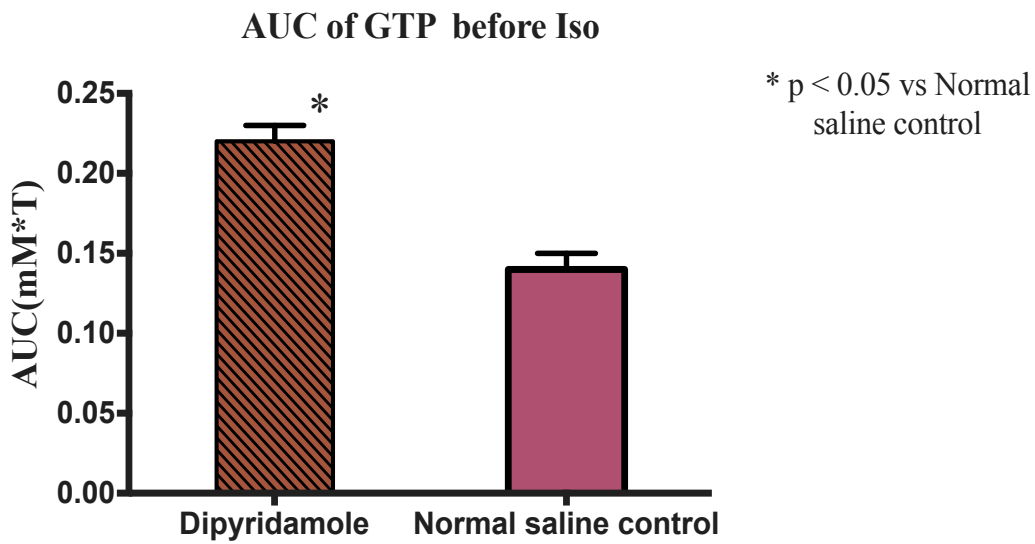


Figure 15: Cmax of GTP in RBC after isoproterenol injection.

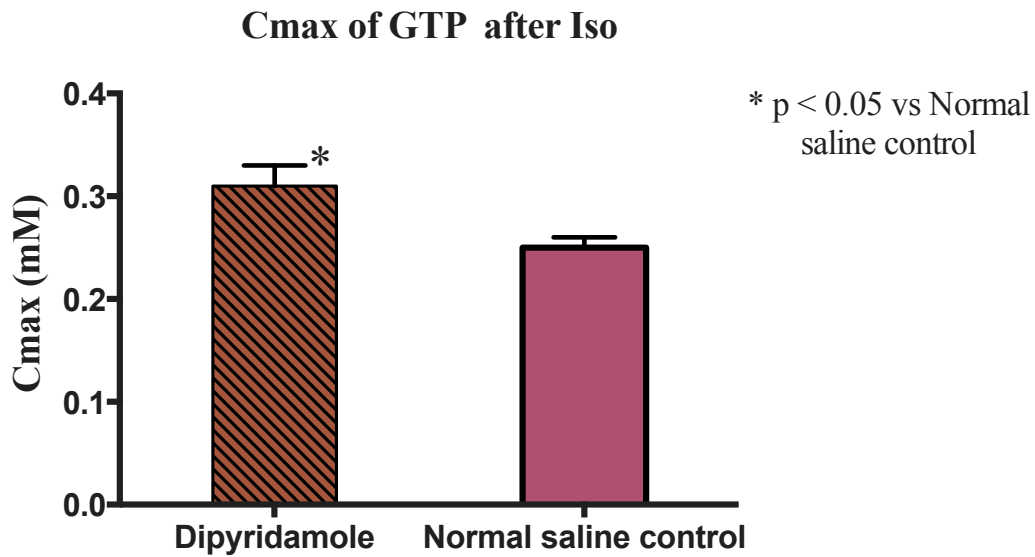
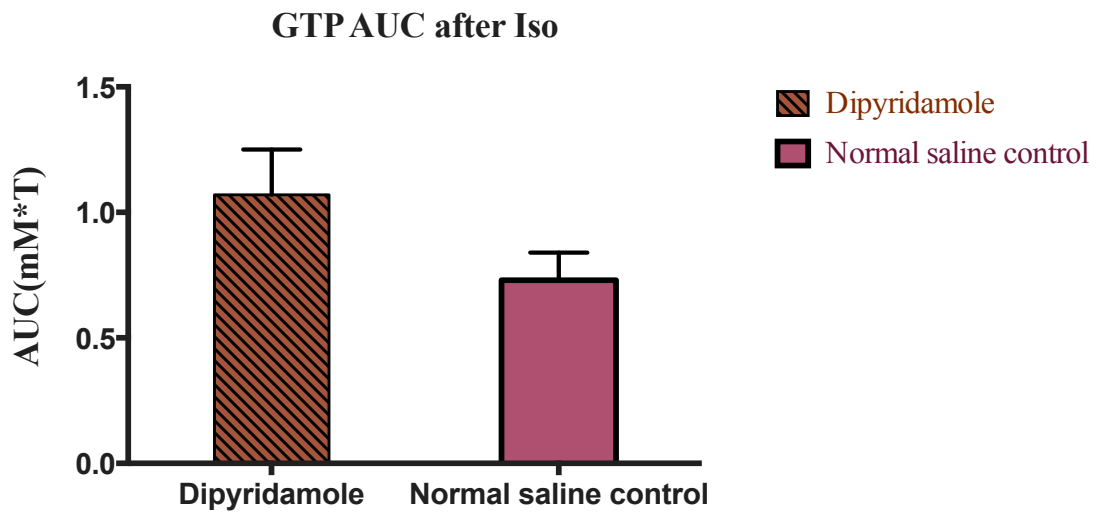


Figure 16: AUC of GTP in RBC after isoproterenol injection.



The RBC GDP concentrations in normal saline control group increased

shortly after isoproterenol injection, whereas its concentrations remained stable in treatment group (Figure 17). The Cmax of GDP after isoproterenol was also higher in control than in the treatment group (0.09 ± 0.04 vs. 0.07 ± 0.02). However the difference was not statistically significant (Figure 18).

Figure 17: Effect of dipyridamole on (10 mg/kg) on RBC GDP concentrations vs. normal saline control. (Data presented in mean \pm SEM)

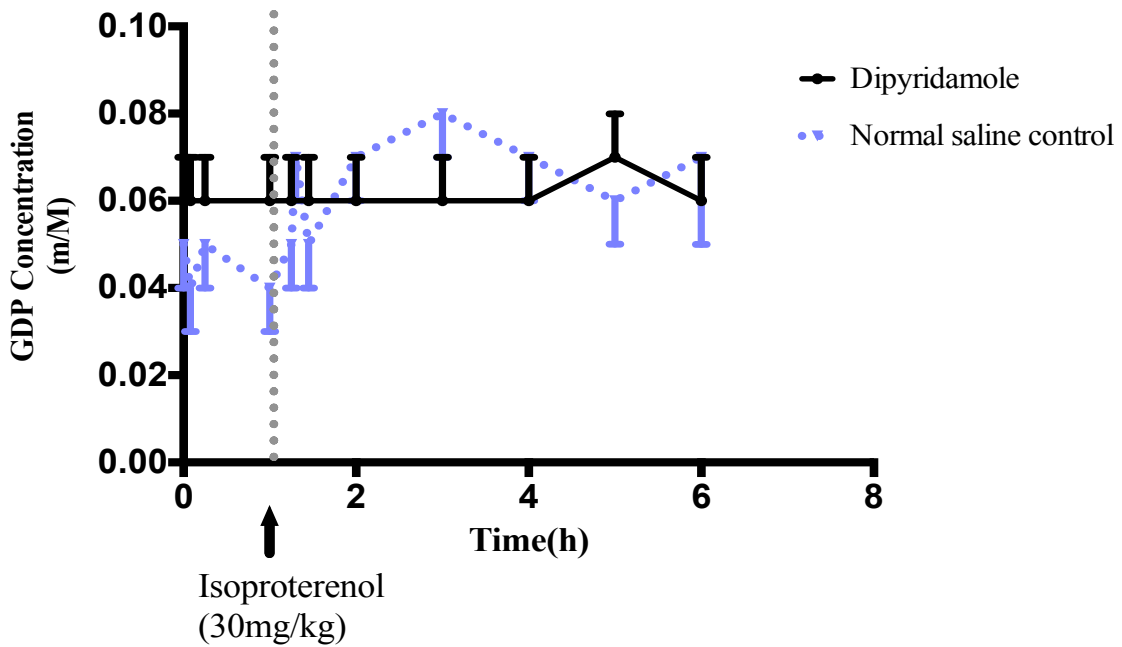
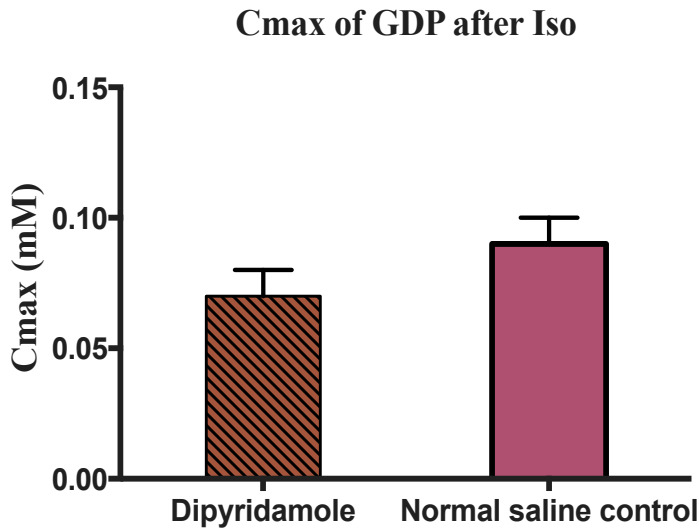


Figure 18: Cmax of GDP in RBC after isoproterenol injection.



4.2.2 Plasma Concentrations of Adenosine and its Purine Metabolites

In normal saline control group, plasma adenosine concentrations were higher and there was a sudden drop in its concentration immediately after isoproterenol injection, which rebounded shortly after (Figure 19). The decline was not observed in the dipyridamole treated group (Figure 19). The AUC of plasma adenosine concentration was higher in the control group both before and after isoproterenol injection, however the difference was statistically significant only for AUC after the isoproterenol injection (AUC after Iso 8.46 ± 4.68 in control vs. 4.73 ± 2.54 in dipyridamole treated group, $p < 0.05$) (Figures 20 & 21). The Cmax of adenosine after isoproterenol was also significantly higher in the control group (4.42 ± 1.88 vs 1.96 ± 0.38 , $p < 0.05$) (Figure 22).

Figure 19: Effect of dipyridamole (10mg/kg) on plasma adenosine concentrations vs. normal saline control. (Data presented in mean \pm SEM).

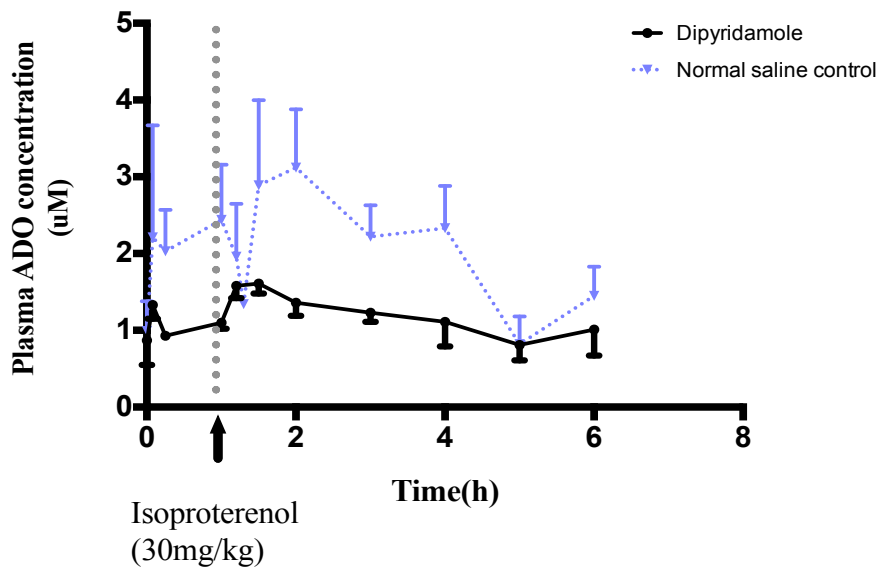


Figure 20: AUC of adenosine in plasma before isoproterenol injection.

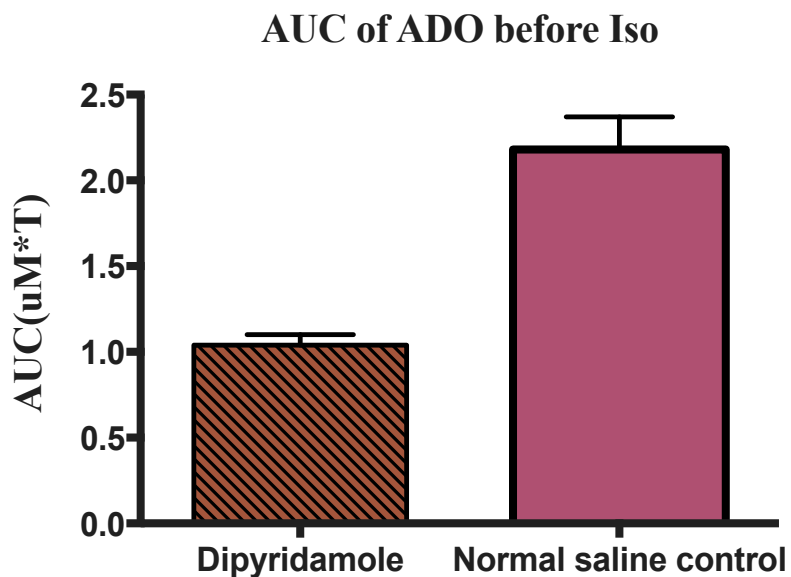


Figure 21: AUC of adenosine in plasma after isoproterenol injection.

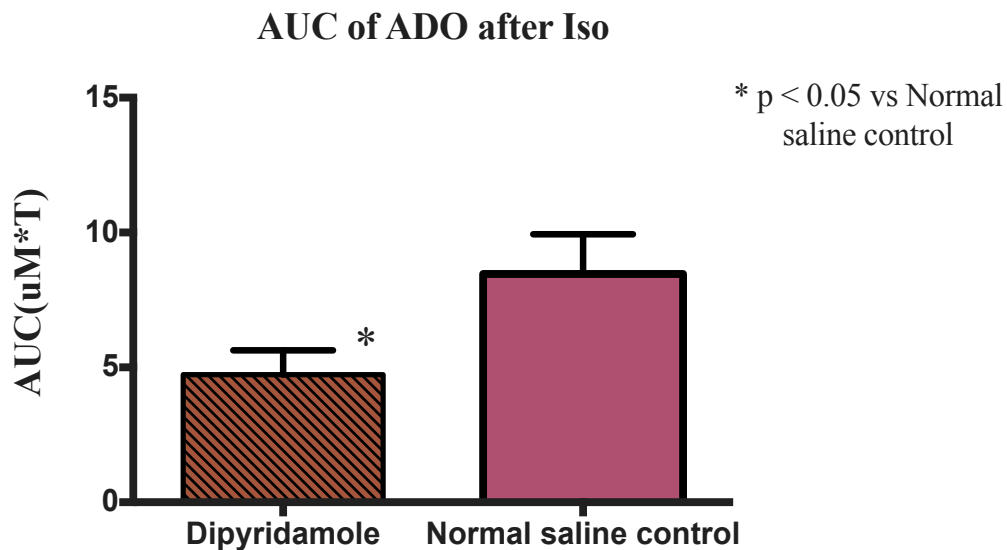
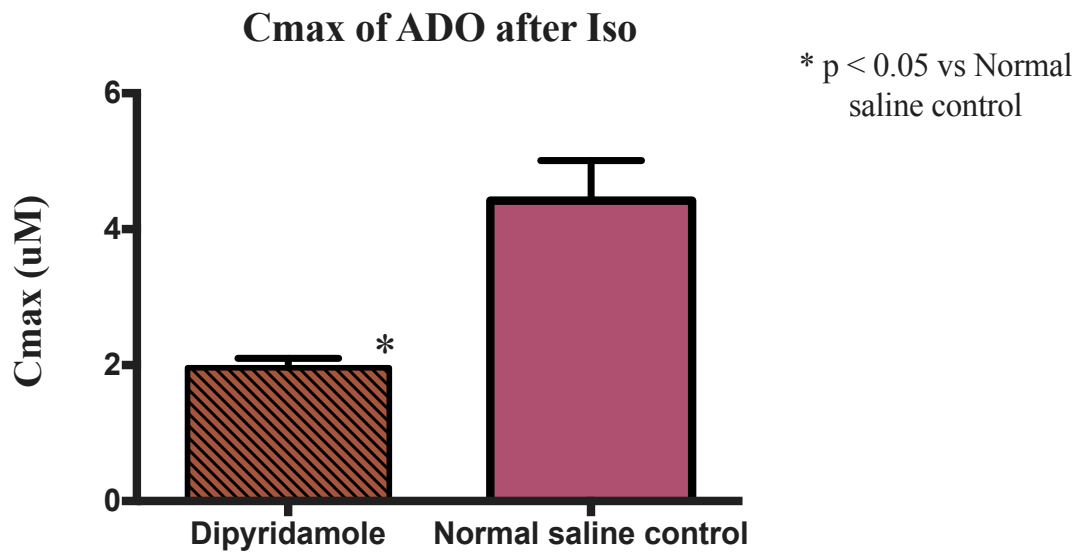


Figure 22: Cmax of adenosine in plasma after isoproterenol injection.



In normal saline control group plasma inosine was not detected, on the other hand it was measurable in the dipyridamole treated group and appeared to increase after the isoproterenol injection (Figure 23). The AUC of plasma inosine concentrations both before and after isoproterenol were significantly higher in treatment group (AUC before 1.49 ± 0.20 vs. non-detectable in the control; AUC after Iso 7.34 ± 4.44 vs. non-detectable in the control; $p < 0.05$ for both) (Figure 24 & 25).

Figure 23: Effect of dipyridamole (10mg/kg) on plasma inosine concentrations vs. normal saline control. (Data presented in mean \pm SEM).

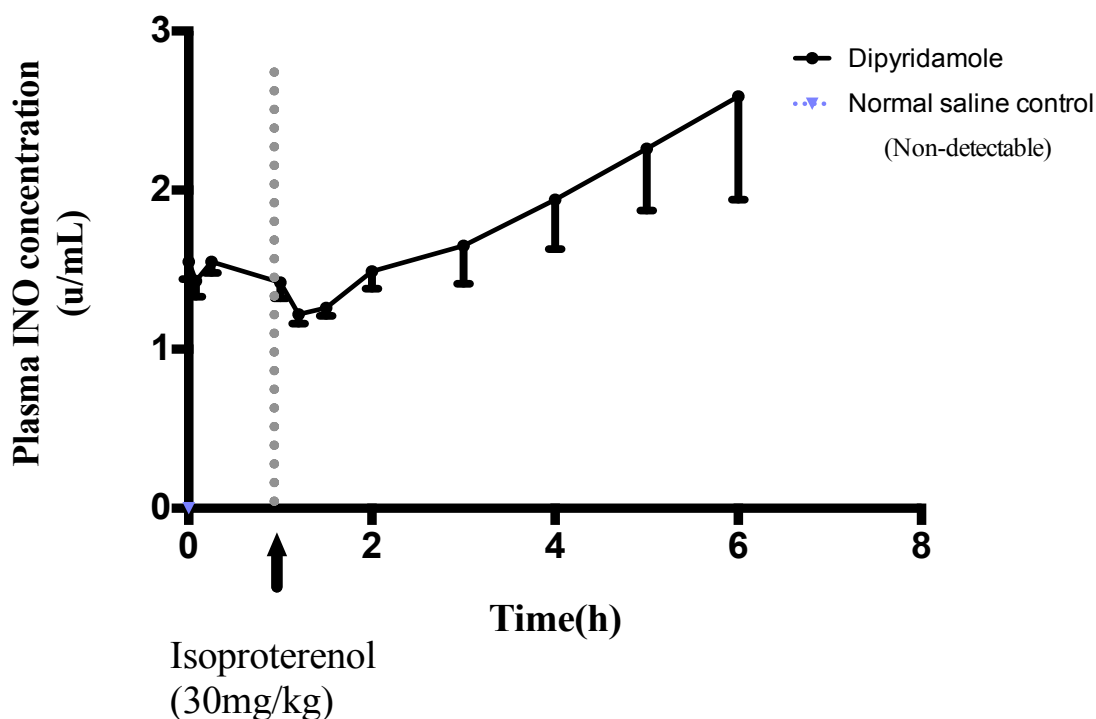


Figure 24: AUC of inosine in plasma before isoproterenol injection.

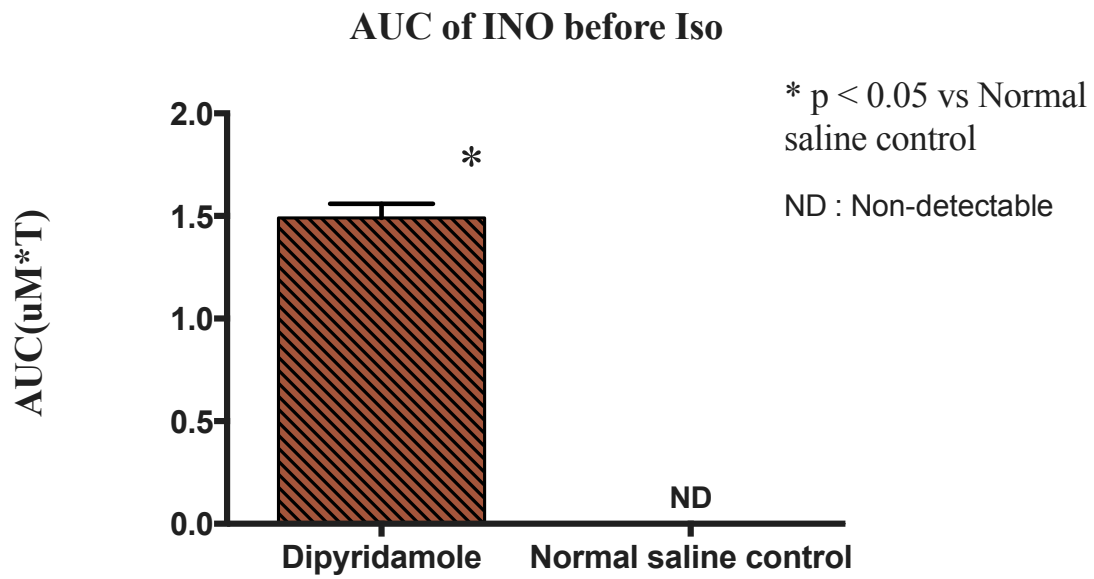
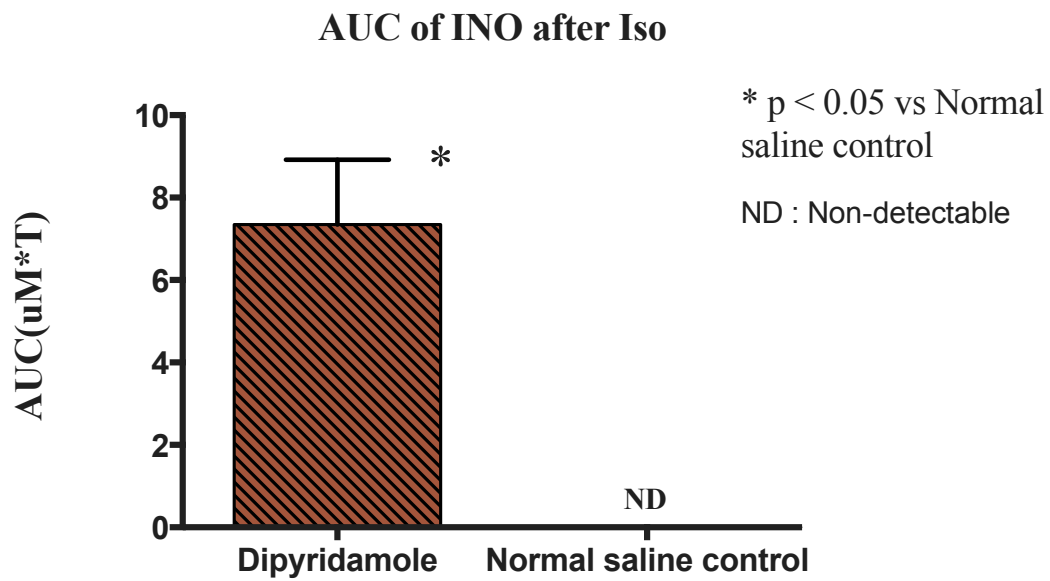


Figure 25: AUC of inosine in plasma after isoproterenol injection.



Similar to adenosine, plasma concentration of hypoxanthine fell immediately after the isoproterenol injection in the control group and which was quickly rebound close to the baseline concentration. This was not observed in the dipyridamole treatment group (Figure 26). There was no significant difference in the AUC of plasma concentration of hypoxanthine before (AUC 21.90 ± 1.31 in dipyridamole treated vs. 21.36 ± 9.28 $\mu\text{M}\cdot\text{T}$ in control group) and after isoproterenol (AUC 90.92 ± 41.93 in dipyridamole treated vs. 73.65 ± 35.68 $\mu\text{M}\cdot\text{T}$ in control group) between the control and dipyridamole treated groups (Figures 27 & 28).

Figure 26: Effect of dipyridamole (10mg/kg) on plasma hypoxanthine concentrations vs. normal saline control. (Data presented in mean \pm SEM).

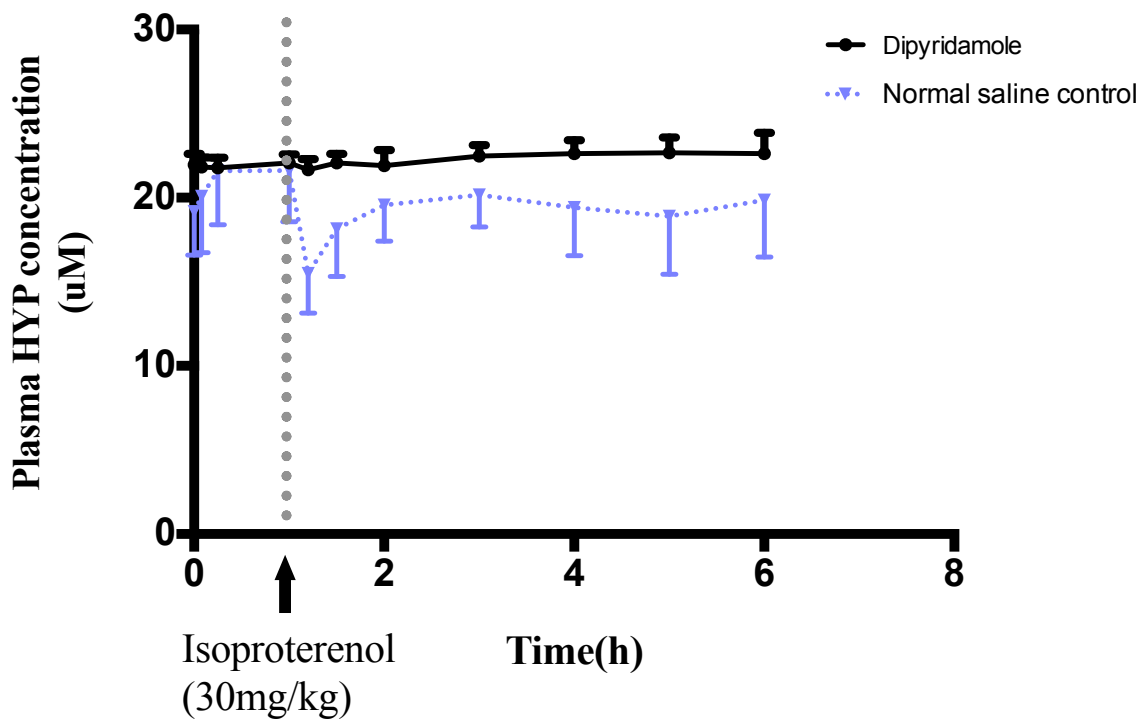


Figure 27: AUC of hypoxanthine in plasma before isoproterenol injection.

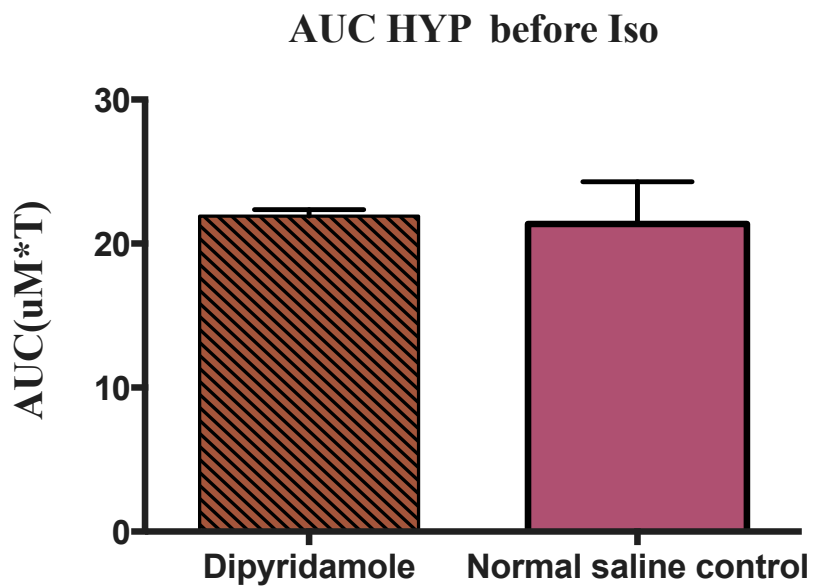
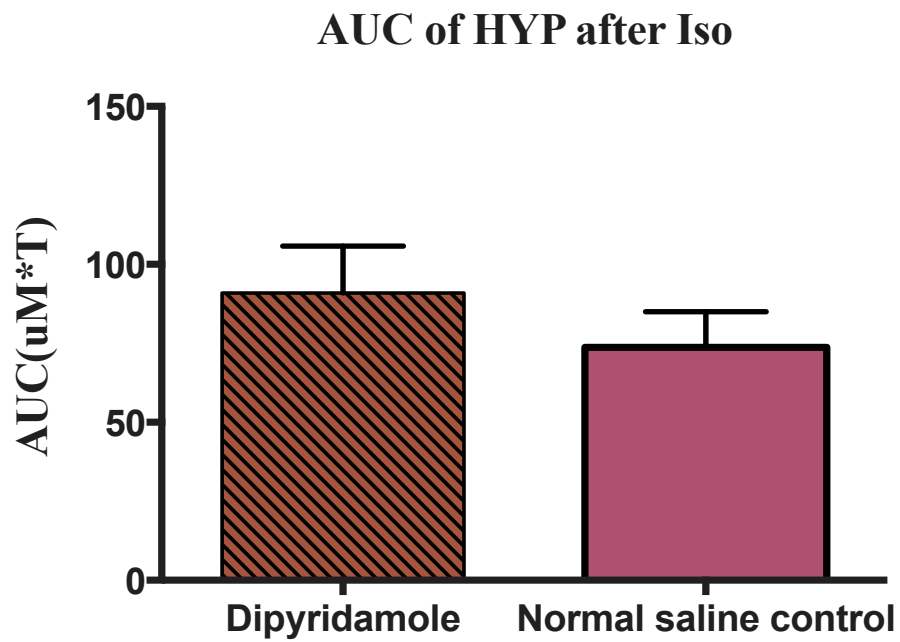


Figure 28: AUC of hypoxanthine in plasma after isoproterenol injection.



Shortly after isoproterenol injection there was an increase in plasma uric acid concentration both in normal saline control and dipyridamole treated group, however the increase was considerably greater in control group (Figure 29). There was no significant difference in the AUC of plasma concentrations of uric acid before or after isoproterenol between the dipyridamole treated and control groups (before Iso 32.85 ± 7.53 in dipyridamole vs. 19.52 ± 25.22 $\mu\text{M}\cdot\text{T}$ in control) and (after Iso 254.33 ± 119.82 in dipyridamole vs. 389.36 ± 239.95 $\mu\text{M}\cdot\text{T}$ in control) respectively (Figures 30 & 31). However, the C_{max} of plasma concentrations of uric acid after isoproterenol was significantly lower in treatment group (112.02 ± 36.28 vs. control 251 ± 152.94 μM , $p < 0.05$) (Figure 32).

Figure 29: Effect of dipyridamole (10mg/kg) on plasma uric acid concentrations vs. normal saline control. (Data presented in mean \pm SEM).

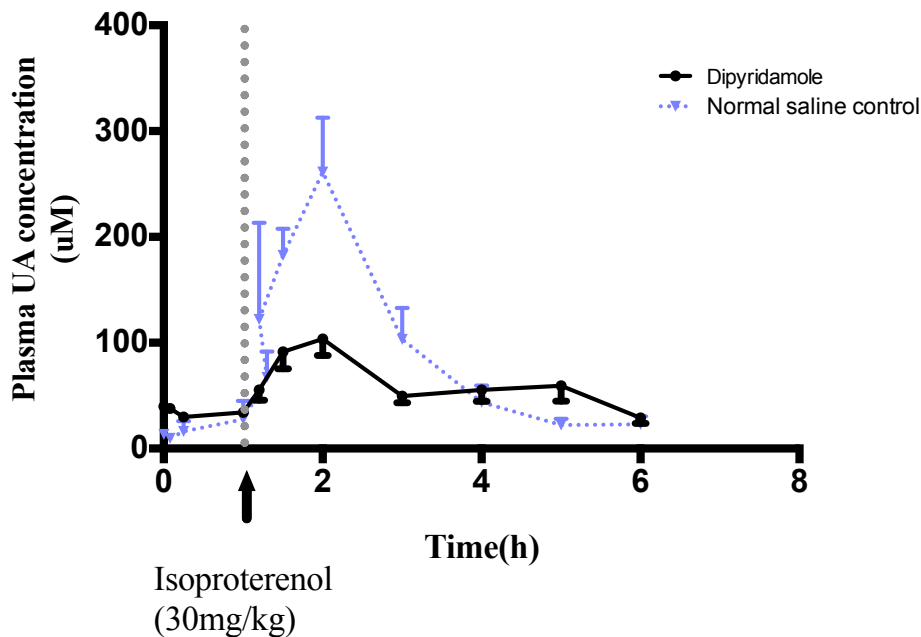


Figure 30: AUC of uric acid in plasma before isoproterenol injection.

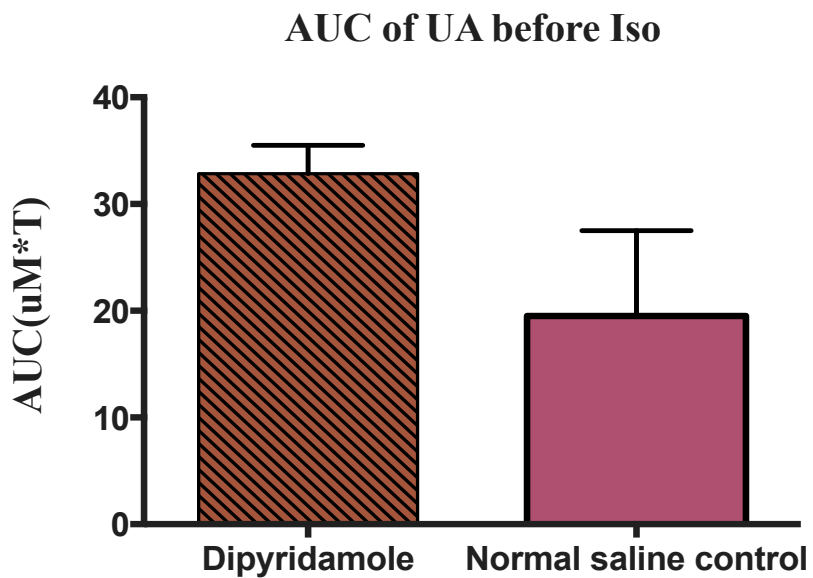


Figure 31: AUC of uric acid in plasma after isoproterenol injection.

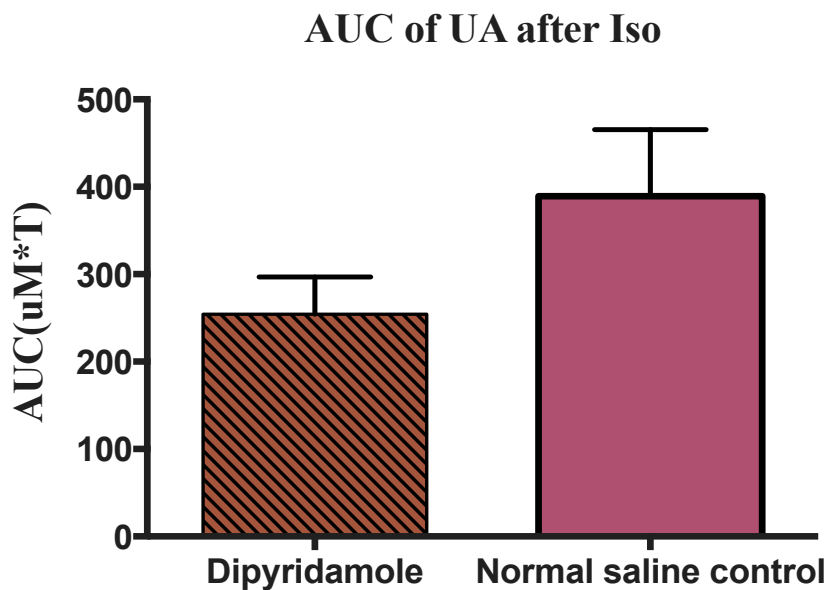
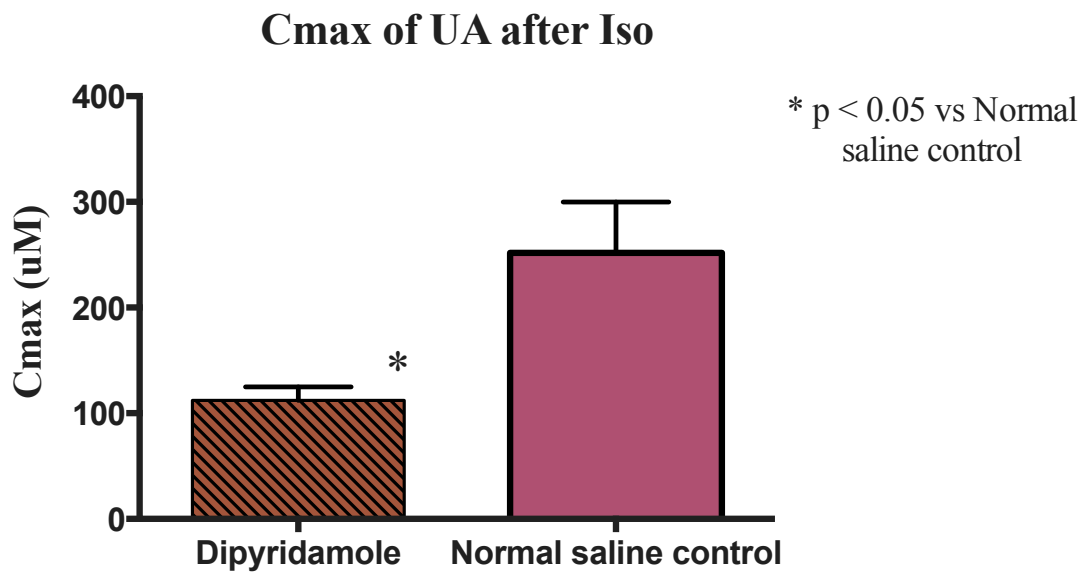


Figure 32: Cmax of uric acid in plasma after isoproterenol injection.



In the normal saline control group, plasma guanosine concentrations were not detected. However in the dipyridamole treated group the concentration were measurable and appeared to gradually decrease after isoproterenol injection (Figure 33). The AUC of guanosine concentrations in plasma both before and after isoproterenol were significantly higher in treatment group (AUC before Iso 1.10 ± 0.10 in dipyridamole vs. non-detectable in control; AUC after Iso 4.47 ± 2.03 in dipyridamole vs. non-detectable in control; $p < 0.05$ for both) (Figures 34 & 35).

Figure 33: Effect of dipyridamole (10mg/kg) on plasma guanosine concentrations vs. normal saline control. (Data presented in mean \pm SEM).

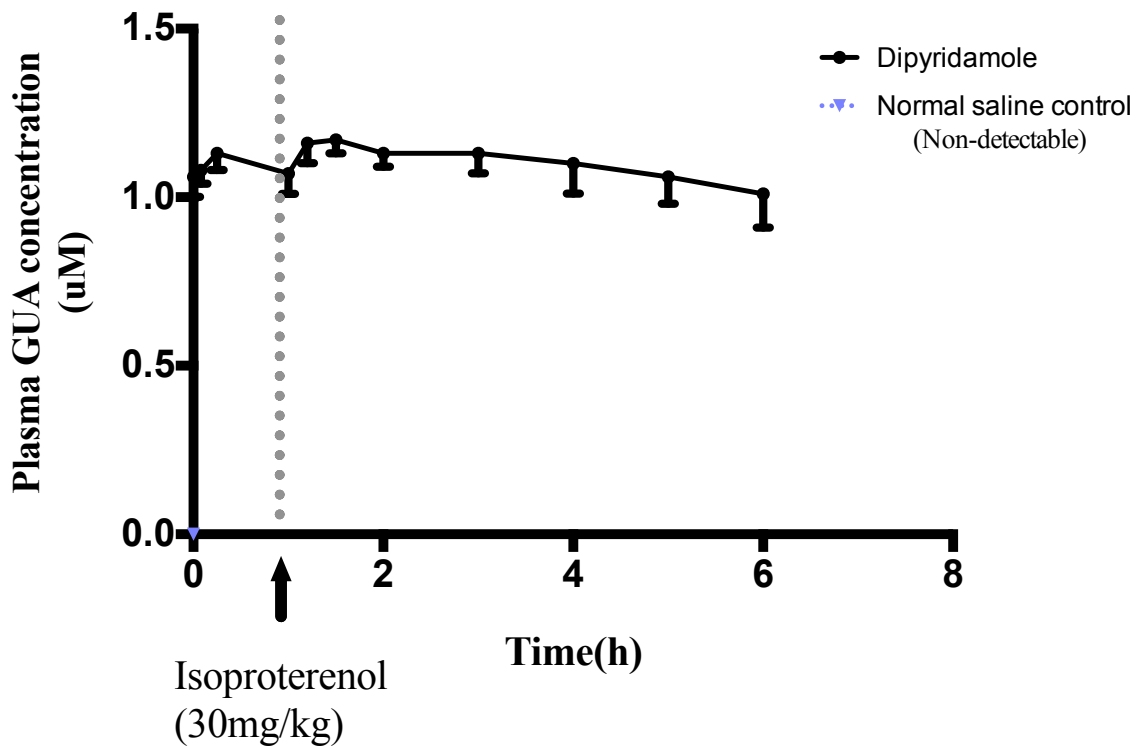


Figure 34: AUC of guanosine concentrations in plasma before isoproterenol injection.

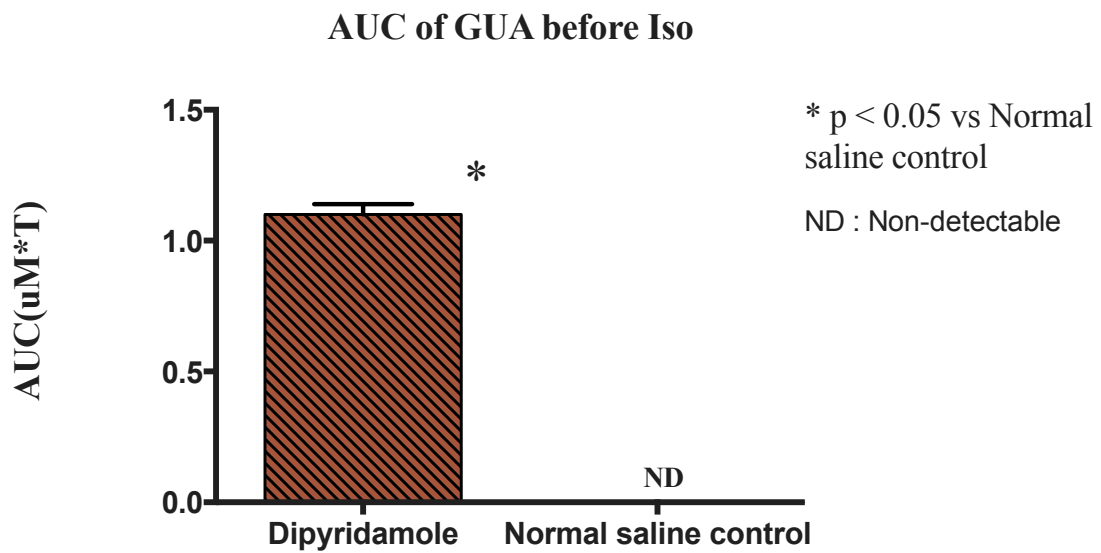
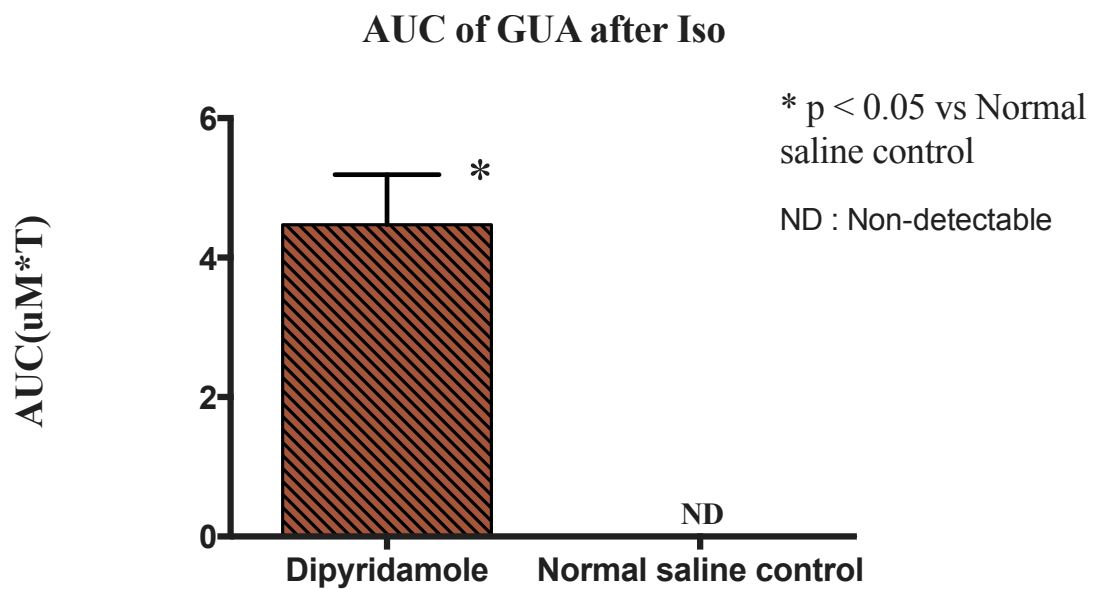


Figure 35: AUC of guanosine concentrations in plasma after isoproterenol injection.



4.2.3 Plasma Concentrations of Dipyridamole

Following the final and 5th doses of 10 mg/kg of dipyridamole administered via subcutaneous injection, plasma concentrations of dipyridamole rose rapidly reaching a maximum concentration (C_{max}) of 1.09 ± 0.42 ug/mL in 0.5 hr, and then followed a biphasic decline which was adequately characterized by a two compartment model with a first order input yielding a goodness of fit R-square value of 0.90 (Figure 36). The AUC of dipyridamole from T₀ to last measurable concentration as calculated was 2.79 ug*T with a terminal (elimination) half-life of 7.74 ± 6.02 hr. The pharmacokinetic variables of dipyridamole are summarized in Table 2.

Figure 36: Plasma concentration time profile of dipyridamole in rats after the 5th subcutaneous injection of dipyridamole (10 mg/kg).

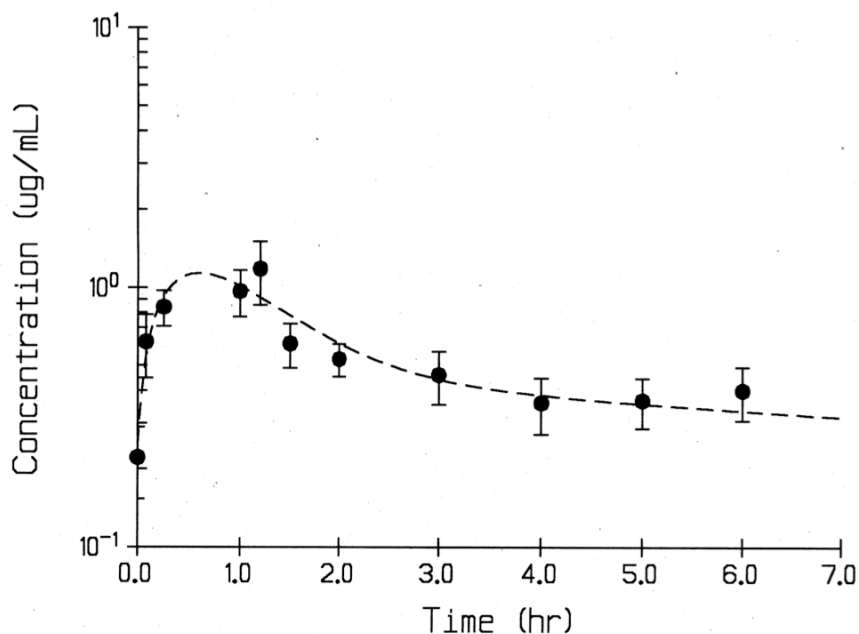


Table 2: Pharmacokinetic of dipyridamole in rats following the last subcutaneous (sc) injection (10 mg/kg).

Pharmacokinetic parameter	Data of dipyridamole in rats. (Data in mean values)
AUC last (uM*T)	2.79 ± 1.21
AUC infinity (uM*T)	6.97 ± 5.48
AUC trap (uM*T)	2.89 ± 1.26
CL last at SS (L/hr/kg)	4.25 ± 2.18
Half- life (absorption)	0.24 ± 0.10
Half- life (distribution - alpha)	1.18 ± 1.96
Half- life (elimination - beta)	7.74 ± 6.02
MRT (h)	1.97 ± 0.75
Cmax (ug/mL)	1.09 ± 0.42
Tmax (h)	0.51 ± 0.24
R-square	0.90 ± 0.09
MSQ	0.25 ± 0.48

4.2.4 Assay Reproducibility During Analysis

The reproducibility of the HPLC assay for the purine nucleotides was determined by using spiked RBC QC lysate samples prepared and analyzed in duplicates at 250 and 100 ug/mL for ATP and ADP; and at 50 and 20 ug/mL for AMP, GTP and GDP over a 8-month period. The mean coefficient of variation (% CV) of the peak height ratios for each batch of spiked RBC QC lysate standard (n = 8 -10) were used to determine the intra-assay variations. The coefficient of variation calculated from the mean peak height ratios from each separate batch (n = 8 - 10) were used to calculate inter-assay variations. The day-to-day reproducibility data of the assay are summarized in Table 3. The intra-assay variations of analytes were mostly smaller than 10 % and the inter-assay variations were smaller than 15 %.

Table 3: Summary of the reproducibility data of HPLC Assay for Purine nucleotides in RBC.

Assay variation			
(November 2013 – July 2014)			
Compound	Concentration of spiked QC RBC lysate (ug/mL)	Intra-assay variation *(%CV)	Inter-assay-variation **(%CV)
ATP	250	7.80	13.55
	100	11.19	10.15
ADP	250	6.97	11.94
	100	8.99	15.24
AMP	50	5.69	13.22
	20	10.64	14.30
GTP	50	8.28	14.02
	20	12.30	11.67
GDP	50	6.83	13.32
	20	10.23	15.61

**The mean CV's of the peak height ratios for each batch of spiked QC RBC lysate standards (n=8 - 10) were used to calculate the intra-assay variations.*

***The mean peak height ratios from each separate batch (n=8-10) were used to calculate the inter-assay variations.*

The reproducibility of the adenosine assay was determined by using spiked plasma QC samples at 2.5 and 0.5 ug/mL for ADO, INO and GUA and at 25 and 5 ug/mL for HYP, XAN, and UA over 3 months. The day-to-day reproducibility data of the assay are summarized in Table 2. The intra-assay and the inter-assay variations of analytes were

mostly smaller than 15 % with few exceptions (Table 4).

Table 4: Summary of the reproducibility data of HPLC Assay for adenosine and its metabolites in plasma.

Assay variation			
(January 2015 – March 2015)			
Compound	Concentration of Spiked Plasma (ug/mL)	Intra-Assay Variation *(%CV)	Inter-Assay-Variation **(%CV)
ADO	2.5	12.52	10.66
	0.5	12.44	10.73
INO	2.5	13.29	5.32
	0.5	10.09	14.82
HYP	25	9.56	9.37
	5	6.46	7.55
XAN	25	20.64***	13.85
	5	17.46	10.74
UA	25	11.15	9.31
	0.5	10.38	10.57
GUA	2.5	11.15	5.62
	0.5	14.32	22.22***

**The mean CV's of the peak height ratios for each batch of spiked plasma QC samples (n=4 - 5) were used to calculate the intra-assay variations.*

*** The mean peak height ratios from each separate batch (n=4-5) were used to calculate the inter-assay variations.*

****The large variation was due to different plasma blank used during the course of the analysis some of which had more interference than others*

The reproducibility of the dipyridamole assay was determined by spiked plasma QC samples at 1 and 0.1 ug/mL. The intra- and inter-assay variations for this assay were calculated using a similar procedure over a 2-month period (n = 5 – 7), and was determined to be less than 15% and 20%, respectively (Table 5).

Table 5: Summary of the reproducibility data of HPLC Assay for dipyridamole in plasma.

Assay Variation (September 2014 – October 2014)		
Concentration of Dipyridamole Spiked Plasma (ug/mL)	Intra-Assay Variation *(%CV)	Inter-Assay-Variation **(%CV)
1	14.62	17.64
0.1	8.49	7.78

**The mean CV's of the peak height ratios for each batch of spiked plasma standards (n=5 - 7) were used to express the intra-assay variations.*

*** The mean peak height ratios from each separate batch (n=5 - 7) were used to calculate the inter-assay variations.*

CHAPTER: 5 DISCUSSION

In adopting an HPLC assay to measure plasma concentrations of dipyridamole for the pharmacokinetic study, we optimized the solid phase extraction using different eluting solvent compositions and pH of the phosphate buffer. As shown in Table 6 with the composition of acetonitrile: methanol (1:9) at a pH of 6.5 recoveries of dipyridamole and losartan were 100 % and 20 % respectively. Also at this composition and pH there was minimal interference from endogenous plasma materials, which eluted after losartan.

The T_{1/2} and T_{max} reported in the present study as shown in Table 4 were comparable with the results previously reported from an earlier study [73]. In our earlier study isoproterenol (30 mg/kg) injected subcutaneously induced ischemia injury resulting in 50 % mortality in rats [6]. In the current study reported in thesis, mortality in the dipyridamole (10 mg/kg) treated rats was reduced to 25 % compared to the normal saline control group although the reduction in mortality was not statistically significant because of the small number of animal used in the study (n = 8 in dipyridamole treated group vs. n = 10 in normal saline control). The results never the less concur with the finding from earlier studies, which showed cardiovascular protective effect of dipyridamole against ischemia and myocardial injury in experimental animal models [52, 74].

It has also been shown that during ischemia/hypoxia ATP is broken down to ADP, AMP and adenosine in myocardium, endothelium and RBC [19, 25, 75]. In the current study there was a decline of ATP concentrations in the RBC shortly after isoproterenol in

the control group, which was not observed in the dipyridamole treatment group (Figure 3). There was also a significant increase in RBC ATP concentrations immediately after isoproterenol injection in both control and dipyridamole treated rats ($p < 0.05$ paired t-test), and the increase was significantly higher in the treatment group ($p < 0.05$ t-test). On the other hand, there was a significant increase in the RBC AMP concentrations after isoproterenol in the control group, but no increase in the dipyridamole treatment group (Table 7). The AUC ratios of ADP to ATP and AMP to ATP after isoproterenol were significantly lower in the dipyridamole treatment group ($p < 0.05$) suggesting dipyridamole reduced the breakdown/metabolism of ATP in the RBC induced by ischemia injury (Table 9). In our earlier study, diltiazem was shown to significantly decrease RBC ADP and AMP concentrations ($p < 0.05$) and a moderate increase in RBC ATP concentrations after ischemia injury [72]. These results suggest that during ischemia / hypoxia dipyridamole preserves RBC ATP concentrations by attenuating its catabolism to ADP and AMP in the RBC. Similar results were also observed with RBC GTP concentrations (Table 7, 8 & 9) suggesting that the effect of dipyridamole is not specific only to moderate ATP metabolism, and that other factors may be involved in the cardiovascular protection. On the other hand, there were no differences in the purine nucleotide ratios before isoproterenol between the control and dipyridamole treated rats (Table 8) suggesting that dipyridamole had no effect on catabolism of purine nucleotides in normal physiologic conditions.

Table 6. Optimization Of SPE Of Dipyridamole And Losartan From Plasma Samples			
Eluting solvent composition and pH of the phosphate buffer	Dipyridamole Recovery (%)	Losartan Recovery (%)	Plasma interference eluted after losartan*
Acetonitrile (pH 6.5)	60	89	++++
Acetonitrile: Methanol (1:1) (pH 6.5)	85	57	+++
Acetonitrile: Methanol (3:7) (pH 6.5)	83	41	++
Acetonitrile: Methanol (1:9) (pH 6.5)	100	20	+
Acetonitrile: Methanol (2:8) (pH 6.5)	92	17	+
Acetonitrile: Methanol (3:7) (0.7 mL) (pH 6.5)	59	38	+++
Acetonitrile: Methanol (7:3) (pH 6.5)	179	65	++
Acetonitrile: Methanol:water (3:5:2) (pH 6.5)	124	91.61	+++
Acetonitrile: Methanol (3:7) (pH 3.2)	123	112	++++
Acetonitrile: Methanol (3:7) (pH 8)	98	100	++++
Methanol (6.5)	90	73	++++
*Amount of interference depend on the integrity of the plasma samples (e.g. amount increase after more freeze-thaw cycles)			

Table 7. Effect Of Dipyridamole On Changes Of RBC ATP And Adenine Nucleotides Concentration After Isoproterenol Injection⁺

Adenine nucleotides/Treatment Group	Dipyridamole (10 mg/kg) twice daily for 4 doses (n = 8)	Normal saline control 1mL/kg twice daily for 4 doses (n = 10)
ATP (mM)	0.55 + 0.26* [*] **	0.20 + 0.28**
ADP (mM)	0.01 + 0.13*	0.20 + 0.18**
AMP (mM)	0.00 + 0.02*	0.13 + 0.19 **
GTP (mM)	0.07 + 0.04**	0.04 + 0.06
GDP (mM)	0.01 + 0.02	0.02 + 0.02**

⁺Determined from the difference between the average concentration before and 0.5 – 2 hrs after isoproterenol.

*p < 0.05 vs. normal saline control.

**p < 0.05 vs. before isoproterenol injection (paired t-test).

Table 8. Effect Of Dipyridamole On RBC ATP And Adenine Nucleotide Ratios Before Isoproterenol Injection		
Adenine Nucleotides / Treatment Group	Dipyridamole (10 mg/kg) twice daily for 4 doses (n = 8)	Normal saline control (1 mL/kg) twice daily for 4 doses (n = 10)
ADP/ATP AUC Ratio	0.18± 0.06	0.36 ± 0.36
AMP/ATP AUC Ratio	0.02 ± 0.02	0.04 ± 0.05
AMP/ADP AUC Ratio	0.10± 0.08	0.12 ± 0.04
GDP/GTP AUC Ratio	0.28 ± 0.09	0.32 ± 0.11
*p < 0.05 vs. Normal saline control		

Table 9. Effect Of Dipyridamole On RBC ATP And Adenine Nucleotide Ratios After Isoproterenol Injection		
Adenine Nucleotides / Treatment Group	Dipyridamole (10 mg/kg) twice daily for 4 doses (n = 8)	Vehicle Normal saline 1 mL/kg twice daily for 4 doses (n = 10)
ADP/ATP AUC Ratio	0.16 ± 0.08*	0.34 ± 0.16
AMP/ATP AUC Ratio	0.01 ± 0.01*	0.09 ± 0.09
AMP/ADP AUC Ratio	0.13 ± 0.11	0.22 ± 0.11
GDP/GTP AUC Ratio	0.25 ± 0.09	0.39 ± 0.19
*p < 0.05 vs. Normal saline control		

There was a significant increase in plasma concentrations of adenosine and uric acid shortly after isoproterenol ($p < 0.05$ paired t-test) in both dipyridamole treated rats and in the control group. The increase was significantly higher in the control group ($p < 0.05$) (Table 10). The fact that the increase of plasma adenosine and uric acid levels was less in the treatment group could be attributed to the effect of dipyridamole on preserving ATP in the RBC after isoproterenol. As shown in Table 9, the ratios of ADP/ATP and AMP/ATP were significantly lower in the dipyridamole treatment group. As a result of a decreased ATP breakdown in the RBC in the dipyridamole treatment group plasma concentration of adenosine was lower. Consequently the uric acid concentration was also lower in the dipyridamole treatment vs. control groups (Figure 28). However, as shown in Table 12, there was no significant difference in the AUC ratios of UA/ADO or HYP/ADO after isoproterenol suggesting that the effect of dipyridamole on breakdown of adenosine to its oxypurine metabolites was minimal. The lower plasma concentrations of adenosine and uric acid were attributed mainly to an indirect effect from less breakdown of ATP in the RBC.

It is interesting to note that before isoproterenol the concentrations and AUC of adenosine in plasma were higher in the control vs. treatment group although the difference was not statistically significant (Figure 19 and 20, $p > 0.05$). The lower adenosine concentrations in the dipyridamole treatment group before isoproterenol could be attributed to an increased oxidative metabolism of adenosine to hypoxanthine and uric acid which was supported by the significantly higher AUC ratios of HYP/ADO and

UA/ADO in the dipyridamole treatment group (Table 11). The clinical significance of the effect of dipyridamole on adenosine metabolism in plasma is not clear and warrants further investigation.

Table 10. Effect Of Dipyridamole On Changes Of Adenosine And Its Metabolites After Isoproterenol Injection⁺		
Adenosine and its metabolites /Treatment Group	Dipyridamole (10 mg/kg) twice daily for 4 doses (n = 8)	Normal Saline Control 1mL/kg twice daily for 4 doses (n = 10)
ADO (uM) ⁺⁺	0.90 + 0.65* ^{***}	2.50 + 1.98**
INO (uM)	-0.24 + 0.16**	ND
HYP (uM)	0.47 + 1.45	0.68 + 5.22
UA (uM)	64.61 + 41.32* ^{***}	221.44 + 165.64**
GUA (uM)	-0.05 + 0.08	ND
⁺ Determined from the difference between the average concentration before and 0.5 – 2 hrs after isoproterenol. ⁺⁺ Determined from the difference between the average concentration before and Cmax after isoproterenol * ^p < 0.05 vs normal saline control. ** ^p < 0.05 vs before isoproterenol injection (paired t-test).		

Table 11. Effect Of Dipyridamole On Adenosine And Its Purine Metabolite Ratios In Plasma Before Isoproterenol (Iso) Injection In Rats		
Adenine Nucleotides/Treatment Group	Dipyridamole (10 mg/kg) twice daily for 4 doses (n = 8)	Normal saline control 1mL/kg twice daily for 4 doses (n = 10)
HYP / ADO AUC Ratio	21.68 ± 4.64*	11.64 ± 6.08
UA / ADO AUC Ratio	32.95 ± 11.79*	9.31 ± 13.59
UA / HYP AUC Ratio	1.49 ± 0.30	1.12 ± 1.57
*p < 0.05 vs normal saline control		

Table 12. Effect Of Dipyridamole On Adenosine And Its Purine Metabolite Ratios In Plasma After Isoproterenol (Iso) Injection In Rats		
Adenine Nucleotides/Treatment Group	Dipyridamole (10 mg/kg) twice daily for 4 doses (n = 8)	Normal saline control 1mL/kg twice daily for 4 doses (n = 10)
HYP / ADO AUC Ratio	19.93 ± 7.77	15.67 ± 21.98
UA / ADO AUC Ratio	53.63 ± 18.80	66.18 ± 54.10
UA / HYP AUC Ratio	2.90 ± 1.42	6.89 ± 6.69

CHAPTER: 6 LIMITATIONS AND FUTURE DIRECTIONS

In the current study the vehicle used to prepare the dipyridamole injectable was PEG: normal saline (8:2). In the current study, the control group received normal saline (1mL/kg) for comparison. Thus the cardiovascular protective effect of dipyridamole observed from the current study could be confounded by the effect of the vehicle albeit that it may be very small. A future study should include a control group, which will receive PEG: normal saline (8: 2) for comparison. Such studies can further tease out the cardiovascular protective effect of dipyridamole without the vehicle effect. The relationship between the purine nucleotide concentrations in the RBC and that occurring in other tissues such as in the endothelium and myocardium, and the effect of other protective agents on ATP metabolism warrant further investigation. Understanding these concepts and the linkages would enable us to explore further the potential of nucleoside transport inhibitors (NTI) and other cardiovascular agents for myocardial ischemia[15]. Finally, the central question whether or not ATP metabolism in the RBC may be used as a surrogate biomarker for energy balance in the body warrants for management of cardiovascular disease warrants further studies.

CHAPTER 7: CONCLUSION

In conclusion, we have shown dipyridamole reduced mortality from acute myocardial injury induced by isoproterenol in an experimental rat model. However, due to the small number of rats employed in the experiment, the difference in mortality between the dipyridamole treatment group and the control was not statistically significant. The study has also showed dipyridamole preserved ATP concentrations in the RBC after isoproterenol. However, whether or not ATP metabolism in the RBC may be used, as a prognostic biomarker for cardiovascular protective effect of dipyridamole could not be confirmed by the study. The effect may be attributed to preserving ATP concentrations in the RBC and possibly also via other indirect mechanisms which will require further studies including using larger number of animals, other models of cardiovascular injury and anti-ischemia agents in addition to dipyridamole.

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APPENDIX 1: Rat 371

Title: Measurement of RBC Concentrations of ATP in Rat 371 extracted by Shyam Sundar
 Based on SOP NO.: SOP/STD/2005-005-0* (With Stopping Solution)
 Experiment Date: 15/04/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered	% Recovery
ATP 3 ng										
a250	250 µg/mL	62.86	14.91	4.22	35.00	4.22	4.22	0.35	14378.46	48.63
b250	250 µg/mL	48.02	11.95	4.02	35.00	4.02	4.02	0.35	10983.99	35.05
Mean		55.44	13.43	4.12	35.00	4.12	4.12		12681.22	41.84
SD		10.48	2.09	0.14	0.00	0.14	0.14		2403.25	5.50
%CV		18.93	15.58	3.39	0.00	3.39	3.39		18.93	22.95
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a100										
a100	100 µg/mL	21.46	11.88	1.81	35.00	1.81	1.81	0.35	4908.71	26.87
b100	100 µg/mL	28.86	14.09	2.05	35.00	2.05	2.05	0.35	6601.37	43.80
100*	100 µg/mL									
Mean		25.16	12.99	1.93	35.00	1.93	1.93		5755.04	35.34
SD		5.23	1.56	0.17	0.00	0.17	0.17		1196.89	11.97
%CV		20.80	12.03	8.87	0.00	8.87	8.87		20.80	33.87
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a0										
a0	0 µg/mL (a)	55.55	97.90	0.57	35.00	0.57	0.57	2.00	2232.62	
b0	0 µg/mL (a)	55.44	98.32	0.56	35.00	0.56	0.56	2.00	2219.21	
Mean		55.50	98.11	0.57	35.00	0.57	0.57		2225.42	
SD		0.08	0.30	0.00	0.00	0.00	0.00		3.11	
%CV		0.14	0.30	0.44	0.00	0.44	0.44		0.14	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRB
250.00	4.12	0.57	3.55
100.00	1.93	0.57	1.36
0.00	0.57	0.57	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0233
Std Err of Y Est	0.0478
R Squared	0.0996
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0142
Std Err of Coef.	0.0003

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc(mM) RBC	Conc(mM) RBC
R37110	0.00	123.96	123.36	1.00	35.00	1.00	1.00	2.00	-	72.21	0.1424	1.831
R37110.05	0.05	111.95	91.17	1.23	35.00	1.23	1.23	2.00	-	97.88	0.1733	2.228
R37110.25	0.25	136.25	122.75	1.03	35.00	1.03	1.03	2.00	-	73.88	0.1457	1.873
R37111	1.00	89.79	59.81	1.50	35.00	1.50	1.50	1.00	-	107.08	0.2111	2.714
Regression (µg/ml/kg bw)												
R37111.2	1.20	81.06	50.50	1.61	35.00	1.61	1.61	1.00	-	114.38	0.2255	2.899
R37111.5	1.50	80.98	65.60	1.23	35.00	1.23	1.23	1.00	-	88.34	0.1742	2.239
R37112	2.00	61.50	53.20	1.16	35.00	1.16	1.16	1.00	-	82.83	0.1633	2.160
R37113	3.00	50.39	73.81	1.22	35.00	1.22	1.22	1.00	-	87.65	0.1728	2.222
R37114	4.00	107.38	66.79	1.61	35.00	1.61	1.61	1.00	-	114.56	0.2259	2.904
R37115	5.00	97.81	69.73	1.40	35.00	1.40	1.40	1.00	-	100.16	0.1975	2.539
R37116	6.00	97.29	57.86	1.68	35.00	1.68	1.68	1.00	-	119.49	0.2355	3.029
Mean		97.12	75.88	1.33	35.00	1.33	1.33	1.27		95.31	0.19	2.42
SD		16.44	25.79	0.24	0.00	0.24	0.24	0.47		16.71	0.03	0.42
%CV		20.02	33.99	17.84	0.00	17.84	17.84	36.70		17.53	17.53	17.53
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00		11.00	11.00	11.00

ATP (5 ng)

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on May 31, 2011.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 15/04/2014

Checked by: Date:

Approved by: Pollen Young Date: 17/04/2014

Title: Measurement of RBC Concentrations of ADP in Rat 371 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 10/04/2014

Sample/standard ID	Standard C (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj VoL (µL)	Amount Reco	% Recovery
ADP 3 ng		74.16						3.00		
a250	250 ug/mL	114.17	14.91	7.66	35.00	7.66	7.66	0.35	7257.69	28.20
b250	250 ug/mL	89.69	11.95	7.51	35.00	7.51	7.51	0.35	5701.51	21.97
Mean		101.93	13.43	7.58	35.00	7.58	7.58		6479.60	25.09
SD		17.31	2.09	0.11	0.00	0.11	0.11		1100.38	4.40
% CV		16.98	15.58	1.42	0.00	1.42	1.42		16.98	17.54
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
100a	100 ug/mL	31.45	11.88	2.65	35.00	2.65	2.65	0.35	1999.25	17.91
100b	100 ug/mL	37.41	14.09	2.66	35.00	2.66	2.66	0.35	2378.12	21.70
100*	100 ug/mL									
Mean		34.43	12.99	2.65	35.00	2.65	2.65		2188.68	19.81
SD		4.21	1.56	0.01	0.00	0.01	0.01		267.90	2.68
% CV		12.24	12.03	0.21	0.00	0.21	0.21		12.24	13.52
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
aB	0 ug/mL (a)	18.83	97.90	0.19	35.00	0.19	0.19	2.00	209.48	
bB	0 ug/mL (a)	18.53	98.32	0.19	35.00	0.19	0.19	2.00	206.14	
Mean		18.68	98.11	0.19	35.00	0.19	0.19		207.81	
SD		0.21	0.30	0.00	0.00	0.00	0.00		2.36	
% CV		1.14	0.30	1.44	0.00	1.44	1.44		1.14	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Valu (PHRV)	Blank (PHRb)	PHRV-PHRb
250.00	7.58	0.19	7.39
100.00	2.65	0.19	2.46
0.00	0.19	0.19	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.1956
Std Err of Y Est	0.4020
R Squared	0.9943
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0298
Std Err of Coef.	0.0023

Sample ID	Time post c	Peak Ht. # (nm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj VoL (µL)	Hemolysis Degree	Conc(µg/mL)	Conc.(mM) Lysate	Conc.(mM) RBC
R371T0	0.00	62.45	123.36	0.51	35.00	0.51	0.51	2.00	-	23.53	0.0551	0.708
R371T0.08	0.08	29.60	91.17	0.32	35.00	0.32	0.32	2.00	-	17.45	0.0408	0.525
R371T0.25	0.25	31.70	122.75	0.26	35.00	0.26	0.26	2.00	-	15.22	0.0356	0.458
R371T1	1.00	24.47	59.81	0.41	35.00	0.41	0.41	1.00	-	20.28	0.0475	0.610
Metamizolol (30 mg/kg sc)												
R371T1.2	1.20	16.54	50.50	0.33	35.00	0.33	0.33	1.00	-	17.54	0.0411	0.528
R371T1.5	1.50	15.56	65.60	0.24	35.00	0.24	0.24	1.00	-	14.51	0.0340	0.437
R371T2	2.00	15.97	53.20	0.30	35.00	0.30	0.30	1.00	-	16.62	0.0389	0.500
R371T3	3.00	26.18	73.81	0.35	35.00	0.35	0.35	1.00	-	18.45	0.0432	0.555
R371T4	4.00	18.34	66.79	0.27	35.00	0.27	0.27	1.00	-	15.77	0.0389	0.474
R371T5	5.00	13.52	69.73	0.19	35.00	0.19	0.19	1.00	-	13.06	0.0306	0.393
R371T6	6.00	12.89	57.98	0.22	35.00	0.22	0.22	1.00	-	14.01	0.0328	0.422
Mean		41.25	112.43	0.36	35.00	0.36		2.00		18.73	0.04	0.56
SD		18.39	18.41	0.13	0.00	0.13		0.00		4.30	0.01	0.13
% CV		44.58	16.38	35.36	0.00	35.36		0.00		22.98	22.98	22.98
n		3.00	3.00	3.00	3.00	3.00		3.00		3.00	2.00	3.00

ADP (5 ng)

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on May 31, 2011.
 *Repeated injections of a or b at 0.5 -1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj VoL = Injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K

Date: 15/04/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date: 17/04/2014

Title: Measurement of RBC Concentrations of AMP in Rat 371 extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date 10/04/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
AMP 3 ng										
a50	50 ug/mL	58.44	14.91	3.92	35.00	3.92	3.92	0.35	2315.34	45.99
b50	50 ug/mL	45.37	11.95	3.80	35.00	3.80	3.80	0.35	1797.52	35.63
Mean		51.91	13.43	3.86	35.00	3.86	3.86		2056.43	40.81
SD		9.24	2.09	0.09	0.00	0.09	0.09		366.16	7.32
%CV		17.81	15.58	2.25	0.00	2.25	2.25		17.81	17.95
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20										
a20	20 ug/mL	16.17	11.88	1.36	35.00	1.36	1.36	0.35	640.64	31.23
b20	20 ug/mL	19.75	14.09	1.40	35.00	1.40	1.40	0.35	782.48	38.32
20*	20 ug/mL									
Mean		17.96	12.99	1.38	35.00	1.38	1.38		711.56	34.77
SD		2.53	1.56	0.03	0.00	0.03	0.03		100.29	5.01
%CV		14.09	12.03	2.08	0.00	2.08	2.08		14.09	14.42
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a8										
a8	0 ug/mL (a)	2.47	97.90	0.03	35.00	0.03	0.03	2.00		17.13
b8	0 ug/mL (a)	2.17	98.32	0.02	35.00	0.02	0.02	2.00		15.05
Mean		2.32	98.11	0.02	35.00	0.02	0.02			16.09
SD		0.21	0.30	0.00	0.00	0.00	0.00			1.47
%CV		9.14	0.30	9.45	0.00	9.45	9.45			9.14
n		2.00	2.00	2.00	2.00	2.00	2.00			2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRB
50.00	3.86	0.02	3.83
20.00	1.38	0.02	1.36
0.00	0.02	0.02	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0695
Std Err of Y Est	0.1428
R Squared	0.9973
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0772
Std Err of Coef.	0.0040

Sample ID	Time post dose	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL)	Conc.(mM) Lysate	Conc.(mM) RBC
R371T0	0.00	ND	123.36	0.00	35.00	0.00	0.00	2.00	-	0.90	0.0026	0.033
R371T0.08	0.08	1.30	91.17	0.01	35.00	0.01	0.01	2.00	-	1.09	0.0031	0.040
R371T0.25	0.25	1.60	122.75	0.01	35.00	0.01	0.01	2.00	-	1.07	0.0031	0.040
R371T1	1.00	3.78	59.81	0.06	35.00	0.06	0.06	1.00	-	1.72	0.0050	0.054
Isoproterenol (10 mg/kg sc)												
R371T1.2	1.20	0.63	50.50	0.01	35.00	0.01	0.01	1.00	-	1.06	0.0031	0.039
R371T1.5	1.50	0.58	65.60	0.01	35.00	0.01	0.01	1.00	-	1.02	0.0029	0.038
R371T2	2.00	1.00	53.20	0.02	35.00	0.02	0.02	1.00	-	1.14	0.0033	0.042
R371T3	3.00	1.94	73.81	0.03	35.00	0.03	0.03	1.00	-	1.24	0.0036	0.046
R371T4	4.00	0.86	66.79	0.01	35.00	0.01	0.01	1.00	-	1.07	0.0031	0.040
R371T5	5.00	0.60	69.73	0.01	35.00	0.01	0.01	1.00	-	1.01	0.0029	0.037
R371T6	6.00	0.43	57.98	0.01	35.00	0.01	0.01	1.00	-	1.00	0.0029	0.037
Mean		1.16	75.88	0.02	35.00	0.02	0.02	1.27	0.00	1.12	0.00	0.04
SD		1.03	25.79	0.02	0.00	0.02	0.02	0.47	0.00	0.22	0.00	0.01
%CV		89.06	33.99	99.11	0.00	99.11	99.11	36.70	EPH	19.38	19.38	19.38
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00		11.00	11.00	11.00

AMP (5 ng)

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on May 31, 2011.
 *Repeated injections of "a" or "b" at 0.5 - 1 ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar Date: 15/04/2014

Checked by: Date:

Approved by: Pallen Yeung Date: 16/04/2014

Title: Measurement of RBC Concentrations of GTP in Rat 371 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 10/04/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recove %	Recovery
GTP 3 ng		36.11						3.00		
a50	50 ug/mL	19.83	14.91	1.33	35.00	1.33	1.33	0.35	2588.88	48.52
b50	50 ug/mL	15.97	11.95	1.34	35.00	1.34	1.34	0.35	2084.94	38.44
Mean		17.90	13.43	1.33	35.00	1.33	1.33		2336.91	43.48
SD		2.73		0.00	0.00	0.00	0.00		356.34	7.13
%CV		15.25	0.00	0.34	0.00	0.34	0.34		15.25	16.39
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20	20 ug/mL	6.64	11.88	0.56	35.00	0.56	0.56	0.35	868.88	35.19
b20	20 ug/mL	7.86	14.09	0.56	35.00	0.56	0.56	0.35	1026.15	43.16
20*	20 ug/mL									
Mean		7.25	12.99	0.56	35.00	0.56	0.56		946.51	39.18
SD		0.86	1.56	0.00	0.00	0.00	0.00		112.62	5.63
%CV		11.90	12.03	0.14	0.00	0.14	0.14		11.90	14.37
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
aB	0 ug/mL (a)	7.16	97.90	0.07	35.00	0.07	0.07	2.00	163.58	
bB	0 ug/mL (a)	7.11	98.32	0.07	35.00	0.07	0.07	2.00	162.44	
Mean		7.14	98.11	0.07	35.00	0.07	0.07		163.01	
SD		0.04	0.30	0.00	0.00	0.00	0.00		0.81	
%CV		0.50	0.30	0.80	0.00	0.65	0.80		0.50	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRB
50.00	1.33	0.07	1.26
20.00	0.56	0.07	0.49
0.00	0.07	0.07	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0073
Std Err of Y Est	0.0150
R Squared	0.9987
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0253
Std Err of Coef.	0.0004

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(nM) RBC	Conc.(nM)
R371T0	0.00	15.74	123.36	0.13	35.00	0.13	0.13	2.00	-	5.34	0.0102	0.131
R371T0.08	0.08	19.63	91.17	0.22	35.00	0.22	0.22	2.00	-	8.81	0.0198	0.217
R371T0.25	0.25	23.72	122.75	0.19	35.00	0.19	0.19	2.00	-	7.94	0.0152	0.195
R371T1	1.00	16.61	59.81	0.28	35.00	0.28	0.28	1.00	-	11.28	0.0216	0.277
Isoproterenol (30 µg/kg sc)												
R371T1.2	1.20	15.54	50.50	0.31	35.00	0.31	0.31	1.00	-	12.47	0.0238	0.306
R371T1.5	1.50	15.92	65.60	0.24	35.00	0.24	0.24	1.00	-	9.90	0.0199	0.243
R371T2	2.00	10.69	53.20	0.20	35.00	0.20	0.20	1.00	-	8.24	0.0157	0.202
R371T3	3.00	16.37	73.81	0.22	35.00	0.22	0.22	1.00	-	9.07	0.0173	0.223
R371T4	4.00	24.66	66.79	0.37	35.00	0.37	0.37	1.00	-	14.91	0.0285	0.366
R371T5	5.00	21.92	69.73	0.31	35.00	0.31	0.31	1.00	-	12.74	0.0243	0.313
R371T6	6.00	22.55	57.98	0.39	35.00	0.39	0.39	1.00	-	15.69	0.0300	0.386
Mean		18.49	75.88	0.26	35.00	0.26	0.26	1.27	0.00	10.58	0.02	0.26
SD		4.32	25.79	0.08	0.00	0.08	0.08	0.47	0.00	3.15	0.01	0.08
%CV		23.38	33.99	30.62	0.00	30.62	30.62	36.70	EPH	29.78	29.78	29.78
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

GTP (5 ng)

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on May 31, 2011.
 *Repeated injections of a or b at 0.5 -1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 15/04/2014

Checked by: Date:

Approved by: Pollen Yeung Date:16/04/2014

Title: Measurement of RBC Concentrations of GDP in Rat 371 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 10/04/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
GDP 3 ng										
a50	50 ug/mL	32.27	14.91	2.16	35.00	2.16	2.16	0.35	1480.73	29.22
b50	50 ug/mL	24.84	11.95	2.08	35.00	2.08	2.08	0.35	1139.80	22.40
Mean		28.56	13.43	2.12	35.00	2.12	2.12		1310.26	25.81
SD		5.25	2.09	0.06	0.00	0.06	0.06		241.07	4.82
%CV		18.40	15.58	2.86	0.00	2.86	2.86		18.40	18.68
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20	20 ug/mL	8.47	11.88	0.71	35.00	0.71	0.71	0.35	388.65	18.44
b20	20 ug/mL	9.99	14.09	0.71	35.00	0.71	0.71	0.35	458.40	21.93
20*	20 ug/mL									
Mean		9.23	12.99	0.71	35.00	0.71	0.71		423.52	20.19
SD		1.07	1.56	0.00	0.00	0.00	0.00		49.32	2.47
%CV		11.64	12.03	0.39	0.00	0.39	0.39		11.64	12.21
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
aB	0 ug/mL (a)	2.60	97.90	0.03	35.00	0.03	0.03	2.00		20.88
bB	0 ug/mL (a)	2.32	98.32	0.02	35.00	0.02	0.02	2.00		18.63
Mean		2.46	98.11	0.03	35.00	0.03	0.03			19.75
SD		0.20	0.30	0.00	0.00	0.00	0.00			1.59
%CV		8.05	0.30	8.35	0.00	8.35	8.35			8.05
n		2.00	2.00	2.00	2.00	2.00	2.00			2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	2.12	0.03	2.10
20.00	0.71	0.03	0.69
0.00	0.03	0.03	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0603
Std Err of Y Est	0.1238
R Squared	0.9933
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0423
Std Err of Coef.	0.0035

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc (µg/mL) Lyse	Conc (mM) RBC	Conc (mM) RBC
R371T0	0.00	7.55	123.36	0.06	35.00	0.06	0.06	2.00	-	2.87	0.0065	0.083
R371T0.08	0.08	5.37	91.17	0.06	35.00	0.06	0.06	2.00	-	2.82	0.0064	0.082
R371T0.25	0.25	7.53	122.75	0.06	35.00	0.06	0.06	2.00	-	2.87	0.0065	0.083
R371T1	1.00	4.80	59.81	0.08	35.00	0.08	0.08	1.00	-	3.32	0.0075	0.096
Not detected (SD mg/kg sd)												
R371T1.2	1.20	3.63	60.50	0.07	35.00	0.07	0.07	1.00	-	3.12	0.0070	0.091
R371T1.5	1.50	3.73	65.60	0.06	35.00	0.06	0.06	1.00	-	2.77	0.0062	0.080
R371T2	2.00	3.64	53.20	0.07	35.00	0.07	0.07	1.00	-	3.04	0.0069	0.088
R371T3	3.00	5.10	73.81	0.07	35.00	0.07	0.07	1.00	-	3.06	0.0069	0.089
R371T4	4.00	5.26	66.79	0.08	35.00	0.08	0.08	1.00	-	3.28	0.0074	0.095
R371T5	5.00	4.36	69.73	0.06	35.00	0.06	0.06	1.00	-	2.90	0.0065	0.084
R371T6	6.00	3.63	57.98	0.06	35.00	0.06	0.06	1.00	-	2.90	0.0065	0.084
Mean		40.42	75.88	0.07	35.00	0.07	0.07	1.27	0.00	3.00	0.01	0.09
SD		40.44	25.79	0.01	0.00	0.01	0.01	0.47	0.00	0.19	0.00	0.01
%CV		100.03	33.99	11.79	0.00	11.79	11.79	36.70	ERR	6.19	6.19	6.19
n		22.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

GDP (5 ng)

Comments: RBC Lyseate from R 338 was used for QC Samples. New calibration solution was prepared on May 31, 2011.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 15/04/2014

Checked by: Date:

Approved by: Pallen Yeung Date: 16/04/2014

Title: Measurement of RBC Concentrations of GMP in Rat 371 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 10/04/2014

Sample/standard ID	Standard Concentrations (µg/mL)	Peak Ht. # (nm)	Peak Ht. L.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
GMP 3 ng										
a50	50 ug/mL	off scale	14.91	0.00	35.00	0.00		0.35	ERR	ERR
b50	50 ug/mL	125.98	11.95	10.54	35.00	10.54	10.54	0.35	ERR	ERR
Mean		62.99	13.43	5.27	35.00	5.27	10.54		ERR	ERR
SD		89.08	12.69	7.45	0.00	7.45	ERR		ERR	ERR
%CV		141.42	13.06	141.42	0.00	141.42	ERR		ERR	ERR
n		2.00	13.06	2.00	2.00	2.00	1.00		2.00	2.00
a20										
b20	20 ug/mL	off scale	14.09	0.00	35.00	0.00		0.35	ERR	ERR
20*	20 ug/mL							0.35	ERR	ERR
Mean		55.95	12.99	4.71	35.00	4.71	9.42		ERR	ERR
SD		78.13	1.56	6.66	0.00	6.66	ERR		ERR	ERR
%CV		141.42	12.03	141.42	0.00	141.42	ERR		ERR	ERR
n		2.00	2.00	2.00	2.00	2.00	1.00		2.00	2.00
a8										
b8	0 ug/mL (a)	Off scale	97.90	0.00	35.00	0.00	0.00	2.00	0.00	0.00
	0 ug/mL (a)	Off scale	98.32	0.00	35.00	0.00	0.00	2.00	0.00	0.00
Mean		0.00	98.11	0.00	35.00	0.00	0.00		0.00	0.00
SD		0.00	0.30	0.00	0.00	0.00	0.00		0.00	0.00
%CV		ERR	0.30	ERR	0.00	ERR	ERR		ERR	ERR
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRB
50.00	10.54	0.00	10.54
20.00	9.42	0.00	9.42
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	3.4165
R Squared	0.6520
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.2467
Std Err of Coef.	0.0634

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. L.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R371T0		0.00 off scale	123.36	0.00	35.00	0.00	0.00	2.00	-	0.00	0.0000
R371T08		0.08 off scale	61.17	0.00	35.00	0.00	0.00	2.00	-	0.00	0.0000
R371T025		0.25 off scale	122.75	0.00	35.00	0.00	0.00	2.00	-	0.00	0.0000
R371T1		1.00 off scale	59.81	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000
Not Reported (20 mg/kg sc)											
R371T1.2	1.20 off scale	50.50	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	
R371T1.5	1.50 off scale	65.60	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	
R371T2	2.00 off scale	53.20	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	
R371T3	3.00 off scale	73.81	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	
R371T4	4.00 off scale	66.79	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	
R371T5	5.00 off scale	69.73	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	
R371T6	6.00 off scale	57.98	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	
Mean		0.00	75.88	0.00	35.00	0.00	0.00	1.27	0.00	0.00	0.00
SD		0.00	25.79	0.00	0.00	0.00	0.00	0.47	0.00	0.00	0.00
%CV		ERR	33.99	ERR	0.00	ERR	ERR	36.70	ERR	ERR	ERR
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

GMP (5 ng)

Comments: RBC Lysate from Rat 338 was used for QC Samples. New calibration solution was prepared on May 31, 2011.
 *Repeated injections of "a" or "b" at 0.5-1ul injection volume

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 15/04/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 16/04/2014

Plasma Concentrations of Adenosine in Rat 371
Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
Experiment Date: 23/01/2015 - 29/01/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Adenosine 5 ng									
a2.5	2.5ug/ml (a)	27.43	14.89	27.08	0.55	0.55	10	162.85	65.14
b2.5	2.5ug/ml (b)		14.42	38.13	0.38	0.38	10	157.71	63.08
Mean			14.66	32.61	0.46	0.46		160.28	64.11
SD			0.33	7.81	0.12	0.12		3.63	1.45
%CV			2.27	23.96	26.16	26.16		2.27	2.27
N			2.00	2.00	2.00	2.00		2.00	2.00
a0.5	0.5ug/ml (a)		7.16	72.90	0.10	0.10	30.00	26.10	52.21
b0.5	0.5ug/ml (b)		4.36	74.96	0.06	0.06	30.00	15.90	31.79
	0.5 ug/ml		4.29	70.43	0.06	0.06	30.00	15.64	31.28
	0.5 ug/ml		7.38	74.50	0.10	0.10	30.00	26.90	53.81
	0.5 ug/ml		5.87	74.51	0.08	0.08	30.00	21.40	42.80
Mean			5.81	73.46	0.08	0.08		21.19	42.38
SD			1.48	1.87	0.02	0.02		5.38	10.76
%CV			25.38	2.54	24.77	24.77		25.38	25.38
N			5.00	5.00	5.00	5.00		5.00	5.00
BLANKS:									
aB	0ug/mL (a)		N/D	8.40	0.00	0.00	2	0.00	
bB	0 ug/mL (b)		N/D	9.23	0.00	0.00	2	0.00	
Mean			0.00	8.82	0.00	0.00		0.00	
SD			0.00	0.59	0.00	0.00		0.00	
%CV			ERR	6.66	ERR	ERR		ERR	
N			2.00	2.00	2.00	2.00		2.00	
Adenosine 5 ng 26/01/2015			28.18	10.14			5.00		
Adenosine 5 ng 28/01/2015			30.32	10.70			5.00		
Adenosine 5 ng 29/01/2015			29.67	9.54			5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.46	0.00	0.46
0.50	0.08	0.00	0.08
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0066
Std Err of Y Est	0.0106
R Squared	0.9991
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.1876
Std Err of Coef.	0.0057

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	Conc.(µM)	Conc (µM) Corrected for dilution
T0 R371	0.00	ND	95.05	0.00	0.00	35	-	0.03	0.13	0.17
T0.08 R371	0.08	2.96	88.75	0.03	0.03	35	-	0.21	0.80	1.06
T0.25 R371	0.25	2.24	93.30	0.02	0.02	35	-	0.16	0.61	0.81
T1 R371	1.00	2.34	96.17	0.02	0.02	35	-	0.16	0.62	0.82
Adenosine (30 mg/kg)										
T1.2 R371	1.20	5.54	97.23	0.06	0.06	35	-	0.34	1.27	1.69
T1.5 R371	1.50	7.42	104.67	0.07	0.07	35	-	0.41	1.55	2.06
T2 R371	2.00	5.40	111.88	0.05	0.05	35	-	0.29	1.09	1.46
T3 R371	3.00	6.50	116.23	0.06	0.06	35	-	0.33	1.25	1.66
T4 R371	4.00	10.10	109.51	0.09	0.09	40	-	0.53	1.97	2.63
T5 R371	5.00	4.75	100.09	0.05	0.05	35	-	0.29	1.08	1.44
T6 R371	6.00	8.83	98.81	0.09	0.09	35	-	0.51	1.91	2.55
Mean		5.10	101.06	0.05	0.05			0.30	1.12	1.49
SD		3.05	8.51	0.03	0.03			0.15	0.56	0.75
%CV		59.83	8.42	57.21	57.21			50.50	50.50	50.50
n		11.00	11.00	11.00	11.00			11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
The Spiking solutions were made on: 22/02/2006
***Repeat injections of QC a or b**

Submitted by: Shyam Sundar Date: 02/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 09/02/2015

Plasma Concentrations of Inosine in Rat 371
 Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
 Experiment Date: 23/01/2015 - 29/01/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Inosine 5 ng		41.05						
a2.5	2.5ug/ml (a)	27.51	27.08	1.02	1.02	10	201.05	80.42
b2.5	2.5ug/ml (b)	25.87	38.13	0.68	0.68	10	189.06	75.62
Mean		26.69	32.61	0.85	0.85		195.05	78.02
SD		1.16	7.81	0.24	0.24		8.47	3.39
%CV		4.34	23.96	28.16	28.16		4.34	4.34
N		2.00	2.00	2.00	2.00		2.00	2.00
a0.5	0.5ug/ml (a)	3.85	72.90	0.05	0.07	30.00	9.38	18.76
b0.5	0.5ug/ml (b)	4.79	74.96	0.06	0.06	30.00	11.67	23.34
	0.5ug/ml	4.41	70.43	0.06	0.07	30.00	10.74	21.49
	0.5ug/ml	3.20	74.50	0.04	0.07	30.00	7.80	15.59
	0.5ug/ml	3.24	74.51	0.04	0.07	30.00	7.89	15.79
Mean		3.90	73.46	0.05	0.07		9.50	18.99
SD		0.70	1.87	0.01	0.00		1.71	3.43
%CV		18.05	2.54	18.88	5.15		18.05	18.05
N		5.00	5.00	5.00	5.00		5.00	5.00
BLANKS:								
aB	0ug/mL (a)	ND	8.40	0.00	0.00	2	0.00	
bB	0 ug/mL (b)	ND	9.23	0.00	0.00	2	0.00	
Mean		0.00	8.82	0.00	0.00		0.00	
SD		0.00	0.59	0.00	0.00		0.00	
%CV		ERR	6.66	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00		2.00	
Inosine 5ng 26/01/15		41.91	10.14			5.00		
Inosine 5ng 28/01/15		40.98	10.70			5.00		
Inosine 5ng 29/01/15		40.62	9.54			5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.85	0.00	0.85
0.50	0.07	0.00	0.07
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0477
Std Err of Y Est	0.0772
R Squared	0.9865
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3532
Std Err of Coef.	0.0413

Sample ID	Time post-dos	Peak #	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	Conc.(µM)	Conc (µM) Corrected for dilution
T0 R371	0.00	6.05	95.05	0.06	0.06	35	-	0.32	1.18	1.57
T0.08 R371	0.08	5.24	88.75	0.06	0.06	35	-	0.30	1.13	1.50
T0.25 R371	0.25	6.55	93.30	0.07	0.07	35	-	0.33	1.24	1.66
T1 R371	1.00	6.57	96.17	0.07	0.07	35	-	0.33	1.22	1.63
Propofol (30 mg/kg)										
T1.2 R371	1.20	5.85	97.23	0.06	0.06	35	-	0.31	1.14	1.52
T1.5 R371	1.50	4.82	104.67	0.05	0.05	35	-	0.27	0.99	1.32
T2 R371	2.00	8.66	111.88	0.08	0.08	35	-	0.35	1.32	1.76
T3 R371	3.00	18.51	116.23	0.16	0.16	35	-	0.59	2.18	2.91
T4 R371	4.00	20.39	109.51	0.19	0.19	35	-	0.66	2.47	3.29
T5 R371	5.00	24.70	100.09	0.25	0.25	35	-	0.83	3.11	4.14
T6 R371	6.00	32.82	98.81	0.33	0.33	35	-	1.08	4.01	5.35
Mean		12.74	101.06	0.12	0.12			0.49	1.82	2.42
SD		9.71	8.51	0.09	0.09			0.27	1.00	1.33
%CV		76.18	8.42	76.08	76.08			55.02	55.02	55.02
n		11.00	11.00	11.00	11.00			11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
 The Spiking solutions were made on: 22/02/2006
 *Repeat injections of QC a or b

Submitted by: Shyam Sundar Date: 26/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 26/02/2015

Plasma Concentrations of Hypoxanthine in Rat 371
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 23/01/2015 - 29/01/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Hypoxanthine 5 ng								
a25	25ug/ml (a)	105.46	5.97	17.66	17.66	2	1588.18	63.45
b25	25ug/ml (b)	113.50	8.23	13.79	13.79	2	1707.11	68.28
Mean		109.48	7.10	15.73	15.73		1646.65	65.87
SD		5.69	1.60	2.74	2.74		85.51	3.42
%CV		5.19	22.51	17.42	17.42		5.19	5.19
N		2.00	2.00	2.00	2.00		2.00	2.00
a5								
a5	5ug/ml (a)	13.13	6.37	2.06	2.06	2	197.48	39.50
b5	5ug/ml (b)	14.06	8.70	1.62	1.62	2	211.47	42.29
	5ug/ml	14.04	8.15	1.72	1.72	2	211.17	42.23
	5ug/ml	14.86	8.39	1.77	1.77	2	223.50	44.70
	5ug/ml	12.95	7.21	1.80	1.80	2	194.78	38.96
Mean		13.81	7.76	1.79	1.79		207.68	41.54
SD		0.78	0.96	0.16	0.16		11.70	2.34
%CV		5.63	12.33	9.19	9.19		5.63	5.63
N		5.00	5.00	5.00	5.00		5.00	5.00
BLANKS:								
aB	0ug/ml (a)	ND	8.40	0.00	0.55	2	0.00	
bB	0ug/ml (b)	ND	9.23	0.00	0.40	2	0.00	
Mean		0.00	8.82	0.00	0.47		0.00	
SD		0.00	0.59	0.00	0.11		0.00	
%CV		ERR	6.66	ERR	22.61		ERR	
N		2.00	2.00	2.00	2.00		2.00	
Hypoxanthine 5 ng 26/01/15		103.80	10.14				5.00	
Hypoxanthine 5 ng 28/01/15		105.22	10.70				5.00	
Hypoxanthine 5 ng 29/01/15		103.74	9.54				5.00	

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	15.73	0.47	15.26
5.00	1.79	0.47	1.32
0.00	0.47	0.47	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.8232
Std Err of Y Est	1.3337
R Squared	0.9876
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.6350
Std Err of Coef.	0.0713

Sample ID	Time post-dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	ic. (µg/mL):onc. (µM)	Conc. (µM)	Corrected for dilution
T0 R371	0.00	14.30	18.36	0.74	0.74	5	-	2.46	18.07	24.10
T0.08 R371	0.08	14.07	19.01	0.74	0.74	5	-	2.46	18.09	24.12
T0.25 R371	0.25	13.00	16.48	0.79	0.79	5	-	2.54	18.65	24.87
T1 R371	1.00	12.95	18.30	0.71	0.71	5	-	2.41	17.71	23.62
Isoproterenol (30 mg/kg)										
T1.2 R371	1.20	13.86	18.93	0.73	0.73	5	-	2.45	18.00	24.00
T1.5 R371	1.50	13.45	17.79	0.76	0.76	5	-	2.49	18.27	24.36
T2 R371	2.00	16.46	18.46	0.89	0.89	5	-	2.70	19.84	26.46
T3 R371	3.00	16.36	20.75	0.79	0.79	5	-	2.54	18.65	24.86
T4 R371	4.00	16.83	19.91	0.85	0.85	5	-	2.63	19.31	25.74
T5 R371	5.00	13.98	16.43	0.85	0.85	5	-	2.64	19.37	25.83
T6 R371	6.00	14.41	14.84	0.97	0.97	5	-	2.83	20.76	27.68
Mean		14.31	18.64	0.77	0.77			2.51	18.41	24.55
SD		1.38	1.24	0.06	0.06			0.09	0.66	0.88
%CV		9.67	6.64	7.44	7.44			3.59	3.59	3.59
n		8.00	8.00	8.00	8.00			8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 02/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/02/2015

Plasma Concentrations of Xanthine in Rat 371
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 23/01/2015 -29/01/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Reco (%)	Recovery (%)
Xanthine 5 ng		36.30					5			
a25	25ug/ml (a)	43.03	5.97	7.21	7.21	2	1778.10	71.12		
b25	25ug/ml (b)	45.28	8.23	5.50	5.50	2	1871.07	74.84		
Mean		44.16	7.10	6.35	6.35		1824.59	72.98		
SD		1.59	1.60	1.21	1.21		65.74	2.63		
%CV		3.60	22.51	18.98	18.98		3.60	3.60		
N		2.00	2.00	2.00	2.00		2.00	2.00		
a5	5ug/ml (a)	4.54	6.37	0.71	0.71	2	187.60	37.52		
b5	5ug/ml (b)	6.48	8.70	0.74	0.74	2	267.77	53.55		
	5ug/ml	4.35	8.15	0.53	0.53	2	179.75	35.95		
	5ug/ml	4.09	8.39	0.49	0.49	2	169.01	33.80		
	5ug/ml	3.98	7.21	0.55	0.55	2	164.46	32.89		
Mean		4.69	7.76	0.61	0.61		193.72	38.74		
SD		1.03	0.96	0.11	0.11		42.37	8.47		
%CV		21.87	12.33	18.96	18.96		21.87	21.87		
N		5.00	5.00	5.00	5.00		5.00	5.00		
BLANKS:										
a5	0ug/ml (a)	ND	8.40	0.00	0.00	2	0.00			
b5	0ug/ml (b)	ND	9.23	0.00	0.00	2	0.00			
Mean		0.00	8.82	0.00	0.00		0.00			
SD		0.00	0.59	0.00	0.00		0.00			
%CV		ERR	6.66	ERR	ERR		ERR			
N		2.00	2.00	2.00	2.00		2.00			

Xanthine 5 ng 26/01/15	36.86	10.14		5.00
Xanthine 5 ng 28/01/15	37.55	10.70		5.00
Xanthine 5 ng 29/01/15	37.39	9.54		5.00

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	6.35	0.00	6.35
5.00	0.61	0.00	0.61
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.4610
R Squared	0.9827
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.2491
Std Err of Coef.	0.0181

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c. (ug/mL)	Conc. (µM)	Conc. (µM) Corrected for dilution
T0 R371	0.00	INT	19.36	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T0.08 R371	0.08	INT	19.01	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T0.25 R371	0.25	INT	16.48	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T1 R371	1.00	INT	18.30	0.00	0.00	0.00	5	-	0.00	0.00	0.00
Spiking control (30 mg/kg)											
T1.2 R371	1.20	INT	18.93	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T1.5 R371	1.50	INT	17.79	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T2 R371	2.00	INT	18.46	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T3 R371	3.00	INT	20.75	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T4 R371	4.00	INT	19.91	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T5 R371	5.00	INT	16.43	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T6 R371	6.00	INT	14.84	0.00	0.00	0.00	5	-	0.00	0.00	0.00
Mean		0.00	18.64	0.00	0.00	0.00			0.00	0.00	0.00
SD		0.00	1.24	0.00	0.00	0.00			0.00	0.00	0.00
%CV		ERR	6.64	ERR	ERR	ERR			ERR	ERR	ERR
n		8.00	8.00	8.00	8.00	8.00			8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 02/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/02/2015

Plasma Concentrations of Uric Acid in Rat 371
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 23/01/2015 - 29/01/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Uric Acid 5 ng		28.69					5		
a25	25ug/ml (a)	50.65	5.97	8.48	8.48		2	2648.14	99.29
b25	25ug/ml (b)	51.89	8.23	6.30	6.30		2	2712.97	101.88
Mean		51.27	7.10	7.39	7.39			2680.55	100.58
SD		0.88	1.60	1.54	1.54			45.84	1.83
%CV		1.71	22.51	20.84	20.84			1.71	1.82
N		2.00	2.00	2.00	2.00			2.00	2.00
a5	5ug/ml (a)	9.59	6.37	1.51	1.51		2	501.39	67.08
b5	5ug/ml (b)	9.68	8.70	1.11	1.11		2	506.10	68.02
	5ug/ml	10.13	8.15	1.24	1.24		2	529.63	72.73
	5ug/ml	10.66	8.39	1.27	1.27		2	557.34	78.27
	5ug/ml	8.55	7.21	1.19	1.19		2	447.02	56.20
Mean		9.72	7.76	1.26	1.26			508.30	68.46
SD		0.78	0.96	0.15	0.15			40.83	8.17
%CV		8.03	12.33	11.73	11.73			8.03	11.93
N		5.00	5.00	5.00	5.00			5.00	5.00
BLANKS:									
aB	0ug/ml (a)	2.69	8.40	0.32	0.32		2	140.64	
bB	0ug/ml (b)	3.66	9.23	0.40	0.40		2	191.36	
Mean		3.18	8.82	0.36	0.36			166.00	
SD		0.69	0.59	0.05	0.05			35.86	
%CV		21.60	6.66	15.05	15.05			21.60	
N		2.00	2.00	2.00	2.00			2.00	
Uric Acid 5 ng 26/01/15		31.14	9.54					5.00	
Uric Acid 5 ng 28/01/15		31.60	10.70					5.00	
Uric Acid 5 ng 29/01/15		28.26	10.14					5.00	

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	7.39	0.36	7.04
5.00	1.26	0.36	0.91
0.00	0.36	0.36	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.2391
Std Err of Y Est	0.3874
R Squared	0.9949
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.2886
Std Err of Coef.	0.0207

Sample ID	Time post-dose	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	ic. (µg/mL); conc. (µM)	Conc. (µM) Corrected for dilution
T0 R371	0.00	23.08	19.36	1.19	1.19	5	-	4.96	29.50	39.33
T0.08 R371	0.08	18.76	19.01	0.88	0.88	5	-	3.88	23.10	30.80
T0.25 R371	0.25	21.66	18.48	1.31	1.31	5	-	5.38	32.02	42.69
T1 R371	1.00	18.13	18.30	0.99	0.99	5	-	4.26	25.35	33.80
Isoproterenol (30 mg/kg)										
T1.2 R371	1.20	17.40	18.93	0.92	0.92	5	-	4.01	29.49	39.32
T1.5 R371	1.50	42.92	17.79	2.41	2.41	5	-	9.19	67.51	90.01
T2 R371	2.00	91.32	18.46	4.95	4.95	5	-	17.97	106.89	142.52
T3 R371	3.00	34.32	20.75	1.65	1.65	5	-	6.56	39.02	52.03
T4 R371	4.00	48.29	19.91	2.43	2.43	5	-	9.23	54.92	73.23
T5 R371	5.00	67.16	16.43	4.09	4.09	5	-	14.99	89.18	118.91
T6 R371	6.00	25.23	14.84	1.70	1.70	5	-	6.72	39.97	53.29
Mean		33.20	18.64	1.79	1.79			7.03	44.11	58.81
SD		25.23	1.24	1.37	1.37			4.75	28.99	38.65
%CV		76.00	6.64	76.70	76.70			67.66	65.72	65.72
n		8.00	8.00	8.00	8.00			8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No. 181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 02/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/02/2015

Plasma Concentrations of Guanosine in Rat 371
Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
Experiment Date: 23/1/2015 - 29/01/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Guanosine 5 ng								
a2.5	2.5ug/ml (a)	24.48	27.08	0.90	0.90	10	142.82	57.13
b2.5	2.5ug/ml (b)	22.91	38.13	0.60	0.60	10	133.66	53.47
Mean		23.70	32.61	0.75	0.75		138.24	55.30
SD		1.11	7.81	0.21	0.21		6.48	2.59
%CV		4.69	23.96	28.49	28.49		4.69	4.69
N		2.00	2.00	2.00	2.00		2.00	2.00
a0.5								
a0.5	0.5ug/ml (a)	3.65	72.90	0.05	0.05	30.00	7.10	14.20
b0.5	0.5ug/ml (b)	3.87	74.96	0.05	0.05	30.00	7.53	15.05
	0.5ug/ml	3.85	70.43	0.05	0.05	30.00	7.49	14.97
	0.5ug/ml	3.23	74.50	0.04	0.04	30.00	6.28	12.96
	0.5ug/ml	4.50	74.51	0.06	0.06	30.00	8.75	17.50
Mean		3.82	73.46	0.05	0.05		7.43	14.86
SD		0.46	1.87	0.01	0.01		0.89	1.79
%CV		12.02	2.54	12.01	12.01		12.02	12.02
N		5.00	5.00	5.00	5.00		5.00	5.00
BLANKS:								
aB	0ug/mL (a)	ND	8.40	0.00	0.00	2	0.00	
bB	0 ug/mL (b)	ND	9.23	0.00	0.00	2	0.00	
Mean		0.00	8.82	0.00	0.00		0.00	
SD		0.00	0.59	0.00	0.00		0.00	
%CV		ERR	6.66	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00		2.00	
Guanosine 5 ng 26/01/15		52.22	10.14			5.00		
Guanosine 5 ng 28/01/15		54.11	10.70			5.00		
Guanosine 5 ng 29/01/15		51.63	9.54			5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.75	0.00	0.75
0.50	0.05	0.00	0.05
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0469
Std Err of Y Est	0.0760
R Squared	0.9837
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3150
Std Err of Coef.	0.0406

Sample ID	Time post-dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	ic.(µg/mL)	conc.(µM)	Conc.(µM) Corrected for dilution
T0 371	0.00	ND	95.05	0.000	0.000	35	-	0.15	0.53	0.70
T0.08 371	0.08	2.72	98.75	0.031	0.031	35	-	0.25	0.87	1.16
T0.25 371	0.25	1.99	93.30	0.021	0.021	35	-	0.22	0.76	1.02
T1371	1.00	2.61	96.17	0.027	0.027	35	-	0.23	0.83	1.11
Isoproterenol (30 mg/kg)										
T1.2 371	1.20	2.59	97.23	0.027	0.027	35	-	0.23	0.87	1.16
T1.5 371	1.50	2.81	104.67	0.027	0.027	35	-	0.23	0.88	1.17
T2 371	2.00	1.98	111.88	0.018	0.018	35	-	0.21	0.72	0.97
T3 371	3.00	2.97	116.23	0.026	0.026	35	-	0.23	0.81	1.08
T4 371	4.00	2.85	109.51	0.026	0.026	35	-	0.23	0.82	1.09
T5 371	5.00	2.88	100.09	0.029	0.029	35	-	0.24	0.85	1.13
T6 371	6.00	2.57	98.81	0.026	0.026	35	-	0.23	0.82	1.09
Mean		2.36	101.06	0.02	0.02			0.22	0.80	1.06
SD		0.85	8.51	0.01	0.01			0.03	0.10	0.13
%CV		36.00	8.42	36.38	36.38			12.09	12.69	12.69
n		11.00	11.00	11.00	11.00			11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
The Spiking solutions were made on: 22/02/2006
*Repeat injections of QC a or b

Submitted by: Shyam Sundar Date: 02/02/2015
Checked by: Date:
Approved by: Polen Yeung Date: 10/02/2015

Title: Measurement of Plasma Concentrations of Diprydamole in Rat 371

According to SOP No: SOP/STD/2008-001-1 (Plasma with no Stopping Solution) Experiment Date:11/09/2014- 17/09/2014

Abs.amt ng	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Diprydamole (1ng)		90.77				1		
a1000	1 ug/mL(a)	115.14	5.39	21.36	21.36	5	50.74	101.48
b1000	1ug/mL(b)	121.91	6.74	18.09	18.09	5	53.72	107.45
1000*	1ug/mL(c)	110.34	5.12	21.55	21.55	5	48.62	97.25
1000*	1ug/mL(c)	103.16	3.68	28.03	28.03	5	45.46	90.92
Mean		112.64	5.23	22.26	22.26		49.64	99.27
SD		7.90	1.25	4.17	4.17		3.48	6.96
%CV		7.02	23.97	18.71	18.71		7.02	7.02
n		4.00	4.00	4.00	4.00		1.00	1.00
a100	0.1 ug/mL (a)	25.74	7.73	3.33	3.33	10	5.67	113.43
b100	0.1ug/mL (b)	29.91	10.56	2.83	2.83	10	6.59	131.81
Mean		27.63	9.15	3.08	3.08		6.13	122.62
SD		2.95	2.00	0.35	0.35		0.65	12.99
%CV		10.60	21.88	11.42	11.42		10.60	10.60
n		2.00	2.00	2.00	2.00		2.00	2.00
aB	0 ug/mL (a)	5.01	6.61	0.76	0.76	10	1.10	22.08
bB	0 ug/mL (b)	6.67	12.01	0.56	0.56	10	1.47	29.39
Mean		5.84	9.31	0.66	0.66		1.29	25.74
SD		1.17	3.82	0.14	0.14		0.26	5.17
%CV		20.10	41.01	21.81	21.81		20.10	20.10
n		2.00	2.00	2.00	2.00		2.00	2.00

Plasma Conc. (ug/mL)	Peak Ht.Ratio (PHR)	Blank (PHRb)	PHRV-PHRb
0.00	0.66	0.66	0.60
0.10	3.08	0.66	2.42
1.00	22.26	0.66	21.60

Regression Output Begins Here:

Regression Output:	
Constant	0.1307
Std Err of Y Est	0.1959
R Squared	0.9999
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	21.4853
Std Err of Coef.	0.2516

Sample ID	Time Post-dose (h)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)*	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc.(ug/mL)
R371T0	0.00	10.02	26.32	0.38	0.38	10	-	0.12
R371T0.08	0.08	42.01	26.52	1.58	1.58	10	-	0.68
R371T0.25	0.25	27.67	12.13	2.28	2.28	10	-	1.00
R371T1	1.00	21.23	9.55	2.22	2.22	10	-	0.97
Standard Error (30 mg/kg sc)								
R371T1.2	1.20	21.99	3.17	6.94	6.94	10	-	3.16
R371T1.5	1.50	19.80	34.50	0.57	0.57	10	-	0.21
R371T2	2.00	20.59	35.71	0.58	0.58	10	-	0.21
R371T3	3.00	12.70	18.04	0.70	0.70	10	-	0.27
R371T4	4.00	5.20	12.16	0.43	0.43	10	-	0.14
R371T5	5.00	19.12	23.22	0.82	0.82	10	-	0.32
R371T6	6.00	5.69	15.40	0.37	0.37	10	-	0.11
Mean		18.73	19.70	1.53	1.53			0.65
SD		10.53	10.44	1.93	1.93			0.90
%CV		56.22	52.99	125.63	125.63			137.33
n		11.00	11.00	11.00	11.00			11.00

Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 Corr. PHR = (PHR - RGB PHR)

Diprydamole (1ng)(12/09/2014)	96.79	9.53	1.00
Diprydamole(1ng)(16/09/2014)	81.87	8.54	1.00
Diprydamole(1ng)(17/09/2014)	86.94	9.28	1.00

Comments: Plasma from Rat 163 was used for extraction QC's.
***A repeat injection of a or b**

Submitted by: Shyam Sundar Date: 18/09/2014

Checked by: Pollen Yeung Date: 29/09/2014

Approved by: Date:

APPENDIX 2: Rat 372

Title: Measurement of RBC Concentrations of ATP in Rat 372 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
Experiment Date 23/04/2014

Sample/standard ID	Standard Concentra (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recov %	Recovery
ATP 4 ng										
a250	250 ug/mL	47.34	8.37	5.66	35.00	5.66	5.66	0.35	14804.26	44.71
b250	250 ug/mL	45.44	7.72	5.89	35.00	5.89	5.89	0.35	14210.09	42.33
Mean		46.39	8.05	5.77	35.00	5.77	5.77		14507.18	43.52
SD		1.34	0.46	0.16	0.00	0.16	0.16		420.14	1.68
%CV		2.90	5.71	2.82	0.00	2.82	2.82		2.90	3.86
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a100	100 ug/mL	28.23	8.75	3.23	35.00	3.23	3.23	0.35	8828.14	52.01
b100	100 ug/mL	28.37	11.31	2.51	35.00	2.51	2.51	0.35	8871.93	52.45
100'	100 ug/mL	29.80	8.99	3.31	35.00	3.31	3.31	0.35	9319.12	56.92
Mean		28.80	9.68	3.02	35.00	3.02	3.02		8850.04	52.23
SD		0.87	1.41	0.44	0.00	0.44	0.44		30.96	0.31
%CV		3.02	14.60	14.66	0.00	14.66	14.66		0.35	0.59
n		3.00	3.00	3.00	3.00	3.00	3.00		2.00	2.00
aB	0 ug/mL (a)	61.96	80.02	0.77	35.00	0.77	0.77	2.00	3368.01	
bB	0 ug/mL (a)	71.20	96.44	0.74	35.00	0.74	0.74	2.00	3896.52	
Mean		66.28	88.23	0.75	35.00	0.75	0.75		3627.26	
SD		6.96	11.61	0.02	0.00	0.02	0.02		380.78	
%CV		10.50	13.16	2.68	0.00	2.68	2.68		10.50	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
250.00	5.77	0.75	5.02
100.00	3.02	0.75	2.26
0.00	0.75	0.75	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.1013
Std Err of Y Est	0.2081
R Squared	0.9966
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0199
Std Err of Coef.	0.0012

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R37270	0.00	102.89	58.30	1.76	35.00	1.76	1.76	1.50	-	83.43	0.1845
R37270.08	0.08	103.62	76.76	1.35	35.00	1.35	1.35	1.50	-	82.62	0.1235
R37270.25	0.25	103.26	55.38	1.86	35.00	1.86	1.86	1.50	-	88.44	0.1744
R37271	1.00	56.70	27.76	2.04	35.00	2.04	2.04	1.00	-	97.36	0.1820
Repeated (10 mg/kg sc)											
R37271.2	1.20							1.00	-		
R37271.5	1.50	86.50	39.66	2.18	35.00	2.18	2.18	1.00	-	104.31	0.2057
R37272	2.00	90.45	39.39	2.30	35.00	2.30	2.30	1.00	-	110.09	0.2170
R37273	3.00	53.28	28.75	1.85	35.00	1.85	1.85	1.00	-	87.87	0.1732
R37274	4.00	35.60	44.31	0.80	35.00	0.80	0.80	1.00	-	35.22	0.0694
R37275	5.00							1.00	-		
R37276	6.00							1.00	-		
Mean		79.04	46.29	1.77	35.00	1.77	1.77	1.14		83.67	0.16
SD		26.71	16.48	0.49	0.00	0.49	0.49	0.23		24.33	0.05
%CV		33.80	35.61	27.41	0.00	27.41	27.41	20.55		29.08	29.08
n		8.00	8.00	8.00	8.00	8.00	8.00	11.00		8.00	8.00

ATP (4 ng) 24/04/2014

20.04

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
*Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
Peak Ht. = peak height
Peak Ht. R. (or: PHR) = peak height ratio
I.S. = internal standard
Inj Vol = injection volume
ND = not detected or determined
NS = no sample
INT = interference
PCV = packed cell volume (haematocrit)
CorPHR = corrected peak height ratio
Hemolysis Degree:
-: no visible hemolysis
+: slight hemolysis
++: intermediate hemolysis
+++: serious hemolysis

Submitted by: Shyam Sundar K Date: 29/04/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 02/05/2014

Title: Measurement of RBC Concentrations of ADP in Rat 372 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)

Experiment Date: 23/04/2014

Sample/standard ID	Standard Co (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
ADP 4 ng		41.58						4.00		
a250	250 µg/mL	75.34	8.37	9.00	35.00	9.00	9.00	0.35	11389.27	43.10
b250	250 µg/mL	71.39	7.72	9.25	35.00	9.25	9.25	0.35	10792.14	40.72
Mean		73.37	8.05	9.12	35.00	9.12	9.12		11090.70	41.91
SD		2.79	0.46	0.17	0.00	0.17	0.17		422.23	1.69
%CV		3.81	5.71	1.91	0.00	1.91	1.91		3.81	4.03
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
100a	100 µg/mL	40.94	8.75	4.68	35.00	4.68	4.68	0.35	6188.96	55.76
100b	100 µg/mL	39.71	11.31	3.51	35.00	3.51	3.51	0.35	6003.02	53.90
100*	100 µg/mL	43.25	8.99	4.81	35.00	4.81	4.81	0.35	6538.17	59.25
Mean		41.30	9.68	4.33	35.00	4.33	4.33		6243.39	56.30
SD		1.80	1.41	0.72	0.00	0.72	0.72		271.69	2.72
%CV		4.35	14.60	16.51	0.00	16.51	16.51		4.35	4.83
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB	0 µg/mL (a)	21.20	80.02	0.26	35.00	0.26	0.26	2.00	560.85	
bB	0 µg/mL (a)	25.15	96.44	0.26	35.00	0.26	0.26	2.00	665.34	
Mean		23.18	88.23	0.26	35.00	0.26	0.26		613.10	
SD		2.79	11.51	0.00	0.00	0.00	0.00		73.89	
%CV		12.05	13.16	1.12	0.00	1.12	1.12		12.05	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
250.00	9.12	0.26	8.86
100.00	4.33	0.26	4.07
0.00	0.26	0.26	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.3454
R Squared	0.9939
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.0362
Std Err of Coef.	0.0013

Sample ID	Time post	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) Lysate	Conc.(mM) RBC
R37210	0.00	22.71	59.30	0.39	35.00	0.39	0.39	1.50	-	10.77	0.0252	0.324
R37210B	0.08	20.59	76.76	0.27	35.00	0.27	0.27	1.50	-	7.42	0.0174	0.223
R37210.25	0.25	20.09	55.38	0.36	35.00	0.36	0.36	1.50	-	10.03	0.0235	0.302
R37211	1.00	11.98	27.76	0.43	35.00	0.43	0.43	1.00	-	11.93	0.0279	0.359
Ibuprofenolol (30 mg/kg sc)												
R37211.2	1.20							1.00	-			
R37211.5	1.50	14.35	39.66	0.36	35.00	0.36	0.36	1.00	-	10.00	0.0234	0.301
R37212	2.00	13.70	39.39	0.35	35.00	0.35	0.35	1.00	-	9.62	0.0225	0.289
R37213	3.00	12.29	28.75	0.43	35.00	0.43	0.43	1.00	-	11.82	0.0277	0.356
R37214	4.00	8.48	44.31	0.19	35.00	0.19	0.19	1.00	-	5.28	0.0124	0.159
R37215	5.00							1.00	-			
R37216	6.00							1.00	-			
Mean		21.13	63.48	0.34	35.00	0.34	0.34	1.50		9.40	0.02	0.28
SD		1.39	11.59	0.06	0.00	0.06	0.06	0.00		1.76	0.00	0.05
%CV		6.58	18.26	18.73	0.00	18.73	18.73	0.00		18.73	18.73	18.73
n		3.00	3.00	3.00	3.00	3.00	3.00	3.00		3.00	2.00	3.00

ADP (4 ng) 24/04/2014

40.94

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.

*Repeated injections of a or b at 0.5-1ul injection volume.

PL = plasma; RBC = red blood cells

Peak Ht. = peak height

Peak Ht. R. (or: PHR) = peak height ratio

I.S. = internal standard

Inj Vol = injection volume

ND = not detected or determined

NS = no sample

INT = interference

PCV = packed cell volume (haematocrit)

CorPHR = corrected peak height ratio

Hemolysis Degree:

-: no visible hemolysis

+: slight hemolysis

++: intermediate hemolysis

+++: serious hemolysis

Submitted by: Shyam Sundar K

Date: 29/04/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date: 06/05/2014

Title: Measurement of RBC Concentrations of AMP in Rat 372 extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date 23/04/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount	Recovery %	Recovery
AMP 4 ng											
		62.86						4.00			
a50	50 ug/mL	39.97	8.37	4.78	35.00	4.78	4.78	0.35	3996.82		79.01
b50	50 ug/mL	38.32	7.72	4.96	35.00	4.96	4.96	0.35	3831.83		75.71
Mean		39.15	8.05	4.87	35.00	4.87	4.87		3914.32		77.36
SD		1.17	0.46	0.13	0.00	0.13	0.13		116.87		2.33
%CV		2.98	5.71	2.73	0.00	2.73	2.73		2.98		3.02
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00		2.00
a20	20 ug/mL	20.74	8.75	2.37	35.00	2.37	2.37	0.35	2073.91		101.37
b20	20 ug/mL	20.03	11.31	1.77	35.00	1.77	1.77	0.35	2002.91		97.82
20*	20 ug/mL	21.81	8.99	2.43	35.00	2.43	2.43	0.35	2190.90		106.72
Mean		20.86	9.68	2.19	35.00	2.19	2.19		2085.91		101.97
SD		0.90	1.41	0.36	0.00	0.36	0.36		89.60		4.48
%CV		4.30	14.60	16.59	0.00	16.59	16.59		4.30		4.39
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00		3.00
aB	0 ug/mL (a)	2.60	80.02	0.03	35.00	0.03	0.03	2.00			45.59
bB	0 ug/mL (a)	2.71	96.44	0.03	35.00	0.03	0.03	2.00			47.42
Mean		2.66	88.23	0.03	35.00	0.03	0.03				46.46
SD		0.08	11.81	0.00	0.00	0.00	0.00				1.36
%CV		2.93	13.16	10.25	0.00	10.25	10.25				2.93
n		2.00	2.00	2.00	2.00	2.00	2.00				2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	4.87	0.03	4.84
20.00	2.19	0.03	2.16
0.00	0.03	0.03	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.1465
R Squared	0.9963
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.0983
Std Err of Coef.	0.0027

Sample ID	Time post dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL)	Conc.(mM) Lysate	Conc.(mM) RBC
R37670	0.00	0.50	58.30	0.01	35.00	0.01	0.01	1.50	-	0.09	0.0003	0.003
R37670.08	0.08	0.46	76.76	0.01	35.00	0.01	0.01	1.50	-	0.06	0.0002	0.002
R37670.25	0.25	0.41	55.38	0.01	35.00	0.01	0.01	1.00	-	0.08	0.0002	0.003
R37671	1.00	0.83	27.76	0.03	35.00	0.03	0.03	1.00	-	0.30	0.0009	0.011
Saprofenenol (30 mg/kg sc)												
R37671.2	1.20											
R37671.5	1.50	0.37	39.66	0.01	35.00	0.01	0.01	1.00	-	0.09	0.0003	0.004
R37672	2.00	0.28	39.39	0.01	35.00	0.01	0.01	1.00	-	0.07	0.0002	0.002
R37673	3.00	2.56	28.75	0.09	35.00	0.09	0.09	1.00	-	0.91	0.0028	0.034
R37674	4.00	0.36	44.31	0.01	35.00	0.01	0.01	1.00	-	0.08	0.0002	0.003
R37675	5.00											
R37676	6.00											
Mean		0.72	46.29	0.02	35.00	0.02	0.02	1.13	0.00	0.21	0.00	0.01
SD		0.76	16.48	0.03	0.00	0.03	0.03	0.23	0.00	0.29	0.00	0.01
%CV		106.13	35.61	139.37	0.00	139.37	139.37	20.57	0.00	136.37	136.37	136.37
n		8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00

AMP (4 ng)24/04/2014

64.63

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of "a" or "b" at 0.5 - 1 ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar

Date: 29/04/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date: 06/05/2014

Title: Measurement of RBC Concentrations of GTP in Rat 372 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 23/04/2014

Sample/standard ID	Standard Concentra (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recove (%)	Recovery
GTP 4 ng										
a50	50 ug/mL	14.63	8.37	1.75	35.00	1.75	1.75	0.35	2351.32	42.87
b50	50 ug/mL	14.15	7.72	1.83	35.00	1.83	1.83	0.35	2274.17	41.32
Mean		14.39	8.05	1.79	35.00	1.79	1.79		2312.74	42.10
SD		0.34	0.06	0.00	0.06	0.06	0.06		54.55	1.09
%CV		2.36	0.00	3.36	0.00	3.36	3.36		2.36	2.59
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20	20 ug/mL	8.19	8.75	0.94	35.00	0.94	0.94	0.35	1310.20	55.41
b20	20 ug/mL	7.48	11.31	0.66	35.00	0.66	0.66	0.35	1263.78	49.79
20*	20 ug/mL	7.84	8.99	0.87	35.00	0.87	0.87	0.35	1260.04	52.60
Mean		7.84	9.68	0.82	35.00	0.82	0.80		1260.04	52.60
SD		0.35	1.44	0.14	0.00	0.14	0.05		55.25	2.81
%CV		4.46	14.60	17.39	0.00	17.39	5.00		4.46	5.35
n		3.00	3.00	3.00	3.00	3.00	2.00		3.00	3.00
a8	0 ug/mL (a)	6.84	80.02	0.09	35.00	0.09	0.09	2.00	192.38	
b8	0 ug/mL (a)	7.95	96.44	0.08	35.00	0.08	0.08	2.00	223.60	
Mean		7.40	88.23	0.08	35.00	0.08	0.08		207.99	
SD		0.78	11.61	0.00	0.00	0.00	0.00		22.08	
%CV		10.61	13.16	2.56	0.00	2.56	2.56		10.61	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	1.79	0.08	1.71
20.00	0.90	0.08	0.82
0.00	0.08	0.08	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0543
Std Err of Y Est	0.1115
R Squared	0.9915
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0338
Std Err of Coef.	0.0031

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R37210	0.00	18.35	58.30	0.31	35.00	0.31	0.31	1.50	-	7.71	0.0147
R37210.08	0.08	18.99	78.76	0.25	35.00	0.25	0.25	1.50	-	5.72	0.0109
R37210.25	0.25	19.32	65.38	0.35	35.00	0.35	0.35	1.50	-	8.72	0.0167
R37211	1.00	10.99	27.76	0.40	35.00	0.40	0.40	1.00	-	10.12	0.0193
Suprofenol (20 mg/kg sc)											
R37211.2	1.20							1.00	-		
R37211.5	1.50	15.99	39.66	0.40	35.00	0.40	0.40	1.00	-	10.33	0.0197
R37212	2.00	18.12	39.39	0.46	35.00	0.46	0.46	1.00	-	12.02	0.0230
R37213	3.00	10.98	28.75	0.38	35.00	0.38	0.38	1.00	-	9.70	0.0185
R37214	4.00	5.82	44.31	0.13	35.00	0.13	0.13	1.00	-	2.28	0.0044
R37215	5.00							1.00	-		
R37216	6.00							1.00	-		
Mean		14.82	46.29	0.34	35.00	0.34	0.34	1.14	0.00	8.33	0.02
SD		4.97	16.48	0.10	0.00	0.10	0.10	0.23	0.00	3.06	0.01
%CV		33.52	35.61	31.05	0.00	31.05	31.05	20.55	0.00	37.05	37.05
n		8.00	8.00	8.00	8.00	8.00	8.00	11.00	11.00	8.00	8.00

GTP (4 ng) 24/04/2014

33.75

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K

Date: 29/04/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date:06/05/2014

Title: Measurement of RBC Concentrations of GDP in Rat 372 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 23/04/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recove	% Recovery
GDP 4 ng										
		57.79						4.00		
a50	50 ug/mL	21.61	8.37	2.58	35.00	2.58	2.58	0.35	2350.48	45.85
b50	50 ug/mL	20.22	7.72	2.62	35.00	2.62	2.62	0.35	2199.29	42.83
Mean		20.92	8.05	2.60	35.00	2.60	2.60		2274.89	44.34
SD		0.98	0.46	0.03	0.00	0.03	0.03		106.81	2.14
%CV		4.70	5.71	1.02	0.00	1.02	1.02		4.70	4.82
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20										
	20 ug/mL	11.50	8.75	1.31	35.00	1.31	1.31	0.35	1250.83	59.64
b20	20 ug/mL	10.58	11.31	0.94	35.00	0.94	0.94	0.35	1150.77	54.64
20*	20 ug/mL	11.86	8.99	1.32	35.00	1.32	1.32	0.35	1289.99	61.60
Mean		11.31	9.68	1.19	35.00	1.19	1.19		1230.53	58.63
SD		0.66	1.41	0.22	0.00	0.22	0.22		71.80	3.59
%CV		5.83	14.60	18.51	0.00	18.51	18.51		5.83	6.12
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB										
	0 ug/mL (a)	3.01	80.02	0.04	35.00	0.04	0.04	2.00		57.29
bB	0 ug/mL (a)	3.08	96.44	0.03	35.00	0.03	0.03	2.00		58.63
Mean		3.05	88.23	0.03	35.00	0.03	0.03			57.96
SD		0.05	11.61	0.00	0.00	0.00	0.00			0.94
%CV		1.63	13.16	11.55	0.00	11.55	11.55			1.63
n		2.00	2.00	2.00	2.00	2.00	2.00			2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRB
50.00	2.60	0.03	2.57
20.00	1.19	0.03	1.15
0.00	0.03	0.03	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.0844
R Squared	0.9957
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.0022
Std Err of Coef.	0.0016

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(nM) RBC
R37270	0.00	4.77	58.30	0.08	35.00	0.08	0.08	1.50	-	1.57	0.0035
R37270.08	0.08	4.75	76.76	0.06	35.00	0.06	0.06	1.50	-	1.19	0.0027
R37270.25	0.25	4.27	55.38	0.08	35.00	0.08	0.08	1.50	-	1.48	0.0033
R37271	1.00	2.58	27.76	0.09	35.00	0.09	0.09	1.00	-	1.78	0.0040
Isoproterenol (30 µg/kg sc)											
R37271.2	1.20							1.00	-		
R37271.5	1.50	3.26	39.66	0.08	35.00	0.08	0.08	1.00	-	1.57	0.0036
R37272	2.00	3.16	39.59	0.08	35.00	0.08	0.08	1.00	-	1.54	0.0035
R37273	3.00	2.60	28.75	0.09	35.00	0.09	0.09	1.00	-	1.73	0.0039
R37274	4.00	1.29	44.31	0.03	35.00	0.03	0.03	1.00	-	0.56	0.0013
R37275	5.00							1.00	-		
R37276	6.00							1.00	-		
Mean		24.81	46.29	0.07	35.00	0.07	0.07	1.14	0.00	1.43	0.00
SD		24.89	16.48	0.02	0.00	0.02	0.02	0.23	0.00	0.39	0.00
%CV		100.31	35.61	27.64	0.00	27.64	27.64	20.55	ERR	27.64	27.64
n		16.00	8.00	8.00	8.00	8.00	8.00	11.00	11.00	8.00	8.00

GDP (4 ng) 24/04/2014 55.38

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date:28/04/2014

Checked by: Date:

Approved by: Pallen Yeung Date:02/05/2014

Title: Measurement of RBC Concentrations of GMP in Rat 372 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 23/04/2014

Sample/standard	ID/Standard Concentr (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (µL)	% Recovery
GMP 4 ng										
a50	50 ug/mL	off scale	8.37	0.00	35.00	0.00		0.35		
b50	50 ug/mL	123.47	7.72	15.99	35.00	15.99	15.99	0.35	9265.73	185.31
Mean		61.74	8.05	8.00	35.00	8.00	15.99		9265.73	185.31
SD		87.21	7.88	11.31	0.00	11.31	ERR		ERR	ERR
%CV		141.42	7.96	141.42	0.00	141.42	ERR		ERR	ERR
n		2.00	7.96	2.00	2.00	2.00	1.00		1.00	1.00
a20	20 ug/mL	116.53	8.75	13.36	35.00	13.36	13.36	0.35	8774.94	438.75
b20	20 ug/mL	off scale	11.31	0.00	35.00	0.00		0.35		
20*	20 ug/mL	116.64	8.99	12.97	35.00	12.97	12.97	0.35	8753.17	437.66
Mean		77.86	9.68	8.78	35.00	8.78	13.17		8764.05	438.20
SD		67.43	1.41	7.61	0.00	7.61	0.28		15.39	0.77
%CV		86.60	14.60	86.63	0.00	86.63	2.09		0.18	0.18
n		3.00	3.00	3.00	3.00	3.00	2.00		2.00	2.00
aB	0 ug/mL (a)	Off scale	80.02	0.00	35.00	0.00	0.00	2.00		0.00
bB	0 ug/mL (a)	Off scale	96.44	0.00	35.00	0.00	0.00	2.00		0.00
Mean		0.00	88.23	0.00	35.00	0.00	0.00			0.00
SD		0.00	11.61	0.00	0.00	0.00	0.00			0.00
%CV		ERR	13.16	ERR	0.00	ERR	ERR			ERR
n		2.00	2.00	2.00	2.00	2.00	2.00			2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	15.99	0.00	15.99
20.00	13.17	0.00	13.17
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	3.6730
Std Err of Y Est	5.4924
R Squared	0.7930
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3021
Std Err of Coef.	0.1543

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R37270		0.00 off scale	58.30	0.00	35.00	0.00	0.00	1.50	-	-8.85	-0.0244
R37270.08		0.08 off scale	76.76	0.00	35.00	0.00	0.00	1.50	-	-8.85	-0.0244
R37270.25		0.25 off scale	55.38	0.00	35.00	0.00	0.00	1.50	-	-8.85	-0.0244
R37111		1.00 off scale	27.76	0.00	35.00	0.00	0.00	1.00	-	-8.85	-0.0244
Not detected (80 mg/kg sc)											
R37211.2		1.20 off scale						1.00	-		
R37211.5		1.50 off scale	39.66	0.00	35.00	0.00	0.00	1.00	-	-8.85	-0.0244
R37212		2.00 off scale	39.39	0.00	35.00	0.00	0.00	1.00	-	-8.85	-0.0244
R37273		3.00 off scale	28.75	0.00	35.00	0.00	0.00	1.00	-	-8.85	-0.0244
R37274		4.00 off scale	44.31	0.00	35.00	0.00	0.00	1.00	-	-8.85	-0.0244
R37275		5.00 off scale							-		
R37276		6.00 off scale							-		
Mean		0.00	46.29	0.00	35.00	0.00	0.00	1.17		0.00	-8.85
SD		0.00	16.48	0.00	0.00	0.00	0.25			0.00	0.00
%CV		ERR	35.61	ERR	0.00	ERR	ERR	21.43		ERR	-0.00
n			11.00	8.00	8.00	8.00	8.00	9.00		11.00	8.00

GMP (4ng) 24/04/2014 88.77

Comments: RBC Lysate from Rat 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of "a" or "b" at 0.5-1ul injection volume

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 28/04/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 08/05/2014

Plasma Concentrations of Adenosine in Rat 372
Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
Experiment Date: 4/02/2015 - 09/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Adenosine 5 ng								
a2.5	2.5ug/ml (a)	12.87	32.71	0.39	0.39	10	143.48	57.39
b2.5	2.5ug/ml (b)	15.06	36.38	0.41	0.41	10	167.89	67.16
Mean		13.97	34.55	0.40	0.40		155.69	62.27
SD		1.55	2.60	0.01	0.01		17.26	6.91
%CV		11.09	7.51	3.59	3.59		11.09	11.09
N		2.00	2.00	2.00	2.00		2.00	2.00
a0.5	0.5ug/ml (a)	5.63	83.97	0.07	0.07	30.00	20.92	41.84
b0.5	0.5ug/ml (b)	5.67	84.27	0.07	0.07	30.00	21.07	42.14
	0.5 ug/ml	6.24	86.37	0.07	0.07	30.00	23.19	46.38
	0.5 ug/ml	3.63	89.60	0.04	0.04	30.00	13.49	26.98
Mean		5.29	86.05	0.06	0.07		19.67	39.33
SD		1.14	2.59	0.01	0.00		4.25	8.49
%CV		21.59	3.02	23.27	4.26		21.59	21.59
N		4.00	4.00	4.00	3.00		4.00	4.00
BLANKS:								
aB	0ug/mL (a)	N/D	7.59	0.00	0.00	2	0.00	
bB	0 ug/mL (b)	N/D	7.84	0.00	0.00	2	0.00	
Mean		0.00	7.72	0.00	0.00		0.00	
SD		0.00	0.18	0.00	0.00		0.00	
%CV		ERR	2.29	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00		2.00	
Adenosine 5 ng 05/02/2015		26.91	10.32			5.00		
Adenosine 5 ng 09/02/2015		26.91	8.66			5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.40	0.00	0.40
0.50	0.07	0.00	0.07
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.0082
R Squared	0.9985
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.1606
Std Err of Coef.	0.0032

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	Conc.(µM)	Conc (µM) Corrected for dilution
T0 R372	0.00	ND	123.28	0.00	0.00	35	-	0.00	0.00	0.00
T0.08 R372	0.08	4.79	97.93	0.05	0.05	35	-	0.30	1.14	1.52
T0.25 R372	0.25	4.34	130.62	0.03	0.03	35	-	0.21	0.77	1.03
T1 R372	1.00	3.54	128.66	0.03	0.03	35	-	0.17	0.64	0.86
Regadenosin (30 mg/kg)										
T1.2 R372	1.20	4.61	83.99	0.05	0.05	35	-	0.34	1.28	1.71
T1.5 R372	1.50	5.36	114.97	0.05	0.05	35	-	0.29	1.09	1.45
T2 R372	2.00	7.59	125.74	0.06	0.06	35	-	0.38	1.41	1.88
T3 R372	3.00	4.11	126.53	0.03	0.03	35	-	0.20	0.76	1.01
T4 R372	4.00	1.97	103.20	0.02	0.02	35	-	0.12	0.44	0.59
T5 R372	5.00	1.93	94.66	0.02	0.02	35	-	0.13	0.48	0.63
T6 R372	6.00	2.68	121.51	0.02	0.02	35	-	0.14	0.51	0.69
Mean		3.72	113.74	0.03	0.03			0.21	0.77	1.03
SD		2.03	16.06	0.02	0.02			0.11	0.42	0.56
%CV		54.67	14.12	54.31	54.31			54.31	54.31	54.31
n		11.00	11.00	11.00	11.00			11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
The Spiking solutions were made on: 22/02/2006
***Repeat injections of QC a or b**

Submitted by: Shyam Sundar Date: 17/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 24/02/2015

Plasma Concentrations of Inosine in Rat 372
 Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
 Experiment Date: 04/02/2015 - 09/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Inosine 5 ng		43.31				5		
a2.5	2.5ug/ml (a)	26.69	32.71	0.82	0.82	10	184.88	73.95
b2.5	2.5ug/ml (b)	27.94	36.38	0.77	0.77	10	193.53	77.41
Mean		27.32	34.55	0.79	0.79		189.21	75.68
SD		0.88	2.60	0.03	0.03		6.12	2.45
%CV		3.24	7.51	4.28	4.28		3.24	3.24
N		2.00	2.00	2.00	2.00		2.00	2.00
a0.5	0.5ug/ml (a)	7.71	83.97	0.09	0.09	30.00	17.80	35.60
b0.5	0.5ug/ml (b)	3.86	84.27	0.05	0.05	30.00	8.91	17.82
	0.5ug/ml	8.08	86.37	0.09	0.09	30.00	18.66	37.31
	0.5ug/ml	7.50	89.60	0.08	0.08	30.00	17.32	34.63
Mean		6.79	86.05	0.08	0.09		15.67	31.34
SD		1.97	2.59	0.02	0.01		4.54	9.08
%CV		28.97	3.02	28.40	5.96		28.97	28.97
N		4.00	4.00	4.00	3.00		4.00	4.00
BLANKS:								
aB	0ug/mL (a)	ND	7.59	0.00	0.00	2	0.00	
bB	0 ug/mL (b)	ND	7.84	0.00	0.00	2	0.00	
Mean		0.00	7.72	0.00	0.00		0.00	
SD		0.00	0.18	0.00	0.00		0.00	
%CV		ERR	2.29	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00		2.00	
Inosine 5ng 05/02/15		42.06	10.32			5.00		
Inosine 5ng 09/02/15		38.83	8.66			5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.79	0.00	0.79
0.50	0.09	0.00	0.09
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0327
Std Err of Y Est	0.0530
R Squared	0.9925
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3266
Std Err of Coef.	0.0283

Sample ID	Time post-dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	ic. (µg/mL)	Conc. (µM)	Conc. (µM) Corrected for dilution
T0 R372	0.00	4.67	123.28	0.04	0.04	35	-	0.22	0.81	1.07
T0.08 R372	0.08	4.94	97.93	0.05	0.05	35	-	0.25	0.95	1.27
T0.25 R372	0.25	5.75	130.62	0.04	0.04	35	-	0.23	0.88	1.17
T1 R372	1.00	5.35	128.66	0.04	0.04	35	-	0.23	0.85	1.13
Isoproterenol (30 mg/kg)										
T1.2 R372	1.20	3.92	83.99	0.05	0.05	35	-	0.24	0.91	1.21
T1.5 R372	1.50	6.42	114.97	0.06	0.06	35	-	0.27	1.01	1.35
T2 R372	2.00	6.08	125.74	0.05	0.05	35	-	0.25	0.93	1.23
T3 R372	3.00	6.46	126.53	0.05	0.05	35	-	0.26	0.96	1.28
T4 R372	4.00	6.58	103.20	0.06	0.06	35	-	0.30	1.10	1.47
T5 R372	5.00	10.17	94.66	0.11	0.11	35	-	0.43	1.60	2.13
T6 R372	6.00	24.40	121.51	0.20	0.20	35	-	0.71	2.67	3.55
Mean		7.70	113.74	0.07	0.07			0.31	1.15	1.53
SD		5.77	16.06	0.05	0.05			0.15	0.55	0.73
%CV		74.85	14.12	70.50	70.50			47.60	47.60	47.60
n		11.00	11.00	11.00	11.00			11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
 The Spiking solutions were made on: 22/02/2006
 *Repeat injections of QC a or b

Submitted by: Shyam Sundar Date: 17/02/2015
 Checked by: Date:
 Approved by: Pollen Yeung Date: 26/02/2015

Plasma Concentrations of Hypoxanthine in Rat 372
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 04/02/2015 - 09/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Hypoxanthine 5 ng		105.43				5		
a25	25ug/ml (a)	105.02	7.86	13.36	13.36	2	1494.17	58.08
b25	25ug/ml (b)	85.12	6.88	12.37	12.37	2	1211.04	46.75
Mean		95.07	7.37	12.87	12.87		1352.60	52.42
SD		14.07	0.69	0.70	0.70		200.20	8.01
%CV		14.80	9.40	5.44	5.44		14.80	15.28
N		2.00	2.00	2.00	2.00		2.00	2.00
a5	5ug/ml (a)	14.39	8.56	1.68	1.68	2	204.73	32.51
b5	5ug/ml (b)	13.97	8.90	1.57	1.57	2	198.76	31.31
	5ug/ml	15.27	9.55	1.60	1.60	2	217.25	35.01
	5ug/ml	15.08	9.95	1.52	1.52	2	214.55	34.47
Mean		14.68	9.24	1.59	1.59		208.82	33.33
SD		0.60	0.63	0.07	0.07		8.60	1.72
%CV		4.12	6.78	4.34	4.34		4.12	5.16
N		4.00	4.00	4.00	4.00		4.00	4.00
BLANKS:								
aB	0ug/ml (a)	2.65	7.59	0.35	0.35	2	37.70	
bB	0ug/ml (b)	3.28	9.23	0.36	0.36	2	46.67	
Mean		2.97	8.41	0.35	0.35		42.18	
SD		0.45	1.16	0.00	0.00		6.34	
%CV		15.02	13.79	1.25	1.25		15.02	
N		2.00	2.00	2.00	2.00		2.00	
Hypoxanthine 5 ng 05/02/15		102.69	10.32				5.00	
Hypoxanthine 5 ng 09/02/15		101.39	8.66				5.00	

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	12.87	0.35	12.51
5.00	1.59	0.35	1.24
0.00	0.35	0.35	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.6018
Std Err of Y Est	0.9751
R Squared	0.9900
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.5186
Std Err of Coef.	0.0521

Sample ID	Time post-dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	ic. (µg/mL); conc. (µM)	Conc. (µM)
T0 R372	0.00	12.41	22.26	0.56	0.56	5	-	2.24	16.42
T0.08 R372	0.08	11.03	18.37	0.60	0.60	5	-	2.32	17.03
T0.25 R372	0.25	13.84	24.04	0.58	0.58	5	-	2.27	16.68
T1 R372	1.00	13.95	22.31	0.63	0.63	5	-	2.37	17.38
Isoproterenol (30 mg/kg)									
T1.2 R372	1.20	11.68	22.03	0.53	0.53	5	-	2.18	16.04
T1.5 R372	1.50	11.56	20.50	0.56	0.56	5	-	2.25	16.51
T2 R372	2.00	11.67	23.64	0.49	0.49	5	-	2.11	15.52
T3 R372	3.00	13.72	20.86	0.66	0.66	5	-	2.43	17.84
T4 R372	4.00	13.29	19.60	0.68	0.68	5	-	2.47	18.13
T5 R372	5.00	11.25	16.55	0.68	0.68	5	-	2.47	18.16
T6 R372	6.00	13.86	22.58	0.61	0.61	5	-	2.34	17.22
Mean		12.48	21.75	0.58	0.58			2.27	16.68
SD		1.18	1.82	0.05	0.05			0.10	0.74
%CV		9.48	8.38	9.07	9.07			4.43	4.43
n		8.00	8.00	8.00	8.00			8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 17/02/2015
 Checked by: Date:
 Approved by: Pollen Yeung Date: 26/02/2015

Plasma Concentrations of Xanthine in Rat 372
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 04/02/2015 -09/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Reco (%)	Recovery (%)
Xanthine 5 ng									
a25	25ug/ml (a)	54.57	7.86	6.94	6.94	2	2168.34	86.73	
b25	25ug/ml (b)	25.86	6.88	3.76	3.76	2	1027.55	41.10	
Mean		40.22	7.37	5.35	5.35		1597.95	63.92	
SD		20.30	0.69	2.25	2.25		806.66	32.27	
%CV		50.48	9.40	42.08	42.08		50.48	50.48	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a5	5ug/ml (a)	2.33	8.56	0.27	0.27	2	92.58	18.52	
b5	5ug/ml (b)	4.92	8.90	0.55	0.55	2	195.50	39.10	
	5ug/ml	3.77	9.55	0.39	0.39	2	149.80	29.96	
	5ug/ml	3.77	9.95	0.38	0.38	2	149.80	29.96	
Mean		3.70	9.24	0.40	0.35		146.92	29.38	
SD		1.06	0.63	0.12	0.07		42.15	8.43	
%CV		28.69	6.78	28.95	19.12		28.69	28.69	
N		4.00	4.00	4.00	3.00		4.00	4.00	
BLANKS:									
aB	0ug/ml (a)	ND	7.59	0.00	0.00	2	0.00	0.00	
bB	0ug/ml (b)	ND	7.84	0.00	0.00	2	0.00	0.00	
Mean		0.00	7.72	0.00	0.00		0.00	0.00	
SD		0.00	0.18	0.00	0.00		0.00	0.00	
%CV		ERR	2.29	ERR	ERR		ERR	ERR	
N		2.00	2.00	2.00	2.00		2.00	2.00	

Xanthine 5 ng 05/02/15	36.70	10.32	5.00
Xanthine 5 ng 09/02/15	34.64	8.66	5.00

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	5.35	0.00	5.35
5.00	0.35	0.00	0.35
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.5003
R Squared	0.9721
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.2085
Std Err of Coef.	0.0196

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c. (µg/mL)	Conc. (µM)	Conc. (µM) Corrected for dilution
T0 R372	0.00	INT	22.26	0.00	0.00	5	-	0.00	0.00	0.00
T0.08 R372	0.08	INT	18.37	0.00	0.00	5	-	0.00	0.00	0.00
T0.25 R372	0.25	INT	24.04	0.00	0.00	5	-	0.00	0.00	0.00
T1 R372	1.00	INT	22.31	0.00	0.00	5	-	0.00	0.00	0.00

T1.2 R372	1.20	INT	22.03	0.00	0.00	5	-	0.00	0.00	0.00
T1.5 R372	1.50	INT	20.50	0.00	0.00	5	-	0.00	0.00	0.00
T2 R372	2.00	INT	23.64	0.00	0.00	5	-	0.00	0.00	0.00
T3 R372	3.00	INT	20.86	0.00	0.00	5	-	0.00	0.00	0.00
T4 R372	4.00	INT	19.60	0.00	0.00	5	-	0.00	0.00	0.00
T5 R372	5.00	INT	16.55	0.00	0.00	5	-	0.00	0.00	0.00
T6 R372	6.00	INT	22.58	0.00	0.00	5	-	0.00	0.00	0.00
Mean		0.00	21.75	0.00	0.00			0.00	0.00	0.00
SD		0.00	1.82	0.00	0.00			0.00	0.00	0.00
%CV		ERR	8.38	ERR	ERR			ERR	ERR	ERR
n		8.00	8.00	8.00	8.00			8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 17/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 26/02/2015

Plasma Concentrations of Uric Acid in Rat 372
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 04/02/2015 - 09/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Uric Acid 5 ng		29.71				5		
a25	25ug/ml (a)	49.63	7.86	6.31	6.31	2	2505.72	90.25
b25	25ug/ml (b)	39.88	6.88	5.80	5.80	2	2013.46	70.56
Mean		44.76	7.37	6.06	6.06		2259.59	80.41
SD		6.89	0.69	0.37	0.37		348.08	13.92
%CV		15.40	9.40	6.05	6.05		15.40	17.32
N		2.00	2.00	2.00	2.00		2.00	2.00
a5	5ug/ml (a)	9.97	8.56	1.16	1.16	2	503.37	50.79
b5	5ug/ml (b)	9.55	8.90	1.07	1.07	2	482.16	46.55
	5ug/ml	9.11	9.55	0.95	0.95	2	459.95	42.11
	5ug/ml	10.33	9.95	1.04	1.04	2	521.54	54.43
Mean		9.74	9.24	1.06	1.06		491.75	48.47
SD		0.53	0.63	0.09	0.09		26.62	5.32
%CV		5.41	6.78	8.25	8.25		5.41	10.98
N		4.00	4.00	4.00	4.00		4.00	4.00
BLANKS:								
aB	0ug/ml (a)	3.34	7.59	0.44	0.44	2	168.63	
bB	0ug/ml (b)	6.54	7.84	0.83	0.83	2	330.19	
Mean		4.94	7.72	0.64	0.64		249.41	
SD		2.26	0.18	0.28	0.28		114.24	
%CV		45.80	2.29	43.74	43.74		45.80	
N		2.00	2.00	2.00	2.00		2.00	
Uric Acid 5 ng 05/01/15		9.11	9.55			5.00		
Uric Acid 5 ng 09/01/15		10.33	9.95			5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	6.06	0.64	5.42
5.00	1.06	0.64	0.42
0.00	0.64	0.64	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.3159
Std Err of Y Est	0.5117
R Squared	0.9856
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.2262
Std Err of Coef.	0.0274

Sample ID	Time post-dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	ic. (µg/mL);onc. (µM)	Conc. (µM)	Corrected for dilution
T0 R372	0.00	11.78	22.26	0.53	0.53	5	-	3.74	22.22	29.63
T0.08 R372	0.08	25.02	18.37	1.36	1.36	5	-	7.42	44.12	58.83
T0.25 R372	0.25	16.79	24.04	0.70	0.70	5	-	4.48	26.67	35.57
T1 R372	1.00	5.03	22.31	0.23	0.23	5	-	2.39	14.24	18.98
Isoproterenol (30 mg/kg)										
T1.2 R372	1.20	36.06	22.03	1.64	1.64	5	-	8.63	63.43	84.57
T1.5 R372	1.50	54.34	20.50	2.65	2.65	5	-	13.11	96.36	128.48
T2 R372	2.00	54.78	23.64	2.32	2.32	5	-	11.64	69.25	92.33
T3 R372	3.00	17.47	20.86	0.84	0.84	5	-	5.10	30.33	40.44
T4 R372	4.00	37.56	19.60	1.92	1.92	5	-	9.87	58.70	78.27
T5 R372	5.00	35.44	16.55	2.14	2.14	5	-	10.86	64.62	86.16
T6 R372	6.00	7.75	22.58	0.34	0.34	5	-	2.91	17.33	23.11
Mean		27.66	21.75	1.28	1.28			7.06	45.83	61.10
SD		18.94	1.82	0.87	0.87			3.85	28.23	37.63
%CV		68.48	8.38	67.86	67.86			54.44	61.59	61.59
n		8.00	8.00	8.00	8.00			8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No. 181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 17/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 26/02/2015

Plasma Concentrations of Guanosine in Rat 372
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 04/02/2015 - 09/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio	Inj Vol. (µL)	Amount Reco (ng)	Recovery (%)
Guanosine 5 ng		53.29				5		
a2.5	2.5ug/ml (a)	22.93	32.71	0.70	0.70	10	129.09	51.63
b2.5	2.5ug/ml (b)	25.34	36.38	0.70	0.70	10	142.65	57.06
Mean		24.14	34.55	0.70	0.70		135.87	54.35
SD		1.70	2.60	0.00	0.00		9.59	3.84
%CV		7.06	7.51	0.45	0.45		7.06	7.06
N		2.00	2.00	2.00	2.00		2.00	2.00
a0.5	0.5ug/ml (a)	4.11	83.97	0.05	0.05	30.00	7.71	15.43
b0.5	0.5ug/ml (b)	3.97	84.27	0.05	0.05	30.00	7.45	14.90
	0.5ug/ml	4.11	86.37	0.05	0.05	30.00	7.71	15.43
	0.5ug/ml	2.52	89.60	0.03	0.03	30.00	4.73	9.46
Mean		3.68	86.05	0.04	0.05		6.90	13.80
SD		0.77	2.59	0.01	0.00		1.45	2.91
%CV		21.06	3.02	23.07	1.99		21.06	21.06
N		4.00	4.00	4.00	3.00		4.00	4.00
BLANKS:								
aB	0ug/mL (a)	ND	7.59	0.00	0.00	2	0.00	
bB	0 ug/mL (b)	ND	7.84	0.00	0.00	2	0.00	
Mean		0.00	7.72	0.00	0.00		0.00	
SD		0.00	0.18	0.00	0.00		0.00	
%CV		ERR	2.29	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00		2.00	
Guanosine 5 ng 05/02/15		51.74	10.32			5.00		
Guanosine 5 ng 09/02/15		50.40	8.66			5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.70	0.00	0.70
0.50	0.05	0.00	0.05
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output	
Constant	-0.0437
Std Err of Y Est	0.0709
R Squared	0.9835
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.2926
Std Err of Coef.	0.0379

Sample ID	Time post-dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	ic (µg/mL)	Conc. (µM)	Conc. (µM) Corrected for dilution
T0 R372	0.00	1.71	123.28	0.014	0.014	35	-	0.20	0.70	0.93
T0.08 R372	0.08	1.64	97.93	0.017	0.017	35	-	0.21	0.73	0.97
T0.25 R372	0.25	1.92	130.62	0.015	0.015	35	-	0.20	0.71	0.94
T1 R372	1.00	1.91	128.66	0.015	0.015	35	-	0.20	0.71	0.94
Isoproterenol (30 mg/kg)										
T1.2 R372	1.20	2.52	83.99	0.030	0.030	35	-	0.25	0.94	1.26
T1.5 R372	1.50	2.70	114.97	0.023	0.023	35	-	0.23	0.86	1.15
T2 R372	2.00	3.05	125.74	0.024	0.024	35	-	0.23	0.82	1.09
T3 R372	3.00	1.27	126.53	0.010	0.010	35	-	0.18	0.65	0.87
T4 R372	4.00	2.29	103.20	0.022	0.022	35	-	0.23	0.80	1.06
T5 R372	5.00	1.97	94.66	0.021	0.021	35	-	0.22	0.78	1.04
T6 R372	6.00	ND	121.51	0.000	0.000	35	-	0.15	0.53	0.70
Mean		1.91	113.74	0.02	0.02			0.21	0.75	1.00
SD		1.93	16.06	0.01	0.01			0.03	0.11	0.15
%CV		100.94	14.12	46.84	46.84			13.31	14.91	14.91
n		11.00	11.00	11.00	11.00			11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
 The Spiking solutions were made on: 22/02/2006
 *Repeat injections of QC a or b

Submitted by: Shyam Sundar Date: 17/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 26/02/2015

Title: Measurement of Plasma Concentrations of Dipyridamole in Rat 372

According to SOP No: SOP/STD/2008-001-1 (Plasma with no Stopping Solution) Experiment Date:22/09/2014- 24/09/2014

Abs.amt ng	STD ID	Peak Ht. #	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Dipyridamole (1ng)		72.38				1		
a1000	1 ug/mL(a)	85.06	5.90	14.42	14.42	3	78.35	156.69
b1000	1ug/mL(b)	111.21	8.83	12.59	12.59	3	102.43	204.86
1000*	1ug/mL(c)	120.86	7.67	15.76	15.76	3	111.32	222.64
Mean		105.71	7.47	14.26	14.26		97.37	194.73
SD		18.52	1.48	1.59	1.59		17.06	34.12
%CV		17.52	19.76	11.14	11.14		17.52	17.52
n		3.00	3.00	3.00	3.00		1.00	1.00
a100	0.1 ug/mL (a)	42.66	13.45	3.17	3.17	10	11.79	235.76
b100	0.1ug/mL (b)	15.58	5.55	2.81	2.81	10	4.31	86.10
Mean		29.12	9.50	2.99	2.99		8.05	160.93
SD		19.15	5.59	0.26	0.26		5.29	105.82
%CV		65.76	58.80	8.62	8.62		65.76	65.76
n		2.00	2.00	2.00	2.00		2.00	2.00
aB	0 ug/mL (a)	5.44	8.31	0.65	0.65	10	1.50	30.06
bB	0 ug/mL (b)	5.13	2.52	2.04	2.04	10	1.42	28.35
Mean		5.29	5.42	1.35	0.65		1.46	29.21
SD		0.22	4.09	0.98	ERR		0.06	1.21
%CV		4.15	75.61	72.60	ERR		4.15	4.15
n		2.00	2.00	2.00	1.00		2.00	2.00

Plasma Conc. (ug/mL)	Peak Ht.Ratio (PHR)	Blank (PHRb)	PHRV-PHRb
0.00	0.65	0.65	0.60
0.10	2.99	0.65	2.33
1.00	14.26	0.65	13.60

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.6858
R Squared	0.9911
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	13.6982
Std Err of Coef.	0.6824

Sample ID	Time Post-dose (h)	Peak Ht. #	Peak Ht. I.S. (mm)*	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc.(ug/mL)
R372T0	0.00	3.70	13.28	0.28	0.28	10	-	0.13
R372T0.08	0.08	9.53	11.41	0.84	0.84	20	-	0.39
R372T0.25	0.25	13.65	10.06	1.36	1.36	20	-	0.63
R372T1	1.00	13.03	9.71	1.34	1.34	20	-	0.62
Internal Standard (30 mg/kg sc)						20		
R372T1.2	1.20					20		
R372T1.5	1.50	22.51	8.53	2.64	2.64	20	-	1.23
R372T2	2.00	14.70	8.52	1.73	1.73	20	-	0.80
R372T3	3.00	18.28	9.28	1.97	1.97	20	-	0.92
R372T4	4.00	6.15	9.54	0.64	0.64	20	-	0.30
R372T5	5.00	8.70	6.91	1.26	1.26	20	-	0.59
R372T6	6.00	8.24	5.33	1.55	1.55	20	-	0.72
Mean		11.85	9.26	1.36	1.36			0.63
SD		5.73	2.20	0.68	0.68			0.32
%CV		48.32	23.82	49.90	49.90			49.90
n		10.00	10.00	10.00	10.00			10.00

Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 Corr. PHR = (PHR - RGB PHR)

Dipyridamole (1ng)/23/09/2014)	61.63	5.30		1.00
Dipyridamole(1ng)/24/09/2014)	89.60	9.36		1.00

Comments: Plasma from Rat 163 was used for extraction QC's.
 *A repeat injection of a or b

Submitted by: Shyam Sundar Date: 25/09/2014

Checked by: Pollen Yeung Date: 03/10/2014

Approved by: Date:

APPENDIX 3: Rat 373

Title: Measurement of RBC Concentrations of ATP in Rat 373 extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date 24/04/2014

Sample/standard ID	Standard Concentra (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recov %	Recovery
ATP 4 ng		20.10						4.00		
a250	250 ug/mL	47.34	8.37	5.66	35.00	5.66	5.66	0.35	14804.26	44.71
b250	250 ug/mL	45.44	7.72	5.89	35.00	5.89	5.89	0.35	14210.09	42.33
Mean		46.39	8.05	5.77	35.00	5.77	5.77		14507.18	43.52
SD		1.34	0.46	0.16	0.00	0.16	0.16		420.14	1.68
%CV		2.90	5.71	2.82	0.00	2.82	2.82		2.90	3.86
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a100	100 ug/mL	28.23	8.75	3.23	35.00	3.23	3.23	0.35	8828.14	52.01
b100	100 ug/mL	28.37	11.31	2.51	35.00	2.51	2.51	0.35	8871.93	52.45
100'	100 ug/mL	29.80	8.99	3.31	35.00	3.31	3.31	0.35	9319.12	56.92
Mean		28.80	9.68	3.02	35.00	3.02	3.02		8850.04	52.23
SD		0.87	1.41	0.44	0.00	0.44	0.44		30.96	0.31
%CV		3.02	14.60	14.66	0.00	14.66	14.66		0.35	0.59
n		3.00	3.00	3.00	3.00	3.00	3.00		2.00	2.00
a8	0 ug/mL (a)	61.36	80.02	0.77	35.00	0.77	0.77	2.00	3368.01	
b8	0 ug/mL (a)	71.20	96.44	0.74	35.00	0.74	0.74	2.00	3896.52	
Mean		66.28	88.23	0.75	35.00	0.75	0.75		3627.26	
SD		6.96	11.61	0.02	0.00	0.02	0.02		380.78	
%CV		10.50	13.16	2.68	0.00	2.68	2.68		10.50	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
250.00	5.77	0.75	5.02
100.00	3.02	0.75	2.26
0.00	0.75	0.75	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.1013
Std Err of Y Est	0.2081
R Squared	0.9966
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0199
Std Err of Coef.	0.0012

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R37370	0.00	80.80	46.54	1.85	35.00	1.85	1.85	1.50	-	92.77	0.1029
R37370.08	0.08	110.01	47.32	2.32	35.00	2.32	2.32	1.50	-	111.52	0.2199
R37370.25	0.25	110.16	53.36	2.06	35.00	2.06	2.06	1.50	-	98.46	0.1941
R37371	1.00	80.80	33.79	2.39	35.00	2.39	2.39	1.00	-	114.85	0.2264
Regression Method (10 mg/kg sc)											
R37371.2	1.20	102.16	35.86	2.85	35.00	2.85	2.85	1.00	-	137.80	0.2717
R37371.5	1.50	85.60	33.76	2.54	35.00	2.54	2.54	1.00	-	122.09	0.2407
R37372	2.00	55.62	37.41	1.49	35.00	1.49	1.49	1.00	-	69.49	0.1370
R37373	3.00	57.68	26.81	2.15	35.00	2.15	2.15	1.00	-	102.82	0.2027
R37374	4.00	76.73	32.88	2.33	35.00	2.33	2.33	1.00	-	111.96	0.2207
R37375	5.00							1.00	-		
R37376	6.00							1.00	-		
Mean		85.51	38.64	2.23	35.00	2.23	2.23	1.14		106.86	0.21
SD		20.25	8.54	0.39	0.00	0.39	0.39	0.23		19.31	0.04
%CV		23.68	22.12	17.25	0.00	17.25	17.25	20.55		18.07	18.07
n		9.00	9.00	9.00	9.00	9.00	9.00	11.00		9.00	9.00

ATP (4 ng) 24/04/2014 20.04

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 29/04/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 06/05/2014

Title: Measurement of RBC Concentrations of ADP in Rat 373 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)

Experiment Date: 24/04/2014

Sample/standard ID	Standard Co (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered	Recovery %
ADP 4 ng		41.58						4.00		
a250	250 µg/mL	75.34	8.37	9.00	35.00	9.00	9.00	0.35	11989.27	43.10
b250	250 µg/mL	71.39	7.72	9.25	35.00	9.25	9.25	0.35	10792.14	40.72
Mean		73.37	8.05	9.12	35.00	9.12	9.12		11090.70	41.91
SD		2.79	0.46	0.17	0.00	0.17	0.17		422.23	1.69
%CV		3.81	5.71	1.91	0.00	1.91	1.91		3.81	4.03
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
100a	100 µg/mL	40.94	8.75	4.68	35.00	4.68	4.68	0.35	6188.96	55.76
100b	100 µg/mL	39.71	11.31	3.51	35.00	3.51	3.51	0.35	6003.02	53.90
100*	100 µg/mL	43.25	8.99	4.81	35.00	4.81	4.81	0.35	6538.17	59.25
Mean		41.30	9.68	4.33	35.00	4.33	4.33		6243.39	56.30
SD		1.80	1.41	0.72	0.00	0.72	0.72		271.69	2.72
%CV		4.35	14.60	16.51	0.00	16.51	16.51		4.35	4.83
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB	0 µg/mL (a)	21.20	80.02	0.26	35.00	0.26	0.26	2.00	560.85	
bB	0 µg/mL (a)	25.15	96.44	0.26	35.00	0.26	0.26	2.00	665.34	
Mean		23.18	88.23	0.26	35.00	0.26	0.26		613.10	
SD		2.79	11.51	0.00	0.00	0.00	0.00		73.89	
%CV		12.05	13.16	1.12	0.00	1.12	1.12		12.05	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
250.00	9.12	0.26	8.86
100.00	4.33	0.26	4.07
0.00	0.26	0.26	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.2077
Std Err of Y Est	0.4288
R Squared	0.9954
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0352
Std Err of Coef.	0.0024

Sample ID	Time post	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) Lysate	Conc.(mM) RBC
R37310	0.00	20.42	46.54	0.44	35.00	0.44	0.44	1.50	-	6.57	0.0154	0.198
R37310B	0.06	21.76	47.32	0.46	35.00	0.46	0.46	1.50	-	7.17	0.0168	0.216
R37310.25	0.25	20.54	53.36	0.38	35.00	0.38	0.38	1.50	-	5.04	0.0118	0.152
R37311	1.00	13.80	33.79	0.41	35.00	0.41	0.41	1.00	-	5.71	0.0134	0.172
Isoproterenol (30 mg/kg sc)												
R37311.2	1.20	15.92	35.86	0.44	35.00	0.44	0.44	1.00	-	6.72	0.0157	0.202
R37311.5	1.50	13.10	33.76	0.39	35.00	0.39	0.39	1.00	-	5.13	0.0120	0.154
R37312	2.00	12.14	37.41	0.32	35.00	0.32	0.32	1.00	-	3.32	0.0078	0.100
R37313	3.00	10.21	26.81	0.38	35.00	0.38	0.38	1.00	-	4.92	0.0115	0.148
R37314	4.00	13.93	32.88	0.42	35.00	0.42	0.42	1.00	-	6.14	0.0144	0.185
R37315	5.00							1.00	-			
R37316	6.00							1.00	-			
Mean		20.91	49.07	0.43	35.00	0.43	0.43	1.50		6.26	0.01	0.19
SD		0.74	3.73	0.04	0.00	0.04	0.04	0.00		1.10	0.00	0.03
%CV		3.55	7.61	9.03	0.00	9.03	9.03	0.00		17.55	17.55	17.55
n		3.00	3.00	3.00	3.00	3.00	3.00	3.00		3.00	2.00	3.00

ADP (4 ng)24/04/2014

40.94

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.

*Repeated injections of a or b at 0.5-1ul injection volume.

PL = plasma; RBC = red blood cells

Peak Ht. = peak height

Peak Ht. R. (or: PHR) = peak height ratio

I.S. = internal standard

Inj Vol = injection volume

ND = not detected or determined

NS = no sample

INT = interference

PCV = packed cell volume (haematocrit)

CorPHR = corrected peak height ratio

Hemolysis Degree:

-: no visible hemolysis

+: slight hemolysis

++: intermediate hemolysis

+++ : serious hemolysis

Submitted by: Shyam Sundar K

Date: 29/04/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date: 06/05/2014

Title: Measurement of RBC Concentrations of AMP in Rat 373 extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date 24/04/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
AMP 4 ng										
		62.86						4.00		
a50	50 µg/mL	39.97	8.37	4.78	35.00	4.78	4.78	0.35	3996.82	78.01
b50	50 µg/mL	38.32	7.72	4.96	35.00	4.96	4.96	0.35	3831.83	75.71
Mean		39.15	8.05	4.87	35.00	4.87	4.87		3914.32	77.36
SD		1.17	0.46	0.13	0.00	0.13	0.13		116.87	2.33
%CV		2.98	5.71	2.73	0.00	2.73	2.73		2.98	3.02
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20	20 µg/mL	20.74	8.75	2.37	35.00	2.37	2.37	0.35	2073.91	101.37
b20	20 µg/mL	20.03	11.31	1.77	35.00	1.77	1.77	0.35	2002.91	97.82
20*	20 µg/mL	21.81	8.99	2.43	35.00	2.43	2.43	0.35	2180.90	106.72
Mean		20.86	9.68	2.19	35.00	2.19	2.19		2085.91	101.97
SD		0.90	1.41	0.36	0.00	0.36	0.36		89.60	4.48
%CV		4.30	14.60	16.59	0.00	16.59	16.59		4.30	4.39
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB	0 µg/mL (a)	2.60	80.02	0.03	35.00	0.03	0.03	2.00		45.59
bB	0 µg/mL (a)	2.71	96.44	0.03	35.00	0.03	0.03	2.00		47.42
Mean		2.66	88.23	0.03	35.00	0.03	0.03			46.46
SD		0.08	11.81	0.00	0.00	0.00	0.00			1.36
%CV		2.93	13.16	10.25	0.00	10.25	10.25			2.93
n		2.00	2.00	2.00	2.00	2.00	2.00			2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	4.87	0.03	4.84
20.00	2.19	0.03	2.16
0.00	0.03	0.03	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.1465
R Squared	0.9963
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.0983
Std Err of Coef.	0.0027

Sample ID	Time post dose	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R37310	0.00	0.54	46.54	0.01	35.00	0.01	0.01	1.50	-	0.12	0.0003
R37310.08	0.08	0.50	47.32	0.01	35.00	0.01	0.01	1.50	-	0.11	0.0003
R37310.25	0.25	0.28	53.36	0.01	35.00	0.01	0.01	1.50	-	0.05	0.0002
R37311	1.00	0.31	33.79	0.01	35.00	0.01	0.01	1.00	-	0.09	0.0003
Soprolefenol (30 mg/kg sc)											
R37311.2	1.20	0.37	35.66	0.01	35.00	0.01	0.01	1.00	-	0.10	0.0003
R37311.5	1.50	0.16	33.76	0.00	35.00	0.00	0.00	1.00	-	0.05	0.0001
R37312	2.00	0.22	37.41	0.01	35.00	0.01	0.01	1.00	-	0.06	0.0002
R37313	3.00	0.23	26.81	0.01	35.00	0.01	0.01	1.00	-	0.09	0.0003
R37314	4.00	0.28	32.88	0.01	35.00	0.01	0.01	1.00	-	0.09	0.0002
R37315	5.00							1.00	-		
R37316	6.00							1.00	-		
Mean		0.32	38.64	0.01	35.00	0.01	0.01	1.14	0.00	0.08	0.00
SD		0.13	8.54	0.00	0.00	0.00	0.00	0.23	0.00	0.03	0.00
%CV		39.74	22.12	29.80	0.00	29.80	29.80	20.15	0.00	26.80	29.80
n		9.00	9.00	9.00	9.00	9.00	9.00	11.00	9.00	9.00	9.00

AMP (4 ng)24/04/2014

64.63

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of "a" or "b" at 0.5 - 1 ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar

Date: 29/04/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date: 06/05/2014

Title: Measurement of RBC Concentrations of GTP in Rat 373 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 24/04/2014

Sample/standard ID	Standard Concentra (ug/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (uL)	Hemolysis Degree	Amount Recove %	Recovery
GTP 4 ng											
a50	50 ug/mL	14.63	8.37	1.75	35.00	1.75	1.75	0.35	2351.32	42.87	
b50	50 ug/mL	14.15	7.72	1.83	35.00	1.83	1.83	0.35	2274.17	41.32	
Mean		14.39	8.05	1.79	35.00	1.79	1.79		2312.74	42.10	
SD		0.34	0.06	0.00	0.06	0.06	0.06		54.55	1.09	
%CV		2.36	0.00	3.36	0.00	3.36	3.36		2.36	2.59	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00	
a20	20 ug/mL	8.19	8.75	0.94	35.00	0.94	0.94	0.35	1310.20	55.41	
b20	20 ug/mL	7.48	11.31	0.66	35.00	0.66	0.66	0.35	1263.78	49.79	
20*	20 ug/mL	7.84	8.99	0.87	35.00	0.87	0.87	0.35	1260.04	52.60	
Mean		7.84	9.68	0.82	35.00	0.82	0.80		1260.04	52.60	
SD		0.35	1.41	0.14	0.00	0.14	0.05		55.25	2.81	
%CV		4.46	14.60	17.39	0.00	17.39	5.00		4.46	5.35	
n		3.00	3.00	3.00	3.00	3.00	2.00		3.00	3.00	
a0	0 ug/mL (a)	6.84	80.02	0.09	35.00	0.09	0.09	2.00	192.38		
b0	0 ug/mL (a)	7.95	96.44	0.08	35.00	0.08	0.08	2.00	223.60		
Mean		7.40	88.23	0.08	35.00	0.08	0.08		207.99		
SD		0.78	11.61	0.00	0.00	0.00	0.00		22.08		
%CV		10.61	13.16	2.56	0.00	2.56	2.56		10.61		
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	Peak Height Ratio Value (PHRV)	Blank PHRV	PHRV-PHRb
50.00	1.79	0.08	1.71
20.00	0.90	0.08	0.82
0.00	0.08	0.08	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0543
Std Err of Y Est	0.1115
R Squared	0.9915
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0338
Std Err of Coef.	0.0031

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (uL)	Hemolysis Degree	Conc.(ug/mL) Lysate	Conc.(mM) RBC
R37310	0.00	15.37	46.54	0.33	35.00	0.33	0.44	1.50	-	11.39	0.0218
R37310.08	0.08	19.72	47.32	0.42	35.00	0.42	0.42	1.50	-	10.73	0.0205
R37310.25	0.25	19.30	53.36	0.36	35.00	0.36	0.36	1.50	-	9.10	0.0174
R37311	1.00	13.26	33.79	0.39	35.00	0.39	0.39	1.00	-	10.01	0.0191
Supranal (30 mg/kg sc)											
R37311.2	1.20	18.77	35.86	0.52	35.00	0.52	0.52	1.00	-	13.89	0.0266
R37311.5	1.50	15.88	33.76	0.47	35.00	0.47	0.47	1.00	-	12.32	0.0236
R37312	2.00	8.34	37.41	0.22	35.00	0.22	0.22	1.00	-	4.99	0.0095
R37313	3.00	12.33	26.81	0.46	35.00	0.46	0.46	1.00	-	12.01	0.0230
R37314	4.00	14.59	32.88	0.44	35.00	0.44	0.44	1.00	-	11.53	0.0220
R37315	5.00							1.00	-		
R37316	6.00							1.00	-		
Mean		15.28	38.64	0.40	35.00	0.40	0.41	1.14	0.00	10.67	0.02
SD		3.71	8.54	0.09	0.00	0.09	0.09	0.23	0.00	2.53	0.00
%CV		24.24	22.12	22.17	0.00	22.17	20.63	20.55	0.00	23.74	23.74
n		9.00	9.00	9.00	9.00	9.00	9.00	11.00	11.00	9.00	9.00

GTP (4 ng)/24/04/2014 33.75

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K

Date: 29/04/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date:06/05/2014

Title: Measurement of RBC Concentrations of GDP in Rat 373 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 24/04/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recove %	Recovery
GDP 4 ng										
		57.79						4.00		
a50	50 ug/mL	21.61	8.37	2.58	35.00	2.58	2.58	0.35	2350.48	45.85
b50	50 ug/mL	20.22	7.72	2.62	35.00	2.62	2.62	0.35	2199.29	42.83
Mean		20.92	8.05	2.60	35.00	2.60	2.60		2274.89	44.34
SD		0.98	0.46	0.03	0.00	0.03	0.03		106.81	2.14
%CV		4.70	5.71	1.02	0.00	1.02	1.02		4.70	4.82
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20	20 ug/mL	11.50	8.75	1.31	35.00	1.31	1.31	0.35	1250.83	59.64
b20	20 ug/mL	10.58	11.31	0.94	35.00	0.94	0.94	0.35	1150.77	54.64
20*	20 ug/mL	11.86	8.99	1.32	35.00	1.32	1.32	0.35	1289.99	61.60
Mean		11.31	9.68	1.19	35.00	1.19	1.19		1230.53	58.03
SD		0.66	1.41	0.22	0.00	0.22	0.22		71.80	3.59
%CV		5.83	14.60	18.51	0.00	18.51	18.51		5.83	6.12
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB	0 ug/mL (a)	3.01	80.02	0.04	35.00	0.04	0.04	2.00		57.29
bB	0 ug/mL (a)	3.08	96.44	0.03	35.00	0.03	0.03	2.00		58.03
Mean		3.05	88.23	0.03	35.00	0.03	0.03			57.96
SD		0.05	11.81	0.00	0.00	0.00	0.00			0.94
%CV		1.63	13.16	11.55	0.00	11.55	11.55			1.63
n		2.00	2.00	2.00	2.00	2.00	2.00			2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRb
50.00	2.60	0.03	2.57
20.00	1.19	0.03	1.15
0.00	0.03	0.03	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.0844
R Squared	0.9957
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.0022
Std Err of Coef.	0.0016

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL)	Conc.(nM) Lysate	Conc.(nM) RBC
R37370	0.00	2.96	46.54	0.06	35.00	0.06	0.06	1.50	-	1.05	0.0024	0.031
R37370.08	0.08	2.39	47.32	0.05	35.00	0.05	0.05	1.50	-	0.97	0.0022	0.028
R37370.25	0.25	3.21	53.36	0.06	35.00	0.06	0.06	1.50	-	1.15	0.0026	0.033
R37371	1.00	2.01	33.79	0.06	35.00	0.06	0.06	1.00	-	1.14	0.0026	0.033
Isoproterenol (30 µg/kg sc)												
R37371.2	1.20	2.86	35.86	0.08	35.00	0.08	0.08	1.00	-	1.53	0.0034	0.044
R37371.5	1.50	1.90	33.76	0.06	35.00	0.06	0.06	1.00	-	1.08	0.0024	0.031
R37372	2.00	1.11	37.61	0.03	35.00	0.03	0.03	1.00	-	0.57	0.0013	0.016
R37373	3.00	0.85	26.81	0.03	35.00	0.03	0.03	1.00	-	0.61	0.0014	0.018
R37374	4.00	1.52	32.88	0.06	35.00	0.06	0.06	1.00	-	1.12	0.0025	0.032
R37375	5.00							1.00	-			
R37376	6.00							1.00	-			
Mean		20.36	38.64	0.05	35.00	0.05	0.05	1.14	0.00	1.02	0.00	0.03
SD		19.70	8.54	0.02	0.00	0.02	0.23	0.00	0.29	0.00	0.00	0.01
%CV		96.76	22.12	28.50	0.00	28.50	28.50	20.55	EPH	28.50	28.50	28.50
n		18.00	9.00	9.00	9.00	9.00	9.00	11.00	11.00	9.00	9.00	9.00

GDP (4ng)24/04/2014

55.38

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date:28/04/2014

Checked by: Date:

Approved by: Pallen Yeung Date:06/05/2014

Title: Measurement of RBC Concentrations of GMP in Rat 373 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 24/04/2014

Sample/standard	ID/Standard Concentr (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recov. (µL)	% Recovery
GMP 4 ng										
a50	50 ug/mL	off scale	8.37	0.00	35.00	0.00		0.35		
b50	50 ug/mL	123.47	7.72	15.99	35.00	15.99	15.99	0.35	9265.73	185.31
Mean		61.74	8.05	8.00	35.00	8.00	15.99		9265.73	185.31
SD		87.21	7.88	11.31	0.00	11.31	ERR		ERR	ERR
%CV		141.42	7.96	141.42	0.00	141.42	ERR		ERR	ERR
n		2.00	7.96	2.00	2.00	2.00	1.00		1.00	1.00
a20										
b20	20 ug/mL	116.63	8.75	13.36	35.00	13.36	13.36	0.35	8774.04	438.75
20*	20 ug/mL	116.64	8.99	12.97	35.00	12.97	12.97	0.35	8753.17	437.66
Mean		77.86	9.68	8.78	35.00	8.78	13.17		8764.05	438.20
SD		67.43	1.41	7.61	0.00	7.61	0.28		15.39	0.77
%CV		86.60	14.60	86.63	0.00	86.63	2.09		0.18	0.18
n		3.00	3.00	3.00	3.00	3.00	2.00		2.00	2.00
aB										
bB	0 ug/mL (a)	Off scale	80.02	0.00	35.00	0.00	0.00	2.00	0.00	0.00
	0 ug/mL (a)	Off scale	96.44	0.00	35.00	0.00	0.00	2.00	0.00	0.00
Mean		0.00	88.23	0.00	35.00	0.00	0.00		0.00	0.00
SD		0.00	11.61	0.00	0.00	0.00	0.00		0.00	0.00
%CV		ERR	13.16	ERR	0.00	ERR	ERR		ERR	ERR
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	15.99	0.00	15.99
20.00	13.17	0.00	13.17
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	3.6730
Std Err of Y Est	5.4924
R Squared	0.7930
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3021
Std Err of Coef.	0.1543

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc (µg/mL) Lysate	Conc (mM) RBC
R37370		0.00 off scale	46.54	0.00	35.00	0.00	0.00	1.50	-	-8.85	-0.0244
R37370.08		0.08 off scale	47.32	0.00	35.00	0.00	0.00	1.50	-	-8.85	-0.0244
R37370.25		0.25 off scale	53.36	0.00	35.00	0.00	0.00	1.50	-	-8.85	-0.0244
R37371		1.00 off scale	33.79	0.00	35.00	0.00	0.00	1.00	-	-8.85	-0.0244
Not detected (ND mg/kg sat)											
R37371.2		1.20 off scale	35.66	0.00	35.00	0.00	0.00	1.00	-	-8.85	-0.0244
R37371.5		1.50 off scale	33.76	0.00	35.00	0.00	0.00	1.00	-	-8.85	-0.0244
R37372		2.00 off scale	37.41	0.00	35.00	0.00	0.00	1.00	-	-8.85	-0.0244
R37373		3.00 off scale	26.81	0.00	35.00	0.00	0.00	1.00	-	-8.85	-0.0244
R37374		4.00 off scale	32.68	0.00	35.00	0.00	0.00	1.00	-	-8.85	-0.0244
R37375		5.00						1.00	-		
R37376		6.00						1.00	-		
Mean		0.00	38.64	0.00	35.00	0.00	0.00	1.14	0.00	-8.85	-0.02
SD		0.00	8.54	0.00	0.00	0.00	0.23	0.00	0.00	0.00	0.00
%CV		ERR	22.12	ERR	0.00	ERR	ERR	20.55	ERR	-0.00	-0.00
n		9.00	9.00	9.00	9.00	9.00	9.00	11.00	11.00	9.00	9.00

GMP (4 ng)24/04/2014 88.77

Comments: RBC Lysate from Rat 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of "a" or "b" at 0.5-1ul injection volume

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 28/04/2014
 Checked by: Date:
 Approved by: Pollen Yeung Date: 08/05/2014

Plasma Concentrations of Adenosine in Rat 373
Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
Experiment Date: 4/02/2015 - 11/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount Reco (ng)	Recovery (%)
Adenosine 5 ng								
a2.5	2.5ug/ml (a)	12.87	32.71	0.39	0.39	10	143.48	57.39
b2.5	2.5ug/ml (b)	15.06	36.38	0.41	0.41	10	167.89	67.16
Mean		13.97	34.55	0.40	0.40		155.69	62.27
SD		1.55	2.60	0.01	0.01		17.26	6.91
%CV		11.09	7.51	3.59	3.59		11.09	11.09
N		2.00	2.00	2.00	2.00		2.00	2.00
a0.5	0.5ug/ml (a)	5.63	83.97	0.07	0.07	30.00	20.92	41.84
b0.5	0.5ug/ml (b)	5.67	84.27	0.07	0.07	30.00	21.07	42.14
	0.5 ug/ml	3.63	89.60	0.04	0.04	30.00	13.49	26.98
	0.5 ug/ml	4.56	90.89	0.05	0.05	30.00	16.95	33.89
	0.5 ug/ml	6.05	88.61	0.07	0.07	30.00	22.48	44.96
Mean		5.11	87.47	0.06	0.07		18.98	37.96
SD		1.00	3.16	0.01	0.00		3.70	7.40
%CV		19.48	3.62	21.54	0.97		19.48	19.48
N		5.00	5.00	5.00	3.00		5.00	5.00
BLANKS:								
aB	0ug/mL (a)	N/D	7.59	0.00	0.00	2	0.00	
bB	0 ug/mL (b)	N/D	7.84	0.00	0.00	2	0.00	
Mean		0.00	7.72	0.00	0.00		0.00	
SD		0.00	0.18	0.00	0.00		0.00	
%CV		ERR	2.29	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00		2.00	
Adenosine 5 ng 09/02/2015		26.91	8.66			5.00		
Adenosine 5 ng 10/02/2015		29.16	10.93			5.00		
Adenosine 5 ng 11/02/2015		31.06	10.23			5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.40	0.00	0.40
0.50	0.07	0.00	0.07
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	0.0000
Std Err of Y Est	0.0092
R Squared	0.9982
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.1605
Std Err of Coef.	0.0036

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	Conc.(µM)	Conc (µM) Corrected for dilution
T0 R373	0.00	8.76	99.70	0.09	0.09	35	-	0.55	2.05	2.73
T0.08 R373	0.08	7.67	117.48	0.07	0.07	35	-	0.41	1.52	2.03
T0.25 R373	0.25	3.96	119.74	0.03	0.03	35	-	0.21	0.77	1.03
T1 R373	1.00	4.08	103.00	0.04	0.04	35	-	0.25	0.92	1.23
Regosol (30 mg/kg)										
T1.2 R373	1.20	3.31	127.15	0.03	0.03	35	-	0.16	0.61	0.81
T1.5 R373	1.50	5.65	132.84	0.04	0.04	35	-	0.27	0.99	1.32
T2 R373	2.00	2.00	99.48	0.02	0.02	35	-	0.13	0.47	0.63
T3 R373	3.00	4.05	105.28	0.04	0.04	35	-	0.24	0.90	1.20
T4 R373	4.00	2.02	124.02	0.02	0.02	35	-	0.10	0.38	0.51
T5 R373	5.00	ND	111.82	0.00	0.00	35	-	0.00	0.00	0.00
T6 R373	6.00	ND	127.96	0.00	0.00	35	-	0.00	0.00	0.00
Mean		3.77	115.32	0.03	0.03			0.21	0.78	1.04
SD		2.81	12.11	0.03	0.03			0.16	0.61	0.82
%CV		74.35	10.50	78.17	78.17			78.17	78.17	78.17
n		11.00	11.00	11.00	11.00			11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
The Spiking solutions were made on: 22/02/2006
***Repeat injections of QC a or b**

Submitted by: Shyam Sundar Date: 17/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 27/02/2015

Plasma Concentrations of Inosine in Rat 373
Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
Experiment Date: 04/02/2015 - 11/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Inosine 5 ng		43.31						
a2.5	2.5ug/ml (a)	26.69	32.71	0.82	0.82	10	184.88	73.95
b2.5	2.5ug/ml (b)	27.94	36.38	0.77	0.77	10	193.53	77.41
Mean		27.32	34.55	0.79	0.79		189.21	75.68
SD		0.88	2.60	0.03	0.03		6.12	2.45
%CV		3.24	7.51	4.28	4.28		3.24	3.24
N		2.00	2.00	2.00	2.00		2.00	2.00
a0.5	0.5ug/ml (a)	7.71	83.97	0.09	0.09	30.00	17.80	35.60
b0.5	0.5ug/ml (b)	3.86	84.27	0.05	0.05	30.00	8.91	17.82
	0.5ug/ml	7.50	89.60	0.08	0.08	30.00	17.32	34.63
	0.5ug/ml	6.56	89.60	0.07	0.07	30.00	15.15	30.29
	0.5ug/ml	6.32	88.61	0.07	0.07	30.00	14.59	29.18
Mean		6.39	87.21	0.07	0.08		14.75	29.51
SD		1.53	2.85	0.02	0.01		3.54	7.08
%CV		24.00	3.27	23.78	11.96		24.00	24.00
N		5.00	5.00	5.00	4.00		5.00	5.00
BLANKS:								
aB	0ug/mL (a)	ND	7.59	0.00	0.00	2	0.00	
bB	0 ug/mL (b)	ND	7.84	0.00	0.00	2	0.00	
Mean		0.00	7.72	0.00	0.00		0.00	
SD		0.00	0.18	0.00	0.00		0.00	
%CV		ERR	2.29	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00		2.00	
Inosine Sng 09/02/15		38.83	8.66			5.00		
Inosine Sng 10/02/15		40.59	10.93			5.00		
Inosine Sng 11/02/15		40.83	10.23			5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.79	0.00	0.79
0.50	0.08	0.00	0.08
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0373
Std Err of Y Est	0.0605
R Squared	0.9904
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3280
Std Err of Coef.	0.0323

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	Conc.(µM)	Conc (µM) Corrected for dilution
T0 R373	0.00	7.56	99.70	0.08	0.08	35	-	0.34	1.29	1.72
T0.08 R373	0.08	7.88	117.48	0.07	0.07	35	-	0.32	1.19	1.58
T0.25 R373	0.25	8.08	119.74	0.07	0.07	35	-	0.32	1.19	1.59
T1 R373	1.00	6.10	103.00	0.06	0.06	35	-	0.29	1.10	1.46
Regosol (30 mg/kg)										
T1.2 R373	1.20	6.52	127.15	0.05	0.05	35	-	0.27	1.01	1.34
T1.5 R373	1.50	6.23	132.84	0.05	0.05	35	-	0.26	0.96	1.28
T2 R373	2.00	7.80	99.48	0.08	0.08	35	-	0.35	1.32	1.75
T3 R373	3.00	7.87	105.28	0.07	0.07	35	-	0.34	1.27	1.70
T4 R373	4.00	9.20	124.02	0.07	0.07	35	-	0.34	1.27	1.69
T5 R373	5.00	9.62	111.82	0.09	0.09	35	-	0.38	1.40	1.87
T6 R373	6.00	7.52	127.96	0.06	0.06	35	-	0.29	1.09	1.46
Mean		7.67	115.32	0.07	0.07			0.32	1.19	1.59
SD		1.11	12.11	0.01	0.01			0.04	0.14	0.18
%CV		14.46	10.50	17.97	17.97			11.56	11.56	11.56
n		11.00	11.00	11.00	11.00			11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
The Spiking solutions were made on: 22/02/2006
***Repeat injections of QC a or b**

Submitted by: Shyam Sundar Date: 17/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 27/02/2015

Plasma Concentrations of Hypoxanthine in Rat 373
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 04/02/2015 - 11/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Hypoxanthine 5 ng		105.43				5		
a25	25ug/ml (a)	105.02	7.86	13.36	13.36	2	1494.17	58.08
b25	25ug/ml (b)	85.12	6.88	12.37	12.37	2	1211.04	46.75
Mean		95.07	7.37	12.87	12.87		1352.60	52.42
SD		14.07	0.69	0.70	0.70		200.20	8.01
%CV		14.80	9.40	5.44	5.44		14.80	15.28
N		2.00	2.00	2.00	2.00		2.00	2.00
a5	5ug/ml (a)	14.39	8.56	1.68	1.68	2	204.73	32.51
b5	5ug/ml (b)	13.97	8.90	1.57	1.57	2	198.76	31.31
	5ug/ml	15.08	9.95	1.52	1.52	2	214.55	34.47
	5ug/ml	13.61	7.86	1.73	1.73	2	193.64	30.29
	5ug/ml	14.06	8.12	1.73	1.73	2	200.04	31.57
Mean		14.22	8.68	1.65	1.65		202.34	32.03
SD		0.55	0.82	0.10	0.10		7.89	1.58
%CV		3.90	9.40	5.98	5.98		3.90	4.92
N		5.00	5.00	5.00	5.00		5.00	5.00
BLANKS:								
aB	0ug/ml (a)	2.65	7.59	0.35	0.35	2	37.70	
bB	0ug/ml (b)	3.28	9.23	0.36	0.36	2	46.67	
Mean		2.97	8.41	0.35	0.35		42.18	
SD		0.45	1.16	0.00	0.00		6.34	
%CV		15.02	13.79	1.25	1.25		15.02	
N		2.00	2.00	2.00	2.00		2.00	
Hypoxanthine 5 ng 09/02/15		101.39	8.66				5.00	
Hypoxanthine 5 ng 10/02/15		107.69	10.93				5.00	
Hypoxanthine 5 ng 11/02/15		106.74	10.23				5.00	

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	12.87	0.35	12.51
5.00	1.65	0.35	1.29
0.00	0.35	0.35	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.5758
Std Err of Y Est	0.9330
R Squared	0.9908
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.5179
Std Err of Coef.	0.0499

Sample ID	Time post-dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	ic. (µg/mL);onc. (µM)	Conc. (µM)	Corrected for dilution
T0 R373	0.00	7.94	14.28	0.56	0.56	5	-	2.19	16.06	21.41
T0.08 R373	0.08	14.46	23.76	0.61	0.61	5	-	2.29	16.81	22.41
T0.25 R373	0.25	13.31	22.63	0.59	0.59	5	-	2.25	16.52	22.02
T1 R373	1.00	12.49	23.36	0.53	0.53	5	-	2.14	15.76	21.01
Isoproterenol (30 mg/kg)										
T1.2 R373	1.20	14.09	22.43	0.63	0.63	5	-	2.33	17.08	22.78
T1.5 R373	1.50	15.53	23.24	0.67	0.67	5	-	2.40	17.65	23.54
T2 R373	2.00	13.17	19.23	0.68	0.68	5	-	2.43	17.89	23.85
T3 R373	3.00	13.20	19.41	0.68	0.68	5	-	2.43	17.82	23.76
T4 R373	4.00	13.50	22.67	0.60	0.60	5	-	2.26	16.62	22.16
T5 R373	5.00	10.61	20.00	0.53	0.53	5	-	2.14	15.70	20.93
T6 R373	6.00	9.88	23.20	0.43	0.43	5	-	1.93	14.21	18.95
Mean		13.02	21.04	0.62	0.62			2.31	16.95	22.60
SD		2.26	3.25	0.06	0.06			0.11	0.81	1.08
%CV		17.35	15.42	9.21	9.21			4.77	4.77	4.77
n		8.00	8.00	8.00	8.00			8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 17/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 27/02/2015

Plasma Concentrations of Xanthine in Rat 373
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 04/02/2015 -11/02/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Reco (%)	Recovery (%)
Xanthine 5 ng									
a25	25ug/ml (a)	54.57	7.86	6.94	6.94	2	2168.34	86.73	
b25	25ug/ml (b)	25.86	6.88	3.76	3.76	2	1027.55	41.10	
Mean		40.22	7.37	5.35	5.35		1597.95	63.92	
SD		20.30	0.69	2.25	2.25		806.66	32.27	
%CV		50.48	9.40	42.08	42.08		50.48	50.48	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a5									
	5ug/ml (a)	2.33	8.56	0.27	0.27	2	92.58	18.52	
b5	5ug/ml (b)	4.92	8.90	0.55	0.55	2	195.50	39.10	
	5ug/ml	3.77	9.95	0.38	0.38	2	149.80	29.96	
	5ug/ml	2.83	7.86	0.36	0.36	2	112.45	22.49	
	5ug/ml	3.36	8.12	0.41	0.41	2	133.51	26.70	
Mean		3.44	8.68	0.40	0.36		136.77	27.35	
SD		0.99	0.82	0.10	0.06		39.28	7.86	
%CV		28.72	9.40	25.85	16.92		28.72	28.72	
N		5.00	5.00	5.00	4.00		5.00	5.00	
BLANKS:									
aB	0ug/ml (a)	ND	7.59	0.00	0.00	2	0.00	0.00	
bB	0ug/ml (b)	ND	7.84	0.00	0.00	2	0.00	0.00	
Mean		0.00	7.72	0.00	0.00		0.00	0.00	
SD		0.00	0.18	0.00	0.00		0.00	0.00	
%CV		ERR	2.29	ERR	ERR		ERR	ERR	
N		2.00	2.00	2.00	2.00		2.00	2.00	

Xanthine 5 ng 09/02/15	34.64	8.66	5.00
Xanthine 5 ng 10/02/15	38.39	10.93	5.00
Xanthine 5 ng 11/02/15	37.35	10.23	5.00

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	5.35	0.00	5.35
5.00	0.36	0.00	0.36
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	0.0000
Std Err of Y Est	0.4950
R Squared	0.9726
No. of Observations	3.0000
Degrees of Freedom	2.0000

X Coefficient(s)	0.2085
Std Err of Coef.	0.0194

Sample ID	Time post-dos	Peak #	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c. (ug/mL)	Conc. (µM)	Conc. (µM) Corrected for dilution
T0 R373	0.00	INT	14.28	0.00	0.00	5	-	0.00	0.00	0.00
T0.08 R373	0.08	INT	23.76	0.00	0.00	5	-	0.00	0.00	0.00
T0.25 R373	0.25	INT	22.63	0.00	0.00	5	-	0.00	0.00	0.00
T1 R373	1.00	INT	23.36	0.00	0.00	5	-	0.00	0.00	0.00
Spiking level (30 mg/kg)										
T1.2 R373	1.20	INT	22.43	0.00	0.00	5	-	0.00	0.00	0.00
T1.5 R373	1.50	INT	23.24	0.00	0.00	5	-	0.00	0.00	0.00
T2 R373	2.00	INT	19.23	0.00	0.00	5	-	0.00	0.00	0.00
T3 R373	3.00	INT	19.41	0.00	0.00	5	-	0.00	0.00	0.00
T4 R373	4.00	INT	22.67	0.00	0.00	5	-	0.00	0.00	0.00
T5 R373	5.00	INT	20.00	0.00	0.00	5	-	0.00	0.00	0.00
T6 R373	6.00	INT	23.20	0.00	0.00	5	-	0.00	0.00	0.00
Mean		0.00	21.04	0.00	0.00			0.00	0.00	0.00
SD		0.00	3.25	0.00	0.00			0.00	0.00	0.00
%CV		ERR	15.42	ERR	ERR			ERR	ERR	ERR
n		8.00	8.00	8.00	8.00			8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 17/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 27/02/2015

Plasma Concentrations of Uric Acid in Rat 373
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 04/02/2015 - 11/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Uric Acid 5 ng		29.71				5		
a25	25ug/ml (a)	49.63	7.86	6.31	6.31	2	2505.72	90.25
b25	25ug/ml (b)	39.88	6.88	5.80	5.80	2	2013.46	70.56
Mean		44.76	7.37	6.06	6.06		2259.59	80.41
SD		6.89	0.69	0.37	0.37		348.08	13.92
%CV		15.40	9.40	6.05	6.05		15.40	17.32
N		2.00	2.00	2.00	2.00		2.00	2.00
a5	5ug/ml (a)	9.97	8.56	1.16	1.16	2	503.37	50.79
b5	5ug/ml (b)	9.55	8.90	1.07	1.07	2	482.16	46.55
	5ug/ml	10.33	9.95	1.04	1.04	2	521.54	54.43
	5ug/ml	10.41	7.86	1.32	1.32	2	525.58	55.23
	5ug/ml	8.91	8.12	1.10	1.10	2	449.85	40.09
Mean		9.83	8.68	1.14	1.09		496.50	49.42
SD		0.62	0.82	0.11	0.05		31.23	6.25
%CV		6.29	9.40	9.94	4.89		6.29	12.64
N		5.00	5.00	5.00	4.00		5.00	5.00
BLANKS:								
aB	0ug/ml (a)	3.34	7.59	0.44	0.44	2	168.63	
bB	0ug/ml (b)	6.54	7.84	0.83	0.83	2	330.19	
Mean		4.94	7.72	0.64	0.64		249.41	
SD		2.26	0.18	0.28	0.28		114.24	
%CV		45.80	2.29	43.74	43.74		45.80	
N		2.00	2.00	2.00	2.00		2.00	
Uric Acid 5 ng 09/02/2015		10.33	9.95				5.00	
Uric Acid 5 ng 10/02/2015		31.03	10.93				5.00	
Uric Acid 5 ng 11/02/2015		30.25	10.23				5.00	

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	6.06	0.64	5.42
5.00	1.09	0.64	0.46
0.00	0.64	0.64	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.2988
Std Err of Y Est	0.4841
R Squared	0.9870
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.2257
Std Err of Coef.	0.0259

Sample ID	Time post-dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	ic. (µg/mL); conc. (µM)	Conc. (µM) Corrected for dilution
T0 R373	0.00	22.54	14.28	1.58	1.58	5	-	8.32	49.48 65.97
T0.08 R373	0.08	15.86	23.76	0.67	0.67	5	-	4.28	25.47 33.96
T0.25 R373	0.25	6.27	22.63	0.23	0.23	5	-	2.36	14.01 18.68
T1 R373	1.00	26.86	23.36	1.15	1.15	5	-	6.42	38.18 50.91
Isoproterenol (30 mg/kg)									
T1.2 R373	1.20	4.89	22.43	0.22	0.22	5	-	2.29	16.82 22.43
T1.5 R373	1.50	11.04	23.24	0.48	0.48	5	-	3.43	25.19 33.59
T2 R373	2.00	33.33	19.23	1.73	1.73	5	-	9.00	53.56 71.41
T3 R373	3.00	29.01	19.41	1.49	1.49	5	-	7.95	47.27 63.03
T4 R373	4.00	7.99	22.67	0.35	0.35	5	-	2.89	17.17 22.89
T5 R373	5.00	20.98	20.00	1.05	1.05	5	-	5.97	35.53 47.37
T6 R373	6.00	5.23	23.20	0.23	0.23	5	-	2.32	13.82 18.42
Mean		18.60	21.04	0.94	0.94			5.51	33.75 45.00
SD		10.95	3.25	0.62	0.62			2.75	15.40 20.53
%CV		58.87	15.42	65.81	65.81			49.99	45.63 45.63
n		8.00	8.00	8.00	8.00			8.00	8.00 8.00

NOTE: QC samples were prepared with plasma from healthy rat No. 181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 17/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 27/02/2015

Plasma Concentrations of Guanosine in Rat 373
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 04/02/2015 - 11/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount Reco (ng)	Recovery (%)
Guanosine 5 ng								
a2.5	2.5ug/ml (a)	22.93	32.71	0.70	0.70	10	129.09	51.63
b2.5	2.5ug/ml (b)	25.34	36.38	0.70	0.70	10	142.65	57.06
Mean		24.14	34.55	0.70	0.70		135.87	54.35
SD		1.70	2.60	0.00	0.00		9.59	3.84
%CV		7.06	7.51	0.45	0.45		7.06	7.06
N		2.00	2.00	2.00	2.00		2.00	2.00
a0.5	0.5ug/ml (a)	4.11	83.97	0.05	0.05	30.00	7.71	15.43
b0.5	0.5ug/ml (b)	3.97	84.27	0.05	0.05	30.00	7.45	14.90
	0.5ug/ml	2.52	89.60	0.03	0.03	30.00	4.73	9.46
	0.5ug/ml	5.69	90.89	0.06	0.06	30.00	10.68	21.35
	0.5ug/ml	4.85	88.61	0.05	0.05	30.00	9.10	18.20
Mean		4.23	87.47	0.05	0.05		7.93	15.87
SD		1.17	3.16	0.01	0.01		2.20	4.41
%CV		27.79	3.62	26.49	26.49		27.79	27.79
N		5.00	5.00	5.00	5.00		5.00	5.00
BLANKS:								
aB	0ug/mL (a)	ND	7.59	0.00	0.00	2	0.00	
bB	0 ug/mL (b)	ND	7.84	0.00	0.00	2	0.00	
Mean		0.00	7.72	0.00	0.00		0.00	
SD		0.00	0.18	0.00	0.00		0.00	
%CV		ERR	2.29	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00		2.00	
*Guanosine 5 ng 09/02/15		50.40	8.66			5.00		
*Guanosine 5 ng 10/02/15		54.50	10.93			5.00		
*Guanosine 5 ng 11/02/15		53.36	10.23			5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.70	0.00	0.70
0.50	0.05	0.00	0.05
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0435
Std Err of Y Est	0.0706
R Squared	0.9837
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.2926
Std Err of Coef.	0.0377

Sample ID	Time post-dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	ic.(µg/mL)	onc.(µM)	Conc.(µM) Corrected for dilution
T0 R373	0.00	2.44	99.70	0.024	0.024	35	-	0.23	0.82	1.09
T0.08 R373	0.08	2.46	117.48	0.021	0.021	35	-	0.22	0.78	1.04
T0.25 R373	0.25	3.19	119.74	0.027	0.027	35	-	0.24	0.85	1.13
T1 R373	1.00	2.32	103.00	0.023	0.023	35	-	0.23	0.80	1.06
Isoproterenol (30 mg/kg)										
T1.2 R373	1.20	2.39	127.15	0.019	0.019	35	-	0.21	0.80	1.06
T1.5 R373	1.50	1.67	132.84	0.013	0.013	35	-	0.19	0.72	0.96
T2 R373	2.00	3.05	99.48	0.031	0.031	35	-	0.25	0.90	1.19
T3 R373	3.00	3.95	105.28	0.038	0.038	35	-	0.28	0.98	1.30
T4 R373	4.00	ND	124.02	0.000	0.000	35	-	0.15	0.53	0.70
T5 R373	5.00	ND	111.82	0.000	0.000	35	-	0.15	0.53	0.70
T6 R373	6.00	ND	127.96	0.000	0.000	35	-	0.15	0.53	0.70
Mean		1.95	115.32	0.02	0.02			0.21	0.75	1.00
SD		1.38	12.11	0.01	0.01			0.04	0.16	0.21
%CV		70.74	10.50	73.61	73.61			21.23	20.98	20.98
n		22.00	22.00	11.00	11.00			11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
 The Spiking solutions were made on: 22/02/2006
 *Repeat injections of QC a or b

Submitted by: Shyam Sundar Date: 17/02/2015

Checked by: Date:

Approved by: Polen Yeung Date:

Title: Measurement of Plasma Concentrations of Dipyridamole in Rat 373

According to SOP No: SOP/STD/2008-001-1 (Plasma with no Stopping Solution) Experiment Date:29/09/2014- 30/09/2014

Abs.amt ng	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Peak Ht. Ratio	Peak Ht. Ratio	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Dipyridamole (1ng)			86.18			1		
a1000	1 ug/mL(a)	99.31	3.36	29.56	29.56	5	46.09	92.19
b1000	1ug/mL(b)	100.10	2.76	36.27	36.27	5	46.46	92.92
1000*	1ug/mL(c)	93.91	2.96	31.73	31.73	5	43.59	87.18
Mean		97.77	3.03	32.52	32.52		45.38	90.76
SD		3.37	0.31	3.42	3.42		1.56	3.13
%CV		3.45	10.09	10.53	10.53		3.45	3.45
n		3.00	3.00	3.00	3.00		1.00	1.00
a100	0.1 ug/mL (a)	16.61	9.09	1.83	1.83	10	3.85	77.09
b100	0.1ug/mL (b)	26.19	13.97	1.87	1.87	20	3.04	60.78
Mean		21.40	11.53	1.85	1.85		3.45	68.94
SD		6.77	3.45	0.03	0.03		0.58	11.54
%CV		31.65	29.93	1.81	1.81		16.73	16.73
n		2.00	2.00	2.00	2.00		2.00	2.00
aB	0 ug/mL (a)	0.00	10.71	0.00	0.00	20	0.00	0.00
bB	0 ug/mL (b)	0.00	18.23	0.00	0.00	20	0.00	0.00
Mean		0.00	14.47	0.00	0.00		0.00	0.00
SD		0.00	5.32	0.00	0.00		0.00	0.00
%CV		ERR	36.75	ERR	ERR		ERR	ERR
n		2.00	2.00	2.00	2.00		2.00	2.00

Plasma Conc. (ug/mL)	Peak Ht.Ratio (PHR)	Blank (PHRb)	PHRV-PHRb
0.00	0.00	0.00	0.00
0.10	1.85	0.00	1.85
1.00	32.52	0.00	32.52

Regression Output Begins Here:

Regression Output:	
Constant	-0.6927
Std Err of Y Est	1.0383
R Squared	0.9984
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	33.1327
Std Err of Coef.	1.3330

Sample ID	Time Post-dose (h)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)*	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc.(ug/mL)
R373T0	0.00	2.45	17.66	0.14	0.14	20	-	0.39
R373T0.08	0.08	10.01	16.06	0.62	0.62	20	-	0.61
R373T0.25	0.25	12.34	19.37	0.64	0.64	20	-	0.62
R373T1	1.00	11.30	20.53	0.55	0.55	20	-	0.58
Metoprolol (30 mg/kg sc)						20		
R373T1.2	1.20	20.31	20.63	0.98	0.98	20	-	0.85
R373T1.5	1.50	11.56	14.33	0.81	0.81	20	-	0.70
R373T2	2.00	9.91	21.71	0.46	0.46	20	-	0.53
R373T3	3.00	5.80	11.95	0.49	0.49	20	-	0.55
R373T4	4.00	6.50	24.94	0.26	0.26	20	-	0.44
R373T5	5.00	2.60	16.90	0.15	0.15	20	-	0.39
R373T6	6.00	3.65	14.64	0.25	0.25	20	-	0.44
Mean		8.77	18.07	0.49	0.49			0.55
SD		5.30	3.79	0.27	0.27			0.14
%CV		60.48	20.97	55.81	55.81			25.47
n		11.00	11.00	11.00	11.00			11.00

Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 Corr. PHR = (PHR - RGB PHR)

Dipyridamole (1ng)/30/09/2014) 66.09 6.04 1.00

Comments: Plasma from Rat 156 was used for extraction QC's.
 *A repeat injection of a or b

Submitted by: Shyam Sundar Date: 01/10/2014

Checked by: Pollen Yeung Date:03/10/2014

Approved by: Date:

APPENDIX 4: Rat 374

Title: Measurement of RBC Concentrations of ATP in Rat 374 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
Experiment Date 04-05/06/2014

Sample/standard ID	Standard Concentra (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recov (µL)	% Recovery
ATP 4 ng										
a250	250 ug/mL	33.60	8.68	3.87	35.00	3.87	3.87	0.35	23783.78	81.30
b250	250 ug/mL	32.11	8.90	3.61	35.00	3.61	3.61	0.35	22729.09	77.08
Mean		32.86	8.79	3.74	35.00	3.74	3.74		23256.44	79.19
SD		1.85	0.16	0.19	0.00	0.19	0.19		745.78	2.98
%CV		3.21	1.77	4.98	0.00	4.98	4.98		3.21	3.77
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a100										
b100	100 ug/mL	14.83	9.40	1.59	35.00	1.59	1.59	0.35	10588.21	71.10
100'	100 ug/mL	19.01	8.37	2.27	35.00	2.27	2.27	0.35	13458.24	99.98
Mean		16.99	9.05	1.85	35.00	1.85	1.85		10883.20	74.25
SD		2.15	0.89	0.37	0.00	0.37	0.37		445.47	4.45
%CV		12.63	6.54	20.00	0.00	20.00	20.00		4.09	6.00
n		3.00	3.00	3.00	3.00	3.00	3.00		2.00	2.00
aB										
bB	0 ug/mL (a)	5.75	12.06	0.48	35.00	0.48	0.48	0.35	4070.14	
	0 ug/mL (a)	4.02	7.96	0.51	35.00	0.51	0.51	0.35	2845.56	
Mean		4.89	10.01	0.49	35.00	0.49	0.49		3457.85	
SD		1.22	2.90	0.02	0.00	0.02	0.02		865.91	
%CV		25.04	28.96	4.07	0.00	4.07	4.07		25.04	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
250.00	3.74	0.49	3.25
100.00	1.85	0.49	1.36
0.00	0.49	0.49	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0228
Std Err of Y Est	0.0489
R Squared	0.9996
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0130
Std Err of Coef.	0.0003

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R37470	0.00	94.06	92.91	1.01	35.00	1.01	1.01	1.50	-	76.33	0.1555
R37470.08	0.08	93.22	94.29	0.99	35.00	0.99	0.99	1.50	-	74.50	0.1469
R37470.25	0.25	102.85	93.11	1.10	35.00	1.10	1.10	1.50	-	83.45	0.1645
R37471	1.00	50.79	59.69	0.85	35.00	0.85	0.85	1.00	-	63.88	0.1259
Regression of (10 mg/kg set)											
R37471.2	1.20	38.07	34.18	1.11	35.00	1.11	1.11	1.00	-	84.10	0.1659
R37471.5	1.50	64.13	62.73	1.22	35.00	1.22	1.22	1.00	-	92.06	0.1815
R37472	2.00	72.23	59.18	1.22	35.00	1.22	1.22	1.00	-	92.39	0.1822
R37473	3.00	48.12	53.78	0.89	35.00	0.89	0.89	1.00	-	67.26	0.1326
R37474	4.00	31.56	43.18	0.73	35.00	0.73	0.73	1.00	-	54.62	0.1077
R37475	5.00	45.83	45.54	1.01	35.00	1.01	1.01	1.00	-	75.87	0.1456
R37476	6.00	50.89	53.37	0.95	35.00	0.95	0.95	1.00	-	71.79	0.1415
Mean		62.89	62.00	1.01	35.00	1.01	1.01	1.14		76.03	0.15
SD		24.44	21.46	0.15	0.00	0.15	0.15	0.23		11.61	0.02
%CV		38.87	34.62	14.93	0.00	14.93	14.93	20.55		15.28	15.28
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00		11.00	11.00

ATP (4 ng) 5/06/2014

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.
*Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
Peak Ht. = peak height
Peak Ht. R. (or: PHR) = peak height ratio
I.S. = internal standard
Inj Vol = injection volume
ND = not detected or determined
NS = no sample
INT = interference
PCV = packed cell volume (haematocrit)
CorPHR = corrected peak height ratio
Hemolysis Degree:
-: no visible hemolysis
+: slight hemolysis
++: intermediate hemolysis
+++: serious hemolysis

Submitted by: Shyam Sundar K Date: 16/06/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 14/07/2014

Title: Measurement of RBC Concentrations of ADP in Rat 374 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 04-05/06/2014

Sample/standard ID	Standard Co (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered	% Recovery
ADP 4 ng		21.82						4.00		
s250	250 µg/mL	58.14	8.68	6.70	35.00	6.70	6.70	0.35	16748.46	64.41
b250	250 µg/mL	54.72	8.90	6.15	35.00	6.15	6.15	0.35	15763.26	60.47
Mean		56.43	8.79	6.42	35.00	6.42			16255.86	62.44
SD		2.42	0.16	0.39	0.00	0.39			696.64	2.79
%CV		4.29	1.77	6.05	0.00	6.05			4.29	4.46
n		2.00	2.00	2.00	2.00	2.00			2.00	2.00
100a	100 µg/mL	23.96	9.39	2.55	35.00	2.55	2.55	0.35	6902.19	62.57
100b	100 µg/mL	22.83	9.40	2.43	35.00	2.43	2.43	0.35	6576.67	59.31
100*	100 µg/mL	21.53	8.37	2.57	35.00	2.57	2.57	0.35	6202.17	55.57
Mean		22.77	9.05	2.52	35.00	2.52			6560.34	59.15
SD		1.22	0.59	0.08	0.00	0.08			350.29	3.50
%CV		5.34	6.54	3.08	0.00	3.08			5.34	5.92
n		3.00	3.00	3.00	3.00	3.00			3.00	3.00
aB	0 µg/mL (a)	2.54	12.06	0.21	35.00	0.21	0.21	0.35	731.70	
bB	0 µg/mL (a)	1.94	7.96	0.24	35.00	0.24	0.24	0.35	558.86	
Mean		2.24	10.01	0.23	35.00	0.23			645.28	
SD		0.42	2.90	0.02	0.00	0.02			122.22	
%CV		18.94	28.96	10.30	0.00	10.30			18.94	
n		2.00	2.00	2.00	2.00	2.00			2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
250.00	6.42	0.23	6.20
100.00	2.52	0.23	2.29
0.00	0.23	0.23	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0742
Std Err of Y Est	0.1525
R Squared	0.9988
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0249
Std Err of Coef.	0.0009

Sample ID	Time post	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. Hemolysis (µL) Degree	Conc(µg/mL) Lysate	Conc.(mM) Lysate	Conc.(mM) RBC
R37470	0.00	28.41	92.91	0.31	35.00	0.31	0.31	1.50 -	15.27	0.0357	0.460
R37470B	0.06	22.23	94.29	0.24	35.00	0.24	0.24	1.50 -	12.46	0.0292	0.375
R37470.25	0.25	20.67	93.11	0.22	35.00	0.22	0.22	1.50 -	11.90	0.0279	0.358
R37471	1.00	16.61	99.69	0.28	35.00	0.28	0.28	1.00 -	14.17	0.0332	0.426
Isoproterenol (30 mg/kg sc)											
R37471.2	1.20	6.66	34.18	0.19	35.00	0.19	0.19	1.00 -	10.81	0.0253	0.325
R37471.5	1.50	10.96	52.73	0.21	35.00	0.21	0.21	1.00 -	11.34	0.0269	0.341
R37472	2.00	12.13	59.18	0.20	35.00	0.20	0.20	1.00 -	11.22	0.0263	0.338
R37473	3.00	7.81	53.78	0.15	35.00	0.15	0.15	1.00 -	8.82	0.0206	0.265
R37474	4.00	4.95	43.18	0.11	35.00	0.11	0.11	1.00 -	7.59	0.0178	0.228
R37475	5.00	6.29	45.54	0.14	35.00	0.14	0.14	1.00 -	8.53	0.0200	0.257
R37476	6.00	6.71	53.37	0.13	35.00	0.13	0.13	1.00 -	8.04	0.0188	0.242
Mean		23.77	93.44	0.25	35.00	0.25		1.50	13.21	0.03	0.40
SD		4.09	0.75	0.04	0.00	0.04		0.00	1.81	0.00	0.05
%CV		17.22	0.80	17.65	0.00	17.65		0.00	13.67	13.67	13.67
n		3.00	3.00	3.00	3.00	3.00		3.00	3.00	2.00	3.00

ADP (4 ng) 05/06/2014 22.39

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5-1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 16/06/2014
 Checked by: Date:
 Approved by: Pollen Yeung Date: 14/07/2014

Title: Measurement of RBC Concentrations of AMP in Rat 374 extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date 04-05/06/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
AMP 4 ng										
		5254						4.00		
a50	50 ug/mL	32.80	8.68	3.78	35.00	3.78	3.78	0.35	3924.09	76.56
b50	50 ug/mL	31.20	8.90	3.51	35.00	3.51	3.51	0.35	3732.67	72.73
Mean		32.00	8.79	3.64	35.00	3.64	3.64		3828.38	74.64
SD		1.13	0.16	0.19	0.00	0.19	0.19		135.35	2.71
%CV		3.54	1.77	5.30	0.00	5.30	5.30		3.54	3.63
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20	20 ug/mL	12.89	9.39	1.37	35.00	1.37	1.37	0.35	1542.12	72.29
b20	20 ug/mL	12.01	9.40	1.28	35.00	1.28	1.28	0.35	1436.84	67.03
20*	20 ug/mL	11.37	8.37	1.36	35.00	1.36	1.36	0.35	1360.27	63.20
Mean		12.89	9.05	1.34	35.00	1.34	1.34		1446.41	67.51
SD		0.78	0.59	0.05	0.00	0.05	0.05		91.30	4.57
%CV		6.31	6.54	3.84	0.00	3.84	3.84		6.31	6.76
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB	0 ug/mL (a)	0.96	12.06	0.08	35.00	0.08	0.08	0.35	114.85	
bB	0 ug/mL (a)	0.65	7.96	0.08	35.00	0.08	0.08	0.35	77.76	
Mean		0.81	10.01	0.08	35.00	0.08	0.08		96.31	
SD		0.22	2.90	0.00	0.00	0.00	0.00		26.22	
%CV		27.23	28.96	1.80	0.00	1.80	1.80		27.23	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	3.64	0.08	3.56
20.00	1.34	0.08	1.26
0.00	0.08	0.08	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0667
Std Err of Y Est	0.1371
R Squared	0.9971
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0717
Std Err of Coef.	0.0039

Sample ID	Time post dose	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL)	Conc.(mM) Lysate	Conc.(mM) RBC
R37470	0.00	19.68	92.91	0.21	35.00	0.21	0.21	1.50	-	3.89	0.0112	0.144
R37470.08	0.08	2.06	94.29	0.02	35.00	0.02	0.02	1.50	-	1.24	0.0036	0.046
R37470.25	0.25	1.91	93.11	0.02	35.00	0.02	0.02	1.00	-	1.22	0.0035	0.045
R37471	1.00	3.15	59.69	0.05	35.00	0.05	0.05	1.00	-	1.67	0.0048	0.062
Soprolefenol (30 mg/kg sc)												
R37471.2	1.20	0.70	34.18	0.02	35.00	0.02	0.02	1.00	-	1.22	0.0035	0.045
R37471.5	1.50	0.64	52.73	0.01	35.00	0.01	0.01	1.00	-	1.10	0.0032	0.041
R37472	2.00	0.53	59.18	0.01	35.00	0.01	0.01	1.00	-	1.06	0.0030	0.039
R37473	3.00	0.68	53.78	0.01	35.00	0.01	0.01	1.00	-	1.11	0.0032	0.041
R37474	4.00	0.56	43.18	0.01	35.00	0.01	0.01	1.00	-	1.11	0.0032	0.041
R37475	5.00	0.68	45.54	0.01	35.00	0.01	0.01	1.00	-	1.14	0.0033	0.042
R37476	6.00	ND	53.37	0.00	35.00	0.00	0.00	1.00	-	0.93	0.0027	0.034
Mean		2.78	62.00	0.04	35.00	0.04	0.04	1.09	0.00	1.42	0.00	0.05
SD		5.68	21.46	0.06	0.00	0.06	0.06	0.20	0.00	0.84	0.00	0.03
%CV		204.20	34.62	169.60	0.00	166.60	166.60	18.54	0.00	58.77	58.77	58.77
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

AMP (4 ng)/05/06/2014

5416

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of "a" or "b" at 0.5 - 1 ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar

Date: 16/06/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date: 14/07/2014

Title: Measurement of RBC Concentrations of GTP in Rat 374 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 04-05/06/2014

Sample/standard ID	Standard Concentra (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recove (µL)	% Recovery
GTP 4 ng		85.08						4.00		
a50	50 ug/mL	11.28	8.68	1.30	35.00	1.30	1.30	0.35	833.37	15.29
b50	50 ug/mL	10.36	8.90	1.16	35.00	1.16	1.16	0.35	765.40	13.93
Mean		10.62	8.79	1.23	35.00	1.23	1.23		799.38	14.61
SD		0.65	0.10	0.00	0.00	0.10	0.10		48.06	0.96
%CV		6.01	0.00	7.78	0.00	7.78	7.78		6.01	6.58
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20	20 ug/mL	4.46	8.39	0.47	35.00	0.47	0.47	0.35	329.51	13.02
b20	20 ug/mL	4.59	9.40	0.48	35.00	0.48	0.48	0.35	338.11	13.50
20*	20 ug/mL	3.79	8.37	0.45	35.00	0.45	0.45	0.35	280.01	10.55
Mean		4.28	9.05	0.47	35.00	0.47	0.47		316.21	12.36
SD		0.43	0.59	0.02	0.00	0.02	0.02		31.72	1.59
%CV		10.03	6.54	3.80	0.00	3.80	3.80		10.03	12.83
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
a8	0 ug/mL (a)	0.84	12.06	0.07	35.00	0.07	0.07	0.35	62.06	
b8	0 ug/mL (a)	1.53	7.96	0.13	35.00	0.13	0.13	0.35	78.10	
Mean		0.94	10.01	0.10	35.00	0.10	0.10		69.08	
SD		0.13	2.90	0.04	0.00	0.03	0.04		9.93	
%CV		14.37	28.96	42.45	0.00	34.66	42.45		14.37	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	1.23	0.10	1.13
20.00	0.47	0.10	0.37
0.00	0.10	0.10	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0317
Std Err of Y Est	0.0652
R Squared	0.9936
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0229
Std Err of Coef.	0.0018

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R37470	0.00	17.47	92.91	0.19	35.00	0.19	0.19	1.50	-	9.62	0.0194
R37470.08	0.08	18.31	94.29	0.19	35.00	0.19	0.19	1.50	-	9.88	0.0189
R37470.25	0.25	20.16	93.11	0.22	35.00	0.22	0.22	1.50	-	10.86	0.0208
R37471	1.00	8.86	59.69	0.15	35.00	0.15	0.15	1.00	-	7.88	0.0151
Suprofenol (30 mg/kg sc)											
R37471.2	1.20	8.11	34.16	0.24	35.00	0.24	0.24	1.00	-	11.77	0.0225
R37471.5	1.50	13.33	62.73	0.25	35.00	0.25	0.25	1.00	-	12.45	0.0238
R37472	2.00	15.43	59.18	0.26	35.00	0.26	0.26	1.00	-	12.80	0.0245
R37473	3.00	10.04	53.78	0.19	35.00	0.19	0.19	1.00	-	9.56	0.0183
R37474	4.00	6.25	43.18	0.14	35.00	0.14	0.14	1.00	-	7.72	0.0148
R37475	5.00	10.10	45.54	0.22	35.00	0.22	0.22	1.00	-	11.09	0.0212
R37476	6.00	11.75	53.37	0.22	35.00	0.22	0.22	1.00	-	11.02	0.0211
Mean		12.71	62.00	0.21	35.00	0.21	0.21	1.14	0.00	10.42	0.02
SD		4.58	21.46	0.04	0.00	0.04	0.04	0.23	0.00	1.67	0.00
%CV		36.04	34.62	18.52	0.00	18.52	18.52	20.55	0.00	16.05	16.05
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

GTP (4 ng) 05/06/2014

88.27

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 16/06/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 14/07/2014

Title: Measurement of RBC Concentrations of GDP in Rat 374 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 04-05/06/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recove %	Recovery
GDP 4 ng		31.37						4.00		
a50	50 ug/mL	16.98	8.68	1.96	35.00	1.96	1.96	3402.34	64.90	
b50	50 ug/mL	16.21	8.90	1.82	35.00	1.82	1.82	3248.05	61.82	
Mean		16.60	8.79	1.89	35.00	1.89	1.89	3325.20	63.36	
SD		0.54	0.16	0.10	0.00	0.10	0.10	108.10	2.18	
%CV		3.28	1.77	5.05	0.00	5.05	5.05	3.28	3.44	
n		2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	
a20	20 ug/mL	6.75	9.39	0.72	35.00	0.72	0.72	1353.53	59.78	
b20	20 ug/mL	6.75	9.40	0.72	35.00	0.72	0.72	1352.52	59.78	
20*	20 ug/mL	6.14	8.37	0.73	35.00	0.73	0.73	1230.29	53.65	
Mean		6.55	9.05	0.72	35.00	0.72	0.72	1311.78	57.72	
SD		0.35	0.59	0.01	0.00	0.01	0.01	70.57	3.53	
%CV		5.38	6.54	1.21	0.00	1.21	1.21	5.38	6.11	
n		3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	
aB	0 ug/mL (a)	0.88	12.06	0.07	35.00	0.07	0.07	0.35	176.33	
bB	0 ug/mL (a)	0.69	7.96	0.09	35.00	0.09	0.09	0.35	138.26	
Mean		0.79	10.01	0.08	35.00	0.08	0.08		157.29	
SD		0.13	2.90	0.01	0.00	0.01	0.01		26.92	
%CV		17.11	28.96	12.15	0.00	12.15	12.15		17.11	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRb
50.00	1.89	0.08	1.81
20.00	0.72	0.08	0.64
0.00	0.08	0.08	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0315
Std Err of Y Est	0.0648
R Squared	0.9975
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0094
Std Err of Coef.	0.0018

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL)	Conc.(nM) Lysate	Conc.(nM) RBC
R37470	0.00	7.21	92.91	0.08	35.00	0.08	0.08	1.50	-	3.00	0.0088	0.087
R37470.08	0.08	6.48	94.29	0.07	35.00	0.07	0.07	1.50	-	2.76	0.0082	0.080
R37470.25	0.25	6.58	93.11	0.07	35.00	0.07	0.07	1.50	-	2.81	0.0083	0.081
R37471	1.00	4.06	59.69	0.07	35.00	0.07	0.07	1.00	-	2.74	0.0082	0.079
Isoproterenol (30 µg/kg sc)												
R37471.2	1.20	2.90	34.18	0.08	35.00	0.08	0.08	1.00	-	3.20	0.0072	0.093
R37471.5	1.50	3.69	52.73	0.07	35.00	0.07	0.07	1.00	-	2.79	0.0083	0.081
R37472	2.00	4.38	59.18	0.07	35.00	0.07	0.07	1.00	-	2.50	0.0085	0.084
R37473	3.00	3.35	53.78	0.06	35.00	0.06	0.06	1.00	-	2.58	0.0058	0.075
R37474	4.00	1.98	43.18	0.05	35.00	0.05	0.05	1.00	-	2.13	0.0048	0.052
R37475	5.00	1.86	45.64	0.04	35.00	0.04	0.04	1.00	-	1.99	0.0045	0.058
R37476	6.00	2.64	53.37	0.05	35.00	0.05	0.05	1.00	-	2.23	0.0050	0.065
Mean		33.05	62.00	0.06	35.00	0.06	0.06	1.14	0.00	2.65	0.01	0.08
SD		33.15	21.46	0.01	0.00	0.01	0.01	0.23	0.00	0.38	0.00	0.01
%CV		100.30	34.62	21.35	0.00	21.26	21.26	20.55	ERR	14.26	14.36	14.26
n		22.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

GDP (4 ng) 05/06/2014

32.38

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K

Date:16/06/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date:14/07/2014

Title: Measurement of RBC Concentrations of GMP in Rat 374 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 04-05/06/2014

Sample/standard	ID/Standard Concentr (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (µL)	% Recovery
GMP 4 ng										
a50	50 ug/mL	Off scale	8.68	0.00	35.00	0.00	0.00	0.35	0.00	-95.23
b50	50 ug/mL	Off scale	8.90	0.00	35.00	0.00	0.00	0.35	0.00	-95.23
Mean		0.00	8.79	0.00	35.00	0.00	0.00		0.00	-95.23
SD		0.00	8.85	0.00	0.00	0.00	0.00		0.00	0.00
%CV		ERR	8.82	ERR	0.00	ERR	ERR		ERR	-0.00
n		2.00	8.82	2.00	2.00	2.00	2.00		2.00	2.00
a20										
b20	20 ug/mL	105.05	9.39	11.19	35.00	11.19	11.19	0.35	5400.46	31.94
20*	20 ug/mL	95.75	9.40	10.19	35.00	10.19	10.19	0.35	4922.36	8.03
		69.08	8.37	8.25	35.00	8.25	8.25	0.35	3551.30	-60.52
Mean		89.96	9.05	9.88	35.00	9.88	9.88		4624.71	-6.85
SD		18.67	0.59	1.49	0.00	1.49	1.49		959.84	47.99
%CV		20.75	6.54	15.10	0.00	15.10	15.10		20.75	-700.60
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB										
bB	0 ug/mL (a)	112.42	12.06	9.32	35.00	9.32	9.32	0.35	5779.34	
	0 ug/mL (a)	72.83	7.96	9.15	35.00	9.15	9.15	0.35	3744.08	
Mean		92.63	10.01	9.24	35.00	9.24	9.24		4761.71	
SD		27.99	2.90	0.12	0.00	0.12	0.12		1438.15	
%CV		30.22	28.96	1.32	0.00	1.32	1.32		30.22	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	0.00	9.24	-9.24
20.00	9.88	9.24	0.64
0.00	9.24	9.24	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	2.8456
R Squared	0.7349
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	-0.1548
Std Err of Coef.	0.0528

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. Hemolysis (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R37470		0.00 off scale	92.91	0.00	35.00	0.00	0.00	1.50	-	-0.00	-0.0000
R37470.08		0.08 off scale	94.29	0.00	35.00	0.00	0.00	1.50	-	-0.00	-0.0000
R37470.25		0.25 off scale	93.11	0.00	35.00	0.00	0.00	1.50	-	-0.00	-0.0000
R37471		1.00 off scale	59.69	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
Not detected (50 mg/kg sc)											
R37471.2		1.20 off scale	34.18	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
R37471.5		1.50 off scale	52.73	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
R37472		2.00 off scale	59.18	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
R37473		3.00 off scale	53.78	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
R37474		4.00 off scale	43.18	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
R37475		5.00 off scale	45.54	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
R37476		6.00 off scale	53.37	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
Mean		0.00	82.00	0.00	35.00	0.00	0.00	1.14		0.00	0.00
SD		0.00	21.46	0.00	0.00	0.00	0.23			0.00	0.00
%CV		ERR	34.62	ERR	0.00	ERR	ERR	20.55		ERR	ERR
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00		11.00	11.00

GMP (4ng) 05/06/2014 86.77

Comments: RBC Lysate from Rat 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of "a" or "b" at 0.5-1ul injection volume

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 16/06/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 14/07/2014

Plasma Concentrations of Adenosine in Rat 374
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 19/02/2015 - 25/02/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Adenosine 5 ng			29.55						
a2.5	2.5ug/ml (a)	16.73	29.66	0.56	0.56	10	169.85	67.94	
b2.5	2.5ug/ml (b)	16.05	37.57	0.43	0.43	10	162.94	65.18	
Mean		16.39	33.62	0.50	0.50		166.40	66.56	
SD		0.48	5.59	0.10	0.10		4.88	1.95	
%CV		2.93	16.64	19.53	19.53		2.93	2.93	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a0.5	0.5ug/ml (a)	6.73	84.61	0.08	0.08	30.00	22.77	45.55	
b0.5	0.5ug/ml (b)	4.75	76.24	0.06	0.06	30.00	16.07	32.15	
	0.5 ug/ml	7.40	84.50	0.09	0.09	30.00	25.04	50.08	
	0.5 ug/ml	8.08	82.78	0.10	0.10	30.00	27.34	54.69	
	0.5 ug/ml	8.12	83.74	0.10	0.10	30.00	27.48	54.96	
	0.5 ug/ml	8.04	83.93	0.10	0.10	30.00	27.21	54.42	
Mean		7.19	82.63	0.09	0.09		24.32	48.64	
SD		1.31	3.20	0.01	0.01		4.44	8.87	
%CV		18.24	3.87	15.95	15.95		18.24	18.24	
N		6.00	6.00	6.00	6.00		6.00	6.00	
BLANKS:									
aB	0ug/mL (a)	N/D	8.81	0.00	0.00	2	0.00		
bB	0 ug/mL (b)	N/D	8.09	0.00	0.00	2	0.00		
Mean		0.00	8.45	0.00	0.00		0.00		
SD		0.00	0.51	0.00	0.00		0.00		
%CV		ERR	6.03	ERR	ERR		ERR		
N		2.00	2.00	2.00	2.00		2.00		
Adenosine 5 ng 20/02/2015		31.91	10.87			5.00			
Adenosine 5 ng 23/02/2015		31.20	10.32			5.00			
Adenosine 5 ng 24/02/2015		30.73	10.01			5.00			
Adenosine 5 ng 25/02/2015		31.69	10.63			5.00			

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHRV Value (PHRV)	Blank (PHRB)	PHRV-PHRB
2.50	0.50	0.00	0.50
0.50	0.09	0.00	0.09
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	0.0000
Std Err of Y Est	0.0087
R Squared	0.9969
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.1973
Std Err of Coef.	0.0034

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	Conc.(µM)	Conc.(µM) Corrected for dilution
T0 R374	0.00	ND	113.08	0.00	0.00	0.00	35	-	0.00	0.00	0.00
T0.08 R374	0.08	2.45	80.73	0.03	0.03	0.03	35	-	0.15	0.58	0.77
T0.25 R374	0.25	2.90	84.28	0.03	0.03	0.03	35	-	0.17	0.65	0.87
T1 R374	1.00	2.92	82.73	0.04	0.04	0.04	35	-	0.18	0.67	0.89
Isoproterenol (30 mg/kg)											
T1.2 R374	1.20	7.27	110.56	0.07	0.07	0.07	35	-	0.33	1.25	1.66
T1.5R374	1.50	5.40	124.50	0.04	0.04	0.04	35	-	0.22	0.82	1.10
T2 R374	2.00	3.88	89.17	0.04	0.04	0.04	35	-	0.22	0.83	1.10
T3 R374	3.00	5.38	127.53	0.04	0.04	0.04	35	-	0.21	0.80	1.07
T4 R374	4.00	2.62	75.40	0.03	0.03	0.03	35	-	0.18	0.66	0.88
T5 R374	5.00	5.29	112.16	0.05	0.05	0.05	35	-	0.24	0.89	1.19
T6 R374	6.00	5.50	125.80	0.04	0.04	0.04	35	-	0.22	0.83	1.11
Mean		3.96	102.36	0.04	0.04				0.19	0.73	0.97
SD		2.03	20.08	0.02	0.02				0.08	0.30	0.40
%CV		51.17	19.62	41.33	41.33				41.33	41.33	41.33
n		11.00	11.00	11.00	11.00				11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
 The Spiking solutions were made on: 22/02/2006
 *Repeat injections of QC a or b

Submitted by: Shyam Sundar Date: 03/03/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/04/2015

Plasma Concentrations of Inosine in Rat 374
 Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
 Experiment Date: 19/02/2015 - 25/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Rec. (%)	Recovery (%)
Inosine 5 ng		43.64				5			
a2.5	2.5ug/ml (a)	29.29	29.66	0.99	0.99	10	201.35	80.54	
b2.5	2.5ug/ml (b)	27.31	37.57	0.73	0.73	10	187.74	75.10	
Mean		28.30	33.62	0.86	0.86		194.55	77.82	
SD		1.40	5.59	0.18	0.18		9.62	3.85	
%CV		4.95	16.64	21.50	21.50		4.95	4.95	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a0.5	0.5ug/ml (a)	7.85	84.61	0.09	0.09	30.00	17.99	35.98	
b0.5	0.5ug/ml (b)	7.48	76.24	0.10	0.10	30.00	17.14	34.28	
	0.5ug/ml	4.16	84.50	0.05	0.05	30.00	9.53	19.07	
	0.5ug/ml	7.38	82.78	0.09	0.09	30.00	16.91	33.82	
	0.5ug/ml	5.06	83.74	0.06	0.06	30.00	11.59	23.19	
	0.5ug/ml	9.45	83.93	0.11	0.11	30.00	21.65	43.31	
Mean		6.90	82.63	0.08	0.10		15.80	31.61	
SD		1.94	3.20	0.02	0.01		4.45	8.90	
%CV		28.16	3.87	28.69	10.50		28.16	28.16	
N		6.00	6.00	6.00	4.00		6.00	6.00	
BLANKS:									
aB	0ug/mL (a)	ND	8.81	0.00	0.00	2	0.00		
bB	0ug/mL (b)	ND	8.09	0.00	0.00	2	0.00		
Mean		0.00	8.45	0.00	0.00		0.00		
SD		0.00	0.51	0.00	0.00		0.00		
%CV		ERR	6.03	ERR	ERR		ERR		
N		2.00	2.00	2.00	2.00		2.00		
Inosine 5ng 20/02/15		43.40	10.87			5.00			
Inosine 5ng 23/02/15		43.14	10.32			5.00			
Inosine 5ng 24/02/15		41.22	10.01			5.00			
Inosine 5ng 25/02/15		44.35	10.63			5.00			

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.86	0.00	0.86
0.50	0.10	0.00	0.10
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0349
Std Err of Y Est	0.0565
R Squared	0.9927
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3534
Std Err of Coef.	0.0302

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	onc.(µM)	Conc.(µM) Corrected for dilution
T0 R374	0.00	9.79	113.08	0.09	0.09	35	-	0.34	1.28	1.71
T0.08 R374	0.08	5.71	80.73	0.07	0.07	35	-	0.30	1.11	1.49
T0.25 R374	0.25	8.03	84.28	0.10	0.10	35	-	0.37	1.37	1.83
T1 R374	1.00	5.38	82.73	0.07	0.07	35	-	0.28	1.05	1.41
Isoproterenol (30 mg/kg)										
T1.2 R374	1.20	5.30	110.56	0.05	0.05	35	-	0.23	0.87	1.17
T1.5R374	1.50	6.34	124.50	0.05	0.05	35	-	0.24	0.91	1.21
T2 R374	2.00	7.99	89.17	0.09	0.09	35	-	0.35	1.31	1.75
T3 R374	3.00	7.81	127.53	0.06	0.06	35	-	0.27	1.01	1.35
T4 R374	4.00	8.74	75.40	0.12	0.12	35	-	0.43	1.59	2.12
T5 R374	5.00	12.41	112.16	0.11	0.11	35	-	0.41	1.54	2.05
T6 R374	6.00	12.83	125.80	0.10	0.10	35	-	0.39	1.44	1.93
Mean		8.21	102.36	0.08	0.08			0.33	1.23	1.64
SD		2.61	20.08	0.02	0.02			0.07	0.25	0.33
%CV		31.81	19.62	29.04	29.04			20.33	20.33	20.33
n		11.00	11.00	11.00	11.00			11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
 The Spiking solutions were made on: 22/02/2006
 *Repeat injections of QC a or b

Submitted by: Shyam Sundar Date: 04/03/2015
 Checked by: Date:
 Approved by: Pollen Yeung Date: 10/04/2015

Plasma Concentrations of Hypoxanthine in Rat 374
 Based on "SOP NO. - SOP/STD/2004-001-0" (With Stopping Solution)
 Experiment Date: 19/02/2015 - 25/02/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recr (%)	Recovery (%)
Hypoxanthine 5 ng			109.86				5			
a25	25ug/ml (a)	120.42	7.40	16.27	16.27	16.27	2	1644.18	65.77	
b25	25ug/ml (b)	122.45	8.59	14.25	14.25	14.25	2	1671.90	66.88	
Mean		121.44	8.00	15.26	15.26	15.26		1658.04	66.32	
SD		1.44	0.84	1.43	1.43	1.43		19.60	0.78	
%CV		1.18	10.52	9.35	9.35	9.35		1.18	1.18	
N		2.00	2.00	2.00	2.00	2.00		2.00	2.00	
a5	5ug/ml (a)	16.30	10.21	1.60	1.60	1.60	2	222.56	44.51	
b5	5ug/ml (b)	13.21	7.83	1.69	1.69	1.69	2	180.37	36.07	
5ug/ml		13.80	9.41	1.47	1.47	1.47	2	188.42	37.68	
5ug/ml		14.01	8.65	1.58	1.58	1.58	2	191.29	38.26	
5ug/ml		16.13	9.62	1.68	1.68	1.68	2	220.23	44.05	
5ug/ml		13.53	10.07	1.34	1.34	1.34	2	184.74	36.95	
Mean		14.50	9.33	1.56	1.56	1.56		197.93	39.59	
SD		1.36	0.88	0.13	0.13	0.13		18.55	3.71	
%CV		9.37	9.46	8.47	8.47	8.47		9.37	9.37	
N		6.00	6.00	6.00	6.00	6.00		6.00	6.00	
BLANKS:										
aB	0ug/ml (a)	ND	8.81	0.00	0.00	0.00	2	0.00		
bB	0ug/ml (b)	ND	8.09	0.00	0.00	0.00	2	0.00		
Mean		0.00	8.45	0.00	0.00	0.00		0.00		
SD		0.00	0.51	0.00	0.00	0.00		0.00		
%CV		ERR	6.03	ERR	ERR	ERR		ERR		
N		2.00	2.00	2.00	2.00	2.00		2.00		
Hypoxanthine 5 ng 20/02/15		107.74	10.87					5.00		
Hypoxanthine 5 ng 23/02/15		108.67	10.32					5.00		
Hypoxanthine 5 ng 24/02/15		105.73	10.01					5.00		
Hypoxanthine 5 ng 25/02/15		110.76	10.63					5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	15.26	0.00	15.26
5.00	1.56	0.00	1.56
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.7114
Std Err of Y Est	1.1526
R Squared	0.9906
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.6319
Std Err of Coef.	0.0616

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(ug/mL)	Conc.(µM)	Conc.(µM) Corrected for dilution
T0 R374	0.00	13.52	19.52	0.69	0.69	0.69	5	-	2.22	16.33	21.77
T0.08 R374	0.08	9.65	17.10	0.56	0.56	0.56	5	-	2.02	14.83	19.78
T0.25 R374	0.25	13.60	19.13	0.71	0.71	0.71	5	-	2.25	16.54	22.05
T1 R374	1.00	11.72	15.66	0.75	0.75	0.75	5	-	2.31	16.97	22.63
Isoproterenol (30 mg/kg)											
T1.2 R374	1.20	14.59	23.12	0.63	0.63	0.63	5	-	2.12	15.61	20.81
T1.5R374	1.50	16.05	22.15	0.72	0.72	0.72	5	-	2.27	16.70	22.26
T2 R374	2.00	10.79	18.48	0.58	0.58	0.58	5	-	2.05	15.06	20.08
T3 R374	3.00	15.02	23.24	0.65	0.65	0.65	5	-	2.15	15.79	21.05
T4 R374	4.00	9.50	13.79	0.69	0.69	0.69	5	-	2.22	16.28	21.71
T5 R374	5.00	15.77	22.86	0.69	0.69	0.69	5	-	2.22	16.29	21.72
T6 R374	6.00	16.79	23.81	0.71	0.71	0.71	5	-	2.24	16.47	21.96
Mean		13.12	19.80	0.66	0.66	0.66			2.17	15.98	21.30
SD		2.21	2.81	0.07	0.07	0.07			0.11	0.78	1.04
%CV		16.84	14.17	10.13	10.13	10.13			4.89	4.89	4.89
n		8.00	8.00	8.00	8.00	8.00			8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 04/03/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/04/2015

Plasma Concentrations of Xanthine in Rat 374
Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
Experiment Date: 19/02/2015 -25/02/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Rec:Recovery (%)
Xanthine 5 ng		37.40					5		
a25	25ug/ml (a)	61.94	7.40	8.37	8.37		2	2484.22	99.37
b25	25ug/ml (b)	55.25	8.59	6.43	6.43		2	2215.91	88.64
Mean		58.60	8.00	7.40	7.40			2350.07	94.00
SD		4.73	0.84	1.37	1.37			189.73	7.59
%CV		8.07	10.52	18.52	18.52			8.07	8.07
N		2.00	2.00	2.00	2.00			2.00	2.00
a5	5ug/ml (a)	3.71	10.21	0.36	0.36		2	148.80	29.76
b5	5ug/ml (b)	3.59	7.83	0.46	0.46		2	143.98	28.80
	5ug/ml	3.04	9.41	0.32	0.32		2	121.93	24.39
	5ug/ml	4.50	8.85	0.51	0.51		2	180.48	36.10
	5ug/ml	4.92	9.62	0.51	0.51		2	197.33	39.47
	5ug/ml	4.75	10.07	0.47	0.47		2	190.51	38.10
Mean		4.09	9.33	0.44	0.44			163.84	32.77
SD		0.75	0.88	0.08	0.08			29.96	5.99
%CV		18.28	9.46	17.83	17.83			18.28	18.28
N		6.00	6.00	6.00	6.00			6.00	6.00
BLANKS:									
aB	0ug/ml (a)	ND	8.81	0.00	0.00		2	0.00	
bB	0ug/ml (b)	ND	8.09	0.00	0.00		2	0.00	
Mean		0.00	8.45	0.00	0.00			0.00	
SD		0.00	0.51	0.00	0.00			0.00	
%CV		ERR	6.03	ERR	ERR			ERR	
N		2.00	2.00	2.00	2.00			2.00	

Xanthine 5 ng 20/02/15	38.72	10.87	5.00
Xanthine 5 ng 23/02/15	39.70	10.32	5.00
Xanthine 5 ng 20/02/15	37.82	10.01	5.00
Xanthine 5 ng 23/02/15	39.97	10.69	5.00

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	7.40	0.00	7.40
5.00	0.44	0.00	0.44
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	0.0000
Std Err of Y Est	0.7217
R Squared	0.9698
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.2880
Std Err of Coef.	0.0283

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	Conc.(µM)	Conc.(µM) Corrected for dilution
T0 R374	0.00	INT	19.52	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T0.08 R374	0.08	INT	17.10	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T0.25 R374	0.25	INT	19.13	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T1 R374	1.00	INT	15.66	0.00	0.00	0.00	5	-	0.00	0.00	0.00
Isoproterenol (30 mg/kg)											
T1.2 R374	1.20	INT	23.12	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T1.5R374	1.50	INT	22.15	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T2 R374	2.00	INT	18.48	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T3 R374	3.00	INT	23.24	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T4 R374	4.00	INT	13.79	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T5 R374	5.00	INT	22.86	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T6 R374	6.00	INT	23.81	0.00	0.00	0.00	5	-	0.00	0.00	0.00
Mean		0.00	19.80	0.00	0.00	0.00			0.00	0.00	0.00
SD		0.00	2.81	0.00	0.00	0.00			0.00	0.00	0.00
%CV		ERR	14.17	ERR	ERR	ERR			ERR	ERR	ERR
n		8.00	8.00	8.00	8.00	8.00			8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
The Spiking solutions were made on: 22/02/2006
***Repeated injections of QC a or b**

Submitted by: Shyam Sundar Date: 04/03/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/04/2015

Plasma Concentrations of Guanosine in Rat 374
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 19/02/2015 - 25/02/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Guanosine 5 ng		52.68							
a2.5	2.5ug/ml (a)	24.44	29.66	0.82	0.82	10	139.18	55.67	
b2.5	2.5ug/ml (b)	24.75	37.57	0.66	0.66	10	140.95	56.38	
Mean		24.60	33.62	0.74	0.74		140.06	56.03	
SD		0.22	5.59	0.12	0.12		1.25	0.50	
%CV		0.89	16.64	15.76	15.76		0.89	0.89	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a0.5	0.5ug/ml (a)	1.85	84.61	0.02	0.02	30.00	3.51	7.02	
b0.5	0.5ug/ml (b)	1.90	76.24	0.02	0.02	30.00	3.61	7.21	
	0.5ug/ml	2.17	84.50	0.03	0.03	30.00	4.12	8.24	
	0.5ug/ml	2.38	82.78	0.03	0.03	30.00	4.52	9.04	
	0.5ug/ml	2.80	83.74	0.03	0.03	30.00	5.32	10.63	
	0.5ug/ml	2.92	83.93	0.03	0.03	30.00	5.54	11.09	
Mean		2.34	82.63	0.03	0.03		4.44	8.87	
SD		0.45	3.20	0.01	0.01		0.85	1.71	
%CV		19.25	3.87	17.94	17.94		19.25	19.25	
N		6.00	6.00	6.00	6.00		6.00	6.00	
BLANKS:									
aB	0ug/mL (a)	ND	8.81	0.00	0.00	2	0.00		
bB	0 ug/mL (b)	ND	8.09	0.00	0.00	2	0.00		
Mean		0.00	8.45	0.00	0.00		0.00		
SD		0.00	0.51	0.00	0.00		0.00		
%CV		ERR	6.03	ERR	ERR		ERR		
N		2.00	2.00	2.00	2.00		2.00		
'Guanosine 5 ng 20/02/15		56.43	10.87			5.00			
'Guanosine 5 ng 23/02/15		55.25	10.32			5.00			
'Guanosine 5 ng 24/02/15		54.34	10.01			5.00			
'Guanosine 5 ng 25/02/15		54.17	10.63			5.00			

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.74	0.00	0.74
0.50	0.03	0.00	0.03
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0572
Std Err of Y Est	0.0926
R Squared	0.9757
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3137
Std Err of Coef.	0.0495

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	Conc.(µM)	Conc.(µM) Corrected for dilution
T0 R374	0.00	2.38	113.08	0.021	0.021	35	-	-	0.25	0.88	1.17
T0.08 R374	0.08	1.85	80.73	0.023	0.023	35	-	-	0.26	0.90	1.20
T0.25 R374	0.25	2.38	84.28	0.028	0.028	35	-	-	0.27	0.96	1.28
T1 R374	1.00	ND	82.73	0.000	0.000	35	-	-	0.18	0.64	0.86
Isoproterenol (30 mg/kg)											
T1.2 R374	1.20	1.55	110.56	0.014	0.014	35	-	-	0.23	0.85	1.13
T1.5R374	1.50	2.32	124.50	0.019	0.019	35	-	-	0.24	0.90	1.21
T2 R374	2.00	2.38	89.17	0.027	0.027	35	-	-	0.27	0.94	1.26
T3 R374	3.00	1.70	127.53	0.013	0.013	35	-	-	0.22	0.79	1.06
T4 R374	4.00	2.25	75.40	0.030	0.030	35	-	-	0.28	0.98	1.31
T5 R374	5.00	2.21	112.16	0.020	0.020	35	-	-	0.25	0.87	1.15
T6 R374	6.00	2.00	125.80	0.016	0.016	35	-	-	0.23	0.82	1.10
Mean		1.91	102.36	0.02	0.02				0.24	0.87	1.16
SD		0.70	20.08	0.01	0.01				0.03	0.09	0.12
%CV		36.52	19.62	44.06	44.06				11.04	10.80	10.80
n		22.00	22.00	11.00	11.00				11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
 The Spiking solutions were made on: 22/02/2006
 *Repeat injections of QC a or b

Submitted by: Shyam Sundar Date: 04/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/04/2015

Plasma Concentrations of Uric Acid in Rat 374
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 19/02/2015 - 25/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Uric Acid 5 ng		30.40				5		
a25	25ug/ml (a)	58.45	7.40	7.90	7.90	2	2884.05	106.74
b25	25ug/ml (b)	55.77	8.59	6.49	6.49	2	2751.81	101.45
Mean		57.11	8.00	7.20	7.20		2817.93	104.09
SD		1.90	0.84	0.99	0.99		93.51	3.74
%CV		3.32	10.52	13.82	13.82		3.32	3.59
N		2.00	2.00	2.00	2.00		2.00	2.00
a5	5ug/ml (a)	10.27	10.21	1.01	1.01	2	506.74	58.22
b5	5ug/ml (b)	9.88	7.83	1.26	1.26	2	487.50	54.38
	5ug/ml	8.05	9.41	0.86	0.86	2	397.20	36.32
	5ug/ml	9.18	8.85	1.04	1.04	2	452.96	47.47
	5ug/ml	10.23	9.62	1.06	1.06	2	504.77	43.22
	5ug/ml	11.93	10.07	1.18	1.18	2	588.65	60.00
Mean		9.92	9.33	1.07	1.07		489.64	49.93
SD		1.29	0.88	0.14	0.14		63.99	9.23
%CV		12.99	9.46	13.32	13.32		12.99	18.49
N		6.00	6.00	6.00	6.00		6.00	6.00
BLANKS:								
aB	0ug/ml (a)	2.89	8.81	0.33	0.33	2	142.60	
bB	0ug/ml (b)	5.85	8.09	0.72	0.72	2	288.65	
Mean		4.37	8.45	0.53	0.53		215.63	
SD		2.09	0.51	0.28	0.28		103.27	
%CV		47.90	6.03	53.15	53.15		47.90	
N		2.00	2.00	2.00	2.00		2.00	
Uric Acid 5 ng 20/02/2015		33.22	10.87				5.00	
Uric Acid 5 ng 23/02/2015		33.99	10.32				5.00	
Uric Acid 5 ng 24/02/2015		32.95	10.01				5.00	
Uric Acid 5 ng 25/02/2015		32.01	10.63				5.00	

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	7.20	0.53	6.67
5.00	1.07	0.53	0.54
0.00	0.53	0.53	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.3769
Std Err of Y Est	0.6106
R Squared	0.9864
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.2781
Std Err of Coef.	0.0326

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c(µg/mL):onc(µM)	Conc(µM) Corrected for dilution
T0 R374	0.00	23.20	19.52	1.19	1.19	5	-	5.63	33.49
T0.08 R374	0.08	18.24	17.10	1.07	1.07	5	-	5.19	30.88
T0.25 R374	0.25	16.14	19.13	0.84	0.84	5	-	4.39	26.11
T1 R374	1.00	22.48	15.66	1.44	1.44	5	-	6.52	38.77
Isoproterenol (30 mg/kg)									
T1.2 R374	1.20	53.69	23.12	2.32	2.32	5	-	9.71	71.31
T1.5R374	1.50	11.48	22.15	0.52	0.52	5	-	3.22	23.85
T2 R374	2.00	46.77	18.48	2.53	2.53	5	-	10.46	62.20
T3 R374	3.00	17.69	23.24	0.76	0.76	5	-	4.09	24.34
T4 R374	4.00	35.02	13.79	2.54	2.54	5	-	10.49	62.38
T5 R374	5.00	20.12	22.86	0.88	0.88	5	-	4.52	26.89
T6 R374	6.00	16.13	23.81	0.68	0.68	5	-	3.79	22.55
Mean		26.21	19.80	1.33	1.33			6.15	38.84
SD		15.38	2.91	0.73	0.73			2.63	18.11
%CV		58.67	14.17	54.86	54.86			42.77	46.61
n		8.00	8.00	8.00	8.00			8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No. 181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 04/03/2015

Checked by: Date:

Approved by: Pollen Yeung Date:10/04/2015

Title: Measurement of Plasma Concentrations of Dipyridamole in Rat 374

According to SOP No: SOP/STD/2008-001-1 (Plasma with no Stopping Solution) Experiment Date:02/10/2014- 03/10/2014

Abs.amt ng	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Peak Ht. Ratio	Peak Ht. Ratio	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Dipyridamole (1ng)		78.52				1		
a1000	1 ug/mL(a)	98.41	3.25	30.28	30.28	5	50.13	100.26
b1000	1ug/mL(b)	111.64	6.52	17.12		5	56.87	113.74
1000*	1ug/mL(c)	89.65	2.63	34.09	34.09	5	45.67	91.34
Mean		99.90	4.13	27.16	32.18		50.89	101.78
SD		11.07	2.09	8.90	2.69		5.64	11.28
%CV		11.08	50.57	32.77	8.37		11.08	11.08
n		3.00	3.00	3.00	2.00		1.00	1.00
a100	0.1 ug/mL (a)	36.78	22.96	1.60	1.60	20	4.68	93.68
b100	0.1ug/mL (b)	38.45	23.38	1.64	1.64	20	4.90	97.94
Mean		37.62	23.17	1.62	1.62		4.79	95.81
SD		1.18	0.30	0.03	0.03		0.15	3.01
%CV		3.14	1.28	1.86	1.86		3.14	3.14
n		2.00	2.00	2.00	2.00		2.00	2.00
aB	0 ug/mL (a)	0.00	15.33	0.00	0.00	20	0.00	0.00
bB	0 ug/mL (b)	0.00	26.06	0.00	0.00	20	0.00	0.00
Mean		0.00	20.70	0.00	0.00		0.00	0.00
SD		0.00	7.59	0.00	0.00		0.00	0.00
%CV		ERR	36.66	ERR	ERR		ERR	ERR
n		2.00	2.00	2.00	2.00		2.00	2.00

Plasma Conc. (ug/mL)	Peak Ht.Ratio (PHR)	Blank (PHRb)	PHRV-PHRb
0.00	0.00	0.00	0.00
0.10	1.62	0.00	1.62
1.00	32.18	0.00	32.18

Regression Output Begins Here:

Regression Output:	
Constant	-0.7888
Std Err of Y Est	1.1824
R Squared	0.9979
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	32.8849
Std Err of Coef.	1.5180

Sample ID	Time Post-dose (h)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)*	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc.(ug/mL)
R374T0	0.00	2.88	22.01	0.13	0.13	20	-	0.43
R374T0.08	0.08	15.20	19.81	0.77	0.77	20	-	0.72
R374T0.25	0.25	17.00	17.38	0.98	0.98	20	-	0.82
R374T1	1.00	35.02	25.27	1.39	1.39	20	-	1.01
Salicylic Acid (30 mg/kg sc)						20		
R374T1.2	1.20	25.13	29.21	0.86	0.86	20	-	0.86
R374T1.5	1.50	21.17	23.87	0.89	0.89	20	-	0.78
R374T2	2.00	15.33	26.68	0.57	0.57	20	-	0.63
R374T3	3.00	9.01	23.11	0.39	0.39	20	-	0.55
R374T4	4.00	7.79	13.33	0.58	0.58	20	-	0.64
R374T5	5.00	5.43	17.86	0.30	0.30	20	-	0.51
R374T6	6.00	4.74	19.52	0.24	0.24	20	-	0.48
Mean		14.43	21.64	0.65	0.65			0.67
SD		9.87	4.61	0.37	0.37			0.18
%CV		68.39	21.30	57.84	57.84			26.78
n		11.00	11.00	11.00	11.00			11.00

Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 Corr. PHR = (PHR - RGB PHR)

Dipyridamole (1ng)(03/10/2014) 81.21 7.88 1.00

Comments: Plasma from Rat 156 was used for extraction QC's.
 *A repeat injection of a or b

Submitted by: Shyam Sundar Date: 06/10/2014

Checked by: Pollen Yeung Date:08/10/2014

Approved by: Date:

APPENDIX 5: Rat 375

Title: Measurement of RBC Concentrations of ATP in Rat 375 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
Experiment Date 04-05/06/2014

Sample/standard ID	Standard Concentra (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol (µL)	Amount Recov (%)	Recovery
ATP 4 ng										
a250	250 ug/mL	33.60	8.68	3.87	35.00	3.87	3.87	0.35	23783.78	81.30
b250	250 ug/mL	32.11	8.90	3.61	35.00	3.61	3.61	0.35	22729.09	77.08
Mean		32.86	8.79	3.74	35.00	3.74	3.74		23256.44	79.19
SD		1.85	0.16	0.19	0.00	0.19	0.19		745.78	2.98
%CV		3.21	1.77	4.98	0.00	4.98	4.98		3.21	3.77
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a100										
b100	100 ug/mL	15.82	9.39	1.68	35.00	1.68	1.68	0.35	11198.20	77.40
100'	100 ug/mL	19.01	8.37	2.27	35.00	2.27	2.27	0.35	13458.24	99.98
Mean		16.59	9.05	1.85	35.00	1.85	1.85		10883.20	74.25
SD		2.15	0.99	0.37	0.00	0.37	0.37		445.47	4.45
%CV		12.93	6.54	20.00	0.00	20.00	20.00		4.09	6.00
n		3.00	3.00	3.00	3.00	3.00	3.00		2.00	2.00
aB										
bB	0 ug/mL (a)	5.75	12.06	0.48	35.00	0.48	0.48	0.35	4070.14	
	0 ug/mL (a)	4.02	7.96	0.51	35.00	0.51	0.51	0.35	2845.56	
Mean		4.89	10.01	0.49	35.00	0.49	0.49		3457.85	
SD		1.22	2.90	0.02	0.00	0.02	0.02		865.91	
%CV		25.04	28.96	4.07	0.00	4.07	4.07		25.04	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRB
250.00	3.74	0.49	3.25
100.00	1.85	0.49	1.36
0.00	0.49	0.49	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0228
Std Err of Y Est	0.0469
R Squared	0.9996
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0130
Std Err of Coef.	0.0003

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R37570	0.00	47.59	69.25	0.69	35.00	0.69	0.69	1.50	-	61.25	0.1010
R37570.08	0.08	54.59	66.88	0.82	35.00	0.82	0.82	1.50	-	61.20	0.1207
R37570.25	0.25	67.24	81.59	0.82	35.00	0.82	0.82	1.50	-	61.81	0.1219
R37571	1.00	38.53	50.68	0.76	35.00	0.76	0.76	1.00	-	56.89	0.1122
Interim total (10 mg/kg sq)											
R37571.2	1.20	51.06	61.07	0.84	35.00	0.84	0.84	1.00	-	62.74	0.1237
R37571.5	1.50	65.60	62.82	1.04	35.00	1.04	1.04	1.00	-	79.79	0.1553
R37572	2.00	68.44	67.18	1.02	35.00	1.02	1.02	1.00	-	76.83	0.1515
R37573	3.00	44.40	59.63	0.74	35.00	0.74	0.74	1.00	-	55.68	0.1098
R37574	4.00	52.67	56.11	0.94	35.00	0.94	0.94	1.00	-	70.05	0.1363
R37575	5.00	54.67	57.78	0.95	35.00	0.95	0.95	1.00	-	71.23	0.1404
R37576	6.00	56.73	57.55	0.99	35.00	0.99	0.99	1.00	-	74.28	0.1464
Mean		54.68	62.78	0.87	35.00	0.87	0.87	1.14		65.58	0.13
SD		9.51	8.29	0.12	0.00	0.12	0.12	0.23		9.25	0.02
%CV		17.39	13.21	13.73	0.00	13.73	13.73	20.55		14.10	14.10
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00		11.00	11.00

ATP (4 ng) 5/06/2014

8.95

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
*Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 16/06/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 14/07/2014

Title: Measurement of RBC Concentrations of ADP in Rat 375 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)

Experiment Date: 04-05/06/2014

Sample/standard ID	Standard Co (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered	Recovery %
ADP 4 ng		21.82						4.00		
s250	250 µg/mL	58.14	8.68	6.70	35.00	6.70	6.70	0.35	16748.46	64.41
b250	250 µg/mL	54.72	8.90	6.15	35.00	6.15	6.15	0.35	15763.26	60.47
Mean		56.43	8.79	6.42	35.00	6.42	6.42		16255.86	62.44
SD		2.42	0.16	0.39	0.00	0.39	0.39		696.64	2.79
%CV		4.29	1.77	6.05	0.00	6.05	6.05		4.29	4.46
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
100a	100 µg/mL	23.96	9.39	2.55	35.00	2.55	2.55	0.35	6902.19	62.57
100b	100 µg/mL	22.83	9.40	2.43	35.00	2.43	2.43	0.35	6576.67	59.31
100*	100 µg/mL	21.53	8.37	2.57	35.00	2.57	2.57	0.35	6202.17	55.57
Mean		22.77	9.05	2.52	35.00	2.52	2.52		6560.34	59.15
SD		1.22	0.59	0.08	0.00	0.08	0.08		350.29	3.50
%CV		5.34	6.54	3.08	0.00	3.08	3.08		5.34	5.92
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB	0 µg/mL (a)	2.54	12.06	0.21	35.00	0.21	0.21	0.35	731.70	
bB	0 µg/mL (a)	1.94	7.96	0.24	35.00	0.24	0.24	0.35	558.86	
Mean		2.24	10.01	0.23	35.00	0.23	0.23		645.28	
SD		0.42	2.90	0.02	0.00	0.02	0.02		122.22	
%CV		18.94	28.96	10.30	0.00	10.30	10.30		18.94	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
250.00	6.42	0.23	6.20
100.00	2.52	0.23	2.29
0.00	0.23	0.23	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0742
Std Err of Y Est	0.1525
R Squared	0.9988
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0249
Std Err of Coef.	0.0009

Sample ID	Time post	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) Lysate	Conc.(mM) RBC
R37510	0.00	32.07	69.25	0.46	35.00	0.46	0.46	1.50	-	21.59	0.0505	0.650
R37510b	0.06	11.61	66.88	0.17	35.00	0.17	0.17	1.50	-	9.96	0.0233	0.300
R3751025	0.25	17.63	81.59	0.22	35.00	0.22	0.22	1.50	-	11.67	0.0273	0.351
R37511	1.00	12.33	50.68	0.24	35.00	0.24	0.24	1.00	-	12.76	0.0299	0.384
Isoproterenol (30 mg/kg sc)												
R37511.2	1.20	11.71	61.07	0.19	35.00	0.19	0.19	1.00	-	10.69	0.0250	0.322
R37511.5	1.50	16.81	62.62	0.27	35.00	0.27	0.27	1.00	-	13.74	0.0322	0.413
R37512	2.00	15.50	67.18	0.23	35.00	0.23	0.23	1.00	-	12.26	0.0287	0.369
R37513	3.00	18.90	59.63	0.32	35.00	0.32	0.32	1.00	-	15.72	0.0368	0.473
R37514	4.00	11.67	56.11	0.21	35.00	0.21	0.21	1.00	-	11.48	0.0269	0.346
R37515	5.00	9.89	57.78	0.17	35.00	0.17	0.17	1.00	-	9.86	0.0231	0.297
R37516	6.00	10.14	57.55	0.18	35.00	0.18	0.18	1.00	-	10.06	0.0236	0.303
Mean		20.44	72.57	0.28	35.00	0.28	0.28	1.50		14.41	0.03	0.43
SD		10.51	7.90	0.16	0.00	0.16	0.16	0.00		6.28	0.01	0.19
%CV		51.45	10.88	55.00	0.00	55.00	55.00	0.00		43.61	43.61	43.61
n		3.00	3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00	3.00

ADP (4 ng) 05/06/2014

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
*Repeated injections of a or b at 0.5-1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 16/06/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 14/07/2014

Title: Measurement of RBC Concentrations of AMP in Rat 375 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
Experiment Date 04-05/06/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
AMP 4 ng										
		5254						4.00		
a50	50 µg/mL	32.80	8.68	3.78	35.00	3.78	3.78	0.35	3924.09	76.56
b50	50 µg/mL	31.20	8.90	3.51	35.00	3.51	3.51	0.35	3732.67	72.73
Mean		32.00	8.79	3.64	35.00	3.64	3.64		3828.38	74.64
SD		1.13	0.16	0.19	0.00	0.19	0.19		135.35	2.71
%CV		3.54	1.77	5.30	0.00	5.30	5.30		3.54	3.63
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20	20 µg/mL	12.89	9.39	1.37	35.00	1.37	1.37	0.35	1542.12	72.29
b20	20 µg/mL	12.01	9.40	1.28	35.00	1.28	1.28	0.35	1436.84	67.03
20*	20 µg/mL	11.37	8.37	1.36	35.00	1.36	1.36	0.35	1360.27	63.20
Mean		12.89	9.05	1.34	35.00	1.34	1.34		1446.41	67.51
SD		0.78	0.59	0.05	0.00	0.05	0.05		91.30	4.57
%CV		6.31	6.54	3.84	0.00	3.84	3.84		6.31	6.76
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB	0 µg/mL (a)	0.96	12.06	0.08	35.00	0.08	0.08	0.35	114.85	
bB	0 µg/mL (a)	0.65	7.96	0.08	35.00	0.08	0.08	0.35	77.76	
Mean		0.81	10.01	0.08	35.00	0.08	0.08		96.31	
SD		0.22	2.90	0.00	0.00	0.00	0.00		26.22	
%CV		27.23	28.96	1.80	0.00	1.80	1.80		27.23	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	3.64	0.08	3.56
20.00	1.34	0.08	1.26
0.00	0.08	0.08	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0667
Std Err of Y Est	0.1371
R Squared	0.9971
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0717
Std Err of Coef.	0.0039

Sample ID	Time post dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R375T0	0.00	28.13	69.25	0.41	35.00	0.41	0.41	1.50	-	6.60	0.0190
R375T0.08	0.08	1.00	66.88	0.01	35.00	0.01	0.01	1.50	-	1.14	0.0033
R375T0.25	0.25	1.99	81.59	0.02	35.00	0.02	0.02	1.00	-	1.27	0.0037
R375T1	1.00	2.99	50.68	0.06	35.00	0.06	0.06	1.00	-	1.75	0.0051
Soprolefenol (30 mg/kg sc)											
R375T1.2	1.20	1.74	61.07	0.03	35.00	0.03	0.03	1.00	-	1.33	0.0038
R375T1.5	1.50	1.45	62.62	0.02	35.00	0.02	0.02	1.00	-	1.25	0.0036
R375T2	2.00	1.21	67.18	0.02	35.00	0.02	0.02	1.00	-	1.18	0.0034
R375T3	3.00	7.10	59.63	0.12	35.00	0.12	0.12	1.00	-	2.59	0.0075
R375T4	4.00	1.71	56.11	0.03	35.00	0.03	0.03	1.00	-	1.36	0.0039
R375T5	5.00	1.12	57.78	0.02	35.00	0.02	0.02	1.00	-	1.20	0.0035
R375T6	6.00	0.79	57.55	0.01	35.00	0.01	0.01	1.00	-	1.12	0.0032
Mean		4.48	62.78	0.07	35.00	0.07	0.07	1.09	0.00	1.89	0.01
SD		8.04	8.29	0.12	0.00	0.12	0.12	0.20	0.00	1.62	0.00
%CV		179.68	13.21	168.62	0.00	168.62	168.62	18.54	0.00	85.61	85.61
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

AMP (4 ng)/05/06/2014

5416

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
*Repeated injections of "a" or "b" at 0.5 - 1 ul injection volume.

PL = plasma; RBC = red blood cells
Peak Ht. = peak height
Peak Ht. R. (or PHR) = peak height ratio
I.S. = internal standard
Inj Vol = injection volume
ND = not detected or determined
NS = no sample
INT = interference
PCV = packed cell volume (haematocrit)
CorPHR = corrected peak height ratio
Hemolysis Degree:
-: no visible hemolysis
+: slight hemolysis
++: intermediate hemolysis
+++: serious hemolysis

Submitted by: Shyam Sundar

Date: 16/06/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date:14/07/2014

Title: Measurement of RBC Concentrations of GTP in Rat 375 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 04-05/06/2014

Sample/standard ID	Standard Concentra (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recove (%)	Recovery
GTP 4 ng										
a50	50 ug/mL	11.28	8.68	1.30	35.00	1.30	1.30	0.35	833.37	15.29
b50	50 ug/mL	10.36	8.90	1.16	35.00	1.16	1.16	0.35	765.40	13.93
Mean		10.62	8.79	1.23	35.00	1.23	1.23		799.38	14.61
SD		0.65	0.10	0.00	0.10	0.10	0.10		48.06	0.96
%CV		6.01	0.00	7.78	0.00	7.78	7.78		6.01	6.58
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20	20 ug/mL	4.46	8.39	0.47	35.00	0.47	0.47	0.35	329.51	13.02
b20	20 ug/mL	4.59	9.40	0.48	35.00	0.48	0.48	0.35	338.11	13.50
20*	20 ug/mL	3.79	8.37	0.45	35.00	0.45	0.45	0.35	280.01	10.55
Mean		4.28	9.05	0.47	35.00	0.47	0.47		316.21	12.36
SD		0.43	0.59	0.02	0.00	0.02	0.02		31.72	1.59
%CV		10.03	6.54	3.80	0.00	3.80	3.80		10.03	12.83
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB	0 ug/mL (a)	0.84	12.06	0.07	35.00	0.07	0.07	0.35	62.06	
bB	0 ug/mL (a)	1.53	7.96	0.13	35.00	0.13	0.13	0.35	78.10	
Mean		0.94	10.01	0.10	35.00	0.10	0.10		69.08	
SD		0.13	2.90	0.04	0.00	0.03	0.04		9.93	
%CV		14.37	28.96	42.45	0.00	34.66	42.45		14.37	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRv)	Blank PHRv-PHRb
50.00	1.23	0.10
20.00	0.47	0.10
0.00	0.10	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0317
Std Err of Y Est	0.0652
R Squared	0.9936
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0229
Std Err of Coef.	0.0018

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R375T0	0.00	10.63	69.25	0.15	35.00	0.15	0.15	1.50	-	8.10	0.0155
R375T0.08	0.08	11.66	66.88	0.17	35.00	0.17	0.17	1.50	-	9.02	0.0172
R375T0.25	0.25	13.13	81.69	0.16	35.00	0.16	0.16	1.50	-	8.43	0.0161
R375T1	1.00	7.56	50.68	0.15	35.00	0.15	0.15	1.00	-	7.91	0.0151
Supranal (20 mg/kg sc)											
R375T1.2	1.20	10.57	61.07	0.17	35.00	0.17	0.17	1.00	-	8.96	0.0171
R375T1.5	1.50	12.97	62.82	0.21	35.00	0.21	0.21	1.00	-	10.42	0.0159
R375T2	2.00	14.82	67.18	0.22	35.00	0.22	0.22	1.00	-	11.04	0.0211
R375T3	3.00	10.35	59.63	0.17	35.00	0.17	0.17	1.00	-	8.98	0.0172
R375T4	4.00	13.26	56.11	0.24	35.00	0.24	0.24	1.00	-	11.73	0.0224
R375T5	5.00	12.85	57.78	0.22	35.00	0.22	0.22	1.00	-	11.12	0.0213
R375T6	6.00	14.18	57.55	0.25	35.00	0.25	0.25	1.00	-	12.17	0.0233
Mean		12.00	62.78	0.19	35.00	0.19	0.19	1.14	0.00	9.81	0.02
SD		2.99	8.29	0.03	0.00	0.03	0.03	0.23	0.00	1.53	0.00
%CV		17.42	13.21	18.12	0.00	18.12	18.12	20.55	0.00	15.56	15.56
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

GTP (4 ng) 05/06/2014

88.27

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 16/06/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 14/07/2014

Title: Measurement of RBC Concentrations of GDP in Rat 375 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 04-05/06/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
GDP 4 ng										
		31.37						4.00		
a50	50 ug/mL	16.98	8.68	1.96	35.00	1.96	1.96	0.35	3402.34	64.90
b50	50 ug/mL	16.21	8.90	1.82	35.00	1.82	1.82	0.35	3248.05	61.82
Mean		16.60	8.79	1.89	35.00	1.89	1.89		3325.20	63.36
SD		0.54	0.16	0.10	0.00	0.10	0.10		108.10	2.18
%CV		3.28	1.77	5.05	0.00	5.05	5.05		3.28	3.44
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20	20 ug/mL	6.75	9.39	0.72	35.00	0.72	0.72	0.35	1353.53	59.78
b20	20 ug/mL	6.75	9.40	0.72	35.00	0.72	0.72	0.35	1352.52	59.76
20*	20 ug/mL	6.14	8.37	0.73	35.00	0.73	0.73	0.35	1230.29	53.65
Mean		6.55	9.05	0.72	35.00	0.72	0.72		1311.78	57.72
SD		0.35	0.59	0.01	0.00	0.01	0.01		70.57	3.53
%CV		5.38	6.54	1.21	0.00	1.21	1.21		5.38	6.11
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB	0 ug/mL (a)	0.88	12.06	0.07	35.00	0.07	0.07	0.35	176.33	
bB	0 ug/mL (a)	0.69	7.96	0.09	35.00	0.09	0.09	0.35	138.26	
Mean		0.79	10.01	0.08	35.00	0.08	0.08		157.29	
SD		0.13	2.90	0.01	0.00	0.01	0.01		26.92	
%CV		17.11	28.96	12.15	0.00	12.15	12.15		17.11	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRB
50.00	1.89	0.08	1.81
20.00	0.72	0.08	0.64
0.00	0.08	0.08	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0315
Std Err of Y Est	0.0648
R Squared	0.9975
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0094
Std Err of Coef.	0.0018

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(nM) RBC
R37570	0.00	7.38	69.25	0.11	35.00	0.11	0.11	1.50	-	3.80	0.0086
R37570.08	0.08	3.47	86.88	0.05	35.00	0.05	0.05	1.50	-	2.29	0.0032
R37570.25	0.25	4.81	81.59	0.06	35.00	0.06	0.06	1.50	-	2.42	0.0035
R37571	1.00	3.27	50.68	0.06	35.00	0.06	0.06	1.00	-	2.64	0.0060
Isoproterenol (30 µg/kg sc)											
R37571.2	1.20	3.68	61.07	0.06	35.00	0.06	0.06	1.00	-	2.52	0.0057
R37571.5	1.50	4.51	62.62	0.07	35.00	0.07	0.07	1.00	-	2.94	0.0084
R37572	2.00	4.47	57.18	0.07	35.00	0.07	0.07	1.00	-	2.70	0.0081
R37573	3.00	4.55	59.63	0.08	35.00	0.08	0.08	1.00	-	2.96	0.0087
R37574	4.00	3.97	56.11	0.08	35.00	0.08	0.08	1.00	-	3.10	0.0070
R37575	5.00	3.64	57.78	0.07	35.00	0.07	0.07	1.00	-	2.75	0.0062
R37576	6.00	3.54	57.55	0.06	35.00	0.06	0.06	1.00	-	2.60	0.0059
Mean		33.53	62.78	0.07	35.00	0.07	0.07	1.14	0.00	2.78	0.01
SD		30.49	8.29	0.01	0.00	0.01	0.01	0.23	0.00	0.41	0.00
%CV		90.93	13.21	21.25	0.00	21.25	21.25	20.55	ERR	14.63	14.63
n		22.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

GDP (4 ng) 05/06/2014

32.38

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date:16/06/2014

Checked by: Date:

Approved by: Pollen Yeung Date:14/07/2014

Title: Measurement of RBC Concentrations of GMP in Rat 375 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 04-05/06/2014

Sample/standard	ID/Standard Concentr (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (µL)	% Recovery
GMP 4 ng										
a50	50 ug/mL	Off Scale	8.68	0.00	35.00	0.00	0.00	0.35	0.00	-95.23
b50	50 ug/mL	Off Scale	8.90	0.00	35.00	0.00	0.00	0.35	0.00	-95.23
Mean		0.00	8.79	0.00	35.00	0.00	0.00		0.00	-95.23
SD		0.00	8.85	0.00	0.00	0.00	0.00		0.00	0.00
%CV		ERR	8.82	ERR	0.00	ERR	ERR		ERR	-0.00
n		2.00	8.82	2.00	2.00	2.00	2.00		2.00	2.00
a20										
a20	20 ug/mL	105.05	9.39	11.19	35.00	11.19	11.19	0.35	5400.46	31.94
b20	20 ug/mL	95.75	9.40	10.19	35.00	10.19	10.19	0.35	4922.36	3.21
20*	20 ug/mL	69.08	8.37	8.25	35.00	8.25	8.25	0.35	3551.30	-60.52
Mean		89.96	9.05	9.88	35.00	9.88	9.88		4624.71	-4.46
SD		18.67	0.59	1.49	0.00	1.49	1.49		959.84	47.32
%CV		20.75	6.54	15.10	0.00	15.10	15.10		20.75	-59.57
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB										
aB	0 ug/mL (a)	112.42	12.06	9.32	35.00	9.32	9.32	0.35	5779.34	
bB	0 ug/mL (a)	72.83	7.96	9.15	35.00	9.15	9.15	0.35	3744.08	
Mean		92.63	10.01	9.24	35.00	9.24	9.24		4761.71	
SD		27.99	2.90	0.12	0.00	0.12	0.12		1438.15	
%CV		30.22	28.96	1.32	0.00	1.32	1.32		30.22	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	0.00	9.24	-9.24
20.00	9.88	9.24	0.64
0.00	9.24	9.24	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	2.8456
R Squared	0.7349
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	-0.1548
Std Err of Coef.	0.0528

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R375T0		0.00 off scale	69.25	0.00	35.00	0.00	0.00	1.50	-	-0.00	-0.0000
R375T0.08		0.08 off scale	66.88	0.00	35.00	0.00	0.00	1.50	-	-0.00	-0.0000
R375T0.25		0.25 off scale	81.59	0.00	35.00	0.00	0.00	1.50	-	-0.00	-0.0000
R375T1		1.00 off scale	50.68	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
Not detected (ND mg/kg sat)											
R375T1.2		1.20 off scale	61.07	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
R375T1.5		1.50 off scale	62.82	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
R375T2		2.00 off scale	67.18	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
R375T3		3.00 off scale	59.63	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
R375T4		4.00 off scale	56.11	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
R375T5		5.00 off scale	57.78	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
R375T6		6.00 off scale	57.55	0.00	35.00	0.00	0.00	1.00	-	-0.00	-0.0000
Mean		0.00	62.78	0.00	35.00	0.00	0.00	1.14		0.00	0.00
SD		0.00	8.29	0.00	0.00	0.00	0.23			0.00	0.00
%CV		ERR	13.21	ERR	0.00	ERR	ERR	20.55		ERR	ERR
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00		11.00	11.00

GMP (4ng) 05/06/2014 86.77

Comments: RBC Lysate from Rat 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of "a" or "b" at 0.5-1ul injection volume

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 16/06/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 14/07/2014

Plasma Concentrations of Adenosine in Rat 375
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 19/02/2015 - 25/02/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Adenosine 5 ng		29.55							
a2.5	2.5ug/ml (a)	16.73	29.66	0.56	0.56	10	169.85	67.94	
b2.5	2.5ug/ml (b)	16.05	37.57	0.43	0.43	10	162.94	65.18	
Mean		16.39	33.62	0.50	0.50		166.40	66.56	
SD		0.48	5.59	0.10	0.10		4.88	1.95	
%CV		2.93	16.64	19.53	19.53		2.93	2.93	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a0.5	0.5ug/ml (a)	6.73	84.61	0.08	0.08	30.00	22.77	45.55	
b0.5	0.5ug/ml (b)	4.75	76.24	0.06	0.06	30.00	16.07	32.15	
	0.5 ug/ml	7.40	84.50	0.09	0.09	30.00	25.04	50.08	
	0.5 ug/ml	8.08	82.78	0.10	0.10	30.00	27.34	54.69	
	0.5 ug/ml	8.12	83.74	0.10	0.10	30.00	27.48	54.96	
	0.5 ug/ml	8.04	83.93	0.10	0.10	30.00	27.21	54.42	
Mean		7.19	82.63	0.09	0.09		24.32	48.64	
SD		1.31	3.20	0.01	0.01		4.44	8.87	
%CV		18.24	3.87	15.95	15.95		18.24	18.24	
N		6.00	6.00	6.00	6.00		6.00	6.00	
BLANKS:									
aB	0ug/mL (a)	N/D	8.81	0.00	0.00	2	0.00		
bB	0 ug/mL (b)	N/D	8.09	0.00	0.00	2	0.00		
Mean		0.00	8.45	0.00	0.00		0.00		
SD		0.00	0.51	0.00	0.00		0.00		
%CV		ERR	6.03	ERR	ERR		ERR		
N		2.00	2.00	2.00	2.00		2.00		
Adenosine 5 ng 20/02/2015		31.91	10.87			5.00			
Adenosine 5 ng 23/02/2015		31.20	10.32			5.00			
Adenosine 5 ng 24/02/2015		30.73	10.01			5.00			
Adenosine 5 ng 25/02/2015		31.69	10.63			5.00			

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHRV Value (PHRV)	Blank (PHRB)	PHRV-PHRB
2.50	0.50	0.00	0.50
0.50	0.09	0.00	0.09
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0059
Std Err of Y Est	0.0096
R Squared	0.9953
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.2000
Std Err of Coef.	0.0052

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	Conc.(µM)	Conc.(µM) Corrected for dilution
T0 R375	0.00	2.91	110.24	0.03	0.03	35	-	-	0.16	0.61	0.81
T0.08 R375	0.08	2.63	78.92	0.03	0.03	35	-	-	0.20	0.73	0.98
T0.25 R375	0.25	2.23	93.78	0.02	0.02	35	-	-	0.15	0.56	0.74
T1 R375	1.00	3.80	81.78	0.05	0.05	35	-	-	0.26	0.98	1.31
Isoproterenol (30 mg/kg)											
T1.2 R375	1.20	6.34	80.96	0.08	0.08	35	-	-	0.42	1.58	2.10
T1.5 R375	1.50	4.88	89.90	0.05	0.05	35	-	-	0.30	1.13	1.50
T2 R375	2.00	4.26	103.77	0.04	0.04	35	-	-	0.23	0.88	1.17
T3 R375	3.00	3.04	89.00	0.03	0.03	35	-	-	0.20	0.75	1.00
T4 R375	4.00	2.57	84.03	0.03	0.03	35	-	-	0.18	0.68	0.91
T5 R375	5.00	2.56	104.87	0.02	0.02	35	-	-	0.15	0.57	0.76
T6 R375	6.00	2.77	107.80	0.03	0.03	35	-	-	0.16	0.59	0.79
Mean		3.45	93.19	0.04	0.04				0.22	0.82	1.10
SD		1.26	11.61	0.02	0.02				0.08	0.31	0.41
%CV		36.42	12.46	43.48	43.48				37.60	37.60	37.60
n		11.00	11.00	11.00	11.00				11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
 The Spiking solutions were made on: 22/02/2006
 *Repeat injections of QC a or b

Submitted by: Shyam Sundar Date: 03/03/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/04/2015

Plasma Concentrations of Inosine in Rat 375
Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
Experiment Date: 19/02/2015 - 25/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount Rec. (ng)	Recovery (%)
Inosine 5 ng		43.64				5		
a2.5	2.5ug/ml (a)	29.29	29.66	0.99	0.99	10	201.35	80.54
b2.5	2.5ug/ml (b)	27.31	37.57	0.73	0.73	10	187.74	75.10
Mean		28.30	33.62	0.86	0.86		194.55	77.82
SD		1.40	5.59	0.18	0.18		9.62	3.85
%CV		4.95	16.64	21.50	21.50		4.95	4.95
N		2.00	2.00	2.00	2.00		2.00	2.00
a0.5	0.5ug/ml (a)	7.85	84.61	0.09	0.09	30.00	17.99	35.98
b0.5	0.5ug/ml (b)	7.48	76.24	0.10	0.10	30.00	17.14	34.28
	0.5ug/ml	4.16	84.50	0.05	0.05	30.00	9.53	19.07
	0.5ug/ml	7.38	82.78	0.09	0.09	30.00	16.91	33.82
	0.5ug/ml	5.06	83.74	0.06	0.06	30.00	11.59	23.19
	0.5ug/ml	9.45	83.93	0.11	0.11	30.00	21.65	43.31
Mean		6.90	82.63	0.08	0.10		15.80	31.61
SD		1.94	3.20	0.02	0.01		4.45	8.90
%CV		28.16	3.87	28.69	10.50		28.16	28.16
N		6.00	6.00	6.00	4.00		6.00	6.00
BLANKS:								
aB	0ug/mL (a)	ND	8.81	0.00	0.00	2	0.00	
bB	0ug/mL (b)	ND	8.09	0.00	0.00	2	0.00	
Mean		0.00	8.45	0.00	0.00		0.00	
SD		0.00	0.51	0.00	0.00		0.00	
%CV		ERR	6.03	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00		2.00	
Inosine 5ng 20/02/15		43.40	10.87			5.00		
Inosine 5ng 23/02/15		43.14	10.32			5.00		
Inosine 5ng 24/02/15		41.22	10.01			5.00		
Inosine 5ng 25/02/15		44.35	10.63			5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHRV Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.86	0.00	0.86
0.50	0.10	0.00	0.10
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0349
Std Err of Y Est	0.0565
R Squared	0.9927
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3534
Std Err of Coef.	0.0302

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	onc.(µM)	Conc.(µM) Corrected for dilution
T0 R375	0.00	11.72	110.24	0.11	0.11	35	-	0.40	1.49	1.99
T0.08 R375	0.08	8.02	78.92	0.10	0.10	35	-	0.39	1.44	1.92
T0.25 R375	0.25	7.43	93.78	0.08	0.08	35	-	0.32	1.20	1.61
T1 R375	1.00	5.86	81.78	0.07	0.07	35	-	0.30	1.12	1.50
Isoprenalineol (30 mg/kg)										
T1.2 R375	1.20	4.67	80.96	0.06	0.06	35	-	0.26	0.98	1.30
T1.5 R375	1.50	4.84	89.90	0.05	0.05	35	-	0.25	0.94	1.25
T2 R375	2.00	5.62	103.77	0.05	0.05	35	-	0.25	0.94	1.25
T3 R375	3.00	7.61	89.00	0.09	0.09	35	-	0.34	1.27	1.69
T4 R375	4.00	8.62	84.03	0.10	0.10	35	-	0.39	1.45	1.93
T5 R375	5.00	10.44	104.87	0.10	0.10	35	-	0.38	1.42	1.89
T6 R375	6.00	12.52	107.80	0.12	0.12	35	-	0.43	1.59	2.12
Mean		7.94	93.19	0.08	0.08			0.34	1.26	1.68
SD		2.69	11.61	0.02	0.02			0.06	0.24	0.32
%CV		33.88	12.46	26.75	26.75			18.93	18.93	18.93
n		11.00	11.00	11.00	11.00			11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
The Spiking solutions were made on: 22/02/2006
***Repeat injections of QC a or b**

Submitted by: Shyam Sundar Date: 04/03/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/04/2015

Plasma Concentrations of Hypoxanthine in Rat 375
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 19/02/2015 - 25/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Hypoxanthine 5 ng								
a25	25ug/ml (a)	120.42	7.40	16.27	16.27	2	1644.18	65.77
b25	25ug/ml (b)	122.45	8.59	14.25	14.25	2	1671.90	66.88
Mean		121.44	8.00	15.26	15.26		1658.04	66.32
SD		1.44	0.84	1.43	1.43		19.60	0.78
%CV		1.18	10.52	9.35	9.35		1.18	1.18
N		2.00	2.00	2.00	2.00		2.00	2.00
a5								
b5	5ug/ml (a)	16.30	10.21	1.60	1.60	2	222.56	44.51
	5ug/ml (b)	13.21	7.83	1.69	1.69	2	180.37	36.07
	5ug/ml	13.80	9.41	1.47	1.47	2	188.42	37.68
	5ug/ml	14.01	8.85	1.58	1.58	2	191.29	38.26
	5ug/ml	16.13	9.62	1.68	1.68	2	220.23	44.05
	5ug/ml	13.53	10.07	1.34	1.34	2	184.74	36.95
Mean		14.50	9.33	1.56	1.56		197.93	39.59
SD		1.36	0.88	0.13	0.13		18.55	3.71
%CV		9.37	9.46	8.47	8.47		9.37	9.37
N		6.00	6.00	6.00	6.00		6.00	6.00
BLANKS:								
aB	0ug/ml (a)	ND	8.81	0.00	0.00	2	0.00	
bB	0ug/ml (b)	ND	8.09	0.00	0.00	2	0.00	
Mean		0.00	8.45	0.00	0.00		0.00	
SD		0.00	0.51	0.00	0.00		0.00	
%CV		ERR	6.03	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00		2.00	
Hypoxanthine 5 ng 20/02/15								
Hypoxanthine 5 ng 23/02/15								
Hypoxanthine 5 ng 24/02/15								
Hypoxanthine 5 ng 25/02/15								

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	15.26	0.00	15.26
5.00	1.56	0.00	1.56
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output

Constant	-0.7114
Std Err of Y Est	1.1526
R Squared	0.9906
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.6319
Std Err of Coef.	0.0616

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c(µg/mL);onc(µM)	Conc. (µM) Corrected for dilution
T0 R375	0.00	14.27	20.95	0.68	0.68	5	-	2.20	16.19
T0.08 R375	0.08	10.16	14.60	0.70	0.70	5	-	2.23	16.36
T0.25 R375	0.25	13.09	17.32	0.76	0.76	5	-	2.32	17.06
T1 R375	1.00	12.11	15.97	0.76	0.76	5	-	2.33	17.09
Isoproterenol (30 mg/kg)									
T1.2 R375	1.20	13.48	15.87	0.85	0.85	5	-	2.47	18.15
T1.5 R375	1.50	13.99	18.47	0.76	0.76	5	-	2.32	17.08
T2 R375	2.00	14.59	21.01	0.69	0.69	5	-	2.22	16.35
T3 R375	3.00	13.14	17.48	0.75	0.75	5	-	2.32	17.01
T4 R375	4.00	12.20	17.79	0.69	0.69	5	-	2.21	16.25
T5 R375	5.00	16.83	20.98	0.80	0.80	5	-	2.40	17.60
T6 R375	6.00	17.01	20.61	0.83	0.83	5	-	2.43	17.87
Mean		13.10	17.71	0.74	0.74			2.30	16.91
SD		1.42	2.34	0.05	0.05			0.09	0.63
%CV		10.85	13.20	7.27	7.27			3.71	3.71
n		8.00	8.00	8.00	8.00			8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 04/03/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/04/2015

Plasma Concentrations of Xanthine in Rat 375
Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
Experiment Date: 19/02/2015 -25/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio Value	Peak Ht. Ratio	Inj Vol. (µL)	Amount Rec. (ng)	Recovery (%)
Xanthine 5 ng		37.40				5		
a25	25ug/ml (a)	61.94	7.40	8.37	8.37	2	2484.22	99.37
b25	25ug/ml (b)	55.25	8.59	6.43	6.43	2	2215.91	88.64
Mean		58.60	8.00	7.40	7.40		2350.07	94.00
SD		4.73	0.84	1.37	1.37		189.73	7.59
%CV		8.07	10.52	18.52	18.52		8.07	8.07
N		2.00	2.00	2.00	2.00		2.00	2.00
a5	5ug/ml (a)	3.71	10.21	0.36	0.36	2	148.80	29.76
b5	5ug/ml (b)	3.59	7.83	0.46	0.46	2	143.98	28.80
	5ug/ml	3.04	9.41	0.32	0.32	2	121.93	24.39
	5ug/ml	4.50	8.85	0.51	0.51	2	180.48	36.10
	5ug/ml	4.92	9.62	0.51	0.51	2	197.33	39.47
	5ug/ml	4.75	10.07	0.47	0.47	2	190.51	38.10
Mean		4.09	9.33	0.44	0.44		163.84	32.77
SD		0.75	0.88	0.08	0.08		29.96	5.99
%CV		18.28	9.46	17.83	17.83		18.28	18.28
N		6.00	6.00	6.00	6.00		6.00	6.00
BLANKS:								
aB	0ug/ml (a)	ND	8.81	0.00	0.00	2	0.00	0.00
bB	0ug/ml (b)	ND	8.09	0.00	0.00	2	0.00	0.00
Mean		0.00	8.45	0.00	0.00		0.00	0.00
SD		0.00	0.51	0.00	0.00		0.00	0.00
%CV		ERR	6.03	ERR	ERR		ERR	ERR
N		2.00	2.00	2.00	2.00		2.00	2.00

Xanthine 5 ng 20/02/15	38.72	10.87	5.00
Xanthine 5 ng 23/02/15	39.70	10.32	5.00
Xanthine 5 ng 20/02/15	37.82	10.01	5.00
Xanthine 5 ng 23/02/15	39.97	10.69	5.00

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	7.40	0.00	7.40
5.00	0.44	0.00	0.44
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	0.0000
Std Err of Y Est	0.7217
R Squared	0.9698
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.2880
Std Err of Coef.	0.0283

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	Conc.(µM)	Conc.(µM) Corrected for dilution
T0 R375	0.00	INT	20.95	0.00	0.00	5	-	0.00	0.00	0.00
T0.08 R375	0.08	INT	14.60	0.00	0.00	5	-	0.00	0.00	0.00
T0.25 R375	0.25	INT	17.32	0.00	0.00	5	-	0.00	0.00	0.00
T1 R375	1.00	INT	15.97	0.00	0.00	5	-	0.00	0.00	0.00
Isoproterenol (30 mg/kg)										
T1.2 R375	1.20	INT	15.87	0.00	0.00	5	-	0.00	0.00	0.00
T1.5 R375	1.50	INT	18.47	0.00	0.00	5	-	0.00	0.00	0.00
T2 R375	2.00	INT	21.01	0.00	0.00	5	-	0.00	0.00	0.00
T3 R375	3.00	INT	17.48	0.00	0.00	5	-	0.00	0.00	0.00
T4 R375	4.00	INT	17.79	0.00	0.00	5	-	0.00	0.00	0.00
T5 R375	5.00	INT	20.98	0.00	0.00	5	-	0.00	0.00	0.00
T6 R375	6.00	INT	20.61	0.00	0.00	5	-	0.00	0.00	0.00
Mean		0.00	17.71	0.00	0.00			0.00	0.00	0.00
SD		0.00	2.34	0.00	0.00			0.00	0.00	0.00
%CV		ERR	13.20	ERR	ERR			ERR	ERR	ERR
n		8.00	8.00	8.00	8.00			8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
The Spiking solutions were made on: 22/02/2006
***Repeated injections of QC a or b**

Submitted by: Shyam Sundar Date: 04/03/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/04/2015

Plasma Concentrations of Uric Acid in Rat 375
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 19/02/2015 - 25/02/2015

Conc. ug/mL	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Uric Acid 5 ng		30.40				5		
a25	25ug/ml (a)	58.45	7.40	7.90	7.90	2	2884.05	106.74
b25	25ug/ml (b)	55.77	8.59	6.49	6.49	2	2751.81	101.45
Mean		57.11	8.00	7.20	7.20		2817.93	104.09
SD		1.90	0.84	0.99	0.99		93.51	3.74
%CV		3.32	10.52	13.82	13.82		3.32	3.59
N		2.00	2.00	2.00	2.00		2.00	2.00
a5	5ug/ml (a)	10.27	10.21	1.01	1.01	2	506.74	58.22
b5	5ug/ml (b)	9.88	7.83	1.26	1.26	2	487.50	54.38
	5ug/ml	8.05	9.41	0.86	0.86	2	397.20	36.32
	5ug/ml	9.18	8.85	1.04	1.04	2	452.96	47.47
	5ug/ml	10.23	9.62	1.06	1.06	2	504.77	43.22
	5ug/ml	11.93	10.07	1.18	1.18	2	588.65	60.00
Mean		9.92	9.33	1.07	1.07		489.64	49.93
SD		1.29	0.88	0.14	0.14		63.99	9.23
%CV		12.99	9.46	13.32	13.32		12.99	18.49
N		6.00	6.00	6.00	6.00		6.00	6.00
BLANKS:								
aB	0ug/ml (a)	2.89	8.81	0.33	0.33	2	142.60	
bB	0ug/ml (b)	5.85	8.09	0.72	0.72	2	288.65	
Mean		4.37	8.45	0.53	0.53		215.63	
SD		2.09	0.51	0.28	0.28		103.27	
%CV		47.90	6.03	53.15	53.15		47.90	
N		2.00	2.00	2.00	2.00		2.00	
Uric Acid 5 ng 20/02/2015		33.22	10.87				5.00	
Uric Acid 5 ng 23/02/2015		33.99	10.32				5.00	
Uric Acid 5 ng 24/02/2015		32.95	10.01				5.00	
Uric Acid 5 ng 25/02/2015		32.01	10.63				5.00	

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	7.20	0.53	6.67
5.00	1.07	0.53	0.54
0.00	0.53	0.53	0.00

Regression Output Begins Here:

Regression Output

Constant	-0.3769
Std Err of Y Est	0.6106
R Squared	0.9864
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.2781
Std Err of Coef.	0.0326

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c(µg/mL);onc(µM)	Conc. (µM) Corrected for dilution
T0 R375	0.00	6.95	20.95	0.33	0.33	5	-	2.55	15.16
T0.08 R375	0.08	21.41	14.60	1.47	1.47	5	-	6.63	39.43
T0.25 R375	0.25	15.97	17.32	0.92	0.92	5	-	4.67	27.79
T1 R375	1.00	17.58	15.97	1.10	1.10	5	-	5.31	31.61
Isoproterenol (30 mg/kg)									
T1.2 R375	1.20	24.89	15.87	1.57	1.57	5	-	6.99	51.39
T1.5 R375	1.50	49.83	18.47	2.70	2.70	5	-	11.06	81.24
T2 R375	2.00	39.83	21.01	1.90	1.90	5	-	8.17	48.61
T3 R375	3.00	28.74	17.48	1.64	1.64	5	-	7.27	43.23
T4 R375	4.00	22.84	17.79	1.28	1.28	5	-	5.97	35.52
T5 R375	5.00	14.49	20.98	0.69	0.69	5	-	3.84	22.84
T6 R375	6.00	12.02	20.61	0.58	0.58	5	-	3.45	20.54
Mean		25.65	17.71	1.45	1.45			6.58	42.31
SD		13.74	2.34	0.70	0.70			2.53	19.67
%CV		53.58	13.20	48.37	48.37			38.41	46.50
n		8.00	8.00	8.00	8.00			8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No. 181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 04/03/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/04/2015

Plasma Concentrations of Guanosine in Rat 375
Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
Experiment Date: 19/02/2015 - 25/02/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Guanosine 5 ng		52.68							
a2.5	2.5ug/ml (a)	24.44	29.66	0.82	0.82	10	139.18	55.67	
b2.5	2.5ug/ml (b)	24.75	37.57	0.66	0.66	10	140.95	56.38	
Mean		24.60	33.62	0.74	0.74		140.06	56.03	
SD		0.22	5.59	0.12	0.12		1.25	0.50	
%CV		0.89	16.64	15.76	15.76		0.89	0.89	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a0.5	0.5ug/ml (a)	1.85	84.61	0.02	0.02	30.00	3.51	7.02	
b0.5	0.5ug/ml (b)	1.90	76.24	0.02	0.02	30.00	3.61	7.21	
	0.5ug/ml	2.17	84.50	0.03	0.03	30.00	4.12	8.24	
	0.5ug/ml	2.38	82.78	0.03	0.03	30.00	4.52	9.04	
	0.5ug/ml	2.80	83.74	0.03	0.03	30.00	5.32	10.63	
	0.5ug/ml	2.92	83.93	0.03	0.03	30.00	5.54	11.09	
Mean		2.34	82.63	0.03	0.03		4.44	8.87	
SD		0.45	3.20	0.01	0.01		0.85	1.71	
%CV		19.25	3.87	17.94	17.94		19.25	19.25	
N		6.00	6.00	6.00	6.00		6.00	6.00	
BLANKS:									
aB	0ug/mL (a)	ND	8.81	0.00	0.00	2	0.00		
bB	0 ug/mL (b)	ND	8.09	0.00	0.00	2	0.00		
Mean		0.00	8.45	0.00	0.00		0.00		
SD		0.00	0.51	0.00	0.00		0.00		
%CV		ERR	6.03	ERR	ERR		ERR		
N		2.00	2.00	2.00	2.00		2.00		
'Guanosine 5 ng 20/02/15		56.43	10.87			5.00			
'Guanosine 5 ng 23/02/15		55.25	10.32			5.00			
'Guanosine 5 ng 24/02/15		54.34	10.01			5.00			
'Guanosine 5 ng 25/02/15		54.17	10.63			5.00			

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.74	0.00	0.74
0.50	0.03	0.00	0.03
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0572
Std Err of Y Est	0.0926
R Squared	0.9757
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3137
Std Err of Coef.	0.0495

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	Conc.(µM)	Conc.(µM) Corrected for dilution
T0 R375	0.00	2.26	110.24	0.021	0.021	35	-	-	0.25	0.87	1.17
T0.08 R375	0.08	1.83	78.92	0.023	0.023	35	-	-	0.26	0.90	1.21
T0.25 R375	0.25	2.14	93.78	0.023	0.023	35	-	-	0.25	0.90	1.20
T1 R375	1.00	2.94	81.78	0.036	0.036	35	-	-	0.30	1.05	1.40
Isoproterenol (30 mg/kg)											
T1.2 R375	1.20	1.98	80.96	0.024	0.024	35	-	-	0.29	1.09	1.46
T1.5 R375	1.50	2.83	89.90	0.031	0.031	35	-	-	0.28	1.06	1.41
T2 R375	2.00	2.32	103.77	0.022	0.022	35	-	-	0.25	0.90	1.19
T3 R375	3.00	2.13	89.00	0.024	0.024	35	-	-	0.26	0.91	1.22
T4 R375	4.00	2.34	84.03	0.028	0.028	35	-	-	0.27	0.96	1.28
T5 R375	5.00	2.56	104.87	0.024	0.024	35	-	-	0.26	0.92	1.22
T6 R375	6.00	2.99	107.80	0.028	0.028	35	-	-	0.27	0.96	1.27
Mean		2.39	93.19	0.03	0.03				0.27	0.96	1.27
SD		0.39	11.61	0.00	0.01				0.02	0.08	0.10
%CV		16.30	12.46	17.54	19.40				6.19	7.93	7.93
n		22.00	22.00	11.00	11.00				11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
The Spiking solutions were made on: 22/02/2006
***Repeat injections of QC a or b**

Submitted by: Shyam Sundar Date: 04/02/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 10/04/2015

Title: Measurement of Plasma Concentrations of Diprydamole in Rat 375

According to SOP No: SOP/STD/2008-001-1 (Plasma with no Stopping Solution) Experiment Date:08/10/2014 - 10/10/2014

Abs.amt ng	STD ID	Peak Ht. #	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Diprydamole (1ng)		75.64				1		
a1000	1 ug/mL(a)	102.96	6.12	16.82	16.82	5	54.45	108.89
b1000	1ug/mL(b)	109.56	5.43	20.18		5	57.94	115.88
1000*	1ug/mL(c)	97.85	6.63	14.76	14.76	5	51.75	103.49
Mean		103.46	6.06	17.25	15.79		54.71	109.42
SD		5.87	0.60	2.73	1.46		3.10	6.21
%CV		5.67	9.94	15.85	9.25		5.67	5.67
n		3.00	3.00	3.00	2.00		1.00	1.00
a100	0.1 ug/mL (a)	41.22	23.93	1.72	1.72	20	5.45	108.99
b100	0.1ug/mL (b)	37.69	17.14	2.20	2.20	20	4.98	99.66
Mean		39.46	20.54	1.96	1.96		5.22	104.32
SD		2.50	4.80	0.34	0.34		0.33	6.60
%CV		6.33	23.38	17.18	17.18		6.33	6.33
n		2.00	2.00	2.00	2.00		2.00	2.00
aB	0 ug/mL (a)	3.37	19.74	0.17		20	0.45	8.91
bB	0 ug/mL (b)	1.53	18.70	0.08	0.08	20	0.20	4.05
Mean		2.45	19.22	0.13	0.08		0.32	6.48
SD		1.30	0.74	0.06	ERR		0.17	3.44
%CV		53.11	3.83	49.78	ERR		53.11	53.11
n		2.00	2.00	2.00	1.00		2.00	2.00

Plasma Conc. (ug/mL)	Peak Ht.Ratio (PHR)	Blank (PHRb)	PHRV-PHRb
0.00	0.08	0.08	0.00
0.10	1.96	0.08	1.88
1.00	15.79	0.08	15.71

Regression Output Begins Here:

Regression Output:	
Constant	0.1523
Std Err of Y Est	0.2283
R Squared	0.9996
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	15.5739
Std Err of Coef.	0.2931

Sample ID	Time Post-dose (h)	Peak Ht. #	Peak Ht. I.S. (mm)*	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc.(ug/mL)
R375T0	0.00	9.17	23.12	0.40	0.40	20	-	0.11
R375T0.08	0.08	16.78	31.06	0.54	0.54	20	-	0.18
R375T0.25	0.25	41.86	33.36	1.25	1.25	20	-	0.51
R375T1	1.00	22.97	25.62	0.90	0.90	20	-	0.35
Internal Standard (30 mg/kg sc)						20		
R375T1.2	1.20	16.85	28.55	0.59	0.59	20	-	0.86
R375T1.5	1.50	12.35	18.83	0.66	0.66	20	-	0.23
R375T2	2.00	19.05	17.12	1.11	1.11	20	-	0.45
R375T3	3.00	4.88	21.69	0.22	0.22	20	-	0.03
R375T4	4.00	11.15	23.91	0.47	0.47	20	-	0.15
R375T5	5.00	3.18	14.69	0.22	0.22	20	-	0.03
R375T6	6.00	15.76	25.14	0.63	0.63	20	-	0.22
Mean		15.82	23.92	0.63	0.63			0.28
SD		10.48	5.73	0.33	0.33			0.25
%CV		66.28	23.98	52.75	52.75			86.55
n		11.00	11.00	11.00	11.00			11.00

Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 Corr. PHR = (PHR - RGB PHR)

Diprydamole (1ng)(03/10/2014) 90.59 9.29 1.00

Comments: Plasma from Rat 156 was used for extraction QC's.
 *A repeat injection of a or b

Submitted by: Shyam Sundar Date: 14/010/2014

Checked by: Pollen Yeung Date:05/11/2014

Approved by: Date:

APPENDIX 6: Rat 376

Title: Measurement of RBC Concentrations of ATP in Rat 376 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
Experiment Date 08-09/05/2014

Sample/Standard ID	Standard Concentra (ug/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CoPHR	CoPHR Value	Inj Vol. (uL)	Amount Recove %	Recovery
ATP 4 ng										
a250	250 ug/mL	47.21	6.46	7.31	35.00	7.31	7.31	0.35	24936.85	83.38
b250	250 ug/mL	34.91	5.74	6.08	35.00	6.08	6.08	0.35	18439.86	57.40
Mean		41.06	6.10	6.69	35.00	6.69	6.69		21688.36	70.39
SD		8.70	0.51	0.87	0.00	0.87	0.87		4594.07	18.38
%CV		21.18	8.35	12.95	0.00	12.95	12.95		21.18	26.11
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a100										
b100	100 ug/mL	17.80	6.11	2.91	35.00	2.91	2.91	0.35	9402.16	53.11
100*	100 ug/mL	25.12	7.50	3.35	35.00	3.35	3.35	0.35	13268.67	91.78
100*	100 ug/mL	30.10	14.55	2.07	35.00	2.07		0.35	15899.16	118.08
Mean		24.34	9.39	2.78	35.00	2.78	3.13		11335.41	72.44
SD		6.19	4.53	0.65	0.00	0.65	0.31		2734.03	27.34
%CV		25.42	48.21	23.44	0.00	23.44	9.85		24.12	37.74
n		3.00	3.00	3.00	3.00	3.00	2.00		2.00	2.00
a8										
b8	0 ug/mL (a)	9.72	11.47	0.85	35.00	0.85	0.85	0.35	5134.21	
	0 ug/mL (a)	5.77	8.00	0.72	35.00	0.72	0.72	0.35	3047.78	
Mean		7.75	9.74	0.78	35.00	0.78	0.78		4091.00	
SD		2.79	2.45	0.09	0.00	0.09	0.09		1475.33	
%CV		36.06	25.20	11.58	0.00	11.58	11.58		36.06	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
250.00	6.89	0.78	5.91
100.00	3.13	0.78	2.35
0.00	0.78	0.78	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0068
Std Err of Y Est	0.0140
R Squared	1.0000
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0237
Std Err of Coef.	0.0001

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CoPHR	CoPHR Value	Inj Vol. (uL)	Hemolysis Degree	Conc(ug/mL) Lysate	Conc.(mM) RBC	
R37670	0.00	102.42	86.97	1.18	35.00	1.18	1.18	1.50	-	50.08	0.0987	1.269
R37670.08	0.08	126.67	85.63	1.48	35.00	1.48	1.48	1.50	-	62.83	0.1239	1.593
R37670.25	0.25	124.61	86.11	1.88	35.00	1.88	1.88	1.00	-	79.96	0.1577	2.027
R37671	1.00	95.61	88.00	1.40	35.00	1.40	1.40	1.00	-	59.67	0.1177	1.513
Ibuprofenolol (30 mg/kg sc)												
R37671.2	1.20	92.54	63.97	1.45	35.00	1.45	1.45	1.00	-	61.45	0.1212	1.558
R37671.5	1.50	76.88	51.47	1.49	35.00	1.49	1.49	1.00	-	63.44	0.1251	1.606
R37672	2.00	69.19	44.94	1.54	35.00	1.54	1.54	1.00	-	65.36	0.1289	1.657
R37673	3.00	90.41	56.10	1.61	35.00	1.61	1.61	1.00	-	69.43	0.1349	1.735
R37271.2	1.20	56.52	34.78	1.63	35.00	1.63	1.63	1.00	-	69.00	0.1360	1.748
R37275	5.00	76.48	37.30	2.05	35.00	2.05	2.05	1.00	-	86.98	0.1715	2.205
R37276	6.00	61.34	35.06	1.75	35.00	1.75	1.75	1.00	-	74.26	0.1464	1.882
Mean		88.42	57.30	1.59	35.00	1.59	1.59	1.09		67.41	0.13	1.71
SD		23.32	18.73	0.24	0.00	0.24	0.24	0.20		10.14	0.02	0.26
%CV		26.37	32.69	15.11	0.00	15.11	15.11	18.54		15.04	15.04	15.04
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00		11.00	11.00	11.00

ATP (4 ng) 09/05/2014 14.47

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.
*Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
Peak Ht. = peak height
Peak Ht. R. (or: PHR) = peak height ratio
I.S. = internal standard
Inj Vol = injection volume
ND = not detected or determined
NS = no sample
INT = interference
PCV = packed cell volume (haematocrit)
CoPHR = corrected peak height ratio
Hemolysis Degree:
-: no visible hemolysis
+: slight hemolysis
++: intermediate hemolysis
+++: serious hemolysis

Submitted by: Shyam Sundar K Date: 20/05/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 27/05/2014

Title: Measurement of RBC Concentrations of ADP in Rat 376 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)

Experiment Date: 08-09/05/2014

Sample/standard ID	Standard Co (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered	Recovery %
ADP 4 ng		26.98						4.00		
s250	250 µg/mL	69.92	6.48	10.82	35.00	10.82	10.82	0.35	16289.74	62.85
b250	250 µg/mL	52.30	5.74	9.11	35.00	9.11	9.11	0.35	12184.69	46.53
Mean		61.11	6.10	9.97	35.00	9.97	9.97		14237.21	54.74
SD		12.46	0.51	1.21	0.00	1.21	1.21		2902.71	11.61
%CV		20.39	8.35	12.15	0.00	12.15	12.15		20.39	21.21
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
100a	100 µg/mL	22.71	6.11	3.72	35.00	3.72	3.72	0.35	5290.90	47.38
100b	100 µg/mL	33.79	7.50	4.51	35.00	4.51	4.51	0.35	7872.29	73.19
100*	100 µg/mL	43.81	14.55	3.01	35.00	3.01		0.35	10206.71	96.53
Mean		33.44	9.39	3.74	35.00	3.74	4.11		7789.97	72.37
SD		10.55	4.53	0.75	0.00	0.75	0.56		2458.84	24.59
%CV		31.57	48.21	19.96	0.00	19.96	13.56		31.57	33.98
n		3.00	3.00	3.00	3.00	3.00	2.00		3.00	3.00
aB	0 µg/mL (a)	3.04	11.47	0.27	35.00	0.27	0.27	0.35	708.25	
bB	0 µg/mL (a)	1.71	8.00	0.21	35.00	0.21	0.21	0.35	398.39	
Mean		2.38	9.74	0.24	35.00	0.24	0.24		553.32	
SD		0.84	2.45	0.04	0.00	0.04	0.04		219.10	
%CV		39.60	25.20	15.15	0.00	15.15	15.15		39.60	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
250.00	9.97	0.24	9.73
100.00	4.11	0.24	3.87
0.00	0.24	0.24	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0077
Std Err of Y Est	0.0159
R Squared	1.0000
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0389
Std Err of Coef.	0.0001

Sample ID	Time post	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R376T0	0.00	21.81	66.97	0.25	35.00	0.25	0.25	1.50	-	6.64	0.0155
R376T0.08	0.08	33.00	65.63	0.39	35.00	0.39	0.39	1.50	-	10.10	0.0236
R376T0.25	0.25	23.56	66.11	0.36	35.00	0.36	0.36	1.00	-	9.35	0.0219
R376T1	1.00	21.92	68.00	0.32	35.00	0.32	0.32	1.00	-	8.48	0.0198
Isoproterenol (30 mg/kg sc)											
R376T1.2	1.20	18.45	63.97	0.29	35.00	0.29	0.29	1.00	-	7.61	0.0178
R376T1.5	1.50	13.20	51.47	0.26	35.00	0.26	0.26	1.00	-	6.79	0.0159
R376T2	2.00	9.42	44.94	0.21	35.00	0.21	0.21	1.00	-	5.58	0.0131
R376T3	3.00	12.79	56.10	0.23	35.00	0.23	0.23	1.00	-	6.06	0.0142
R372T1.2	1.20	10.24	34.78	0.29	35.00	0.29	0.29	1.00	-	7.76	0.0182
R372T5	5.00	11.89	37.30	0.32	35.00	0.32	0.32	1.00	-	8.39	0.0196
R372T6	6.00	8.30	35.06	0.24	35.00	0.24	0.24	1.00	-	6.28	0.0147
Mean		26.12	79.57	0.33	35.00	0.33		1.33		8.70	0.02
SD		6.02	11.88	0.07	0.00	0.07		0.29		1.82	0.00
%CV		23.04	14.67	21.41	0.00	21.41		21.65		20.92	20.92
n		3.00	3.00	3.00	3.00	3.00		3.00		3.00	3.00

ADP (4 ng)09/05/2014

34.74

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.

*Repeated injections of a or b at 0.5-1ul injection volume.

PL = plasma; RBC = red blood cells

Peak Ht. = peak height

Peak Ht. R. (or: PHR) = peak height ratio

I.S. = internal standard

Inj Vol = injection volume

ND = not detected or determined

NS = no sample

INT = interference

PCV = packed cell volume (haematocrit)

CorPHR = corrected peak height ratio

Hemolysis Degree:

--: no visible hemolysis

+ : slight hemolysis

++ : intermediate hemolysis

+++ : serious hemolysis

Submitted by: Shyam Sundar K

Date: 20/05/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date: 27/05/2014

Title: Measurement of RBC Concentrations of AMP in Rat 376 extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date 08-09/05/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
AMP 4 ng										
a50	50 µg/mL	31.51	6.46	4.88	35.00	4.88	4.88	0.35	4020.76	75.90
b50	50 µg/mL	27.22	5.74	4.74	35.00	4.74	4.74	0.35	3473.35	64.85
Mean		29.37	6.10	4.81	35.00	4.81	4.81		3747.06	70.42
SD		3.03	0.51	0.10	0.00	0.10	0.10		387.08	7.74
%CV		10.33	8.35	1.99	0.00	1.99	1.99		10.33	10.99
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20										
a20	20 µg/mL	10.88	6.11	1.78	35.00	1.78	1.78	0.35	1388.33	58.12
b20	20 µg/mL	15.69	7.50	2.09	35.00	2.09	2.09	0.35	2002.09	88.81
20*	20 µg/mL	22.94	14.55	1.58	35.00	1.58	1.58	0.35	2927.21	135.07
Mean		16.50	9.39	1.82	35.00	1.82	1.82		2105.87	94.00
SD		6.07	4.53	0.26	0.00	0.26	0.26		774.68	38.73
%CV		36.79	48.21	14.29	0.00	14.29	14.29		36.79	41.21
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
a8										
a8	0 µg/mL (a)	1.56	11.47	0.14	35.00	0.14	0.14	0.35	199.06	
b8	0 µg/mL (a)	1.98	6.00	0.25	35.00	0.25	0.25	0.35	252.65	
Mean		1.77	9.74	0.19	35.00	0.19	0.19		225.86	
SD		0.30	2.45	0.08	0.00	0.08	0.08		37.99	
%CV		16.78	25.30	41.11	0.00	41.11	41.11		16.78	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	4.81	0.19	4.62
20.00	1.62	0.19	1.42
0.00	0.19	0.19	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0879
Std Err of Y Est	0.1605
R Squared	0.9970
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0929
Std Err of Coef.	0.0051

Sample ID	Time post dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC	
R37670	0.00	0.58	86.97	0.01	35.00	0.01	0.01	1.50	-	1.02	0.0029	0.038
R37670.08	0.08	4.25	85.63	0.05	35.00	0.05	0.05	1.50	-	1.49	0.0043	0.055
R37670.25	0.25	0.42	66.11	0.01	35.00	0.01	0.01	1.00	-	1.01	0.0029	0.038
R37671	1.00	2.45	68.00	0.04	35.00	0.04	0.04	1.00	-	1.33	0.0038	0.049
Soprolefenol (30 mg/kg sc)												
R37671.2	1.20	0.98	63.97	0.02	35.00	0.02	0.02	1.00	-	1.11	0.0032	0.041
R37671.5	1.50	0.64	51.67	0.01	35.00	0.01	0.01	1.00	-	1.06	0.0031	0.040
R37672	2.00	0.52	44.94	0.01	35.00	0.01	0.01	1.00	-	1.07	0.0031	0.040
R37673	3.00	0.46	56.10	0.01	35.00	0.01	0.01	1.00	-	1.03	0.0030	0.038
R37211.2	1.20	0.35	34.78	0.01	35.00	0.01	0.01	1.00	-	1.06	0.0030	0.038
R37273	5.00	0.49	37.30	0.01	35.00	0.01	0.01	1.00	-	1.09	0.0031	0.040
R37276	6.00	0.43	35.06	0.01	35.00	0.01	0.01	1.00	-	1.08	0.0031	0.040
Mean		1.06	57.30	0.02	35.00	0.02	0.02	1.09	0.00	1.12	0.00	0.04
SD		1.24	16.73	0.01	0.00	0.01	0.01	0.20	0.00	0.15	0.00	0.01
%CV		117.19	32.69	93.78	0.00	93.78	93.78	18.54	0.00	13.33	13.33	13.33
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

AMP (4 ng)/09/05/2014 70.09

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of "a" or "b" at 0.5 - 1 ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 - : no visible hemolysis
 + : slight hemolysis
 ++ : intermediate hemolysis
 +++ : serious hemolysis

Submitted by: Shyam Sundar Date: 09/05/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 27/05/2014

Title: Measurement of RBC Concentrations of GTP in Rat 376 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 08-09/05/2014

Sample/standard ID	Standard Concentra (ug/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (uL)	Hemolysis Degree	Amount Recove (%)	Recovery
GTP 4 ng											
a50	50 ug/mL	14.07	6.46	2.18	35.00	2.18	2.18	0.35	1708.33	30.78	
b50	50 ug/mL	10.53	5.74	1.83	35.00	1.83	1.83	0.35	1278.51	22.18	
Mean		12.30	6.10	2.01	35.00	2.01	2.01		1493.42	26.48	
SD		2.50	0.24	0.00	0.00	0.24	0.24		303.92	6.08	
%CV		20.35	0.00	12.11	0.00	12.11	12.11		20.35	22.95	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00	
a20	20 ug/mL	4.34	8.11	0.71	35.00	0.71	0.71	0.35	538.95	17.88	
b20	20 ug/mL	7.10	7.50	0.95	35.00	0.95	0.95	0.35	862.05	34.83	
20*	20 ug/mL	8.46	14.55	0.58	35.00	0.58	0.58	0.35	1027.18	42.89	
Mean		6.63	9.39	0.75	35.00	0.75	0.75		805.39	31.80	
SD		2.10	4.53	0.19	0.00	0.19	0.19		254.89	12.74	
%CV		31.65	48.21	24.82	0.00	24.82	24.82		31.65	40.08	
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00	
aB	0 ug/mL (a)	1.92	11.47	0.17	35.00	0.17	0.17	0.35	233.12		
bB	0 ug/mL (a)	0.87	8.00	0.11	35.00	0.11	0.11	0.35	195.63		
Mean		1.40	9.74	0.14	35.00	0.14	0.14		169.38		
SD		0.74	2.45	0.04	0.00	0.03	0.04		90.15		
%CV		53.22	25.20	30.03	0.00	24.52	30.03		53.22		
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	Peak Height Ratio Value (PHRV)	Blank PHRV-PHRb	PHRV-PHRb
50.00	2.01	0.14	1.87
20.00	0.75	0.14	0.61
0.00	0.14	0.14	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0549
Std Err of Y Est	0.1129
R Squared	0.9930
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0377
Std Err of Coef.	0.0032

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (uL)	Hemolysis Degree	Conc.(ug/mL) Lysate	Conc.(mM) RBC
R37670	0.00	21.67	86.97	0.25	35.00	0.25	0.25	1.50	-	8.06	0.0154
R37670.08	0.08	26.03	85.63	0.30	35.00	0.30	0.30	1.50	-	9.51	0.0182
R37670.25	0.25	27.36	86.11	0.41	35.00	0.41	0.41	1.00	-	12.43	0.0237
R37671	1.00	19.85	68.00	0.29	35.00	0.29	0.29	1.00	-	9.19	0.0178
Isoproterenol (20 mg/kg sc)											
R37671.2	1.20	20.85	63.97	0.32	35.00	0.32	0.32	1.00	-	10.02	0.0191
R37671.5	1.50	17.17	51.47	0.33	35.00	0.33	0.33	1.00	-	10.30	0.0197
R37672	2.00	16.50	44.94	0.37	35.00	0.37	0.37	1.00	-	11.19	0.0214
R37673	3.00	22.47	56.10	0.40	35.00	0.40	0.40	1.00	-	12.07	0.0231
R37271.2	1.20	11.16	34.78	0.32	35.00	0.32	0.32	1.00	-	9.96	0.0190
R37275	5.00	16.50	37.30	0.44	35.00	0.44	0.44	1.00	-	13.18	0.0252
R37276	6.00	12.15	35.06	0.35	35.00	0.35	0.35	1.00	-	10.64	0.0203
Mean		19.23	57.30	0.34	35.00	0.34	0.34	1.09	0.00	10.60	0.02
SD		5.15	18.73	0.06	0.00	0.06	0.06	0.20	0.00	1.52	0.00
%CV		26.80	32.69	16.58	0.00	16.58	16.58	18.54	0.00	14.30	14.30
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

GTP (4 ng/09/05/2014) 73.30

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 20/05/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 27/05/2014

Title: Measurement of RBC Concentrations of GDP in Rat 376 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 08-09/05/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
GDP 4 ng										
a50	50 ug/mL	16.69	6.46	2.58	35.00	2.58	2.58	0.35	3039.95	60.80
b50	50 ug/mL	14.33	5.74	2.50	35.00	2.50	2.50	0.35	2810.09	52.20
Mean		15.51	6.10	2.54	35.00	2.54	2.54		2825.02	56.50
SD		1.67	0.51	0.06	0.00	0.06	0.06		303.95	6.08
%CV		10.76	8.35	2.42	0.00	2.42	2.42		10.76	10.76
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20										
a20	20 ug/mL	5.63	6.11	0.92	35.00	0.92	0.92	0.35	1025.46	51.27
b20	20 ug/mL	9.12	7.50	1.22	35.00	1.22	1.22	0.35	1661.13	83.06
20*	20 ug/mL	11.66	14.55	0.80	35.00	0.80	0.80	0.35	2123.77	106.19
Mean		8.80	9.39	0.98	35.00	0.98	0.98		1603.46	80.17
SD		3.03	4.53	0.21	0.00	0.21	0.21		551.42	27.57
%CV		34.39	48.21	21.78	0.00	21.78	21.78		34.39	34.39
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
aB										
aB	0 ug/mL (a)	ND	11.47	0.00	35.00	0.00	0.00	0.35		0.00
bB	0 ug/mL (a)	ND	6.00	0.00	35.00	0.00	0.00	0.35		0.00
Mean		0.00	9.74	0.00	35.00	0.00	0.00			0.00
SD		0.00	2.45	0.00	0.00	0.00	0.00			0.00
%CV		ERR	25.30	ERR	0.00	ERR	ERR			ERR
n		2.00	2.00	2.00	2.00	2.00	2.00			2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRB
50.00	2.54	0.00	2.54
20.00	0.98	0.00	0.98
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0144
Std Err of Y Est	0.0295
R Squared	0.9997
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0099
Std Err of Coef.	0.0068

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(nM) RBC
R37670	0.00	6.90	86.97	0.08	35.00	0.08	0.08	1.50	-	1.84	0.0042
R37670.08	0.08	8.79	85.63	0.10	35.00	0.10	0.10	1.50	-	2.30	0.0052
R37670.25	0.25	5.90	86.11	0.09	35.00	0.09	0.09	1.00	-	2.04	0.0046
R37671	1.00	5.90	88.00	0.09	35.00	0.09	0.09	1.00	-	1.99	0.0045
Isoproterenol (30 µg/kg sc)											
R37671.2	1.20	4.49	63.97	0.07	35.00	0.07	0.07	1.00	-	1.66	0.0037
R37671.5	1.50	2.62	51.47	0.05	35.00	0.05	0.05	1.00	-	1.36	0.0031
R37672	2.00	2.46	44.94	0.05	35.00	0.05	0.05	1.00	-	1.36	0.0031
R37673	3.00	4.84	56.10	0.09	35.00	0.09	0.09	1.00	-	1.98	0.0045
R37271.2	1.20	1.94	34.78	0.06	35.00	0.06	0.06	1.00	-	1.38	0.0031
R37275	5.00	2.17	37.30	0.06	35.00	0.06	0.06	1.00	-	1.43	0.0032
R37276	6.00	0.91	35.06	0.03	35.00	0.03	0.03	1.00	-	0.79	0.0018
Mean		30.79	57.30	0.07	35.00	0.07	0.07	1.09	0.00	1.65	0.00
SD		30.10	18.73	0.02	0.00	0.02	0.02	0.20	0.00	0.43	0.00
%CV		97.76	32.69	31.59	0.00	31.59	31.59	18.54	ERR	26.17	26.17
n		22.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

GDP (4ng)09/05/2014 48.22

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date:20/05/2014

Checked by: Date:

Approved by: Pollen Yeung Date:27/05/2014

Title: Measurement of RBC Concentrations of GMP in Rat 376 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 08-09/05/2014

Sample/standard	ID/Standard Concentr (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
GMP 4 ng										
a50	50 ug/mL	130.43	6.46	20.19	35.00	20.19	20.19	0.35	13607.40	272.15
b50	50 ug/mL	103.50	5.74	18.03	35.00	18.03	18.03	0.35	10797.87	215.96
Mean		116.97	6.10	19.11	35.00	19.11	19.11		12202.63	244.05
SD		19.04	5.92	1.53	0.00	1.53	1.53		1986.64	39.73
%CV		16.28	6.01	7.99	0.00	7.99	7.99		16.28	16.28
n		2.00	6.01	2.00	2.00	2.00	2.00		2.00	2.00
a20	20 ug/mL	86.67	6.11	14.23	35.00	14.23	14.23	0.35	9073.34	453.67
b20	20 ug/mL	121.61	7.50	16.21	35.00	16.21	16.21	0.35		
20*	20 ug/mL	124.73	14.55	8.57	35.00	8.57		0.35	13012.73	650.64
Mean		111.10	9.39	13.01	35.00	13.01	15.22		11043.03	552.15
SD		20.96	4.53	3.97	0.00	3.97	1.40		2785.57	139.28
%CV		18.86	48.21	30.49	0.00	30.49	9.20		25.22	25.22
n		3.00	3.00	3.00	3.00	3.00	2.00		2.00	2.00
aB	0 ug/mL (a)	Off scale	11.47	0.00	35.00	0.00		0.35		0.00
bB	0 ug/mL (a)		81.75	8.00	10.22	35.00	10.22	0.35		0.00
Mean		40.88	8.74	5.11	35.00	5.11	10.22			0.00
SD		57.81	2.45	7.23	0.00	7.23	ERR			0.00
%CV		141.42	25.20	141.42	0.00	141.42	ERR			ERR
n		2.00	2.00	2.00	2.00	2.00	2.00			2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	19.11	10.22	8.89
20.00	15.22	10.22	5.01
0.00	10.22	10.22	0.00

Regression Output Begins Here:

Regression Output:

Constant: 0.0000
 Std Err of Y Est: 0.9512
 R Squared: 0.9545
 No. of Observations: 3.0000
 Degrees of Freedom: 2.0000

X Coefficient(s): 0.1878
 Std Err of Coef.: 0.0177

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lyasate	Conc (mM) RBC	Conc (mM) RBC
R37670		0.00 off scale	86.67	0.00	35.00	0.00	0.00	1.50	-	0.00	0.0000	0.000
R37670.08		0.08 off scale	85.63	0.00	35.00	0.00	0.00	1.50	-	0.00	0.0000	0.000
R37670.25		0.25 off scale	86.11	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
R37671		1.00 off scale	68.00	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
Not detected (SD mg/kg sat)												
R37671.2		1.20 off scale	63.97	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
R37671.5		1.50 off scale	51.47	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
R37672		2.00 off scale	44.94	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
R37673		3.00 off scale	56.10	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
R37271.2		1.20 off scale	34.78	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
R37275		5.00 off scale	37.30	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
R37276		6.00 off scale	35.06	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
Mean		0.00	57.30	0.00	35.00	0.00	0.00	1.09	0.00	0.00	0.00	0.00
SD		0.00	18.73	0.00	0.00	0.00	0.00	0.20	0.00	0.00	0.00	0.00
%CV		ERR	32.69	ERR	0.00	ERR	ERR	18.54	ERR	ERR	ERR	ERR
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

GMP 4 ng/09/05/2014 88.07

Comments: RBC Lyasate from Rat 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of "a" or "b" at 0.5-1ul injection volume

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference

PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 20/05/2014
 Checked by: Date:
 Approved by: Pollen Yeung Date: 27/05/2014

Plasma Concentrations of Adenosine in Rat 376
Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. I.S. (mm)	Peak Ht. S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Adenosine 5 ng									
a2.5	2.5ug/ml (a)	15.32	29.55	0.52	0.52	10	143.94	57.58	
b2.5	2.5ug/ml (b)	18.15	40.19	0.45	0.45	10	170.53	68.21	
Mean		16.74	34.87	0.49	0.49		157.23	62.89	
SD		2.00	7.52	0.05	0.05		18.80	7.52	
%CV		11.96	21.58	9.74	9.74		11.96	11.96	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a0.5									
b0.5	0.5ug/ml (a)	6.71	98.80	0.07	0.07	30.00	21.01	42.03	
	0.5ug/ml (b)	7.71	98.81	0.08	0.08	30.00	24.15	48.29	
	0.5 ug/ml	8.08	98.51	0.08	0.08	30.00	25.31	50.61	
	0.5 ug/ml	6.26	100.55	0.06	0.06	30.00	19.61	39.21	
	0.5 ug/ml	8.06	95.49	0.08	0.08	30.00	25.24	50.49	
	0.5 ug/ml	9.96	97.50	0.10	0.10	30.00	31.19	62.39	
	0.5 ug/ml	8.67	99.53	0.09	0.09	30.00	27.15	54.31	
Mean		7.92	98.46	0.08	0.08		24.81	49.62	
SD		1.23	1.61	0.01	0.01		3.84	7.69	
%CV		15.49	1.63	16.24	16.24		15.49	15.49	
N		7.00	7.00	7.00	7.00		7.00	7.00	
BLANKS:									
aB	0ug/mL (a)	N/D	10.66	0.00	0.00	2	0.00		
bB	0 ug/mL (b)	N/D	8.64	0.00	0.00	2	0.00		
Mean		0.00	9.65	0.00	0.00		0.00		
SD		0.00	1.43	0.00	0.00		0.00		
%CV		ERR	14.80	ERR	ERR		ERR		
N		2.00	2.00	2.00	2.00		2.00		
Adenosine 5 ng 09/03/2015									
Adenosine 5 ng 10/03/2015									
Adenosine 5 ng 11/03/2015									
Adenosine 5 ng 12/03/2015									
Adenosine 5 ng 13/03/2015									

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHRV Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.49	0.00	0.49
0.50	0.08	0.00	0.08
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0078
Std Err of Y Est	0.0127
R Squared	0.9988
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.1964
Std Err of Coef.	0.0068

Sample ID	Time post-dos	Peak #	Peak Ht. I.S. (mm)	Peak Ht. S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)onc.(µM)	Conc.(µM) Corrected for dilution
T0 R376	0.00	3.22	88.98	0.04	0.04	35	-	0.22	0.84	1.12
T0.08 R376	0.08	4.13	123.70	0.03	0.03	35	-	0.21	0.79	1.05
T0.25 R376	0.25	4.56	112.97	0.04	0.04	35	-	0.25	0.92	1.22
T1 R376	1.00	3.42	91.78	0.04	0.04	35	-	0.23	0.86	1.15
Sagittaristol (30 mg/kg)										
T1.2 R376	1.20	7.34	134.96	0.05	0.05	35	-	0.32	1.19	1.58
T1.5 R376	1.50	5.69	112.72	0.05	0.05	35	-	0.30	1.11	1.48
T2 R376	2.00	7.83	120.44	0.07	0.07	35	-	0.37	1.39	1.85
T3 R376	3.00	5.43	93.23	0.06	0.06	35	-	0.34	1.26	1.68
T4 R376	4.00									
T5 R376	5.00									
T6 R376	6.00									
Mean		5.20	109.85	0.05	0.05			0.28	1.04	1.39
SD		1.71	16.86	0.01	0.01			0.06	0.22	0.30
%CV		32.84	15.35	24.94	24.94			21.37	21.37	21.37
n		8.00	8.00	8.00	8.00			8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No 181

The Spiking solutions were made on: 22/02/2006

*Repeat injections of QC a or b

Rat died

Submitted by: Shyam Sundar

Date: 02/04/2015

Checked by:

Date:

Approved by: Pollen Yeung

Date: 14/09/2015

Plasma Concentrations of Inosine in Rat 376
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Value	Ratio	Inj Vol. (uL)	Amount (ng)	Recovery (%)
Inosine 5 ng		43.11						5		
a2.5	2.5ug/ml (a)	27.85	29.55	0.94	0.94	10	193.81	77.52		
b2.5	2.5ug/ml (b)	33.72	40.19	0.84	0.84	10	234.86	93.86		
Mean		30.79	34.87	0.89	0.89		214.23	85.69		
SD		4.15	7.52	0.07	0.07		28.88	11.55		
%CV		13.48	21.58	8.21	8.21		13.48	13.48		
N		2.00	2.00	2.00	2.00		2.00	2.00		
a0.5	0.5ug/ml (a)	8.18	98.80	0.08	0.08	30.00	18.97	37.95		
b0.5	0.5ug/ml (b)	8.79	98.81	0.09	0.09	30.00	20.39	40.78		
	0.5ug/ml	11.33	98.51	0.12	0.12	30.00	26.28	52.56		
	0.5ug/ml	13.09	100.55	0.13	0.13	30.00	30.36	60.73		
	0.5ug/ml	9.30	95.49	0.10	0.10	30.00	21.57	43.15		
	0.5ug/ml	10.64	97.50	0.11	0.11	30.00	24.68	49.36		
	0.5ug/ml	5.84	99.53	0.06		30.00	13.55	27.09		
Mean		9.60	98.46	0.10	0.10		22.26	44.52		
SD		2.35	1.61	0.02	0.02		5.45	10.91		
%CV		24.50	1.63	24.09	16.96		24.50	24.50		
N		7.00	7.00	7.00	6.00		7.00	7.00		
BLANKS:										
aB	0ug/mL (a)	ND	10.66	0.00	0.00	2	0.00			
bB	0 ug/mL (b)	ND	8.64	0.00	0.00	2	0.00			
Mean		0.00	9.65	0.00	0.00		0.00			
SD		0.00	1.43	0.00	0.00		0.00			
%CV		ERR	14.80	ERR	ERR		ERR			
N		2.00	2.00	2.00	2.00		2.00			
Inosine 5ng 09/03/2015		43.47	11.29				5.00			
Inosine 5ng 10/03/2015		42.92	9.66				5.00			
Inosine 5ng 11/03/2015		44.40	11.44				5.00			
Inosine 5ng 12/03/2015		42.74	11.47				5.00			
Inosine 5ng 13/03/2015		42.92	9.80				5.00			

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.89	0.00	0.89
0.50	0.10	0.00	0.10
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0354
Std Err of Y Est	0.0573
R Squared	0.9931
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3669
Std Err of Coef.	0.0306

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (uL)	Hemolysis Degree	c.(ug/mL)onc.(uM)	Conc.(uM) Corrected for dilution
T0 R376	0.00	7.44	88.98	0.08	0.08	35	-	-	0.32	1.21 1.61
T0.08 R376	0.08	8.22	123.70	0.07	0.07	35	-	-	0.28	1.03 1.38
T0.25 R376	0.25	8.76	112.97	0.08	0.08	35	-	-	0.31	1.15 1.53
T1 R376	1.00	8.06	91.78	0.09	0.09	35	-	-	0.34	1.25 1.67
Inosine standard (30 mg/kg)										
T1.2 R376	1.20	5.77	134.96	0.04	0.04	35	-	-	0.21	0.79 1.06
T1.5 R376	1.50	7.25	112.72	0.06	0.06	35	-	-	0.27	1.01 1.35
T2 R376	2.00	9.74	120.44	0.08	0.08	35	-	-	0.32	1.18 1.57
T3 R376	3.00	8.85	93.23	0.09	0.09	35	-	-	0.36	1.32 1.77
T4 R376	4.00									
T5 R376	5.00									
T6 R376	6.00									
Mean		8.01	109.85	0.07	0.07				0.30	1.12 1.49
SD		1.21	16.86	0.02	0.02				0.04	0.17 0.22
%CV		15.07	15.35	22.03	22.03				14.96	14.96 14.96
n		8.00	8.00	8.00	8.00				8.00	8.00 8.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
 The Spiking solutions were made on: 22/02/2006
 *Repeat injections of QC a or b

Not died

Submitted by: Shyam Sundar Date: 02/04/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 14/04/2015

Plasma Concentrations of Hypoxanthine in Rat 376
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (uL)	Amount (ng)	Rec: Recovery (%)
Hypoxanthine 5 ng		110.53					5		
a25	25ug/ml (a)	117.85	7.52	15.67	15.67	13.58	2	1599.34	63.97
b25	25ug/ml (b)	129.92	9.57	13.58	13.58	13.58	2	1763.14	70.53
Mean		123.89	8.55	14.62	14.62	14.62		1681.24	67.25
SD		8.53	1.45	1.48	1.48	1.48		115.83	4.63
%CV		6.89	16.96	10.13	10.13	10.13		6.89	6.89
N		2.00	2.00	2.00	2.00	2.00		2.00	2.00
a5	5ug/ml (a)	15.21	9.97	1.53	1.53	1.53	2	206.41	41.28
b5	5ug/ml (b)	16.26	10.09	1.61	1.61	1.61	2	220.66	44.13
	5ug/ml	17.19	10.99	1.56	1.56	1.56	2	233.29	46.66
	5ug/ml	15.52	10.81	1.44	1.44	1.44	2	210.62	42.12
	5ug/ml	15.73	10.57	1.49	1.49	1.49	2	213.47	42.69
	5ug/ml	14.64	10.18	1.44	1.44	1.44	2	196.66	39.74
	5ug/ml	15.55	10.47	1.49	1.49	1.49	2	211.03	42.21
Mean		15.73	10.44	1.51	1.51	1.51		213.45	42.69
SD		0.81	0.38	0.06	0.06	0.06		11.01	2.20
%CV		5.16	3.64	4.30	4.30	4.30		5.16	5.16
N		7.00	7.00	7.00	7.00	7.00		7.00	7.00
BLANKS:									
aB	0ug/ml (a)	ND	10.66	0.00	0.00	0.00	2	0.00	
bB	0ug/ml (b)	ND	8.64	0.00	0.00	0.00	2	0.00	
Mean		0.00	9.65	0.00	0.00	0.00		0.00	
SD		0.00	1.43	0.00	0.00	0.00		0.00	
%CV		ERR	14.80	ERR	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00	2.00		2.00	
Hypoxanthine 5 ng 09/03/15		106.55	11.29				5.00		
Hypoxanthine 5 ng 10/03/15		108.38	9.66				5.00		
Hypoxanthine 5 ng 11/03/15		112.03	11.44				5.00		
Hypoxanthine 5 ng 12/03/15		108.94	11.47				5.00		
Hypoxanthine 5 ng 13/03/15		104.17	9.80				5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	14.62	0.00	14.62
5.00	1.51	0.00	1.51
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.6751
Std Err of Y Est	1.0939
R Squared	0.9908
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.6052
Std Err of Coef.	0.0585

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (uL)	Hemolysis Degree	c.(ug/mL)onc.(uM)	Conc.(uM) Corrected for dilution
T0 R376	0.00	11.09	16.79	0.66	0.66	5	-	2.21	16.22
T0.08 R376	0.08	13.17	25.20	0.52	0.52	5	-	1.98	14.54
T0.25 R376	0.25	13.76	23.32	0.59	0.59	5	-	2.09	15.36
T1 R376	1.00	12.71	19.60	0.65	0.65	5	-	2.19	16.07
Sawadiphenol (30 mg/kg)									
T1.2 R376	1.20	14.89	24.34	0.61	0.61	5	-	2.13	15.62
T1.5 R376	1.50	13.74	21.53	0.64	0.64	5	-	2.17	15.94
T2 R376	2.00	12.79	20.10	0.64	0.64	5	-	2.17	15.92
T3 R376	3.00	15.57	25.58	0.61	0.61	5	-	2.12	15.59
T4 R376	4.00								
T5 R376	5.00								
T6 R376	6.00								
Mean		13.47	22.06	0.61	0.61			2.13	15.66
SD		1.38	3.09	0.04	0.04			0.07	0.53
%CV		10.27	14.02	7.12	7.12			3.39	3.39
n		8.00	8.00	8.00	8.00			8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181

The Spiking solutions were made on: 22/02/2006

*Repeated injections of QC a or b

Rat died

Submitted by: Shyam Sundar Date: 02/04/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 15/04/2015

Plasma Concentrations of Xanthine in Rat 376
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (uL)	Amount (ng)	Rec Recovery (%)
Xanthine 5 ng		39.10					5		
a25	25ug/ml (a)	48.18	7.52	6.41	6.41	2	1848.34	73.93	
b25	25ug/ml (b)	58.78	9.57	6.14	6.14	2	2254.99	90.20	
Mean		53.48	8.55	6.27	6.27		2051.66	82.07	
SD		7.50	1.45	0.19	0.19		287.54	11.50	
%CV		14.02	16.96	2.98	2.98		14.02	14.02	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a5	5ug/ml (a)	3.36	9.97	0.34	0.34	2	128.90	25.78	
b5	5ug/ml (b)	3.20	10.09	0.32	0.32	2	122.76	24.55	
	5ug/ml	4.88	10.99	0.44	0.44	2	187.21	37.44	
	5ug/ml	4.36	10.81	0.40	0.40	2	167.26	33.45	
	5ug/ml	5.82	10.57	0.55	0.55	2	223.27	44.65	
	5ug/ml	5.20	10.18	0.51	0.51	2	199.49	39.90	
	5ug/ml	3.56	10.47	0.34	0.34	2	136.57	27.31	
Mean		4.34	10.44	0.41	0.37		166.50	33.30	
SD		1.01	0.38	0.09	0.05		38.65	7.73	
%CV		23.21	3.64	21.97	14.47		23.21	23.21	
N		7.00	7.00	7.00	5.00		7.00	7.00	
BLANKS:									
aB	0ug/ml (a)	ND	10.66	0.00	0.00	2	0.00	0.00	
bB	0ug/ml (b)	ND	8.64	0.00	0.00	2	0.00	0.00	
Mean		0.00	9.65	0.00	0.00		0.00	0.00	
SD		0.00	1.43	0.00	0.00		0.00	0.00	
%CV		ERR	14.80	ERR	ERR		ERR	ERR	
N		2.00	2.00	2.00	2.00		2.00	2.00	

Xanthine 5 ng 09/03/15	37.00	11.29	5.00
Xanthine 5 ng 10/03/15	38.29	9.66	5.00
Xanthine 5 ng 11/03/15	39.49	11.44	5.00
Xanthine 5 ng 12/03/15	38.74	11.47	5.00
Xanthine 5 ng 13/03/15	37.26	9.80	5.00

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	6.27	0.00	6.27
5.00	0.37	0.00	0.37
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.6147
R Squared	0.9695
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.2442
Std Err of Coef.	0.0241

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (uL)	Hemolysis Degree	c.(ug/mL)	onc.(uM)	Conc.(uM) Corrected for dilution
T0 R376	0.00	INT	16.79	0.00	0.00	5	-	0.00	0.00	0.00	0.00
T0.08 R376	0.08	INT	25.20	0.00	0.00	5	-	0.00	0.00	0.00	0.00
T0.25 R376	0.25	INT	23.32	0.00	0.00	5	-	0.00	0.00	0.00	0.00
T1 R376	1.00	INT	19.60	0.00	0.00	5	-	0.00	0.00	0.00	0.00
Spontaneous (30 mg/kg)											
T1.2 R376	1.20	INT	24.34	0.00	0.00	5	-	0.00	0.00	0.00	0.00
T1.5 R376	1.50	INT	21.53	0.00	0.00	5	-	0.00	0.00	0.00	0.00
T2 R376	2.00	INT	23.41	0.00	0.00	5	-	0.00	0.00	0.00	0.00
T3 R376	3.00	INT	25.58	0.00	0.00	5	-	0.00	0.00	0.00	0.00
T4 R376	4.00										
T5 R376	5.00										
T6 R376	6.00										
Mean		0.00	22.47	0.00	0.00			0.00	0.00	0.00	0.00
SD		0.00	3.01	0.00	0.00			0.00	0.00	0.00	0.00
%CV		ERR	13.41	ERR	ERR			ERR	ERR	ERR	ERR
n		8.00	8.00	8.00	8.00			8.00	8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Rat died

Submitted by: Shyam Sundar Date: 02/04/2015
 Checked by: Date:
 Approved by: Pollen Yeung Date: 15/04/2015

Plasma Concentrations of Guanosine in Rat 376
 Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Guanosine 5 ng									
a2.5	2.5ug/ml (a)	25.46	29.55	0.86	0.86	10	132.12	52.85	
b2.5	2.5ug/ml (b)	29.79	40.19	0.74	0.74	10	154.59	61.84	
Mean		27.63	34.87	0.80	0.80		143.36	57.34	
SD		3.06	7.52	0.09	0.09		15.89	6.36	
%CV		11.08	21.58	10.62	10.62		11.08	11.08	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a0.5	0.5ug/ml (a)	3.26	98.80	0.03	0.03	30.00	5.64	11.28	
b0.5	0.5ug/ml (b)	3.71	98.81	0.04	0.04	30.00	6.42	12.84	
	0.5ug/ml	2.54	98.51	0.03	0.03	30.00	4.39	8.79	
	0.5ug/ml	3.20	100.55	0.03	0.03	30.00	5.54	11.07	
	0.5ug/ml	3.23	95.49	0.03	0.03	30.00	5.59	11.17	
	0.5ug/ml	1.99	97.50	0.02	0.02	30.00	3.44	6.88	
	0.5ug/ml	4.49	99.53	0.05	0.05	30.00	7.77	15.53	
Mean		3.20	98.46	0.03	0.03		5.54	11.08	
SD		0.80	1.61	0.01	0.00		1.38	2.76	
%CV		24.94	1.63	24.43	13.18		24.94	24.94	
N		7.00	7.00	7.00	5.00		7.00	7.00	
BLANKS:									
aB	0ug/mL (a)	ND	10.66	0.00	0.00	2	0.00		
bB	0 ug/mL (b)	ND	8.64	0.00	0.00	2	0.00		
Mean		0.00	9.65	0.00	0.00		0.00		
SD		0.00	1.43	0.00	0.00		0.00		
%CV		ERR	14.80	ERR	ERR		ERR		
N		2.00	2.00	2.00	2.00		2.00		
'Guanosine 5 ng 09/03/15		55.16	11.29			5.00			
'Guanosine 5 ng 10/03/15		54.94	9.66			5.00			
'Guanosine 5 ng 11/03/15		58.06	11.44			5.00			
'Guanosine 5 ng 12/03/15		55.50	11.47			5.00			
'Guanosine 5 ng 13/03/15		53.33	9.80			5.00			

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHRV Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.80	0.00	0.80
0.50	0.03	0.00	0.03
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0609
Std Err of Y Est	0.0987
R Squared	0.9763
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3388
Std Err of Coef.	0.0527

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(ug/mL)	Conc.(µM)	Conc.(µM) Corrected for dilution
T0 R376	0.00	1.99	88.98	0.022	0.022	35	-	-	0.25	0.87	1.16
T0.08 R376	0.08	2.52	123.70	0.020	0.020	35	-	-	0.24	0.85	1.13
T0.25 R376	0.25	4.03	112.97	0.036	0.036	35	-	-	0.29	1.01	1.34
T1 R376	1.00	1.70	91.78	0.019	0.019	35	-	-	0.23	0.83	1.10
Inj. Standard (30 mg/kg)											
T1.2 R376	1.20	ND	134.96	0.000	0.000	35	-	-	0.18	0.67	0.90
T1.5 R376	1.50	1.93	112.72	0.017	0.017	35	-	-	0.23	0.86	1.15
T2 R376	2.00	1.88	120.44	0.016	0.016	35	-	-	0.23	0.80	1.06
T3 R376	3.00	3.23	93.23	0.035	0.035	35	-	-	0.28	1.00	1.33
T4 R376	4.00										
T5 R376	5.00										
T6 R376	6.00										
Mean		2.16	109.85	0.02	0.02				0.24	0.86	1.15
SD		1.18	16.86	0.01	0.01				0.03	0.11	0.14
%CV		54.80	15.35	55.00	55.00				13.87	12.45	12.45
n		19.00	16.00	8.00	8.00				8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No 181

The Spiking solutions were made on: 22/02/2006

*Repeat injections of QC a or b

Rat died

Submitted by: Shyam Sundar

Date: 02/04/2015

Checked by:

Date:

Approved by: Pollen Yeung

Date: 14/04/2015

Plasma Concentrations of Uric Acid in Rat 376
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Rec: Recovery (%)
Uric Acid 5 ng		32.60					5		
a25	25ug/ml (a)	54.58	7.52	7.26	7.26	2	2511.35	92.37	
b25	25ug/ml (b)	61.14	9.57	6.39	6.39	2	2813.19	104.44	
Mean		57.86	8.55	6.82	6.82		2662.27	98.40	
SD		4.64	1.45	0.61	0.61		213.43	8.54	
%CV		8.02	16.96	9.01	9.01		8.02	8.68	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a5	5ug/ml (a)	9.40	9.97	0.94	0.94	2	432.52	46.06	
b5	5ug/ml (b)	11.72	10.09	1.16	1.16	2	539.26	67.41	
	5ug/ml	9.53	10.99	0.87	0.87	2	438.50	47.25	
	5ug/ml	8.98	10.81	0.83	0.83	2	413.19	42.19	
	5ug/ml	8.95	10.57	0.85	0.85	2	411.81	45.46	
	5ug/ml	10.80	10.18	1.06	1.06	2	496.93	62.46	
	5ug/ml	8.83	10.47	0.84	0.84	2	405.29	44.36	
Mean		9.74	10.44	0.94	0.94		448.36	50.74	
SD		1.10	0.38	0.13	0.13		50.52	9.93	
%CV		11.27	3.64	13.71	13.71		11.27	19.57	
N		7.00	7.00	7.00	7.00		7.00	7.00	
BLANKS:									
aB	0ug/ml (a)	4.78	10.66	0.45	0.45	2	219.94		
bB	0ug/ml (b)	4.01	8.64	0.46	0.46	2	184.51		
Mean		4.40	9.65	0.46	0.46		202.22		
SD		0.54	1.43	0.01	0.01		25.05		
%CV		12.39	14.80	2.44	2.44		12.39		
N		2.00	2.00	2.00	2.00		2.00		
Uric Acid 5 ng 09/03/2015		30.80	11.29				5.00		
Uric Acid 5 ng 10/03/2015		29.98	9.66				5.00		
Uric Acid 5 ng 11/03/2015		32.46	11.44				5.00		
Uric Acid 5 ng 12/03/2015		32.79	11.47				5.00		
Uric Acid 5 ng 13/03/2015		30.21	9.80				5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	6.82	0.46	6.37
5.00	0.94	0.46	0.48
0.00	0.46	0.46	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.3779
Std Err of Y Est	0.6122
R Squared	0.9851
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.2660
Std Err of Coef.	0.0327

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)onc.(µM)	Conc.(µM) Corrected for dilution
T0 R376	0.00	19.56	16.79	1.16	1.16	5	-	5.80	34.50
T0.08 R376	0.08	8.75	25.20	0.35	0.35	5	-	2.73	16.21
T0.25 R376	0.25	13.84	23.32	0.59	0.59	5	-	3.65	21.72
T1 R376	1.00	15.62	19.60	0.80	0.80	5	-	4.42	26.27
Supplemental (30 mg/kg)									
T1.2 R376	1.20	37.39	24.34	1.54	1.54	5	-	7.20	52.87
T1.5 R376	1.50	89.67	21.53	4.16	4.16	5	-	17.08	125.47
T2 R376	2.00	112.96	20.10	5.62	5.62	5	-	22.55	134.12
T3 R376	3.00	53.66	25.58	2.10	2.10	5	-	9.31	55.36
T4 R376	4.00								
T5 R376	5.00								
T6 R376	6.00								
Mean		43.93	22.06	2.04	2.04			9.09	58.32
SD		38.79	3.09	1.88	1.88			7.08	46.29
%CV		88.30	14.02	92.37	92.37			77.93	79.38
n		8.00	8.00	8.00	8.00			8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No. 181

The Spiking solutions were made on: 22/02/2006

*Repeated injections of QC a or b

Rat died

Submitted by: Shyam Sundar Date: 02/04/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 15/04/2015

Title: Measurement of Plasma Concentrations of Dipyridamole in Rat 376
 According to SOP No: SOP/STD/2008-001-1 (Plasma with no Stopping Solution) Experiment Date: 08/10/2014 - 10/10/2014

Abs.amt ng	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	(μ L)	Inj Vol.	Amount (ng)	Recovery (%)
Dipyridamole (1ng)		75.64					1		
a1000	1 ug/mL(a)	102.96	6.12	16.82	16.82		5	54.45	108.89
b1000	1ug/mL(b)	109.56	5.43	20.18			5	57.94	115.88
1000*	1ug/mL(c)	97.85	6.63	14.76	14.76		5	51.75	103.49
Mean		103.46	6.06	17.25	15.79			54.71	109.42
SD		5.87	0.60	2.73	1.46			3.10	6.21
%CV		5.67	9.94	15.85	9.25			5.67	5.67
n		3.00	3.00	3.00	2.00			1.00	1.00
a100	0.1 ug/mL (a)	41.22	23.93	1.72	1.72		20	5.45	108.99
b100	0.1ug/mL (b)	37.69	17.14	2.20	2.20		20	4.98	99.66
Mean		39.46	20.54	1.96	1.96			5.22	104.32
SD		2.50	4.80	0.34	0.34			0.33	6.60
%CV		6.33	23.38	17.18	17.18			6.33	6.33
n		2.00	2.00	2.00	2.00			2.00	2.00
aB	0 ug/mL (a)	3.37	19.74	0.17			20	0.45	8.91
bB	0 ug/mL (b)	1.53	18.70	0.08	0.08		20	0.20	4.05
Mean		2.45	19.22	0.13	0.08			0.32	6.48
SD		1.30	0.74	0.06	ERR			0.17	3.44
%CV		53.11	3.83	49.78	ERR			53.11	53.11
n		2.00	2.00	2.00	1.00			2.00	2.00

Plasma Conc. (ug/mL)	Peak Ht. Ratio (PHR)	Blank (PHRb)	PHRV-PHRb
0.00	0.08	0.08	0.00
0.10	1.96	0.08	1.88
1.00	15.79	0.08	15.71

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.2167
R Squared	0.9994
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	15.7398
Std Err of Coef.	0.2156

Sample ID	Time Post-dose (h)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)*	Peak Ht. Ratio	PHR Value	Inj Vol. (μ L)	Hemolysis Degree	Conc.(ug/mL)
R376T0	0.00	1.22	25.93	0.05	0.05	20	-	0.02
R376T0.08	0.08	8.11	31.85	0.25	0.25	20	-	0.12
R376T0.25	0.25	17.34	13.63	1.27	1.27	20	-	0.59
R376T1	1.00	35.15	11.78	2.98	2.98	20	-	1.39
Internal Standard (30 mg/kg sc)								
R376T1.2	1.20	16.65	19.17	0.87	0.87	20	-	0.86
R376T1.5	1.50	18.79	19.10	0.98	0.98	20	-	0.46
R376T2	2.00	18.40	18.19	1.01	1.01	20	-	0.47
R376T3	3.00	15.73	18.47	0.85	0.85	20	-	0.40
R376T4	4.00							
R376T5	5.00						-	
R376T6	6.00						-	
Mean		16.42	19.77	1.03	1.03			0.54
SD		9.72	6.44	0.89	0.89			0.43
%CV		59.20	32.59	85.74	85.74			80.17
n		8.00	8.00	8.00	8.00			8.00

Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 Corr. PHR = (PHR - RGB PHR)

Dipyridamole (1ng)(03/10/2014) 90.59 9.29 1.00

Comments: Plasma from Rat 156 was used for extraction QC's.

*A repeat injection of a or b

Rat died

Submitted by: Shyam Sundar

Date: 14/01/2014

Checked by: Pollen Yeung

Date: 07/11/2014

Approved by:

Date:

APPENDIX 7: Rat 377

Title: Measurement of RBC Concentrations of ATP in Rat 377 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
Experiment Date 08-09/05/2014

Sample/standard ID	Standard Concentra (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol (µL)	Amount Recovered (%)	Recovery
ATP 4 ng										
a250	250 ug/mL	47.21	6.46	7.31	35.00	7.31	7.31	0.35	24936.85	83.38
b250	250 ug/mL	34.91	5.74	6.08	35.00	6.08	6.08	0.35	18439.86	57.40
Mean		41.06	6.10	6.69	35.00	6.69	6.69		21688.36	70.39
SD		8.70	0.51	0.87	0.00	0.87	0.87		4594.07	18.38
%CV		21.18	8.35	12.85	0.00	12.85	12.85		21.18	26.11
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a100	100 ug/mL	17.80	6.11	2.91	35.00	2.91	2.91	0.35	9402.16	53.11
b100	100 ug/mL	25.12	7.50	3.35	35.00	3.35	3.35	0.35	13288.67	91.78
100'	100 ug/mL	30.10	14.55	2.07	35.00	2.07		0.35	15899.16	118.08
Mean		24.34	9.39	2.78	35.00	2.78	3.13		11335.41	72.44
SD		6.19	4.53	0.65	0.00	0.65	0.31		2734.03	27.34
%CV		25.42	48.21	23.44	0.00	23.44	9.85		24.12	37.74
n		3.00	3.00	3.00	3.00	3.00	2.00		2.00	2.00
a8	0 ug/mL (a)	9.72	11.47	0.85	35.00	0.85	0.85	0.35	5194.21	
b8	0 ug/mL (a)	5.77	8.00	0.72	35.00	0.72	0.72	0.35	3047.78	
Mean		7.75	9.74	0.78	35.00	0.78	0.78		4091.00	
SD		2.79	2.45	0.09	0.00	0.09	0.09		1475.33	
%CV		36.06	25.20	11.38	0.00	11.38	11.38		36.06	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRB
250.00	6.69	0.78	5.91
100.00	3.13	0.78	2.35
0.00	0.78	0.78	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0068
Std Err of Y Est	0.0140
R Squared	1.0000
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0237
Std Err of Coef.	0.0001

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R37770	0.00	28.56	58.21	0.51	35.00	0.51	0.51	1.50	-	21.77	0.0429
R37770.08	0.08	46.80	79.54	0.59	35.00	0.59	0.59	1.50	-	25.17	0.0496
R37770.25	0.25	58.48	63.12	0.93	35.00	0.93	0.93	1.00	-	39.46	0.0778
R37771	1.00	50.48	56.54	0.89	35.00	0.89	0.89	1.00	-	38.04	0.0750
Regression of (10 mg/kg set)											
R37771.2	1.20	50.46	46.16	1.09	35.00	1.09	1.09	1.00	-	46.51	0.0917
R37771.5	1.50	29.78	43.92	0.68	35.00	0.68	0.68	1.00	-	28.96	0.0571
R37772	2.00										
R37773	3.00										
R37774	4.00										
R37375	5.00	113.45	63.79	1.78	35.00	1.78	1.78	1.00	-	75.48	0.1488
R37376	6.00	42.92	56.64	0.76	35.00	0.76	0.76	1.00	-	32.33	0.0637
Mean		52.62	58.24	0.90	35.00	0.90	0.90	1.13		38.46	0.08
SD		26.65	11.14	0.40	0.00	0.40	0.40	0.23		16.99	0.03
%CV		50.65	19.13	44.50	0.00	44.50	44.50	20.57		44.16	44.16
n		8.00	8.00	8.00	8.00	8.00	8.00	8.00		8.00	8.00

ATP (4 ng) 09/05/2014 14.47

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.
*Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
Peak Ht. = peak height
Peak Ht. R. (or: PHRV) = peak height ratio
I.S. = internal standard
Inj Vol = injection volume
ND = not detected or determined
NS = no sample
INT = interference
PCV = packed cell volume (haematocrit)
CorPHR = corrected peak height ratio
Hemolysis Degree:
-: no visible hemolysis
+: slight hemolysis
++: intermediate hemolysis
+++: serious hemolysis

Submitted by: Shyam Sundar K Date: 20/04/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 28/05/2014

Title: Measurement of RBC Concentrations of ADP in Rat 377 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)

Experiment Date: 08-09/05/2014

Sample/standard ID	Standard Co (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered	Recovery %
ADP 4 ng		26.98						4.00		
s250	250 µg/mL	69.92	6.48	10.82	35.00	10.82	10.82	0.35	16289.74	62.85
b250	250 µg/mL	62.30	5.74	9.11	35.00	9.11	9.11	0.35	12184.69	46.53
Mean		61.11	6.10	9.97	35.00	9.97	9.97		14237.21	54.74
SD		12.46	0.51	1.21	0.00	1.21	1.21		2902.71	11.61
%CV		20.39	8.35	12.15	0.00	12.15	12.15		20.39	21.21
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
100a	100 µg/mL	22.71	6.11	3.72	35.00	3.72	3.72	0.35	5290.90	47.38
100b	100 µg/mL	33.79	7.50	4.51	35.00	4.51	4.51	0.35	7872.29	73.19
100*	100 µg/mL	43.81	14.55	3.01	35.00	3.01		0.35	10206.71	96.53
Mean		33.44	9.39	3.74	35.00	3.74	4.11		7789.97	72.37
SD		10.55	4.53	0.75	0.00	0.75	0.56		2458.94	24.59
%CV		31.57	48.21	19.96	0.00	19.96	13.56		31.57	33.98
n		3.00	3.00	3.00	3.00	3.00	2.00		3.00	3.00
aB	0 µg/mL (a)	3.04	11.47	0.27	35.00	0.27	0.27	0.35	708.25	
bB	0 µg/mL (a)	1.71	8.00	0.21	35.00	0.21	0.21	0.35	398.39	
Mean		2.38	9.74	0.24	35.00	0.24	0.24		553.32	
SD		0.84	2.45	0.04	0.00	0.04	0.04		219.10	
%CV		39.60	25.20	15.15	0.00	15.15	15.15		39.60	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
250.00	9.97	0.24	9.73
100.00	4.11	0.24	3.87
0.00	0.24	0.24	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0077
Std Err of Y Est	0.0159
R Squared	1.0000
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0389
Std Err of Coef.	0.0001

Sample ID	Time post	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC	
R37710	0.00	13.62	56.21	0.24	35.00	0.24	0.24	1.50	-	6.42	0.0150	0.193
R37710B	0.08	17.22	79.54	0.22	35.00	0.22	0.22	1.50	-	5.76	0.0135	0.173
R37710.25	0.25	17.42	63.12	0.28	35.00	0.28	0.28	1.00	-	7.29	0.0171	0.219
R37711	1.00	15.34	56.54	0.27	35.00	0.27	0.27	1.00	-	7.17	0.0168	0.216
Ibuprofenolol (30 mg/kg sc)												
R37711.2	1.20	18.42	46.16	0.40	35.00	0.40	0.40	1.00	-	10.45	0.0245	0.315
R37711.5	1.50	28.55	43.92	0.65	35.00	0.65	0.65	1.00	-	16.90	0.0396	0.509
R37712	2.00											
R37713	3.00											
R37714	4.00											
R37315	5.00	15.57	63.79	0.24	35.00	0.24	0.24	1.00	-	6.47	0.0151	0.195
R37316	6.00	8.84	56.64	0.16	35.00	0.16	0.16	1.00	-	4.21	0.0098	0.127
Mean		16.09	66.29	0.24	35.00	0.24	1.33			6.49	0.02	0.20
SD		2.14	11.98	0.03	0.00	0.03	0.29			0.77	0.00	0.02
%CV		13.29	18.08	12.18	0.00	12.18	21.65			11.81	11.81	11.81
n		3.00	3.00	3.00	3.00	3.00	3.00			3.00	2.00	3.00

ADP (4 ng)09/05/2014

34.74

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.

*Repeated injections of a or b at 0.5-1ul injection volume.

PL = plasma; RBC = red blood cells

Peak Ht. = peak height

Peak Ht. R. (or: PHR) = peak height ratio

I.S. = internal standard

Inj Vol = injection volume

ND = not detected or determined

NS = no sample

INT = interference

PCV = packed cell volume (haematocrit)

CorPHR = corrected peak height ratio

Hemolysis Degree:

--: no visible hemolysis

+ : slight hemolysis

++: intermediate hemolysis

+++: serious hemolysis

Submitted by: Shyam Sundar K

Date: 20/05/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date: 26/05/2014

Title: Measurement of RBC Concentrations of AMP in Rat 377 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
Experiment Date 08-09/05/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
AMP 4 ng										
a50	50 µg/mL	31.51	6.46	4.88	35.00	4.88	4.88	4.00	4020.76	75.90
b50	50 µg/mL	27.22	5.74	4.74	35.00	4.74	4.74	0.35	3473.35	64.85
Mean		29.37	6.10	4.81	35.00	4.81	4.81		3747.06	70.42
SD		3.03	0.51	0.10	0.00	0.10	0.10		387.08	7.74
%CV		10.33	8.35	1.99	0.00	1.99	1.99		10.33	10.99
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20										
a20	20 µg/mL	10.88	6.11	1.78	35.00	1.78	1.78	0.35	1388.33	58.12
b20	20 µg/mL	15.69	7.50	2.09	35.00	2.09	2.09	0.35	2002.09	88.81
20*	20 µg/mL	22.94	14.55	1.58	35.00	1.58	1.58	0.35	2927.21	135.07
Mean		16.50	9.39	1.82	35.00	1.82	1.82		2105.87	94.00
SD		6.07	4.53	0.26	0.00	0.26	0.26		774.68	38.73
%CV		36.79	48.21	14.29	0.00	14.29	14.29		36.79	41.21
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00
a8										
a8	0 µg/mL (a)	1.56	11.47	0.14	35.00	0.14	0.14	0.35	199.06	
b8	0 µg/mL (a)	1.98	6.00	0.25	35.00	0.25	0.25	0.35	252.65	
Mean		1.77	9.74	0.19	35.00	0.19	0.19		225.86	
SD		0.30	2.45	0.08	0.00	0.08	0.08		37.99	
%CV		16.78	25.30	41.11	0.00	41.11	41.11		16.78	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	4.81	0.19	4.62
20.00	1.62	0.19	1.42
0.00	0.19	0.19	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0879
Std Err of Y Est	0.1805
R Squared	0.9970
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0929
Std Err of Coef.	0.0051

Sample ID	Time post dose	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC	
R37770	0.00	5.25	56.21	0.09	35.00	0.09	0.09	1.50	-	1.95	0.0056	0.072
R37770.08	0.08	1.54	79.54	0.02	35.00	0.02	0.02	1.50	-	1.17	0.0034	0.043
R37770.25	0.25	0.75	63.12	0.01	35.00	0.01	0.01	1.00	-	1.07	0.0031	0.040
R37771	1.00	2.16	56.54	0.04	35.00	0.04	0.04	1.00	-	1.36	0.0039	0.050
Soprolefenol (30 mg/kg sc)												
R37771.2	1.20	3.60	46.16	0.08	35.00	0.08	0.08	1.00	-	1.78	0.0051	0.066
R37771.5	1.50	28.98	43.92	0.66	35.00	0.66	0.66	1.00	-	8.04	0.0232	0.298
R37772	2.00											
R37773	3.00											
R37774	4.00											
R37775	5.00	1.02	63.79	0.02	35.00	0.02	0.02	1.00	-	1.12	0.0032	0.044
R37776	6.00	0.68	56.64	0.01	35.00	0.01	0.01	1.00	-	1.07	0.0031	0.040
Mean		5.51	58.24	0.12	35.00	0.12	0.12	1.13	0.00	2.20	0.01	0.08
SD		9.61	11.14	0.22	0.00	0.22	0.22	0.23	0.00	2.39	0.01	0.09
%CV		174.67	19.13	190.85	0.00	190.85	190.85	20.57	0.00	108.69	108.69	108.69
n		8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00

AMP (4 ng)/09/05/2014 70.09

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.
*Repeated injections of "a" or "b" at 0.5 - 1 ul injection volume.

PL = plasma; RBC = red blood cells
Peak Ht. = peak height
Peak Ht. R. (or PHR) = peak height ratio
I.S. = internal standard
Inj Vol = injection volume
ND = not detected or determined
NS = no sample
INT = interference
PCV = packed cell volume (haematocrit)
CorPHR = corrected peak height ratio
Hemolysis Degree:
-: no visible hemolysis
+: slight hemolysis
++: intermediate hemolysis
+++: serious hemolysis

Submitted by: Shyam Sundar Date: 20/05/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 28/05/2014

Title: Measurement of RBC Concentrations of GTP in Rat 377 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 08-09/05/2014

Sample/standard ID	Standard Concentra (ug/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (uL)	Hemolysis Degree	Amount Recove %	Recovery
GTP 4 ng		51.77						4.00			
a50	50 ug/mL	14.07	6.46	2.18	35.00	2.18	2.18	0.35	1708.33	30.78	
b50	50 ug/mL	10.53	5.74	1.83	35.00	1.83	1.83	0.35	1278.51	22.18	
Mean		12.30	6.10	2.01	35.00	2.01	2.01		1493.42	26.48	
SD		2.50	0.24	0.24	0.00	0.24	0.24		303.92	6.08	
%CV		20.35	0.00	12.11	0.00	12.11	12.11		20.35	22.95	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00	
a20	20 ug/mL	4.34	8.11	0.71	35.00	0.71	0.71	0.35	538.95	17.88	
b20	20 ug/mL	7.10	7.50	0.95	35.00	0.95	0.95	0.35	862.05	34.83	
20*	20 ug/mL	8.46	14.55	0.58	35.00	0.58	0.58	0.35	1027.18	42.89	
Mean		6.63	9.39	0.75	35.00	0.75	0.75		805.39	31.80	
SD		2.10	4.53	0.19	0.00	0.19	0.19		254.89	12.74	
%CV		31.65	48.21	24.82	0.00	24.82	24.82		31.65	40.08	
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00	
a8	0 ug/mL (a)	1.92	11.47	0.17	35.00	0.17	0.17	0.35	233.12		
b8	0 ug/mL (a)	0.87	8.00	0.11	35.00	0.11	0.11	0.35	195.63		
Mean		1.40	9.74	0.14	35.00	0.14	0.14		169.38		
SD		0.74	2.45	0.04	0.00	0.03	0.04		90.15		
%CV		53.22	25.20	30.03	0.00	24.52	30.03		53.22		
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	Peak Height Ratio Value (PHRV)	Blank PHRV-PHRb	PHRV-PHRb
50.00	2.01	0.14	1.87
20.00	0.75	0.14	0.61
0.00	0.14	0.14	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0549
Std Err of Y Est	0.1129
R Squared	0.9930
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0377
Std Err of Coef.	0.0032

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (uL)	Hemolysis Degree	Conc.(ug/mL) Lysate	Conc.(mM) RBC	
R37770	0.00	4.42	56.21	0.08	35.00	0.08	0.08	1.50	-	3.54	0.0068	0.087
R37770.08	0.08	8.35	79.54	0.08	35.00	0.08	0.08	1.50	-	3.57	0.0068	0.088
R37770.25	0.25	10.70	63.12	0.17	35.00	0.17	0.17	1.00	-	5.95	0.0114	0.146
R37771	1.00	7.41	56.54	0.13	35.00	0.13	0.13	1.00	-	4.93	0.0094	0.121
Propofol (30 mg/kg sc)												
R37771.2	1.20	8.60	46.16	0.21	35.00	0.21	0.21	1.00	-	6.97	0.0133	0.171
R37771.5	1.50	7.24	43.92	0.16	35.00	0.16	0.16	1.00	-	5.83	0.0111	0.143
R37772	2.00											
R37773	3.00											
R37774	4.00											
R37775	5.00	24.38	63.70	0.38	35.00	0.38	0.38	1.00	-	11.54	0.0220	0.283
R37776	6.00	9.73	56.64	0.17	35.00	0.17	0.17	1.00	-	6.01	0.0115	0.148
Mean		9.96	58.24	0.17	35.00	0.17	0.17	1.13	0.00	6.04	0.01	0.15
SD		6.13	11.14	0.10	0.00	0.10	0.10	0.23	0.00	2.53	0.00	0.08
%CV		61.50	18.13	55.11	0.00	55.11	55.11	20.57	0.00	41.92	41.82	41.82
n		8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00

GTP (4 ng)/09/05/2014 73.30

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014 .
 *Repeated injections of a or b at 0.5 -1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 20/05/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 28/05/2014

Title: Measurement of RBC Concentrations of GDP in Rat 377 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 08-09/05/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount	Recovery %	Recovery
GDP 4 ng											
a50	50 ug/mL	16.69	6.46	2.58	35.00	2.58	2.58	0.35	3039.95	60.80	
b50	50 ug/mL	14.33	5.74	2.50	35.00	2.50	2.50	0.35	2810.09	52.20	
Mean		15.51	6.10	2.54	35.00	2.54	2.54		2925.02	56.50	
SD		1.67	0.51	0.06	0.00	0.06	0.06		303.95	6.08	
%CV		10.76	8.35	2.42	0.00	2.42	2.42		10.76	10.76	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00	
a20											
a20	20 ug/mL	5.63	6.11	0.92	35.00	0.92	0.92	0.35	1025.46	51.27	
b20	20 ug/mL	9.12	7.50	1.22	35.00	1.22	1.22	0.35	1661.13	83.06	
20*	20 ug/mL	11.66	14.55	0.80	35.00	0.80	0.80	0.35	2123.77	106.19	
Mean		8.90	9.39	0.98	35.00	0.98	0.98		1603.46	80.17	
SD		3.03	4.53	0.21	0.00	0.21	0.21		551.42	27.57	
%CV		34.39	48.21	21.78	0.00	21.78	21.78		34.39	34.39	
n		3.00	3.00	3.00	3.00	3.00	3.00		3.00	3.00	
aB											
aB	0 ug/mL (a)	ND	11.47	0.00	35.00	0.00	0.00	0.35	0.00	0.00	
bB	0 ug/mL (a)	ND	6.00	0.00	35.00	0.00	0.00	0.35	0.00	0.00	
Mean		0.00	9.74	0.00	35.00	0.00	0.00		0.00	0.00	
SD		0.00	2.45	0.00	0.00	0.00	0.00		0.00	0.00	
%CV		ERR	25.30	ERR	0.00	ERR	ERR		ERR	ERR	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRB
50.00	2.54	0.00	2.54
20.00	0.98	0.00	0.98
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0144
Std Err of Y Est	0.0295
R Squared	0.9997
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0099
Std Err of Coef.	0.0068

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(nM) RBC	Conc.(nM)
R37770	0.00	2.88	56.21	0.05	35.00	0.05	0.05	1.50	-	1.29	0.0029	0.037
R37770.08	0.08	2.78	79.54	0.03	35.00	0.03	0.03	1.50	-	0.97	0.0032	0.028
R37770.25	0.25	3.69	63.12	0.06	35.00	0.06	0.06	1.00	-	1.43	0.0032	0.042
R37771	1.00	2.85	56.54	0.05	35.00	0.05	0.05	1.00	-	1.27	0.0029	0.037
Isoproterenol (30 mg/kg sc)												
R37771.2	1.20	4.19	46.16	0.09	35.00	0.09	0.09	1.00	-	2.07	0.0047	0.060
R37771.5	1.50	5.13	43.62	0.12	35.00	0.12	0.12	1.00	-	2.58	0.0058	0.075
R37772	2.00											
R37773	3.00											
R37774	4.00											
R37375	5.00	5.23	63.79	0.08	35.00	0.08	0.08	1.00	-	1.89	0.0043	0.056
R37376	6.00	2.48	56.64	0.04	35.00	0.04	0.04	1.00	-	1.14	0.0026	0.033
Mean		30.95	58.24	0.07	35.00	0.07	0.07	1.13	0.00	1.58	0.00	0.05
SD		29.21	11.14	0.03	0.00	0.03	0.23	0.00	0.55	0.00	0.00	0.02
%CV		94.38	19.13	42.15	0.00	42.15	42.15	20.57	ERR	34.62	34.62	34.62
n		16.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00

GDP (4ng)09/05/2014 48.22

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date:20/05/2014

Checked by: Date:

Approved by: Pollen Yeung Date:28/05/2014

Title: Measurement of RBC Concentrations of GMP in Rat 377 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 08-09/05/2014

Sample/standard	ID/Standard Concentr (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	Recovery
GMP 4 ng										
a50	50 ug/mL	130.43	6.46	20.19	35.00	20.19	20.19	0.35	13607.40	272.15
b50	50 ug/mL	103.50	5.74	18.03	35.00	18.03	18.03	0.35	10797.87	215.96
Mean		116.97	6.10	19.11	35.00	19.11	19.11		12202.63	244.05
SD		19.04	5.92	1.53	0.00	1.53	1.53		1986.64	39.73
%CV		16.28	6.01	7.99	0.00	7.99	7.99		16.28	16.28
n		2.00	6.01	2.00	2.00	2.00	2.00		2.00	2.00
a20										
a20	20 ug/mL	86.67	6.11	14.23	35.00	14.23	14.23	0.35	9073.34	453.67
b20	20 ug/mL	121.61	7.50	16.21	35.00	16.21	16.21	0.35		
20*	20 ug/mL	124.73	14.55	8.57	35.00	8.57		0.35	13012.73	650.64
Mean		111.10	9.39	13.01	35.00	13.01	15.22		11043.03	552.15
SD		20.96	4.53	3.97	0.00	3.97	1.40		2785.57	139.28
%CV		18.86	48.21	30.49	0.00	30.49	9.20		25.22	25.22
n		3.00	3.00	3.00	3.00	3.00	2.00		2.00	2.00
aB										
aB	0 ug/mL (a)	Off scale	11.47	0.00	35.00	0.00		0.35		0.00
bB	0 ug/mL (a)		81.75	8.00	10.22	35.00	10.22	0.35		0.00
Mean		40.88	8.74	5.11	35.00	5.11	10.22			0.00
SD		57.81	2.45	7.23	0.00	7.23	ERR			0.00
%CV		141.42	25.20	141.42	0.00	141.42	ERR			ERR
n		2.00	2.00	2.00	2.00	2.00	2.00			2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRB
50.00	19.11	10.22	8.89
20.00	15.22	10.22	5.01
0.00	10.22	10.22	0.00

Regression Output Begins Here:

Regression Output:

Constant: 0.0000
 Std Err of Y Est: 1.5081
 R Squared: 0.9538
 No. of Observations: 3.0000
 Degrees of Freedom: 2.0000

X Coefficient(s): 0.2959
 Std Err of Coef.: 0.0280

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lyasate	Conc.(mM) RBC
R37770		0.00 off scale	56.21	0.00	35.00	0.00	0.00	1.50	-	0.00	0.0000
R37770.08		0.08 off scale	79.54	0.00	35.00	0.00	0.00	1.50	-	0.00	0.0000
R37770.25		0.25 off scale	63.12	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000
R37771		1.00 off scale	56.54	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000
Not detected (ND mg/kg sat)											
R37771.2		1.20 off scale	46.16	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000
R37771.5		1.50 off scale	43.92	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000
R37772		2.00									
R37773		3.00									
R37774		4.00									
R37375		5.00 off scale	63.79	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000
R37376		6.00 off scale	56.64	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000
Mean		0.00	58.24	0.00	35.00	0.00	0.00	1.13	0.00	0.00	0.00
SD		0.00	11.14	0.00	0.00	0.00	0.23	0.00	0.00	0.00	0.00
%CV		ERR	19.13	ERR	0.00	ERR	ERR	20.57	ERR	ERR	ERR
n		8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00	8.00

GMP 4 ng/09/05/2014 88.07

Comments: RBC Lyasate from Rat 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of "a" or "b" at 0.5-1ul injection volume

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference

PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date: 20/05/2014
 Checked by: Date:
 Approved by: Pollen Yeung Date: 28/05/2014

Plasma Concentrations of Adenosine in Rat 377
 Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. I.S. (mm)	Peak Ht. L.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Adenosine 5 ng		31.93					5		
a2.5	2.5ug/ml (a)	15.32	29.55	0.52	0.52	10	143.94	57.58	
b2.5	2.5ug/ml (b)	18.15	40.19	0.45	0.45	10	170.53	68.21	
Mean		16.74	34.87	0.49	0.49		157.23	62.89	
SD		2.00	7.52	0.05	0.05		18.80	7.52	
%CV		11.96	21.58	9.74	9.74		11.96	11.96	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a0.5	0.5ug/ml (a)	6.71	98.80	0.07	0.07	30.00	21.01	42.03	
b0.5	0.5ug/ml (b)	7.71	98.81	0.08	0.08	30.00	24.15	48.29	
	0.5 ug/ml	8.08	98.51	0.08	0.08	30.00	25.31	50.61	
	0.5 ug/ml	6.26	100.55	0.06	0.06	30.00	19.61	39.21	
	0.5 ug/ml	8.06	95.49	0.08	0.08	30.00	25.24	50.49	
	0.5 ug/ml	9.96	97.50	0.10	0.10	30.00	31.19	62.39	
	0.5 ug/ml	8.67	99.53	0.09	0.09	30.00	27.15	54.31	
Mean		7.92	98.46	0.08	0.08		24.81	49.62	
SD		1.23	1.61	0.01	0.01		3.84	7.69	
%CV		15.49	1.63	16.24	16.24		15.49	15.49	
N		7.00	7.00	7.00	7.00		7.00	7.00	
BLANKS:									
aB	0ug/mL (a)	N/D	10.66	0.00	0.00	2	0.00		
bB	0 ug/mL (b)	N/D	8.64	0.00	0.00	2	0.00		
Mean		0.00	9.65	0.00	0.00		0.00		
SD		0.00	1.43	0.00	0.00		0.00		
%CV		ERR	14.80	ERR	ERR		ERR		
N		2.00	2.00	2.00	2.00		2.00		
Adenosine 5 ng 09/03/2015		30.02	11.29				5.00		
Adenosine 5 ng 10/03/2015		30.00	9.66				5.00		
Adenosine 5 ng 11/03/2015		31.93	11.44				5.00		
Adenosine 5 ng 12/03/2015		30.96	11.47				5.00		
Adenosine 5 ng 13/03/2015		27.70	9.80				5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.49	0.00	0.49
0.50	0.08	0.00	0.08
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0078
Std Err of Y Est	0.0127
R Squared	0.9988
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.1964
Std Err of Coef.	0.0068

Sample ID	Time post-dos	Peak # (mm)	Peak Ht. I.S. (mm)	Peak Ht. L.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c. (µg/mL)	Conc. (µM)	Conc. (µM) Corrected for dilution
T0 R377	0.00	2.79	95.34	0.03	0.03	35	-	-	0.19	0.71	0.94
T0.08 R377	0.08	3.76	108.46	0.03	0.03	35	-	-	0.22	0.81	1.08
T0.25 R377	0.25	3.04	119.98	0.03	0.03	35	-	-	0.17	0.63	0.84
T1 R377	1.00	6.44	129.53	0.05	0.05	35	-	-	0.29	1.10	1.46
Regenerol (30 mg/kg)											
T1.2 R377	1.20	4.24	126.77	0.03	0.03	35	-	-	0.21	0.79	1.05
T1.5 R377	1.50	10.12	129.68	0.08	0.08	35	-	-	0.44	1.64	2.18
T2 R377	2.00										
T3 R377	3.00										
T4 R377	4.00										
T5 R377	5.00										
T6 R377	6.00										
Mean		5.07	118.29	0.04	0.04				0.25	0.94	1.26
SD		2.80	13.81	0.02	0.02				0.10	0.37	0.50
%CV		55.21	11.68	47.00	47.00				39.57	39.57	39.57
n		6.00	6.00	6.00	6.00				6.00	6.00	6.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
 The Spiking solutions were made on: 22/02/2006
 *Repeat injections of QC a or b

Rat died

Submitted by: Shyam Sundar

Date: 02/04/2015

Checked by:

Date:

Approved by: Pollen Yeung

Date: 14/04/2015

Plasma Concentrations of Inosine in Rat 377
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (uL)	Amount (ng)	Recovery (%)
Inosine 5 ng			43.11				5		
a2.5	2.5ug/ml (a)	27.85	29.55	0.94	0.94	10	193.81	77.52	
b2.5	2.5ug/ml (b)	33.72	40.19	0.84	0.84	10	234.66	93.86	
Mean		30.79	34.87	0.89	0.89		214.23	85.69	
SD		4.15	7.52	0.07	0.07		28.88	11.55	
%CV		13.48	21.58	8.21	8.21		13.48	13.48	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a0.5	0.5ug/ml (a)	8.18	98.80	0.08	0.08	30.00	18.97	37.95	
b0.5	0.5ug/ml (b)	8.79	98.81	0.09	0.09	30.00	20.39	40.78	
	0.5ug/ml	11.33	98.51	0.12	0.12	30.00	26.28	52.56	
	0.5ug/ml	13.09	100.55	0.13	0.13	30.00	30.36	60.73	
	0.5ug/ml	9.30	95.49	0.10	0.10	30.00	21.57	43.15	
	0.5ug/ml	10.64	97.50	0.11	0.11	30.00	24.68	49.36	
	0.5ug/ml	5.84	99.53	0.06		30.00	13.55	27.09	
Mean		9.60	98.46	0.10	0.10		22.26	44.52	
SD		2.35	1.61	0.02	0.02		5.45	10.91	
%CV		24.50	1.63	24.09	16.96		24.50	24.50	
N		7.00	7.00	7.00	6.00		7.00	7.00	
BLANKS:									
aB	0ug/mL (a)	ND	10.66	0.00	0.00	2	0.00		
bB	0 ug/mL (b)	ND	8.64	0.00	0.00	2	0.00		
Mean		0.00	9.65	0.00	0.00		0.00		
SD		0.00	1.43	0.00	0.00		0.00		
%CV		ERR	14.80	ERR	ERR		ERR		
N		2.00	2.00	2.00	2.00		2.00		
Inosine 5ng 09/03/2015		43.47	11.29				5.00		
Inosine 5ng 10/03/2015		42.92	9.66				5.00		
Inosine 5ng 11/03/2015		44.40	11.44				5.00		
Inosine 5ng 12/03/2015		42.74	11.47				5.00		
Inosine 5ng 13/03/2015		42.92	9.80				5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.89	0.00	0.89
0.50	0.10	0.00	0.10
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0354
Std Err of Y Est	0.0573
R Squared	0.9931
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3669
Std Err of Coef.	0.0306

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (uL)	Hemolysis Degree	c.(ug/mL)	onc.(uM)	Conc.(uM) Corrected for dilution
T0 R377	0.00	7.69	95.34	0.08	0.08	35	-	-	0.32	1.21	1.61
T0.08 R377	0.08	8.77	108.46	0.08	0.07	35	-	-	0.28	1.03	1.38
T0.25 R377	0.25	5.25	119.98	0.04	0.08	35	-	-	0.31	1.15	1.53
T1 R377	1.00	5.80	129.53	0.04	0.09	35	-	-	0.34	1.25	1.67
Isoproterenol (30 mg/kg)											
T1.2 R377	1.20	5.60	126.77	0.04	0.04	35	-	-	0.21	0.79	1.06
T1.5 R377	1.50	4.52	129.66	0.03	0.06	35	-	-	0.27	1.01	1.35
T2 R377	2.00										
T3 R377	3.00										
T4 R377	4.00										
T5 R377	5.00										
T6 R377	6.00										
Mean		6.27	118.29	0.05	0.07				0.29	1.07	1.43
SD		1.61	13.81	0.02	0.02				0.04	0.17	0.22
%CV		25.75	11.68	37.19	23.29				15.50	15.50	15.50
n		6.00	6.00	6.00	6.00				6.00	6.00	6.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
 The Spiking solutions were made on: 22/02/2006
 *Repeat injections of QC a or b

Not died

Submitted by: Shyam Sundar Date: 02/04/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 14/04/2015

Plasma Concentrations of Hypoxanthine in Rat 377
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (uL)	Amount (ng)	Rec: Recovery (%)
Hypoxanthine 5 ng		110.53					5		
a25	25ug/ml (a)	117.85	7.52	15.67	15.67	13.58	2	1599.34	63.97
b25	25ug/ml (b)	129.92	9.57	13.58	13.58	13.58	2	1763.14	70.53
Mean		123.89	8.55	14.62	14.62	14.62		1681.24	67.25
SD		8.53	1.45	1.48	1.48	1.48		115.83	4.63
%CV		6.89	16.96	10.13	10.13	10.13		6.89	6.89
N		2.00	2.00	2.00	2.00	2.00		2.00	2.00
a5	5ug/ml (a)	15.21	9.97	1.53	1.53	1.53	2	206.41	41.28
b5	5ug/ml (b)	16.26	10.09	1.61	1.61	1.61	2	220.66	44.13
	5ug/ml	17.19	10.99	1.56	1.56	1.56	2	233.29	46.66
	5ug/ml	15.52	10.81	1.44	1.44	1.44	2	210.62	42.12
	5ug/ml	15.73	10.57	1.49	1.49	1.49	2	213.47	42.69
	5ug/ml	14.64	10.18	1.44	1.44	1.44	2	196.68	39.74
	5ug/ml	15.55	10.47	1.49	1.49	1.49	2	211.03	42.21
Mean		15.73	10.44	1.51	1.51	1.51		213.45	42.69
SD		0.81	0.38	0.06	0.06	0.06		11.01	2.20
%CV		5.16	3.64	4.30	4.30	4.30		5.16	5.16
N		7.00	7.00	7.00	7.00	7.00		7.00	7.00
BLANKS:									
aB	0ug/ml (a)	ND	10.66	0.00	0.00	0.00	2	0.00	
bB	0ug/ml (b)	ND	8.64	0.00	0.00	0.00	2	0.00	
Mean		0.00	9.65	0.00	0.00	0.00		0.00	
SD		0.00	1.43	0.00	0.00	0.00		0.00	
%CV		ERR	14.80	ERR	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00	2.00		2.00	
Hypoxanthine 5 ng 09/03/15		106.55	11.29				5.00		
Hypoxanthine 5 ng 10/03/15		108.38	9.66				5.00		
Hypoxanthine 5 ng 11/03/15		112.03	11.44				5.00		
Hypoxanthine 5 ng 12/03/15		108.94	11.47				5.00		
Hypoxanthine 5 ng 13/03/15		104.17	9.80				5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	14.62	0.00	14.62
5.00	1.51	0.00	1.51
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.6751
Std Err of Y Est	1.0939
R Squared	0.9908
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.6052
Std Err of Coef.	0.0585

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (uL)	Hemolysis Degree	c.(ug/mL)onc.(uM)	Conc.(uM) Corrected for dilution
T0 R377	0.00	14.52	17.16	0.85	0.85	5	-	2.51	18.47	24.63
T0.08 R377	0.08	15.51	20.70	0.75	0.75	5	-	2.35	17.29	23.06
T0.25 R377	0.25	11.33	21.79	0.52	0.52	5	-	1.97	14.51	19.35
T1 R377	1.00	16.97	23.47	0.72	0.72	5	-	2.31	16.96	22.63
Insulin (30 mg/kg)										
T1.2 R377	1.20	13.31	23.62	0.56	0.56	5	-	2.05	15.04	20.05
T1.5 R377	1.50	13.93	24.58	0.57	0.57	5	-	2.05	15.08	20.10
T2 R377	2.00									
T3 R377	3.00									
T4 R377	4.00									
T5 R377	5.00									
T6 R377	6.00									
Mean		14.26	21.89	0.66	0.66			2.21	16.23	21.64
SD		1.93	2.70	0.13	0.13			0.21	1.58	2.10
%CV		13.51	12.35	19.62	19.62			9.71	9.71	9.71
n		6.00	6.00	6.00	6.00			6.00	6.00	6.00

NOTE: QC samples were prepared with plasma from healthy rat No.181

The Spiking solutions were made on: 22/02/2006

*Repeated injections of QC a or b

Rat died

Submitted by: Shyam Sundar

Date: 02/04/2015

Checked by:

Date:

Approved by: Pollen Yeung

Date: 15/04/2015

Plasma Concentrations of Xanthine in Rat 377
 Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (uL)	Amount (ng)	Recovery (%)
Xanthine 5 ng		39.10					5		
a25	25ug/ml (a)	48.18	7.52	6.41	6.41	2	1848.34	73.93	
b25	25ug/ml (b)	58.78	9.57	6.14	6.14	2	2254.99	90.20	
Mean		53.48	8.55	6.27	6.27		2051.66	82.07	
SD		7.50	1.45	0.19	0.19		287.54	11.50	
%CV		14.02	16.96	2.98	2.98		14.02	14.02	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a5	5ug/ml (a)	3.36	9.97	0.34	0.34	2	128.90	25.78	
b5	5ug/ml (b)	3.20	10.09	0.32	0.32	2	122.76	24.55	
	5ug/ml	4.88	10.99	0.44	0.44	2	187.21	37.44	
	5ug/ml	4.36	10.81	0.40	0.40	2	167.26	33.45	
	5ug/ml	5.82	10.57	0.55	0.55	2	223.27	44.65	
	5ug/ml	5.20	10.18	0.51	0.51	2	199.49	39.90	
	5ug/ml	3.56	10.47	0.34	0.34	2	136.57	27.31	
Mean		4.34	10.44	0.41	0.37		166.50	33.30	
SD		1.01	0.38	0.09	0.05		38.65	7.73	
%CV		23.21	3.64	21.97	14.47		23.21	23.21	
N		7.00	7.00	7.00	5.00		7.00	7.00	
BLANKS:									
aB	0ug/ml (a)	ND	10.66	0.00	0.00	2	0.00		
bB	0ug/ml (b)	ND	8.64	0.00	0.00	2	0.00		
Mean		0.00	9.65	0.00	0.00		0.00		
SD		0.00	1.43	0.00	0.00		0.00		
%CV		ERR	14.80	ERR	ERR		ERR		
N		2.00	2.00	2.00	2.00		2.00		

Xanthine 5 ng 09/03/15	37.00	11.29	5.00
Xanthine 5 ng 10/03/15	38.29	9.66	5.00
Xanthine 5 ng 11/03/15	39.49	11.44	5.00
Xanthine 5 ng 12/03/15	38.74	11.47	5.00
Xanthine 5 ng 13/03/15	37.26	9.80	5.00

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHRV Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	6.27	0.00	6.27
5.00	0.37	0.00	0.37
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.6147
R Squared	0.9695
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.2442
Std Err of Coef.	0.0241

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHRV Value	Inj Vol. (uL)	Hemolysis Degree	c.(ug/mL)	onc.(uM)	Conc.(uM) Corrected for dilution
T0 R377	0.00	INT	17.16	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T0.08 R377	0.08	INT	20.70	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T0.25 R377	0.25	INT	21.79	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T1 R377	1.00	INT	23.47	0.00	0.00	0.00	5	-	0.00	0.00	0.00
Spontaneous (30 mg/kg)											
T1.2 R377	1.20	INT	23.62	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T1.5 R377	1.50	INT	24.56	0.00	0.00	0.00	5	-	0.00	0.00	0.00
T2 R377	2.00										
T3 R377	3.00										
T4 R377	4.00										
T5 R377	5.00										
T6 R377	6.00										
Mean		0.00	21.89	0.00	0.00	0.00			0.00	0.00	0.00
SD		0.00	2.70	0.00	0.00	0.00			0.00	0.00	0.00
%CV		ERR	12.35	ERR	ERR	ERR			ERR	ERR	ERR
n		6.00	6.00	6.00	6.00	6.00			6.00	6.00	6.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Rat died

Submitted by: Shyam Sundar Date: 02/04/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 15/04/2015

Plasma Concentrations of Guanosine in Rat 377
 Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Guanosine 5 ng								
a2.5	2.5ug/ml (a)	25.46	29.55	0.86	0.86	10	132.12	52.85
b2.5	2.5ug/ml (b)	29.79	40.19	0.74	0.74	10	154.59	61.84
Mean		27.63	34.87	0.80	0.80		143.36	57.34
SD		3.06	7.52	0.09	0.09		15.89	6.36
%CV		11.08	21.58	10.62	10.62		11.08	11.08
N		2.00	2.00	2.00	2.00		2.00	2.00
a0.5	0.5ug/ml (a)	3.26	98.80	0.03	0.03	30.00	5.64	11.28
b0.5	0.5ug/ml (b)	3.71	98.81	0.04	0.04	30.00	6.42	12.84
	0.5ug/ml	2.54	98.51	0.03	0.03	30.00	4.39	8.79
	0.5ug/ml	3.20	100.55	0.03	0.03	30.00	5.54	11.07
	0.5ug/ml	3.23	95.49	0.03	0.03	30.00	5.59	11.17
	0.5ug/ml	1.99	97.50	0.02	0.02	30.00	3.44	6.88
	0.5ug/ml	4.49	99.53	0.05	0.05	30.00	7.77	15.53
Mean		3.20	98.46	0.03	0.03		5.54	11.08
SD		0.80	1.61	0.01	0.00		1.38	2.76
%CV		24.94	1.63	24.43	13.18		24.94	24.94
N		7.00	7.00	7.00	5.00		7.00	7.00
BLANKS:								
aB	0ug/mL (a)	ND	10.66	0.00	0.00	2	0.00	
bB	0 ug/mL (b)	ND	8.64	0.00	0.00	2	0.00	
Mean		0.00	9.65	0.00	0.00		0.00	
SD		0.00	1.43	0.00	0.00		0.00	
%CV		ERR	14.80	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00		2.00	
'Guanosine 5 ng 09/03/15		55.16	11.29			5.00		
'Guanosine 5 ng 10/03/15		54.94	9.66			5.00		
'Guanosine 5 ng 11/03/15		58.06	11.44			5.00		
'Guanosine 5 ng 12/03/15		55.50	11.47			5.00		
'Guanosine 5 ng 13/03/15		53.33	9.80			5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHRV Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.80	0.00	0.80
0.50	0.03	0.00	0.03
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0609
Std Err of Y Est	0.0987
R Squared	0.9763
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3388
Std Err of Coef.	0.0527

Sample ID	Time post-dos	Peak #	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	Conc.(µM)	Conc.(µM) Corrected for dilution
T0 R377	0.00	1.58	95.34	0.017	0.017	35	-	0.23	0.81	1.08
T0.08 R377	0.08	ND	108.46	0.000	0.000	35	-	0.18	0.63	0.85
T0.25 R377	0.25	1.78	119.98	0.015	0.015	35	-	0.22	0.79	1.05
T1 R377	1.00	1.97	129.53	0.015	0.015	35	-	0.22	0.79	1.06
Mycoplasma (30 mg/kg)										
T1.2 R377	1.20	1.59	126.77	0.013	0.013	35	-	0.22	0.81	1.08
T1.5 R377	1.50	2.08	129.68	0.016	0.016	35	-	0.23	0.85	1.13
T2 R377	2.00									
T3 R377	3.00									
T4 R377	4.00									
T5 R377	5.00									
T6 R377	6.00									
Mean		1.50	118.29	0.01	0.01			0.22	0.78	1.04
SD		0.76	13.81	0.01	0.01			0.02	0.07	0.10
%CV		50.77	11.68	50.23	50.23			8.57	9.58	9.58
n		17.00	12.00	6.00	6.00			6.00	6.00	6.00

NOTE: QC samples were prepared with plasma from healthy rat No 181

The Spiking solutions were made on: 22/02/2006

*Repeat injections of QC a or b

Rat died

Submitted by: Shyam Sundar

Date: 02/04/2015

Checked by:

Date:

Approved by: Pollen Yeung

Date: 14/04/2015

Plasma Concentrations of Uric Acid in Rat 377
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Rec: Recovery (%)
Uric Acid 5 ng		32.60					5		
a25	25ug/ml (a)	54.58	7.52	7.26	7.26	2	2511.35	92.37	
b25	25ug/ml (b)	61.14	9.57	6.39	6.39	2	2813.19	104.44	
Mean		57.86	8.55	6.82	6.82		2662.27	98.40	
SD		4.64	1.45	0.61	0.61		213.43	8.54	
%CV		8.02	16.96	9.01	9.01		8.02	8.68	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a5	5ug/ml (a)	9.40	9.97	0.94	0.94	2	432.52	46.06	
b5	5ug/ml (b)	11.72	10.09	1.16	1.16	2	539.26	67.41	
	5ug/ml	9.53	10.99	0.87	0.87	2	438.50	47.25	
	5ug/ml	8.98	10.81	0.83	0.83	2	413.19	42.19	
	5ug/ml	8.95	10.57	0.85	0.85	2	411.81	45.46	
	5ug/ml	10.80	10.18	1.06	1.06	2	496.93	62.46	
	5ug/ml	8.83	10.47	0.84	0.84	2	405.29	44.36	
Mean		9.74	10.44	0.94	0.94		448.36	50.74	
SD		1.10	0.38	0.13	0.13		50.52	9.93	
%CV		11.27	3.64	13.71	13.71		11.27	19.57	
N		7.00	7.00	7.00	7.00		7.00	7.00	
BLANKS:									
aB	0ug/ml (a)	4.78	10.66	0.45	0.45	2	219.94		
bB	0ug/ml (b)	4.01	8.64	0.46	0.46	2	184.51		
Mean		4.40	9.65	0.46	0.46		202.22		
SD		0.54	1.43	0.01	0.01		25.05		
%CV		12.39	14.80	2.44	2.44		12.39		
N		2.00	2.00	2.00	2.00		2.00		
Uric Acid 5 ng 09/03/2015		30.80	11.29				5.00		
Uric Acid 5 ng 10/03/2015		29.98	9.66				5.00		
Uric Acid 5 ng 11/03/2015		32.46	11.44				5.00		
Uric Acid 5 ng 12/03/2015		32.79	11.47				5.00		
Uric Acid 5 ng 13/03/2015		30.21	9.80				5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	6.82	0.46	6.37
5.00	0.94	0.46	0.48
0.00	0.46	0.46	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.3779
Std Err of Y Est	0.6122
R Squared	0.9851
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.2660
Std Err of Coef.	0.0327

Sample ID	Time post-dos	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)onc.(µM)	Conc.(µM) Corrected for dilution
T0 R377	0.00	16.27	17.16	0.95	0.95	5	-	4.98	29.65
T0.08 R377	0.08	12.08	20.70	0.58	0.58	5	-	3.61	21.50
T0.25 R377	0.25	4.24	21.79	0.19	0.19	5	-	2.15	12.80
T1 R377	1.00	7.55	23.47	0.32	0.32	5	-	2.63	15.64
Benazepril (30 mg/kg)									
T1.2 R377	1.20	5.24	23.62	0.22	0.22	5	-	2.25	16.56
T1.5 R377	1.50	40.70	24.58	1.66	1.66	5	-	7.64	56.17
T2 R377	2.00								
T3 R377	3.00								
T4 R377	4.00								
T5 R377	5.00								
T6 R377	6.00								
Mean		14.35	21.89	0.65	0.65			3.88	25.39
SD		13.67	2.70	0.57	0.57			2.13	16.20
%CV		95.30	12.35	86.55	86.55			54.87	63.81
n		6.00	6.00	6.00	6.00			6.00	6.00

NOTE: QC samples were prepared with plasma from healthy rat No. 181

The Spiking solutions were made on: 22/02/2006

*Repeated injections of QC a or b

Rat died

Submitted by: Shyam Sundar Date: 02/04/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 15/04/2015

Title: Measurement of Plasma Concentrations of Dipyridamole in Rat 377

According to SOP No: SOP/STD/2008-001-1 (Plasma with no Stopping Solution) Experiment Date:26/10/2014

Abs.amt ng	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount Recov. (ng)	Recovery (%)
Dipyridamole (1ng)		65.73				1		
a1000	1 ug/mL(a)	98.17	2.31	42.50	42.50	5	59.74	119.48
b1000	1ug/mL(b)	90.66	3.61	25.11	25.11	5	55.17	110.34
Mean		94.42	2.96	33.81	33.81		57.46	114.91
SD		5.31	0.92	12.29	12.29		3.23	6.46
%CV		5.62	31.06	36.36	36.36		5.62	5.62
n		2.00	2.00	2.00	2.00		1.00	1.00
a100	0.1 ug/mL (a)	39.89	10.08	3.96	3.96	20	6.07	121.38
b100	0.1ug/mL (b)	36.82	9.42	3.91	3.91	20	5.60	112.03
Mean		38.36	9.75	3.93	3.93		5.84	116.70
SD		2.17	0.47	0.03	0.03		0.33	6.61
%CV		5.66	4.79	0.87	0.87		5.66	5.66
n		2.00	2.00	2.00	2.00		2.00	2.00
aB	0 ug/mL (a)	0.00	9.76	0.00	0.00	20	0.00	0.00
bB	0 ug/mL (b)	0.00	9.24	0.00	0.00	20	0.00	0.00
Mean		0.00	9.50	0.00	0.00		0.00	0.00
SD		0.00	0.37	0.00	0.00		0.00	0.00
%CV		ERR	3.87	ERR	ERR		ERR	ERR
n		2.00	2.00	2.00	2.00		2.00	2.00

Plasma Conc. (ug/mL)	Peak Ht.Ratio (PHR)	Blank (PHRb)	PHRV-PHRb
0.00	0.00	0.00	0.00
0.10	3.93	0.00	3.93
1.00	33.81	0.00	33.81

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.3887
R Squared	0.9996
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	33.8604
Std Err of Coef.	0.3868

Sample ID	Time Post-dose (h)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc.(ug/mL)
R3770	0.00	3.82	18.39	0.21	0.21	20	-	0.10
R3770.08	0.08	43.20	12.13	3.56	3.56	20	-	1.66
R3770.25	0.25	24.41	12.39	1.97	1.97	20	-	0.92
R3771	1.00	33.02	20.30	1.63	1.63	20	-	0.76
Isoproterenol (30 mg/kg sc)								
R3771.2	1.20	22.00	11.98	1.84	1.84	20	-	0.86
R3771.3	1.50	21.96	15.76	1.39	1.39	20	-	0.65
R3772	2.00							
R3773	3.00							
R3774	4.00							
R3775	5.00							
R3776	6.00							
Mean		24.74	15.16	1.77	1.77			0.82
SD		13.13	3.58	1.08	1.08			0.50
%CV		53.09	23.64	61.29	61.29			61.28
n		6.00	6.00	6.00	6.00			6.00

Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 Corr. PHR = (PHR - RGB PHR)

Dipyridamole (1ng)

Comments: Plasma from Rat 156 was used for extraction QC's.

*A repeat injection of a or b
Rat died

Submitted by: Shyam Sunder

Date: 27/10/2014

Checked by: Pollen Yeung

Date:07/11/2014

Approved by:

Date:

APPENDIX 8: Rat 378

Title: Measurement of RBC Concentrations of ATP in Rat 378 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)

Experiment Date 10/07/2014

Sample/standard ID	Standard Concentra (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recov %	Recovery
ATP 4 ng										
a250	250 ug/mL	29.24	6.03	4.85	35.00	4.85	4.85	0.35	29740.18	102.12
b250	250 ug/mL	27.31	5.53	4.94	35.00	4.94	4.94	0.35	27777.16	94.27
Mean		28.28	5.78	4.89	35.00	4.89	4.89		28758.67	98.19
SD		1.36	0.35	0.06	0.00	0.06	0.06		1388.06	5.55
%CV		4.83	6.12	1.28	0.00	1.28	1.28		4.83	5.65
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a100										
b100	100 ug/mL	17.41	7.68	2.27	35.00	2.27	2.27	0.35	17707.81	134.97
100'	100 ug/mL	14.44	6.86	2.10	35.00	2.10	2.10	0.35	14687.01	104.76
Mean		15.93	7.27	2.19	35.00	2.19	2.19		16197.41	119.87
SD		2.10	0.58	0.11	0.00	0.11	0.11		2136.03	21.36
%CV		13.19	7.98	5.24	0.00	5.24	5.24		13.19	17.82
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a8										
b8	0 ug/mL (a)	3.96	6.47	0.61	35.00	0.61	0.61	0.35	4027.74	
	0 ug/mL (a)	4.32	8.57	0.50	35.00	0.50	0.50	0.35	4393.90	
Mean		4.14	7.52	0.56	35.00	0.56	0.56		4210.82	
SD		0.25	1.48	0.08	0.00	0.08	0.08		258.91	
%CV		6.15	19.75	13.68	0.00	13.68	13.68		6.15	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank PHRV-PHRb
250.00	4.89	0.56
100.00	2.19	0.56
0.00	0.56	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0420
Std Err of Y Est	0.0863
R Squared	0.9992
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0174
Std Err of Coef.	0.0005

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC
R37870	0.00	62.44	58.32	1.07	35.00	1.07	1.07	1.50	-	63.65	0.1291
R37870.08	0.08	95.51	60.28	1.58	35.00	1.58	1.58	1.50	-	93.48	0.1843
R37870.25	0.25	57.05	51.99	1.10	35.00	1.10	1.10	1.50	-	65.48	0.1291
R37871	1.00	64.17	40.40	1.59	35.00	1.59	1.59	1.00	-	93.71	0.1847
Regression Method (10 mg/kg sq)											
R37871.2	1.20	60.00	42.87	1.40	35.00	1.40	1.40	1.00	-	82.85	0.1634
R37871.5	1.50	64.80	37.47	1.73	35.00	1.73	1.73	1.00	-	101.81	0.2007
R37872	2.00	75.68	40.68	1.86	35.00	1.86	1.86	1.50	-	109.34	0.2156
R37873	3.00	68.01	40.89	1.66	35.00	1.66	1.66	1.00	-	98.01	0.1932
R37874	4.00	70.30	40.52	1.72	35.00	1.72	1.72	1.00	-	101.16	0.1994
R37875	5.00	53.91	37.86	1.42	35.00	1.42	1.42	1.00	-	84.19	0.1660
R37876	6.00	52.22	42.83	1.22	35.00	1.22	1.22	1.00	-	72.49	0.1429
Mean		65.83	44.96	1.49	35.00	1.49	1.49	1.18		87.86	0.17
SD		12.67	8.06	0.27	0.00	0.27	0.27	0.25		15.33	0.03
%CV		18.33	17.93	17.94	0.00	17.94	17.94	21.35		17.44	17.44
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00		11.00	11.00

ATP (4 ng)

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.

*Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma;	RBC = red blood cells
Peak Ht. = peak height	PCV = packed cell volume (haematocrit)
Peak Ht. R. (or: PHR) = peak height ratio	CorPHR = corrected peak height ratio
I.S. = internal standard	Hemolysis Degree:
Inj Vol = injection volume	-: no visible hemolysis
ND = not detected or determined	+ : slight hemolysis
NS = no sample	++ : intermediate hemolysis
INT = interference	+++ : serious hemolysis

Submitted by: Shyam Sundar K Date: 17/07/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 17/07/2014

Title: Measurement of RBC Concentrations of ADP in Rat 378 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)

Experiment Date: 10/07/2014

Sample/standard ID	Standard Co (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered	Recovery %
ADP 4 ng		15.60						4.00		
a250	250 µg/mL	50.48	6.03	8.37	35.00	8.37	8.37	0.35	20339.93	78.52
b250	250 µg/mL	48.39	5.53	8.39	35.00	8.39	8.39	0.35	18661.94	71.92
Mean		48.44	5.78	8.38	35.00	8.38			19515.93	75.22
SD		2.89	0.35	0.01	0.00	0.01			1165.30	4.66
%CV		5.97	6.12	0.15	0.00	0.15			5.97	6.20
n		2.00	2.00	2.00	2.00	2.00			2.00	2.00
100a	100 µg/mL	25.87	7.88	3.37	35.00	3.37	3.37	0.35	10423.81	97.13
100b	100 µg/mL	24.40	6.86	3.56	35.00	3.56	3.56	0.35	9831.50	91.20
100*	100 µg/mL									
Mean		25.14	7.27	3.46	35.00	3.46	3.46		10127.66	94.16
SD		1.04	0.58	0.13	0.00	0.13	0.13		418.82	4.19
%CV		4.14	7.98	3.85	0.00	3.85	3.85		4.14	4.45
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
aB	0 µg/mL (a)	1.50	6.47	0.23	35.00	0.23	0.23	0.35	604.40	
bB	0 µg/mL (a)	2.03	8.57	0.24	35.00	0.24	0.24	0.35	817.95	
Mean		1.77	7.52	0.23	35.00	0.23	0.23		711.17	
SD		0.37	1.48	0.00	0.00	0.00	0.00		151.00	
%CV		21.23	19.75	1.52	0.00	1.52	1.52		21.23	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
250.00	8.38	0.23	8.15
100.00	3.46	0.23	3.23
0.00	0.23	0.23	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0118
Std Err of Y Est	0.0243
R Squared	1.0000
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0326
Std Err of Coef.	0.0001

Sample ID	Time post	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. Hemolysis (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(mM) RBC	
R37870	0.00	16.27	59.32	0.28	35.00	0.28	0.28	1.50	-	8.92	0.0209	0.268
R37870.08	0.08	20.42	60.28	0.34	35.00	0.34	0.34	1.50	-	10.75	0.0252	0.304
R37870.25	0.25	11.25	51.99	0.22	35.00	0.22	0.22	1.50	-	7.00	0.0164	0.211
R37871	1.00	15.18	40.40	0.38	35.00	0.38	0.38	1.00	-	11.89	0.0278	0.358
Ibuprofenolol (30 mg/kg sc)												
R37871.2	1.20	11.26	42.87	0.26	35.00	0.26	0.26	1.00	-	8.42	0.0197	0.253
R37871.5	1.50	10.33	37.47	0.28	35.00	0.28	0.28	1.00	-	8.82	0.0206	0.265
R37872	2.00	12.11	40.88	0.30	35.00	0.30	0.30	1.50	-	9.50	0.0222	0.286
R37873	3.00	11.32	40.89	0.28	35.00	0.28	0.28	1.00	-	8.86	0.0207	0.267
R37874	4.00	9.78	40.92	0.24	35.00	0.24	0.24	1.50	-	7.69	0.0180	0.232
R37875	5.00	10.07	37.89	0.27	35.00	0.27	0.27	1.00	-	8.52	0.0199	0.256
R37876	6.00	7.82	42.83	0.18	35.00	0.18	0.18	1.00	-	5.96	0.0140	0.179
Mean		15.98	56.86	0.28	35.00	0.28		1.50		8.89	0.02	0.27
SD		4.59	4.33	0.06	0.00	0.06		0.00		1.88	0.00	0.06
%CV		28.74	7.62	22.01	0.00	22.01		0.00		21.11	21.11	21.11
n		3.00	3.00	3.00	3.00	3.00		3.00		3.00	2.00	3.00

ADP (4 ng)

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.

*Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells

Peak Ht. = peak height

Peak Ht. R. (or: PHR) = peak height ratio

I.S. = internal standard

Inj Vol = injection volume

ND = not detected or determined

NS = no sample

INT = interference

PCV = packed cell volume (haematocrit)

CorPHR = corrected peak height ratio

Hemolysis Degree:

-: no visible hemolysis

+: slight hemolysis

++: intermediate hemolysis

+++: serious hemolysis

Submitted by: Shyam Sundar K

Date: 17/07/2014

Checked by:

Date:

Approved by: Pollen Yeung

Date: 17/07/2014

Title: Measurement of RBC Concentrations of AMP in Rat 378 extracted by Shyam Sundar

Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)

Experiment Date 10/07/2014

Sample/standard ID	Standard Concentra (ug/mL)	Peak HT. # (mm)	Peak HT. I.S. (mm)	Peak HT. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (uL)	Amount Recove	% Recovery
AMP 4 ng										
a50	50 ug/mL	28.77	6.03	4.77	35.00	4.77	4.77	0.35	4050.17	76.77
b50	50 ug/mL	27.77	5.53	5.02	35.00	5.02	5.02	0.35	3900.39	73.95
Mean		28.27	5.78	4.90	35.00	4.90	4.90		3970.78	75.36
SD		0.71	0.35	0.18	0.00	0.18	0.18		99.54	1.99
%CV		2.50	6.12	3.62	0.00	3.62	3.62		2.50	2.64
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20	20 ug/mL	14.88	7.68	1.94	35.00	1.94	1.94	0.35	2094.77	94.14
b20	20 ug/mL	13.29	6.86	1.94	35.00	1.94	1.94	0.35	1870.93	82.95
20*	20 ug/mL									
Mean		14.09	7.27	1.94	35.00	1.94	1.94		1982.85	88.55
SD		1.12	0.58	0.00	0.00	0.00	0.00		158.28	7.91
%CV		7.98	7.98	0.01	0.00	0.01	0.01		7.98	8.94
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a8	0 ug/mL (a)	1.65	6.47	0.26	35.00	0.26	0.26	0.35	232.28	
b8	0 ug/mL (a)	1.36	8.57	0.16	35.00	0.16	0.16	0.35	191.46	
Mean		1.51	7.52	0.21	35.00	0.21	0.21		211.87	
SD		0.21	1.48	0.07	0.00	0.07	0.07		28.87	
%CV		13.63	19.75	32.93	0.00	32.93	32.93		13.63	
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	4.90	0.21	4.69
20.00	1.94	0.21	1.73
0.00	0.21	0.21	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.0954
R Squared	0.9984
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.0628
Std Err of Coef.	0.0018

Sample ID	Time post dose	Peak HT. # (mm)	Peak HT. I.S. (mm)	Peak HT. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (uL)	Hemolysis Degree	Conc(ug/mL) Lysate	Conc.(mM) RBC
R37870	0.00	2.86	58.32	0.05	35.00	0.05	0.05	1.50	-	0.53	0.0015
R37870.08	0.08	1.88	60.28	0.03	35.00	0.03	0.03	1.50	-	0.34	0.0010
R37870.25	0.25	0.81	51.99	0.02	35.00	0.02	0.02	1.50	-	0.17	0.0005
R37871	1.00	0.92	40.40	0.02	35.00	0.02	0.02	1.00	-	0.25	0.0007
Insufficient (30 mg/kg as)											
R37871.2	1.20	1.38	42.87	0.03	35.00	0.03	0.03	1.00	-	0.35	0.0010
R37871.5	1.50	0.84	37.47	0.02	35.00	0.02	0.02	1.00	-	0.24	0.0007
R37872	2.00	1.37	40.88	0.03	35.00	0.03	0.03	1.50	-	0.38	0.0010
R37873	3.00	1.24	40.89	0.03	35.00	0.03	0.03	1.50	-	0.33	0.0009
R37874	4.00	0.76	40.92	0.02	35.00	0.02	0.02	1.00	-	0.20	0.0006
R37875	5.00	0.92	37.89	0.02	35.00	0.02	0.02	1.00	-	0.26	0.0008
R37876	6.00	ND	42.83	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000
Mean		1.18	44.96	0.03	35.00	0.03	0.03	1.23	0.00	0.27	0.00
SD		0.73	8.06	0.01	0.00	0.01	0.01	0.26	0.00	0.13	0.00
%CV		61.98	17.93	48.69	0.00	48.69	48.69	21.28	ERR	48.69	48.69
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

AMP (4 ng)

Comments: RBC Lysate from R 338 was used for GC Samples. New calibration solution was prepared on April 22, 2014.

*Repeated injections of "a" or "b" at 0.5 - 1 ul injection volume.

PL = plasma;	RBC = red blood cells	PCV = packed cell volume (haematocrit)
Peak HT. = peak height		CorPHR = corrected peak height ratio
Peak HT. R. (or: PHR) = peak height ratio		Hemolysis Degree:
I.S. = internal standard		-: no visible hemolysis
Inj Vol = injection volume		+: slight hemolysis
ND = not detected or determined		++: intermediate hemolysis
NS = no sample		+++: serious hemolysis
INT = interference		

Submitted by: Shyam Sundar Date: 17/07/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 17/07/2014

Title: Measurement of RBC Concentrations of GDP in Rat 378 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 10/07/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recove %	Recovery
GDP 4 ng										
a50	50 ug/mL	15.74	6.03	2.61	35.00	2.61	2.61	0.35	4074.84	80.00
b50	50 ug/mL	13.43	5.53	2.43	35.00	2.43	2.43	0.35	3476.82	68.00
Mean		14.59	5.78	2.52	35.00	2.52	2.52		3775.83	74.00
SD		1.63	0.35	0.13	0.00	0.13	0.13		422.87	8.46
%CV		11.20	6.12	5.10	0.00	5.10	5.10		11.20	11.43
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a20										
b20	20 ug/mL	7.52	7.68	0.98	35.00	0.98	0.98	0.35	1046.81	83.59
20*	20 ug/mL	6.68	6.86	0.97	35.00	0.97	0.97	0.35	1729.35	82.71
Mean		7.10	7.27	0.98	35.00	0.98	0.98		1038.08	88.15
SD		0.59	0.58	0.00	0.00	0.00	0.00		153.77	7.68
%CV		8.37	7.98	0.39	0.00	0.39	0.39		8.37	8.72
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
a8										
b8	0 ug/mL (a)	0.58	6.47	0.09	35.00	0.09	0.09	0.35	150.15	0.00
	0 ug/mL (a)	ND	8.57	0.00	35.00	0.00	0.00	0.35	0.00	0.00
Mean		0.29	7.52	0.04	35.00	0.04	0.09		75.08	
SD		0.41	1.48	0.06	0.00	0.06	ERR		108.17	
%CV		141.42	19.75	141.42	0.00	141.42	ERR		141.42	
n		2.00	2.00	2.00	2.00	2.00	1.00		2.00	

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRB)	PHRV-PHRB
50.00	2.52	0.09	2.43
20.00	0.98	0.09	0.89
0.00	0.09	0.09	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0336
Std Err of Y Est	0.0690
R Squared	0.9984
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.0488
Std Err of Coef.	0.0019

Sample ID	Time post dose hr	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lysate	Conc.(nM) RBC	Conc.(nM)
R37870	0.00	3.88	58.32	0.07	35.00	0.07	0.07	1.50	-	2.05	0.0046	0.059
R37870.08	0.08	5.48	80.28	0.09	35.00	0.09	0.09	1.50	-	2.55	0.0058	0.074
R37870.25	0.25	3.14	51.89	0.06	35.00	0.06	0.06	1.50	-	1.53	0.0043	0.056
R37871	1.00	4.04	40.40	0.10	35.00	0.10	0.10	1.00	-	2.74	0.0082	0.079
Isoproterenol (30 mg/kg sc)												
R37871.2	1.20	2.85	42.87	0.07	35.00	0.07	0.07	1.00	-	2.05	0.0046	0.059
R37871.5	1.50	3.48	37.47	0.09	35.00	0.09	0.09	1.00	-	2.59	0.0058	0.075
R37872	2.00	4.14	40.68	0.10	35.00	0.10	0.10	1.50	-	2.77	0.0083	0.080
R37873	3.00	3.84	40.89	0.09	35.00	0.09	0.09	1.00	-	2.61	0.0059	0.076
R37874	4.00	3.42	40.92	0.08	35.00	0.08	0.08	1.00	-	2.40	0.0054	0.070
R37875	5.00	3.57	37.89	0.09	35.00	0.09	0.09	1.00	-	2.62	0.0059	0.076
R37876	6.00	3.75	42.83	0.09	35.00	0.09	0.09	1.00	-	2.48	0.0056	0.072
Mean		24.37	44.96	0.09	35.00	0.09	0.09	1.18	0.00	2.44	0.01	0.07
SD		21.80	8.06	0.01	0.00	0.01	0.01	0.25	0.00	0.29	0.00	0.01
%CV		89.46	17.93	16.85	0.00	16.85	16.85	21.35	ERR	12.09	12.09	12.09
n		22.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

GDP (4ng)

Comments: RBC Lysate from R 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of a or b at 0.5 - 1ul injection volume.

PL = plasma; RBC = red blood cells
 Peak Ht. = peak height
 Peak Ht. R. (or PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 INT = interference
 PCV = packed cell volume (haematocrit)
 CorPHR = corrected peak height ratio
 Hemolysis Degree:
 -: no visible hemolysis
 +: slight hemolysis
 ++: intermediate hemolysis
 +++: serious hemolysis

Submitted by: Shyam Sundar K Date:17/07/2014

Checked by: Date:

Approved by: Pollen Yeung Date: 18/07/2014

Title: Measurement of RBC Concentrations of GMP in Rat 378 samples extracted by Shyam Sundar
 Based on 'SOP NO.: SOP/STD/2005-005-0' (With Stopping Solution)
 Experiment Date: 10/07/2014

Sample/standard ID	Standard Concentr (µg/mL)	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Amount Recovered (%)	% Recovery
GMP 4 ng										
a50	50 ug/mL	83.56	6.03	13.86	35.00	13.86	13.86	0.35	4151.39	83.03
b50	50 ug/mL	79.58	5.53	14.39	35.00	14.39	14.39	0.35	3953.66	79.07
Mean		81.57	5.78	14.12	35.00	14.12	14.12		4052.53	81.05
SD		2.81	5.66	0.38	0.00	0.38	0.38		139.82	2.80
%CV		3.45	5.72	2.67	0.00	2.67	2.67		3.45	3.45
n		2.00	5.72	2.00	2.00	2.00	2.00		2.00	2.00
a20										
b20	20 ug/mL	84.06	7.68	10.95	35.00	10.95	10.95	0.35	4176.23	208.81
20*	20 ug/mL	84.66	6.66	12.34	35.00	12.34	12.34	0.35	4296.04	210.30
Mean		84.36	7.27	11.64	35.00	11.64	11.64		4191.14	208.56
SD		0.42	0.58	0.99	0.00	0.99	0.99		21.08	1.05
%CV		0.50	7.98	8.48	0.00	8.48	8.48		0.50	0.50
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00
aB										
bB	0 ug/mL (a)	59.60	6.47	9.21	35.00	9.21	9.21	0.35	0.00	0.00
	0 ug/mL (a)	78.42	8.57	9.15	35.00	9.15	9.15	0.35	0.00	0.00
Mean		69.01	7.52	9.18	35.00	9.18	9.18		0.00	0.00
SD		13.31	1.48	0.04	0.00	0.04	0.04		0.00	0.00
%CV		19.28	19.75	0.47	0.00	0.47	0.47		ERR	ERR
n		2.00	2.00	2.00	2.00	2.00	2.00		2.00	2.00

Regression Analysis of Standard Curve Data

Conc. (µg/mL)	Peak Height Ratio Value (PHRV)	Blank (PHRb)	PHRV-PHRb
50.00	14.12	9.18	4.94
20.00	11.64	9.18	2.46
0.00	9.18	9.18	0.00

Regression Output Begins Here:

Regression Output:	
Constant	0.0000
Std Err of Y Est	0.3184
R Squared	0.9834
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.1022
Std Err of Coef.	0.0059

Sample ID	Time post dose hr	Peak Ht. # (nm)	Peak Ht. I.S. (nm)	Peak Ht. Ratio	PCV (%)	CorPHR	CorPHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc(µg/mL) Lyasate	Conc.(mM) RBC	Conc.(mM) RBC
R37870		0.00 off scale	56.21	0.00	35.00	0.00	0.00	1.50	-	0.00	0.0000	0.000
R37870.08		0.08 off scale	79.54	0.00	35.00	0.00	0.00	1.50	-	0.00	0.0000	0.000
R37870.25		0.25 off scale	63.12	0.00	35.00	0.00	0.00	1.50	-	0.00	0.0000	0.000
R37871		1.00 off scale	56.54	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
Not detected (ND mg/kg sat)												
R37871.2		1.20 off scale	46.16	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
R37871.5		1.50 off scale	43.92	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
R37872		2.00 off scale	56.21	0.00	35.00	0.00	0.00	1.50	-	0.00	0.0000	0.000
R37873		3.00 off scale	79.54	0.00	35.00	0.00	0.00	1.50	-	0.00	0.0000	0.000
R37874		4.00 off scale	63.12	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
R37875		5.00 off scale	56.54	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
R37876		6.00 off scale	56.64	0.00	35.00	0.00	0.00	1.00	-	0.00	0.0000	0.000
Mean		0.00	59.78	0.00	35.00	0.00	0.00	1.23	0.00	0.00	0.00	0.00
SD		0.00	11.39	0.00	0.00	0.00	0.26	0.00	0.00	0.00	0.00	0.00
%CV		ERR	19.05	ERR	0.00	ERR	ERR	21.28	ERR	ERR	ERR	ERR
n		11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00	11.00

GMP (4 ng)

Comments: RBC Lyasate from Rat 338 was used for QC Samples. New calibration solution was prepared on April 22, 2014.
 *Repeated injections of "a" or "b" at 0.5-1ul injection volume

PL = plasma;	RBC = red blood cells	PCV = packed cell volume (haematocrit)
Peak Ht. = peak height	CorPHR = corrected peak height ratio	Hemolysis Degree:
Peak Ht. R. (or: PHR) = peak height ratio		-: no visible hemolysis
I.S. = internal standard		+: slight hemolysis
Inj Vol = injection volume		++: intermediate hemolysis
ND = not detected or determined		+++: serious hemolysis
NS = no sample		
INT = interference		

Submitted by: Shyam Sundar K Date: 17/07/2014
 Checked by: Date:
 Approved by: Pollen Yeung Date: 18/07/2014

Plasma Concentrations of Adenosine in Rat 378
Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Value	Ratio	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Adenosine 5 ng										
a2.5	2.5ug/ml (a)	15.32	29.55	0.52	0.52	10	143.94	57.58		
b2.5	2.5ug/ml (b)	18.15	40.19	0.45	0.45	10	170.53	68.21		
Mean		16.74	34.87	0.49	0.49		157.23	62.89		
SD		2.00	7.52	0.05	0.05		18.80	7.52		
%CV		11.96	21.58	9.74	9.74		11.96	11.96		
N		2.00	2.00	2.00	2.00		2.00	2.00		
a0.5										
b0.5	0.5ug/ml (a)	6.71	98.80	0.07	0.07	30.00	21.01	42.03		
	0.5ug/ml (b)	7.71	98.81	0.08	0.08	30.00	24.15	48.29		
	0.5 ug/ml	8.08	98.51	0.08	0.08	30.00	25.31	50.61		
	0.5 ug/ml	6.26	100.55	0.06	0.06	30.00	19.61	39.21		
	0.5 ug/ml	8.06	95.49	0.08	0.08	30.00	25.24	50.49		
	0.5 ug/ml	9.96	97.50	0.10	0.10	30.00	31.19	62.39		
	0.5 ug/ml	8.67	99.53	0.09	0.09	30.00	27.15	54.31		
Mean		7.92	98.46	0.08	0.08		24.81	49.62		
SD		1.23	1.61	0.01	0.01		3.84	7.69		
%CV		15.49	1.63	16.24	16.24		15.49	15.49		
N		7.00	7.00	7.00	7.00		7.00	7.00		
BLANKS:										
aB	0ug/mL (a)	N/D	10.66	0.00	0.00	2	0.00			
bB	0 ug/mL (b)	N/D	8.64	0.00	0.00	2	0.00			
Mean		0.00	9.65	0.00	0.00		0.00			
SD		0.00	1.43	0.00	0.00		0.00			
%CV		ERR	14.80	ERR	ERR		ERR			
N		2.00	2.00	2.00	2.00		2.00			
Adenosine 5 ng 09/03/2015		30.02	11.29				5.00			
Adenosine 5 ng 10/03/2015		30.00	9.66				5.00			
Adenosine 5 ng 11/03/2015		31.93	11.44				5.00			
Adenosine 5 ng 12/03/2015		30.96	11.47				5.00			
Adenosine 5 ng 13/03/2015		27.70	9.80				5.00			

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.49	0.00	0.49
0.50	0.08	0.00	0.08
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0079
Std Err of Y Est	0.0127
R Squared	0.9988
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.1964
Std Err of Coef.	0.0068

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c. (µg/mL) onc. (µM)	Conc. (µM) Corrected for dilution	
T0 R378	0.00	3.55	88.63	0.04	0.04	35	-	-	0.24	0.91	1.22
T0.08 R378	0.08	7.71	99.12	0.08	0.08	35	-	-	0.44	1.63	2.18
T0.25 R378	0.25	2.99	114.18	0.03	0.03	35	-	-	0.17	0.65	0.86
T1 R378	1.00	4.43	126.86	0.03	0.03	35	-	-	0.22	0.81	1.09
Isoproterenol (30 mg/kg)											
T1.2 R378	1.20	6.65	91.88	0.07	0.07	35	-	-	0.41	1.53	2.04
T1.5 R378	1.50	6.92	109.41	0.06	0.06	35	-	-	0.36	1.35	1.81
T2 R378	2.00	5.42	110.63	0.05	0.05	35	-	-	0.29	1.08	1.44
T3 R378	3.00	4.07	129.83	0.03	0.03	35	-	-	0.20	0.75	1.00
T4 R378	4.00	4.30	117.07	0.04	0.04	35	-	-	0.23	0.85	1.13
T5 R378	5.00	2.67	110.00	0.02	0.02	35	-	-	0.16	0.61	0.82
T6 R378	6.00	3.32	113.70	0.03	0.03	35	-	-	0.19	0.71	0.94
Mean		4.73	110.12	0.04	0.04				0.26	0.99	1.32
SD		1.71	12.89	0.02	0.02				0.10	0.36	0.48
%CV		36.13	11.71	42.89	42.89				36.42	36.42	36.42
n		11.00	11.00	11.00	11.00				11.00	11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
The Spiking solutions were made on: 22/02/2006
***Repeat injections of QC a or b**

Submitted by: Shyam Sundar Date: 02/04/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 14/04/2015

Plasma Concentrations of Inosine in Rat 378
Based on "SOP NO.: SOP/STD/2004-001-0" (With Stopping Solution)
Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recx (%)	Recovery (%)
Inosine 5 ng		43.11								
a2.5	2.5ug/ml (a)	27.85	29.55	0.94	0.94	10	193.81	77.52		
b2.5	2.5ug/ml (b)	33.72	40.19	0.84	0.84	10	234.66	93.86		
Mean		30.79	34.87	0.89	0.89		214.23	85.69		
SD		4.15	7.52	0.07	0.07		28.88	11.55		
%CV		13.48	21.58	8.21	8.21		13.48	13.48		
N		2.00	2.00	2.00	2.00		2.00	2.00		
a0.5	0.5ug/ml (a)	8.18	98.80	0.08	0.08	30.00	18.97	37.95		
b0.5	0.5ug/ml (b)	8.79	98.81	0.09	0.09	30.00	20.39	40.78		
	0.5ug/ml	11.33	98.51	0.12	0.12	30.00	26.28	52.56		
	0.5ug/ml	13.09	100.55	0.13	0.13	30.00	30.36	60.73		
	0.5ug/ml	9.30	95.49	0.10	0.10	30.00	21.57	43.15		
	0.5ug/ml	10.64	97.50	0.11	0.11	30.00	24.68	49.36		
	0.5ug/ml	5.84	99.53	0.06	0.06	30.00	13.55	27.09		
Mean		9.60	98.46	0.10	0.10		22.26	44.52		
SD		2.35	1.81	0.02	0.02		5.45	10.91		
%CV		24.50	1.83	24.09	16.96		24.50	24.50		
N		7.00	7.00	7.00	6.00		7.00	7.00		
BLANKS:										
aB	0ug/mL (a)	ND	10.66	0.00	0.00	2	0.00			
bB	0ug/mL (b)	ND	8.64	0.00	0.00	2	0.00			
Mean		0.00	9.65	0.00	0.00		0.00			
SD		0.00	1.43	0.00	0.00		0.00			
%CV		ERR	14.80	ERR	ERR		ERR			
N		2.00	2.00	2.00	2.00		2.00			
Inosine 5ng 09/03/2015		43.47	11.29				5.00			
Inosine 5ng 10/03/2015		42.92	9.66				5.00			
Inosine 5ng 11/03/2015		44.40	11.44				5.00			
Inosine 5ng 12/03/2015		42.74	11.47				5.00			
Inosine 5ng 13/03/2015		42.92	9.80				5.00			

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHRV Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.89	0.00	0.89
0.50	0.10	0.00	0.10
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.0354
Std Err of Y Est	0.0573
R Squared	0.9831
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3669
Std Err of Coef.	0.0306

Sample ID	Time post-dose	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c. (ug/mL)	Conc. (µM)	Conc. (µM) Corrected for dilution
T0 R378	0.00	4.31	88.63	0.05	0.05	35	-	0.23	0.85	1.14	
T0.08 R378	0.08	3.25	99.12	0.03	0.03	35	-	0.19	0.69	0.92	
T0.25 R378	0.25	8.33	114.18	0.07	0.07	35	-	0.30	1.10	1.47	
T1 R378	1.00	4.06	126.86	0.03	0.03	35	-	0.18	0.66	0.91	
Inosineferencel (30 mg/kg)											
T1.2 R378	1.20	3.99	91.88	0.04	0.04	35	-	0.21	0.80	1.07	
T1.5 R378	1.50	3.90	109.41	0.04	0.04	35	-	0.19	0.72	0.96	
T2 R378	2.00	5.24	110.63	0.05	0.05	35	-	0.23	0.84	1.12	
T3 R378	3.00	3.77	129.83	0.03	0.03	35	-	0.18	0.65	0.87	
T4 R378	4.00	5.51	117.07	0.05	0.05	35	-	0.22	0.84	1.12	
T5 R378	5.00	8.22	110.00	0.07	0.07	35	-	0.30	1.12	1.49	
T6 R378	6.00	5.24	113.70	0.05	0.05	35	-	0.22	0.83	1.10	
Mean		5.07	110.12	0.05	0.05			0.22	0.83	1.11	
SD		1.73	12.89	0.02	0.02			0.04	0.16	0.21	
%CV		34.10	11.71	32.96	32.96			18.70	18.70	18.70	
n		11.00	11.00	11.00	11.00			11.00	11.00	11.00	

NOTE: QC samples were prepared with plasma from healthy rat No T81
The Spiking solutions were made on: 22/02/2006
***Repeat injections of QC a or b**

Submitted by: Shyam Sundar Date: 02/04/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 14/04/2015

Plasma Concentrations of Hypoxanthine in Rat 378
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recr (%)	Recovery (%)
Hypoxanthine 5 ng										
a25	25ug/ml (a)	117.85	7.52	15.67	15.67	2	1599.34	63.97		
b25	25ug/ml (b)	129.92	9.57	13.58	13.58	2	1763.14	70.53		
Mean		123.89	8.55	14.62	14.62		1681.24	67.25		
SD		8.53	1.45	1.48	1.48		115.83	4.63		
%CV		6.89	16.96	10.13	10.13		6.89	6.89		
N		2.00	2.00	2.00	2.00		2.00	2.00		
a5										
b5	5ug/ml (a)	15.21	9.97	1.53	1.53	2	206.41	41.28		
	5ug/ml (b)	16.26	10.09	1.61	1.61	2	220.66	44.13		
	5ug/ml	17.19	10.99	1.56	1.56	2	233.29	46.66		
	5ug/ml	15.52	10.81	1.44	1.44	2	210.62	42.12		
	5ug/ml	15.73	10.57	1.49	1.49	2	213.47	42.69		
	5ug/ml	14.64	10.18	1.44	1.44	2	198.68	39.74		
	5ug/ml	15.55	10.47	1.49	1.49	2	211.03	42.21		
Mean		15.73	10.44	1.51	1.51		213.45	42.69		
SD		0.81	0.38	0.06	0.06		11.01	2.20		
%CV		5.16	3.64	4.30	4.30		5.16	5.16		
N		7.00	7.00	7.00	7.00		7.00	7.00		
BLANKS:										
aB	0ug/ml (a)	ND	10.66	0.00	0.00	2	0.00			
bB	0ug/ml (b)	ND	8.64	0.00	0.00	2	0.00			
Mean		0.00	9.65	0.00	0.00		0.00			
SD		0.00	1.43	0.00	0.00		0.00			
%CV		ERR	14.80	ERR	ERR		ERR			
N		2.00	2.00	2.00	2.00		2.00			
Hypoxanthine 5 ng 09/03/15										
Hypoxanthine 5 ng 10/03/15										
Hypoxanthine 5 ng 11/03/15										
Hypoxanthine 5 ng 12/03/15										
Hypoxanthine 5 ng 13/03/15										

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	14.62	0.00	14.62
5.00	1.51	0.00	1.51
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.6751
Std Err of Y Est	1.0839
R Squared	0.9908
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.6052
Std Err of Coef.	0.0585

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	onc.(µM)	Conc.(µM) Corrected for dilution
T0 R378	0.00	8.98	18.98	0.47	0.47	5	-	1.90	13.94	18.59	
T0.08 R378	0.08	10.81	16.88	0.64	0.64	5	-	2.17	15.97	21.30	
T0.25 R378	0.25	13.42	22.82	0.59	0.59	5	-	2.09	15.34	20.45	
T1 R378	1.00	12.97	25.07	0.52	0.52	5	-	1.97	14.48	19.30	
Isoproterenol (30 mg/kg)											
T1.2 R378	1.20	11.73	22.85	0.51	0.51	5	-	1.96	14.43	19.24	
T1.5 R378	1.50	10.82	18.75	0.58	0.58	5	-	2.07	15.20	20.27	
T2 R378	2.00	10.64	20.78	0.51	0.51	5	-	1.96	14.41	19.22	
T3 R378	3.00	14.24	24.19	0.59	0.59	5	-	2.09	15.34	20.46	
T4 R378	4.00	12.81	22.00	0.58	0.58	5	-	2.08	15.27	20.35	
T5 R378	5.00	10.67	19.31	0.55	0.55	5	-	2.03	14.91	19.87	
T6 R378	6.00	12.20	20.84	0.59	0.59	5	-	2.08	15.30	20.41	
Mean		11.70	21.29	0.55	0.55			2.03	14.89	19.85	
SD		1.74	2.90	0.06	0.06			0.09	0.67	0.90	
%CV		14.84	13.63	10.08	10.08			4.53	4.53	4.53	
n		8.00	8.00	8.00	8.00			8.00	8.00	8.00	

NOTE: QC samples were prepared with plasma from healthy rat No.181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 02/04/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 15/04/2015

Plasma Concentrations of Xanthine in Rat 378
Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
Experiment Date: 06/03/2015 -13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Rec: Recovery (%)
Xanthine 5 ng		39.10					5		
a25	25ug/ml (a)	48.18	7.52	6.41	6.41	6.41	2	1848.34	73.93
b25	25ug/ml (b)	58.78	9.57	6.14	6.14	6.14	2	2254.99	90.20
Mean		53.48	8.55	6.27	6.27	6.27		2051.66	82.07
SD		7.50	1.45	0.19	0.19	0.19		287.54	11.50
%CV		14.02	16.96	2.98	2.98	2.98		14.02	14.02
N		2.00	2.00	2.00	2.00	2.00		2.00	2.00
a5	5ug/ml (a)	3.36	9.97	0.34	0.34	0.34	2	128.90	25.78
b5	5ug/ml (b)	3.20	10.09	0.32	0.32	0.32	2	122.76	24.55
	5ug/ml	4.88	10.99	0.44	0.44	0.44	2	187.21	37.44
	5ug/ml	4.36	10.81	0.40	0.40	0.40	2	167.26	33.45
	5ug/ml	5.82	10.57	0.55	0.55	0.55	2	223.27	44.65
	5ug/ml	5.20	10.18	0.51	0.51	0.51	2	199.49	39.90
	5ug/ml	3.56	10.47	0.34	0.34	0.34	2	136.57	27.31
Mean		4.34	10.44	0.41	0.37	0.37		166.50	33.30
SD		1.01	0.38	0.09	0.05	0.05		38.65	7.73
%CV		23.21	3.64	21.97	14.47	14.47		23.21	23.21
N		7.00	7.00	7.00	5.00	5.00		7.00	7.00
BLANKS:									
aB	0ug/ml (a)	ND	10.66	0.00	0.00	0.00	2	0.00	
bB	0ug/ml (b)	ND	8.64	0.00	0.00	0.00	2	0.00	
Mean		0.00	9.65	0.00	0.00	0.00		0.00	
SD		0.00	1.43	0.00	0.00	0.00		0.00	
%CV		ERR	14.80	ERR	ERR	ERR		ERR	
N		2.00	2.00	2.00	2.00	2.00		2.00	

Xanthine 5 ng 09/03/15	37.00	11.29	5.00
Xanthine 5 ng 10/03/15	38.29	9.66	5.00
Xanthine 5 ng 11/03/15	39.49	11.44	5.00
Xanthine 5 ng 12/03/15	38.74	11.47	5.00
Xanthine 5 ng 13/03/15	37.26	9.80	5.00

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	6.27	0.00	6.27
5.00	0.37	0.00	0.37
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:

Constant	0.0000
Std Err of Y Est	0.6147
R Squared	0.9695
No. of Observations	3.0000
Degrees of Freedom	2.0000
X Coefficient(s)	0.2442
Std Err of Coef.	0.0241

Sample ID	Time post-dose	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)/onc.(µM)	Conc.(µM) Corrected for dilution
T0 R378	0.00	INT	18.98	0.00	0.00	0.00	5	-	0.00	0.00
T0.08 R378	0.08	INT	16.88	0.00	0.00	0.00	5	-	0.00	0.00
T0.25 R378	0.25	INT	22.82	0.00	0.00	0.00	5	-	0.00	0.00
T1 R378	1.00	INT	25.07	0.00	0.00	0.00	5	-	0.00	0.00
Isoproterenol (30 mg/kg)										
T1.2 R378	1.20	INT	22.85	0.00	0.00	0.00	5	-	0.00	0.00
T1.5 R378	1.50	INT	18.75	0.00	0.00	0.00	5	-	0.00	0.00
T2 R378	2.00	INT	20.78	0.00	0.00	0.00	5	-	0.00	0.00
T3 R378	3.00	INT	24.19	0.00	0.00	0.00	5	-	0.00	0.00
T4 R378	4.00	INT	22.00	0.00	0.00	0.00	5	-	0.00	0.00
T5 R378	5.00	INT	19.31	0.00	0.00	0.00	5	-	0.00	0.00
T6 R378	6.00	INT	20.84	0.00	0.00	0.00	5	-	0.00	0.00
Mean		0.00	21.29	0.00	0.00	0.00			0.00	0.00
SD		0.00	2.90	0.00	0.00	0.00			0.00	0.00
%CV		ERR	13.63	ERR	ERR	ERR			ERR	ERR
n		8.00	8.00	8.00	8.00	8.00			8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No.181
The Spiking solutions were made on: 22/02/2006
***Repeated injections of QC a or b**

Submitted by: Shyam Sundar Date: 02/04/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 15/04/2015

Plasma Concentrations of Guanosine in Rat 378
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 06/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Value	Ratio	Inj Vol. (µL)	Amount (ng)	Recovery (%)
Guanosine 5 ng		57.81						5		
a2.5	2.5ug/ml (a)	25.46	29.55	0.86	0.86	10	132.12	52.85		
b2.5	2.5ug/ml (b)	29.79	40.19	0.74	0.74	10	154.59	61.84		
Mean		27.63	34.87	0.80	0.80		143.36	57.34		
SD		3.06	7.52	0.09	0.09		15.89	6.36		
%CV		11.08	21.58	10.62	10.62		11.08	11.08		
N		2.00	2.00	2.00	2.00		2.00	2.00		
a0.5	0.5ug/ml (a)	3.26	98.80	0.03	0.03	30.00	5.64	11.28		
b0.5	0.5ug/ml (b)	3.71	98.81	0.04	0.04	30.00	6.42	12.84		
	0.5ug/ml	2.54	98.51	0.03	0.03	30.00	4.39	8.79		
	0.5ug/ml	3.20	100.55	0.03	0.03	30.00	5.54	11.07		
	0.5ug/ml	3.23	95.49	0.03	0.03	30.00	5.59	11.17		
	0.5ug/ml	1.99	97.50	0.02		30.00	3.44	6.88		
	0.5ug/ml	4.49	99.53	0.05		30.00	7.77	15.53		
Mean		3.20	98.46	0.03	0.03		5.54	11.08		
SD		0.80	1.61	0.01	0.00		1.38	2.76		
%CV		24.94	1.63	24.43	13.18		24.94	24.94		
N		7.00	7.00	7.00	5.00		7.00	7.00		
BLANKS:										
aB	0ug/mL (a)	ND	10.66	0.00	0.00	2	0.00			
bB	0 ug/mL (b)	ND	8.64	0.00	0.00	2	0.00			
Mean		0.00	9.65	0.00	0.00		0.00			
SD		0.00	1.43	0.00	0.00		0.00			
%CV		ERR	14.80	ERR	ERR		ERR			
N		2.00	2.00	2.00	2.00		2.00			
'Guanosine 5 ng 09/03/15		55.16	11.29				5.00			
'Guanosine 5 ng 10/03/15		54.84	9.66				5.00			
'Guanosine 5 ng 11/03/15		58.06	11.44				5.00			
'Guanosine 5 ng 12/03/15		55.50	11.47				5.00			
'Guanosine 5 ng 13/03/15		53.33	9.80				5.00			

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
2.50	0.80	0.00	0.80
0.50	0.03	0.00	0.03
0.00	0.00	0.00	0.00

Regression Output Begins Here:

Regression Output:	
Constant	-0.0609
Std Err of Y Est	0.0987
R Squared	0.9763
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.3388
Std Err of Coef.	0.0527

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c. (µg/mL) onc. (µM)	Conc. (µM) Corrected for dilution
T0 R378	0.00	2.02	88.63	0.023	0.023	35	-		0.25	0.87
T0.08 R378	0.08	1.57	99.12	0.016	0.016	35	-		0.23	0.80
T0.25 R378	0.25	1.97	114.18	0.017	0.017	35	-		0.23	0.81
T1 R378	1.00	2.04	126.86	0.016	0.016	35	-		0.23	0.80
Isoproterenol (30 mg/kg)										
T1.2 R378	1.20	2.12	91.88	0.023	0.023	35	-		0.25	0.93
T1.5 R378	1.50	2.26	109.41	0.021	0.021	35	-		0.24	0.90
T2 R378	2.00	2.08	110.63	0.019	0.019	35	-		0.24	0.83
T3 R378	3.00	1.90	129.83	0.015	0.015	35	-		0.22	0.79
T4 R378	4.00	2.13	117.07	0.018	0.018	35	-		0.23	0.87
T5 R378	5.00	2.21	110.00	0.020	0.020	35	-		0.24	0.84
T6 R378	6.00	2.98	113.70	0.026	0.026	35	-		0.26	0.91
Mean		2.12	110.12	0.02	0.02				0.24	0.85
SD		0.34	12.89	0.00	0.00				0.01	0.05
%CV		16.08	11.71	18.30	18.30				4.42	5.69
n		22.00	22.00	11.00	11.00				11.00	11.00

NOTE: QC samples were prepared with plasma from healthy rat No 181
 The Spiking solutions were made on: 22/02/2006
 *Repeat injections of QC a or b

Submitted by: Shyam Sundar Date: 02/04/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 14/04/2015

Plasma Concentrations of Uric Acid in Rat 378
 Based on 'SOP NO.: SOP/STD/2004-001-0' (With Stopping Solution)
 Experiment Date: 09/03/2015 - 13/03/2015

Conc. ug/mL	STD ID	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount (ng)	Recr Recovery (%)
Uric Acid 5 ng		32.60					5		
a25	25ug/ml (a)	54.58	7.52	7.26	7.26	2	2511.35	92.37	
b25	25ug/ml (b)	61.14	9.57	6.39	6.39	2	2813.19	104.44	
Mean		57.86	8.55	6.82	6.82		2662.27	98.40	
SD		4.64	1.45	0.61	0.61		213.43	8.54	
%CV		8.02	16.96	9.01	9.01		8.02	8.68	
N		2.00	2.00	2.00	2.00		2.00	2.00	
a5	5ug/ml (a)	9.40	9.97	0.94	0.94	2	432.52	46.06	
b5	5ug/ml (b)	11.72	10.09	1.16	1.16	2	539.26	67.41	
	5ug/ml	9.53	10.99	0.87	0.87	2	438.50	47.25	
	5ug/ml	8.98	10.81	0.83	0.83	2	413.19	42.19	
	5ug/ml	8.95	10.57	0.85	0.85	2	411.81	45.46	
	5ug/ml	10.80	10.18	1.06	1.06	2	496.93	62.48	
	5ug/ml	8.83	10.47	0.84	0.84	2	406.29	44.36	
Mean		9.74	10.44	0.94	0.94		448.36	50.74	
SD		1.10	0.38	0.13	0.13		50.52	9.93	
%CV		11.27	3.64	13.71	13.71		11.27	19.57	
N		7.00	7.00	7.00	7.00		7.00	7.00	
BLANKS:									
aB	0ug/ml (a)	4.78	10.66	0.45	0.45	2	219.94		
bB	0ug/ml (b)	4.01	8.64	0.46	0.46	2	184.51		
Mean		4.40	9.65	0.46	0.46		202.22		
SD		0.54	1.43	0.01	0.01		25.05		
%CV		12.39	14.80	2.44	2.44		12.39		
N		2.00	2.00	2.00	2.00		2.00		
Uric Acid 5 ng 09/03/2015		30.80	11.29				5.00		
Uric Acid 5 ng 10/03/2015		29.98	9.66				5.00		
Uric Acid 5 ng 11/03/2015		32.46	11.44				5.00		
Uric Acid 5 ng 12/03/2015		32.79	11.47				5.00		
Uric Acid 5 ng 13/03/2015		30.21	9.80				5.00		

Regression Analysis of Standard Curve Data

Conc. (ug/mL)	PHR Value (PHRV)	Blank (PHRb)	PHRV-PHRb
25.00	6.82	0.46	6.37
5.00	0.94	0.46	0.48
0.00	0.46	0.46	0.00

Regression Output Begins Here:

Regression Output:

Constant	-0.3779
Std Err of Y Est	0.6122
R Squared	0.9851
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	0.2660
Std Err of Coef.	0.0327

Sample ID	Time post-dos	Peak #	Peak Ht. (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	PHR Value	Inj Vol. (µL)	Hemolysis Degree	c.(µg/mL)	onc.(µM)	Conc.(µM) Corrected for dilution
T0 R378	0.00	14.01	18.98	0.74	0.74	0.74	5	-	4.20	24.96	33.28
T0.08 R378	0.08	13.65	16.88	0.81	0.81	0.81	5	-	4.46	26.53	35.38
T0.25 R378	0.25	9.55	22.82	0.42	0.42	0.42	5	-	2.99	17.81	23.74
T1 R378	1.00	7.06	25.07	0.28	0.28	0.28	5	-	2.48	14.75	19.66
Isoproterenol (30 mg/kg)											
T1.2 R378	1.20	16.51	22.85	0.72	0.72	0.72	5	-	4.14	30.39	40.52
T1.5 R378	1.50	42.38	18.75	2.26	2.26	2.26	5	-	9.92	72.87	97.15
T2 R378	2.00	56.56	20.78	2.72	2.72	2.72	5	-	11.65	69.32	92.42
T3 R378	3.00	12.71	24.19	0.53	0.53	0.53	5	-	3.40	20.20	26.93
T4 R378	4.00	8.27	22.00	0.38	0.38	0.38	5	-	2.83	20.82	27.76
T5 R378	5.00	16.86	19.31	0.87	0.87	0.87	5	-	4.70	27.98	37.30
T6 R378	6.00	7.17	20.84	0.34	0.34	0.34	5	-	2.71	16.14	21.52
Mean		21.55	21.29	1.06	1.06				5.40	34.60	46.14
SD		17.87	2.90	0.91	0.91				3.42	23.08	30.77
%CV		82.93	13.63	85.90	85.90				63.25	66.70	66.70
n		8.00	8.00	8.00	8.00				8.00	8.00	8.00

NOTE: QC samples were prepared with plasma from healthy rat No. 181
 The Spiking solutions were made on: 22/02/2006
 *Repeated injections of QC a or b

Submitted by: Shyam Sundar Date: 02/04/2015

Checked by: Date:

Approved by: Pollen Yeung Date: 15/04/2015

Title: Measurement of Plasma Concentrations of Dipyridamole in Rat 378

According to SOP No: SOP/STD/2008-001-1 (Plasma with no Stopping Solution)

Experiment Date: 17/10/2014 - 22/10/2014

Abs.amt ng	STD ID	Peak Ht. # (mm)	Peak Ht. I.S. (mm)	Peak Ht. Ratio	Peak Ht. Ratio Value	Inj Vol. (µL)	Amount Recov. (ng)	Recovery (%)
Dipyridamole (1ng)		83.42				1		
a1000	1 ug/mL(a)	95.77	3.15	30.40	30.40	5	45.92	91.84
b1000	1ug/mL(b)	96.62	4.87	19.84	19.84	5	46.33	92.66
1000*	1ug/mL(c)	99.96	8.04	12.43		5	47.93	95.86
Mean		97.45	5.35	20.89	25.12		46.73	93.45
SD		2.21	2.48	9.03	7.47		1.06	2.12
%CV		2.27	46.34	43.23	29.73		2.27	2.27
n		3.00	3.00	3.00	2.00		1.00	1.00
a100	0.1 ug/mL (a)	18.31	11.32	1.62	1.62	20	2.19	43.90
b100	0.1ug/mL (b)	31.43	15.12	2.08	2.08	20	3.77	75.35
Mean		24.87	13.22	1.85	1.85		2.98	59.63
SD		9.28	2.69	0.33	0.33		1.11	22.24
%CV		37.30	20.33	17.65	17.65		37.30	37.30
n		2.00	2.00	2.00	2.00		2.00	2.00
aB	0 ug/mL (a)	0.00	13.47	0.00	0.00	20	0.00	0.00
bB	0 ug/mL (b)	0.00	12.20	0.00	0.00	20	0.00	0.00
Mean		0.00	12.84	0.00	0.00		0.00	0.00
SD		0.00	0.90	0.00	0.00		0.00	0.00
%CV		ERR	7.00	ERR	ERR		ERR	ERR
n		2.00	2.00	2.00	2.00		2.00	2.00

Plasma Conc. (ug/mL)	Peak Ht.Ratio (PHR)	Blank (PHRb)	PHRV-PHRb
0.00	0.00	0.00	0.00
0.10	1.85	0.00	1.85
1.00	25.12	0.00	25.12

Regression Output Begins Here:

Regression Output:	
Constant	-0.3284
Std Err of Y Est	0.4522
R Squared	0.9994
No. of Observations	3.0000
Degrees of Freedom	1.0000
X Coefficient(s)	25.4134
Std Err of Coef.	0.6320

Sample ID	Time Post-dose (h)	Peak Ht. # (mm)	Peak Ht. I.S. (mm)*	Peak Ht. Ratic	PHR Value	Inj Vol. (µL)	Hemolysis Degree	Conc.(ug/mL)
R37870	0.00	11.38	14.25	0.80	0.80	20	-	0.52
R37870.08	0.08	22.78	22.48	1.01	1.01	20	-	0.52
R37870.25	0.25	71.59	21.83	3.28	3.28	20	-	1.68
R37871	1.00	52.46	12.52	4.19	4.19	20	-	2.10
Isoproterenol (30 mg/kg sc)						20		
R37871.2	1.20	44.83	17.19	2.61	2.61	20	-	0.86
R37871.5	1.50	32.64	30.15	1.08	1.08	20	-	0.66
R37872	2.00	22.02	20.50	1.07	1.07	20	-	0.65
R37873	3.00	18.11	20.79	0.87	0.87	20	-	0.56
R37874	4.00	17.46	21.29	0.82	0.82	20	-	0.53
R37875	5.00	10.26	19.02	0.54	0.54	20	-	0.40
R37876	6.00	9.22	13.15	0.70	0.70	20	-	0.48
Mean		28.44	19.38	1.54	1.54			0.82
SD		20.06	5.06	1.23	1.23			0.55
%CV		70.54	26.09	79.67	79.67			66.48
n		11.00	11.00	11.00	11.00			11.00

Peak Ht. = peak height
 Peak Ht. R. (or: PHR) = peak height ratio
 I.S. = internal standard
 Inj Vol = injection volume
 ND = not detected or determined
 NS = no sample
 Corr. PHR = (PHR - RGB PHR)

Dipyridamole (1ng)(03/10/2014)	70.72	8.04		1.00
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Comments: Plasma from Rat 156 was used for extraction QC's.

*A repeat injection of a or b

Rat died

Submitted by: Shyam Sunder

Date: 26/10/2014

Checked by: Pollen Yeung

Date: 05/11/2014

Approved by:

Date: