TITLE PAGE

Protocol Title: An Open-label, Dose-escalation Study to Evaluate the Safety, Tolerability, Pharmacokinetics and Pharmacodynamics of Single Doses of GSK2586881 in Participants with Pulmonary Arterial Hypertension

Protocol Number: 206246 / Amendment 4

Short Title: Study to Evaluate the Safety, Pharmacokinetics and Pharmacodynamics of GSK2586881 in Participants with Pulmonary Arterial Hypertension

Compound Number: GSK2586881

Sponsor Name and Legal Registered Address:

GlaxoSmithKline Research & Development Limited
980 Great West Road
Brentford
Middlesex, TW8 9GS
UK

Medical Monitor Name and Contact Information can be found in the Study Reference Manual

Regulatory Agency Identifying Number(s): EudraCT 2017-000212-41, IND 132281

Approval Date: 10-JUL-2018

SPONSOR SIGNATORY:

PPD

10th July 2018

Tony Cahn, MD
Director, Discovery Medicine
Project Physician Leader, Respiratory Therapy Area

Date

PPD

PROTOCOL AMENDMENT SUMMARY OF CHANGES TABLE

DOCUMENT HISTORY		
Document	**Date**	**DNG Number**
Amendment 4	*10-Jul-2018*	*2016N290015_04*
Amendment 3	*01-Dec-2017*	*2016N290015_03*
Amendment 2	*07-Nov-2017*	*2016N290015_02*
Amendment 1	*03-May-2017*	*2016N290015_01*
Original Protocol	*04-Apr-2017*	*2016N290015_00*

Amendment 4 10-JUL-2018

Overall Rationale for the Amendment:

This is a substantial amendment with changes to the inclusion and exclusion criteria to more accurately reflect the patient population.

Section # and Name	Description of Change ***Bolded** text indicates newly added text, and ~~strikethrough~~ indicates deleted text.	Brief Rationale
6.1 Inclusion Criteria #3	The following change was made: Idiopathic pulmonary arterial hypertension (IPAH), hereditary pulmonary arterial hypertension (HPAH), or PAH associated with collagen vascular disease~~, repaired congenital heart disease,~~ or appetite suppressant use.	PAH associated with repaired congenital heart disease was removed as this differs clinically from other forms of PAH.
6.1 Inclusion Criteria #5	The following change was made: Hemodynamically stable on background therapy with no evidence of **uncontrolled** right heart failure (historic data), **as determined by the investigator.**	This change clarifies that participants with uncontrolled right heart failure must be excluded. However, participants with controlled right heart failure can be included as determined by the investigator.
6.1 Inclusion Criteria #6 and 9.4 Six-minute walk test	The following change was made: Six minute walk (6MW) distance, as performed at screening or within 6 months prior to screening, of **≥**100 meters and ~~≤ 450~~ **≤ 500** meters.	Six-minute walk maximum distance was increased to 500 meters to allow participants to be included that meet all other criteria.
6.1 Inclusion Criteria #10	The following change was made: Body weight ~~≤90~~ **≤ 100** kg and body mass index (BMI) within the range 18~~-30~~ **35** kg/m^2 (inclusive).	Maximum BMI was increased to 35 to more accurately reflect the range observed in the patient population.
6.2 Exclusion criteria #4	The following change was made: Complex **repaired and** unrepaired congenital heart disease.	This change clarifies that both complex repaired and unrepaired congenital heart disease will be excluded from the study.
6.2 Exclusion criteria #9	The following change was made: Estimated glomerular-filtration-rate (eGFR) < ~~60~~ **45** mL/min/1.73m^2.	eGFR rate was reduced to 45ml/min/1.73m^2 to more accurately reflect the range observed in the patient population.
12.2 Appendix 2: Clinical Laboratory Tests	The following change was made: **Activated** Prothrombin time (PT) **a**PTT	Q2 confirmed the assay they use is activated prothrombin time (aPTT)

Section # and Name	Description of Change ***Bolded** text indicates newly added text, and ~~strikethrough~~ indicates deleted text.	Brief Rationale
12.2 Appendix 2: Clinical Laboratory Tests	The following change was made: ~~Local laboratory results are only required in the event that the central laboratory results are not available in time for either study treatment administration and/or response evaluation.~~ If a local sample is required, it is important that the sample for central analysis is obtained at the same time. ~~Additionally, if the local laboratory results are used to make either a study treatment decision or response evaluation, the results must be entered into the CRF.~~	This change allows sites to use local labs, if necessary; however, it ensures samples are always collected and analysed centrally.
12.5 Appendix 5: Contraceptive Guidance and Collection of Pregnancy Information	Pregnancy testing, with a high sensitivity test will be performed using the test kit ~~provided by the central laboratory /and approved by the sponsor and~~ in accordance with instructions provided in its package insert.	This change gives flexibility for sites to use local labs for pregnancy testing.

TABLE OF CONTENTS

PAGE

1. SYNOPSIS

Protocol Title: An Open-label, Dose-escalation Study to Evaluate the Safety, Tolerability, Pharmacokinetics and Pharmacodynamics of Single Doses of GSK2586881 in Participants with Pulmonary Arterial Hypertension

Short Title: Study to Evaluate the Safety, Pharmacokinetics and Pharmacodynamics of GSK2586881 in Participants with Pulmonary Arterial Hypertension

Rationale:

GSK2586881 is a purified intravenous (IV) formulation of soluble recombinant human Angiotensin Converting Enzyme (rhACE2) that is being investigated as a potential treatment for Pulmonary Arterial Hypertension (PAH).

Angiotensin converting enzyme type 2 (ACE2) is a type I, membrane-bound carboxypeptidase that inactivates Angiotensin II (Ang II) by cleaving it to produce Ang(1-7). Ang II is a key effector peptide of the Renin-Angiotensin-System (RAS) that can exert deleterious effects on the pulmonary vasculature resulting in vasoconstriction, proliferation and inflammation, all of which contribute to PAH development. Conversely Ang(1-7) has shown vasodilatory, anti-proliferative and anti-inflammatory properties by signalling through the Mas receptor and recombinant ACE2 has proven effective in blocking or treating established PAH in several rodent models.

ACE2 hydrolyses a number of other biological peptides, including converting Ang I to Ang(1-9) and removing the C-terminal residue from the bradykinin metabolites [des-Arg^9]-bradykinin and lys[des-Arg^9]-bradykinin .

GSK2586881 has been tested in healthy subjects and mechanically ventilated patients with Acute Respiratory Distress Syndrome (ARDS) and has been shown to be well tolerated with the rapid modulation of RAS peptides. This will be the first GlaxoSmithKline (GSK) study to test the safety and preliminary effects on pulmonary hemodynamics of GSK2586881 in participants with PAH.

A detailed description of the biology, pharmacology, efficacy, and safety of GSK2586881 is provided in the Investigator's Brochure (IB).

Objectives and Endpoints:

Objectives	Endpoints
Primary	
• To evaluate changes in the pulmonary hemodynamics after single IV doses of GSK2586881 administered to participants with PAH receiving background PAH therapy.	• Change from baseline in pulmonary vascular resistance (PVR), cardiac output (CO) and mean pulmonary artery pressure (mPAP), as data permit

Objectives	Endpoints
Secondary	
• To evaluate the safety and tolerability of single IV doses of GSK2586881 administered to participants with PAH receiving background PAH therapy.	• Adverse events (AE), clinical laboratory values, vital signs, electrocardiogram (ECG), pulse oximetry and immunogenicity.
• To evaluate the effect of single IV doses of GSK2586881 on RAS peptide responses in participants with PAH receiving background PAH therapy.	• Change from baseline of pulmonary wedge and systemic RAS peptides (e.g. Ang II, Ang(1-7), Ang(1-5), and AngII/Ang(1-7) ratio).
• To evaluate the effect on biomarkers of disease activity after single IV doses of GSK2586881 administered to participants with PAH receiving background PAH therapy.	• Change from baseline in NT pro-BNP, NO and cardiac troponin I.
• To evaluate the pharmacokinetics (PK) of GSK2586881 after single IV doses of GSK2586881 in participants with PAH receiving background PAH therapy.	• Plasma concentrations of GSK2586881 and derived PK parameters.

Overall Design:

This is a Phase IIa, open-label, dose-escalation, single dose study in participants with PAH (as defined in the eligibility criteria) whose symptoms have been clinically stable for 8 weeks prior to enrolment and who have had no change in PAH-specific therapy in the 12 weeks prior to enrolment.

Number of Participants:

The total number of participants recruited will be determined through an ongoing review of the safety, tolerability, PK and PD data with a minimum of 4 participants recruited per cohort. There will be 4 cohorts. At the end of each cohort a dose escalation meeting will occur and a decision will be made whether to dose escalate.

A maximum of 27 participants will complete dosing and critical assessments in this study.

If participants prematurely discontinue the study, additional replacement participants may be recruited and assigned to the same dose level at the discretion of the Sponsor in consultation with the investigator.

Treatment Groups and Duration:

The study will comprise of 4 separate cohorts. Participants will be administered a single dose at the following planned doses: 0.1, 0.2, 0.4, or 0.8mg/kg.

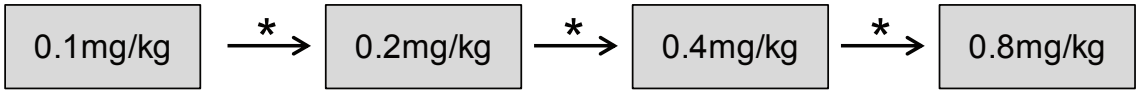
Dose escalation will occur after a minimum of 4 participants have been dosed per cohort and review of safety, tolerability, PK and hemodynamic data up to 24h post dose has taken place. Dose escalation will continue up to a maximum dose of 0.8mg/kg.

Cohort sizes may vary with a minimum of 4 participants per cohort. Additional participants may be enrolled to expand a dosing cohort to further evaluate findings at a given dose level.

It is anticipated that the total duration of participation in the study will be up to a maximum of 59 days from screening to the last study visit.

Study Design

Single Dose
(minimum 4 participants per cohort)



* dose escalation meeting (review of safety, PK and hemodynamic data)

2. SCHEDULE OF ACTIVITIES (SOA)

Procedure	Screening (up to 28 days before dosing)	Treatment Period									Follow-up Day 7-14	Follow-up Day 28 ± 3 days	Notes
		Pre-dose[1]	0h	0.08h (5 min)[2]	0.5h	1h	2h	4h	8h	24h			
Informed consent	X												
Genetics consent	X												Genetics consent is Optional
Inclusion and exclusion criteria	X												Recheck clinical status before randomization and/or 1st dose of study medication
Demography	X												
Full physical examination including height and weight	X												
Medical history (includes substance usage and family history of premature CV disease)	X												Substances: drugs, alcohol, tobacco and caffeine
Past and current medical conditions	X												

Procedure	Screening (up to 28 days before dosing)	Treatment Period									Follow-up Day 7-14	Follow-up Day 28 ± 3 days	Notes
		Pre-dose[1]	0h	0.08h (5 min)[2]	0.5h	1h	2h	4h	8h	24h			
Serum OR urine pregnancy test (WOCBP only)	X	X									X		
FSH and estradiol test (postmenopausal females only as needed)	X												
Human Immunodeficiency Virus (HIV), Hepatitis B and C screening	X												If test performed within 3 months prior to first dose of study treatment, testing at screening is not required
Functional classification	X												
6 Minute Walk Distance	X[3]												
Admission		X											Participants may be admitted the day before dosing to enable completion of required pre-dose time point assessments.
Brief physical		X									X		
Study Treatment			X										

Procedure	Screening (up to 28 days before dosing)	Treatment Period									Follow-up Day 7-14	Follow-up Day 28 ± 3 days	Notes
		Pre-dose[1]	0h	0.08h (5 min)[2]	0.5h	1h	2h	4h	8h	24h			
Vital Signs	X	X			X	X	X	X	X	X	X		Vital signs will be measured after 5 minutes supine at all time points. Triplicate blood pressure (BP) will be taken at screening only.
Pulse Oximetry (SpO_2)	X	X			X	X	X	X	X	X	X		Pulse oximetry will be measured and recorded with each blood pressure assessment
Laboratory assessments (include liver chemistries, haematology panel and coagulation panel)	X	X								X	X		
Urinalysis	X									X			
12-lead ECG	X	X						X		X	X		Triplicate will be performed at Screening and Pre-dose

Procedure	Screening (up to 28 days before dosing)	Treatment Period									Follow-up Day 7-14	Follow-up Day 28 ± 3 days	Notes
		Pre-dose[1]	0h	0.08h (5 min)[2]	0.5h	1h	2h	4h	8h	24h			
Telemetry		←----------								----------→			Monitoring to start 30min prior to treatment administration and continue throughout the study until 24h after dosing
Right heart catheter Insertion		←----------								----------→			RHC inserted prior to dose and removed after the 4h hemodynamic measurement
Blood sample for biomarkers of disease activity		X					X	X		X			
Blood sample for Nitric Oxide		X					X	X		X			
Blood sample for PK		X		X	X	X	X	X	X	X			
Blood sample for immunogenicity		X									X	X	Additional visits may be required. (see Section 9.7 for details).

Procedure	Screening (up to 28 days before dosing)	Treatment Period									Follow-up Day 7-14	Follow-up Day 28 ± 3 days	Notes
		Pre-dose[1]	0h	0.08h (5 min)[2]	0.5h	1h	2h	4h	8h	24h			
Blood sample for renin-angiotensin system biomarkers		X		X	X	X	X	X	X	X	X		
Blood sample for pulmonary wedge RAS biomarkers		X				X	X	X					
Hemodynamic measurements		X				X	X	X					
Genetic sample		X											Can be taken any time after consent has been signed. Only required once and is optional.
Discharge										X			
AE review		←==========→									X		
SAE review	X	←==========→									X		
Concomitant medication review[4]	X	X									X		

1. Pre-dose measurements may be taken any time after admission up until dosing.
2. Procedures will be completed immediately after dosing has completed.

3. If the six minute walk(6MW) has been performed in the last 6 months, and participant has been stable on current medications then there is no need to repeat. Historical data will be databased.
4. Concomitant medications for the 30 days prior (8 weeks prior for PAH medications) will be reviewed/recorded at screening to evaluate eligibility and changes will be recorded throughout the study.

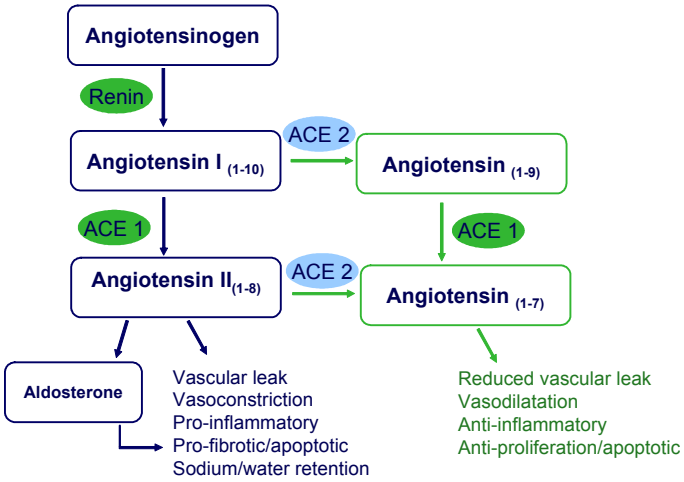
3. INTRODUCTION

While considerable advances have been made in treatment, PAH continues to be a serious, life shortening disease. PAH is characterised by impedance to blood flow through the pulmonary vasculature that occurs as a consequence of enhanced pulmonary vasoconstriction, small pulmonary arteriole remodelling, and loss of vessel compliance/distensibility due to perivascular fibrosis [Maron, 2013; Crosswhite, 2014]. Severe pulmonary hypertension leads to right ventricular overload and right-sided heart failure.

GSK2586881 is a purified intravenous formulation of soluble rhACE2 that is being investigated clinically as a potential treatment for PAH. ACE2 is a type I, membrane-bound carboxypeptidase that inactivates Ang II by cleaving it to produce Ang (1-7). Ang II is a key effector peptide of the RAS that can exert deleterious effects on the pulmonary vasculature resulting in vasoconstriction, inflammation, proliferation and structural remodelling, all of which contribute to PAH development (Lipworth, 1994, Cheng, 2005, Schiffrin, 2003). Conversely Ang (1-7) signals through the Mas receptor and has shown vasodilatory, anti-inflammatory and anti-proliferative properties in experimental models (Clarke, 2011). Vasodilation induced by Ang (1-7) has been attributed to the activation of endothelial nitric oxide synthase (eNOS) and the release of nitric oxide (NO) (Sampaio, 2007). The mechanism of action of ACE2 and the downstream consequences are thus distinct from ACE inhibitors and angiotensin receptor blockers (Figure 1).

Systemic and pulmonary Ang II levels have been reported to be increased in patients with idiopathic (I)PAH and are associated with increased pulmonary vascular remodelling (De Man, 2012) while serum ACE2 levels are reported to be decreased in patients with congenital heart disease-associated PAH, and correlate with mPAP (Dai, 2013). The presence of autoantibodies to ACE2 have also been reported in the serum of patients with autoimmune disease with constrictive vasculopathies, PAH or persistent digital ischemia and correlate with reduced ACE2 activity (Takahashi, 2010). Taken together these studies suggest ACE2 activity may be impaired in PAH patients and administration of exogenous ACE2 may be beneficial. This is supported in rodent models of PAH where recombinant ACE2 has proven effective in blocking or treating established PAH in several rodent models, including in BMPR2-mutation related PAH and pulmonary artery banding (Johnson, 2011; Johnson, 2012).

Figure 1 Renin-Angiotensin System



3.1. Study Rationale

GSK2586881 is a purified intravenous formulation of soluble rhACE2 that is being investigated as a potential treatment for PAH. GSK2586881 has been tested in healthy subjects (Haschke, 2013) and mechanically ventilated patients with Acute Respiratory Distress Syndrome (ARDS) (GSK study ACE114622; GSK Document Number 2014N217963_00) and has been shown to be well tolerated with the rapid modulation of RAS peptides. This will be the first GSK study to test the safety and preliminary hemodynamic effects of GSK2586881 in participants with PAH.

3.2. Background

PAH is defined as mPAP >25 mmHg at rest and a mean pulmonary wedge pressure of ≤15 mmHg (Galie, 2009). Patients may present with shortness of breath, swelling, fatigue, chest pain, and in advanced cases syncope. Women of childbearing age are most commonly affected, although PAH has been reported in children and older adults. Untreated PAH eventually leads to right sided heart failure and death. PAH is a progressive disease, and mortality and morbidity remain high in spite of recent improvements in therapy.

The development of PAH is associated with connective tissue disease, liver disease, HIV infection, use of appetite suppressants, congenital systemic-to-pulmonary shunts (e.g., atrial septal defect), and portal hypertension. PAH may also occur with no known cause, in which case it is considered primary or idiopathic PAH (IPAH), and as an inherited disease, referred to as heritable PAH (HPAH). IPAH is a devastating disease which if untreated has a median life expectancy of 2.8 years (D'Alonzo, 1991). Outcome for PAH associated with other disorders such as scleroderma is even worse.

At the histologic level, all forms of PAH are characterized by marked structural remodeling of small pulmonary arteries (<100µ in diameter); specifically, vascular smooth muscle cell proliferation (medial hyperplasia), intimal fibrosis and hyperplasia, and in situ microthrombosis. In advanced disease, plexiform lesions, likely a disordered attempt at neovascularization, have been identified. These changes in the small

pulmonary arteries lead to narrowing and obliteration of the vessel lumen, increased resistance to blood flow, and increased strain on the right ventricle. The molecular mechanisms underlying this vasculopathy remain poorly understood.

Management strategies for PAH include prevention of microthrombosis using anticoagulants such as warfarin and promoting vasodilation and regression of muscular hypertrophy and intimal fibrosis using endothelin receptor antagonists and phosphodiesterase (PDE) 5 inhibitors (Humbert, 2004). In severe disease or in cases of treatment failure with oral therapy, prostanoids (inhaled, subcutaneous, or intravenous) are used. In selected patients who are not responsive to medical therapy, lung transplant is an option. Despite improvement in medical therapy, only two-thirds of patients are alive 3 years after treatment with the most effective therapy available for PAH, continuous intravenous epoprostenol (Sitbon, 2002). The current therapeutic options remain unsatisfactory, and there remains an urgent need for novel therapies.

A detailed description of the biology, pharmacology, efficacy, and safety of GSK2586881 is provided in the IB (GSK Document Number 2010N108777_03 and GSK Document Number 2016N300457_00)

3.3. Benefit/Risk Assessment

More detailed information about the known and expected benefits and risks and reasonably expected AEs of GSK2586881 may be found in the IB (GSK Document Number 2010N108777_03).

3.3.1. Risk Assessment

Potential Risk of Clinical Significance	Summary of Data/Rationale for Risk	Mitigation Strategy
Investigational Product (IP) GSK2586881		
Unknown potential for adverse hemodynamic effects in patients with PAH	Pre-Clinical: Pharmacology studies in rats showed additive reduction in BP when GSK2586881 at single daily dose of 1 mg/kg was given with an angiotensin type 1 receptor blocker or an ACE inhibitor. Clinical: No adverse hemodynamic or BP effects noted in the first time in humans (FTIH) healthy volunteer study (APN01-1-01). No acute adverse hemodynamic effects related to study drug administration or considered significantly different from those expected of patients with ARDS in study ACE114622.	Participants will be monitored including frequent vital signs. Participants will be excluded if concomitant therapy includes the use of an ACE inhibitor or angiotensin receptor blocker within 14 days prior to dosing.
Potential Cardiovascular(CV) Effects	Pre-Clinical: In the cynomolgus monkey study, twice daily administration of 10.4 mg/kg (20.8mg/kg/day, highest dose given) for 12 consecutive days resulted in a waveform abnormality in a single female (n=12) which was 6 premature ventricular complexes(PVCs) including an episode of non-sustained ventricular tachycardia (NSVT) consisting of 3 consecutive PVCs (triplet). Timing of the NSVT was 2 to 3 hours following the first daily dose while subsequent PVCs occurred at a	Participants will be monitored including telemetry

Potential Risk of Clinical Significance	Summary of Data/Rationale for Risk	Mitigation Strategy
	similar time point after the second daily dose. There was no evidence of QT prolongation or anatomical changes in the heart. Clinical: No adverse effects on ECG were seen in the FTIH study (APN01-1-01). In study ACE114622 in ARDS, no clinically significant effects on ECG that were considered related to study drug were observed.	
Potential for Immunogenicity	Clinical: There was no induction of an immune response to rhACE2 in any of the treated subjects up to 28 days after last administration in the FTIH healthy volunteer study (APN01-1-01) or in ARDS patients (ACE114622).	Participants will have routine monitoring of any immunological response that may occur.
Unknown risk of teratogenicity	Pre-Clinical: Reproductive toxicology studies have not yet been performed with GSK2586881. A review of the literature suggests that ACE2 is present in organs of fetal mice late in gestation, and in the placentae of rats, guinea-pigs and humans. Some of the changes noted in the PD and toxicity studies with ACE2 are similar to those reported with an ACE inhibitor. In addition, the older ACE inhibitors are known to be teratogenic in humans.	Pregnancy testing at screening and prior to dosing. Information on contraception post study will be provided in the informed consent form (ICF).

Potential Risk of Clinical Significance	Summary of Data/Rationale for Risk	Mitigation Strategy
Potential for Rash	Clinical: In study ACE114622, rash was reported more frequently in subjects receiving GSK2586881, although only one event was considered drug-related.	Participants will be monitored for rash in the clinical trials.
Study Procedures		
Pulmonary arterial catheterization (PAC)	More common risks of PAC placement include: bruising at the site of the PAC insertion, excessive bleeding, vein injury or tear or pneumothorax. Less common complications include: thrombosis, infection, hypotension, arrhythmia, cardiac tamponade or pulmonary artery rupture.	Insertion of catheter performed by experienced physicians, continuous monitoring of participants during procedure per site standard operating procedures (SOPs).
Exposure to ionzing radiation	Catheter placement may be confirmed by chest X-ray or fluoroscopy, as clinically indicated. Both chest X-ray and fluoroscopy are assumed to increase the risk of cancer or heritable effects due to radiation exposure. Radiation exposure from chest X-ray is minimal (0.02mSv). Radiation exposure from fluoroscopy used to confirm placement of the catheter is dependent on the equipment and duration of the procedure. The dose will typically be around 2mSv but may range from 2-5mSv. The additional risk of developing a fatal malignancy (cancer) as a result of these exposures has been estimated as between 1 in 10,000 and 1 in 2000 for an adult in normal health.	Negative pregnancy testing prior to procedure. Minimize fluoroscopy time; lead shielding per site SOPs. Maximum exposure will not exceed 10mSv to confirm placement of the catheter.

3.3.2. Benefit Assessment

- All participants will undergo a thorough medical assessment during the study, including ECG, vital signs monitoring, and physical examinations. Monitoring for worsening of their disease will also take place.

- Participants may benefit from the knowledge that they are contributing to the process of developing a new treatment in an area of unmet need, even if not directly beneficial for them.

3.3.3. Overall Benefit:Risk Conclusion

In clinical trials completed to date GSK2586881 has been well-tolerated and most AEs were mild to moderate in intensity. The most commonly observed AE in critically ill patients has been hypokalemia that occurred with equal frequency following administration of GSK2586881 and placebo. Hypernatremia, rash, dysphagia and pneumonia occurred more frequently in subjects receiving GSK2586881 in study ACE114622 (GSK Document Number 2014N217963_00). There have been no treatment related clinically significant changes in vital signs or ECG at any dose of GSK2586881.

Taking into account the measures taken to minimize risks to participants in this study, the potential risks identified in association with GSK2586881 are justified by the anticipated benefits that may be afforded to participants with PAH.

4. OBJECTIVES AND ENDPOINTS

Objectives	Endpoints
Primary	
• To evaluate changes in the pulmonary hemodynamics after single IV doses of GSK2586881 administered to participants with PAH receiving background PAH therapy.	• Change from baseline in pulmonary vascular resistance (PVR), cardiac output (CO) and mean pulmonary artery pressure (mPAP), as data permit
Secondary	
• To evaluate the safety and tolerability of single IV doses of GSK2586881 administered to participants with PAH receiving background PAH therapy.	• Adverse events (AE), clinical laboratory values, vital signs, ECG, pulse oximetry and immunogenicity.
• To evaluate the effect of single IV doses of GSK2586881 on RAS peptide responses in participants with PAH receiving background PAH therapy.	• Change from baseline of pulmonary wedge and systemic RAS peptides (e.g. Ang II, Ang(1-7), Ang(1-5), and AngII/Ang (1-7) ratio).

Objectives	Endpoints
• To evaluate the effect on biomarkers of disease activity after single IV doses of GSK2586881 administered to participants with PAH receiving background PAH therapy.	• Change from baseline in NT pro-BNP, NO and cardiac troponin I.
• To evaluate the pharmacokinetics of GSK2586881 after single IV doses of GSK2586881 in participants with PAH receiving background PAH therapy.	• Plasma concentrations of GSK2586881 and derived PK parameters.
Exploratory	
• To evaluate PK/PD relationships after single IV doses of GSK2586881.	• Pulmonary wedge RAS peptides, and/or pulmonary vascular hemodynamic measurements compared with PK exposure.
• To evaluate pharmacogenetics(PGx).	• Evaluate I/D polymorphisms in the Angiotensin Converting Enzyme (ACE) gene and analyze the impact on Ang II (and possibly other RAS peptides), and responses to GSK2586881 administration.

5. STUDY DESIGN

5.1. Overall Design

This is a Phase IIa, open-label, dose-escalation study in participants with PAH (as defined in the eligibility criteria) whose symptoms have been clinically stable for 8 weeks prior to enrolment and who have had no change in PAH-specific therapy in the 12 weeks prior to enrolment. Eligible participants will undergo baseline assessments before receiving a single dose of GSK2586881.

It is anticipated that the total duration of participation in the study will be up to a maximum of 59 days from screening to last study visit.

5.2. Number of Participants

The total number of participants recruited will be determined through an ongoing review of the safety, tolerability, PK and PD data with a minimum of 4 participants recruited per cohort. There will be 4 cohorts. At the end of each cohort a dose escalation meeting will occur and a decision will be made whether to dose escalate.

Once dose escalation is complete additional participants may be recruited to further evaluate findings at a given dose level. A maximum of 27 participants will complete dosing and critical assessments in this study.

Evaluable participants are defined in Section 10.2.

If participants prematurely discontinue the study, additional replacement participants may be recruited and assigned to the same dose level at the discretion of the Sponsor in consultation with the investigator.

5.3. Participant and Study Completion

A participant is considered to have completed the study if he/she has completed all phases of the study including the follow up visit and the last scheduled procedure shown in the Schedule of Activities.

The end of the study is defined as the date of the last visit of the last participant in the study.

5.4. Scientific Rationale for Study Design

The lack of a placebo arm is justified in that the primary goals of this study are to understand the initial hemodynamic effects, safety and tolerability of GSK2586881 in a small number of participants with PAH.

Due to the open label design it is accepted that there may be potential bias in AE reporting, this will be taken into account when interpreting the study results.

5.5. Dose Justification

GSK2586881 has been studied in 21 healthy subjects as single, intravenous doses over 30 minutes from 0.1 mg/kg to 1.2 mg/kg, and in repeated daily doses for up to six days of 0.4 mg/kg once a day. In addition, ARDS patients have received four doses of GSK2586881: 0.1 mg/kg, 0.2 mg.kg, 0.4 mg/kg and 0.8 mg/kg over a 24 hour period (n=5) or 0.4 mg/kg BID for 3 days (n=19).

A preliminary population PK model for GSK2586881 has been derived from data obtained in healthy subjects and ARDS patients and showed that the systemic PK profile was adequately described by a two-compartment first order elimination model and that the PK profile was independent of population (healthy subjects or ARDS patients). Decreases in Ang II levels occurred rapidly after GSK2586881 administration with Ang II concentrations maximally or near maximally reduced at the first sampling time of 5 minutes post-dose; consistent with maximal plasma concentrations of GSK2586881. The PK/PD response (the relationship between ACE2 and Ang II) in healthy subjects and ARDS patients appeared to be best described with a single direct Emax model after accounting for differences in baseline Ang II concentrations between healthy subjects and ARDS patients. The observation of no significant delay between GSK2586881 concentration and effect is in line with its mechanism of action as an enzyme on a substrate.

Based on the preliminary population PK/PD model described above, it is predicted that following a single dose of ≥0.2 mg/kg, Ang II concentrations will on average be reduced below participant baseline levels for at least 24 hours post-dose (assuming Ang II levels in PAH participants is consistent with levels reported in ARDS participants in GSK study ACE114622 (GSK Document Number 2014N217963_00). The 0.8mg/kg dose has been selected to investigate the safety and tolerability dose range for GSK2586881 in this patient population.

In an investigator-sponsored study (NCT01884051) GSK2586881 has been studied in 3 PAH patients as a single, intravenous dose at 0.2 mg/kg, and in 2 patients at 0.4mg/kg. There were no SAEs and no dose limiting toxicities were observed.

6. STUDY POPULATION

Prospective approval of protocol deviations to recruitment and enrollment criteria, also known as protocol waivers or exemptions, is not permitted.

6.1. Inclusion Criteria

Participants are eligible to be included in the study only if all of the following criteria apply:

Age

1. Participant must be between 18-75 years of age (inclusive), at the time of signing the informed consent.

Type of Participant and Disease Characteristics

2. Documented diagnosis of PAH, defined as mPAP > 25 mmHg and PWP ≤ 15 mmHg.
3. Idiopathic pulmonary arterial hypertension (IPAH), hereditary pulmonary arterial hypertension (HPAH), or PAH associated with collagen vascular disease, or appetite suppressant use.
    - Note: Those with portopulmonary hypertension or pulmonary veno-occlusive diseas (PVOD) are not eligible for the study
4. World Health Organization (WHO) functional class I, II, or III, stable for at least 8 weeks prior to enrollment.
5. Hemodynamically stable on background therapy with no evidence of uncontrolled right heart failure (historic data), as determined by the investigator.
6. Six minute walk (6MW) distance, as performed at screening or within 6 months prior to screening, of ≥100 meters and ≤ 500 meters.
7. Mean BP of >60 mmHg
8. Receiving stable doses of one or more medications that are approved for treatment of PAH, including endothelin receptor antagonists, phosphodiesterase 5 inhibitors,

and/or prostanoids/prostacyclin receptor agonists, for a minimum of 12 consecutive weeks before enrollment.

NOTE: Anticoagulant therapy can be adjusted according to target INR

9. Diuretic dose stable for 8 weeks.

Weight

10. Body weight ≤100 kg and body mass index (BMI) within the range 18-35 kg/m^2 (inclusive).

Sex

11. Male and/or female (following confirmation of negative pregnancy test for WOCBP). Women who are pregnant or breastfeeding are excluded.

Informed Consent

12. Capable of giving signed informed consent as described in Appendix 3 which includes compliance with the requirements and restrictions listed in the ICF and in this protocol.

6.2. Exclusion Criteria

Participants are excluded from the study if any of the following criteria apply:

Medical Conditions

1. History of systemic hypotension, defined as systolic BP <90 mmHg and/or diastolic BP <50 mmHg.
2. Hospitalization for PAH associated deterioration in the previous 6 months.
3. Any additional medical condition, serious intercurrent illness, or other extenuating circumstance that, in the opinion of the Investigator, may significantly interfere with study compliance, including all prescribed evaluations and follow-up activities. Concurrent disease or condition that may interfere with study participation or safety include bleeding disorders, arrhythmia, organ transplant, organ failure, current neoplasm, poorly controlled diabetes mellitus, and serious neurological disorders.
4. Complex repaired and unrepaired congenital heart disease.
5. Subjects with Eisenmenger physiology.
6. Alanine transferase (ALT) >2x upper limit of normal (ULN).
7. Bilirubin >1.5xULN (isolated bilirubin >1.5xULN is acceptable if bilirubin is fractionated and direct bilirubin <35%).
8. Current or chronic history of liver disease, or known hepatic or biliary abnormalities (with the exception of Gilbert's syndrome or asymptomatic gallstones).
9. Estimated glomerular-filtration-rate (eGFR) <45mL/min/1.73m^2.
10. QTc >480 msec or QTc > 500 msec in participants with bundle branch block.

NOTES:

- The QTc is the QT interval corrected for heart rate according to Bazett's formula (QTcB), Fridericia's formula (QTcF), and/or another method. It is either machine-read or manually over-read.

  The specific formula used to determine eligibility and discontinuation for an individual participant should be determined prior to initiation of the study. In other words, several different formulas cannot be used to calculate the QTc for an individual participant and then the lowest QTc value used to include or discontinue the participant from the trial.

11. Any bleeding concerns as evidenced by INR >1.5 (in participants not receiving anticoagulation therapy) or platelet count <80,000.
12. Hb <10 g/dL

Prior/Concomitant Therapy

13. Sensitivity to any of the study treatments, or components thereof, or drug or other allergy that, in the opinion of the investigator or Medical Monitor, contraindicates participation in the study.
14. Any use of an ACE inhibitor, angiotensin receptor blocker, or renin inhibitors within 14 days prior to dosing. Therapy can be stopped to enable inclusion if deemed safe by the participant's treating physician.

Prior/Concurrent Clinical Study Experience

15. Use of any investigational product (IP) or device within 30 days prior to dosing, or known requirement for any investigational agent prior to completion of all scheduled study assessments.

Diagnostic assessments

16. Positive HIV antibody test.
17. Presence of Hepatitis B surface antigen (HBsAg) at screening.
18. Positive Hepatitis C antibody test result at screening or within 3 months prior to starting study treatment.

    - NOTE: Participants with positive Hepatitis C antibody due to prior resolved disease can be enrolled, only if a confirmatory negative Hepatitis C ribonucleic acid (RNA) test is obtained.

19. Positive Hepatitis C RNA test result at screening or within 3 months prior to first dose of study treatment.

    - NOTE: Test is optional and participants with negative Hepatitis C antibody test are not required to also undergo Hepatitis C RNA testing.

Other Exclusions

20. Participation in the study would result in loss of blood or blood products in excess of 300mL within 65 days.

21. Exposure to more than 4 new chemical entities within 12 months prior to the first dosing day.
22. A known or suspected history of alcohol or drug abuse within the 2 years prior to screening
23. Unable to refrain from smoking during the in-house treatment period.

6.3. Lifestyle Restrictions

6.3.1. Caffeine, Alcohol, and Tobacco

- Participants will abstain from ingesting caffeine- or xanthine-containing products (eg, coffee, tea, cola drinks, and chocolate) for 2 hours before the start of dosing.
- Participants will abstain from alcohol for 24 hours before the start of dosing until after collection of the final PK and/or PD sample.
- Participants who use tobacco products will be instructed that use of nicotine-containing products (including nicotine patches) will not be permitted while they are in the clinical unit.

6.3.2. Activity

- Participants will abstain from strenuous exercise for 24 hours before each blood collection for clinical laboratory tests. Participants may participate in light recreational activities during studies (e.g., watching television, reading).

6.3.3. Contraception Requirements

Specific requirements for contraception can be found in Appendix 5 and will be provided in the informed consent form (ICF).

6.4. Screen Failures

Screen failures are defined as participants who consent to participate in the clinical study but are not subsequently enrolled in the study. A minimal set of screen failure information is required to ensure transparent reporting of screen failure participants to meet the Consolidated Standards of Reporting Trials (CONSORT) publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, screen failure details, eligibility criteria, and any serious adverse events (SAEs).

Individuals who do not meet the criteria for participation in this study (screen failure) may be rescreened once. Rescreened participants should be assigned a new participant number.

7. TREATMENTS

Study treatment is defined as any investigational treatment(s), marketed product(s), placebo, or medical device(s) intended to be administered to a study participant according to the study protocol.

7.1. Treatments Administered

Study Treatment Name:	GSK2586881
Dosage formulation:	Clear, colorless liquid
Unit dose strength(s)/Dosage level(s):	Unit dose strength: 5 mg/mL Up to 0.8mg/kg
Route of Administration	IV
Dosing instructions:	IV push over 3-5 minutes
Packaging and Labeling	Study Treatment will be provided in frozen vials. Each vial will be labeled as required per country requirement.
Manufacturer	Polymun/GSK

7.2. Dose Modification

The study will comprise of 4 separate cohorts. Participants will be administered a single dose of one of the following planned doses: 0.1, 0.2, 0.4, or 0.8mg/kg.

Dose escalation will occur after a minimum of 4 participants have been dosed per cohort and review of safety, tolerability, PK and hemodynamic data up to 24h post dose has taken place. Dose escalation will continue up to a maximum dose of 0.8mg/kg.

Cohort sizes may vary with a minimum of 4 participants per cohort. Additional participants may be enrolled to expand a dosing cohort to further evaluate findings at a given dose level.

If a subject experiences a dose limiting toxicity (DLT) that is considered at least possibly related to study drug, dosing of additional participants will be halted and a full safety review will be performed to confirm causality. If 2 participants in any cohort experience

a DLT considered at least possibly related to study drug, the maximum tolerated dose (MTD) has been exceeded.

Dose limiting toxicities are defined as toxicities that, due to their severity or duration, are considered unacceptable, and limit further dose escalation. Examples of DLT's include, but are not limited to:

- Severe headache unrelieved by nonsteroidal anti-inflammatory drugs (NSAIDS)
- Nausea/Vomiting
- Hypotension as defined by mean arterial pressure <60mmHg or a drop in systolic BP of 25%

7.3. Method of Treatment Assignment

As this is an open label study, participants will be assigned to the ongoing treatment group at that time.

Study using IWRS	All participants will be assigned to a treatment group using an Interactive Web Response System (IWRS). Before the study starts, the log in information & directions for the IWRS will be provided to each site. Study treatment will be dispensed at the study visits summarized in SOA.

7.4. Blinding

Open-label using IWRS	This is an open-label study; however, the specific dose level to be taken by a participant will be assigned using an IWRS to aid cohort management. The site will contact the IWRS prior to the start of study treatment administration for each participant. The site will record the treatment assignment on the applicable case report form (CRF), if required.

7.5. Preparation/Handling/Storage/Accountability

- The investigator or designee must confirm appropriate temperature conditions have been maintained during transit for all study treatment received and any discrepancies are reported and resolved before use of the study treatment.
- Only participants enrolled in the study may receive study treatment and only authorized site staff may supply or administer study treatment. All study treatments must be stored in a secure, environmentally controlled, and monitored (manual or automated) area in accordance with the labeled storage conditions with access limited to the investigator and authorized site staff.

- The investigator, institution, or the head of the medical institution (where applicable) is responsible for study treatment accountability, reconciliation, and record maintenance (ie, receipt, reconciliation, and final disposition records).
- Further guidance and information for the final disposition of unused study treatment are provided in the Study Reference Manual.
- Under normal conditions of handling and administration, study treatment is not expected to pose significant safety risks to site staff. Take adequate precautions to avoid direct eye or skin contact and the generation of aerosols or mists. In the case of unintentional occupational exposure notify the monitor, Medical Monitor and/or GSK study contact.
- A Material Safety Data Sheet (MSDS)/equivalent document describing occupational hazards and recommended handling precautions either will be provided to the investigator, where this is required by local laws, or is available upon request from GSK.

7.6. Treatment Compliance

- GSK2586881 will be intravenously administered to participants at the site. Administration will be documented in the source documents and reported in the CRF.
- When the individual dose for a participant is prepared from a bulk supply, the preparation of the dose will be confirmed by a second member of the study site staff.
- When participants are dosed at the site, they will receive study treatment directly from the investigator or designee, under medical supervision. The date and time (start and end time of infusion) of the dose administered in the clinic will be recorded in the source documents. The dose of study treatment and study participant identification will be confirmed at the time of dosing by a member of the study site staff other than the person administering the study treatment.

7.7. Concomitant Therapy

Any medication or vaccine (including over-the-counter or prescription medicines, vitamins, and/or herbal supplements) that the participant is receiving at the time of enrollment or receives during the study must be recorded along with:

- reason for use
- dates of administration including start and end dates
- dosage information including dose and frequency

The Medical Monitor should be contacted if there are any questions regarding concomitant or prior therapy.

No medications will be prohibited except for those listed below:

- ACE inhibitors, angiotensin receptor blockers, and renin inhibitors will be discontinued at least 14 days prior to dosing until 72 hours post dose.

Paracetamol/Acetaminophen, at doses of ≤ 2 grams/day, is permitted for use any time during the study. Other concomitant medication may be considered on a case-by-case basis by the investigator in consultation with the Medical Monitor if required.

7.8. Treatment after the End of the Study

Participants will not receive any additional treatment from GSK after completion of the study because other treatment options are available.

The investigator is responsible for ensuring that consideration has been given to the post-study care of the participants medical condition, whether or not GSK is providing specific post-study treatment.

8. DISCONTINUATION CRITERIA

There are no treatment discontinuation criteria as this is a single dose study.

8.1. Liver Chemistry Monitoring

Liver chemistry monitoring criteria have been designed to assure participant safety and evaluate liver event etiology, (in alignment with the Food and Drug Administration(FDA) premarketing clinical liver safety guidance). These protocol guidelines are in alignment with FDA premarketing clinical liver safety guidance: http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM174090.pdf.

Liver Safety Required Actions and Follow up Assessments Section can be found in Appendix 7.

8.2. QTc Monitoring Criteria

There are no treatment discontinuation criteria as this is a single dose study, however the following criteria described below still apply.

- The *same* QT correction formula *must* be used for *each individual participant* to determine eligibility for and discontinuation from the study. This formula may not be changed or substituted once the participant has been enrolled.
    - For example, if a participant is eligible for the protocol based on QTcB, then QTcB must be used for discontinuation of this individual participant as well.
    - Once the QT correction formula has been chosen for a participant's eligibility, the *same formula* must continue to be used for that participant *for all QTc data being collected for data analysis*. Safety ECGs and other non-protocol specified ECGs are an exception.

- The QTc should be based on averaged QTc values of triplicate ECGs obtained over a brief (e.g., 5-10 minute) recording period.

A participant who meets either bulleted criteria based on the average of triplicate ECG readings will receive additional monitoring as appropriate:

- QTc > 530 msec OR Uncorrected QT > 600 msec
- Change from baseline of QTc > 60 msec

8.3. Withdrawal from the Study

Participants who withdraw from the study after treatment will still be required to complete both follow-up visits as detailed in the SoA.

- A participant may withdraw from the study at any time at his/her own request, or may be withdrawn at any time at the discretion of the investigator for safety, behavioural, compliance or administrative reasons. This applies to withdrawal from the genetic substudy.
- If the participant withdraws consent for disclosure of future information, the sponsor may retain and continue to use any data collected before such a withdrawal of consent. This applies to withdrawal from the genetic substudy.
- If a participant withdraws from the study, he/she may request destruction of any samples taken and not tested, and the investigator must document this in the site study records.
- If a participant is withdrawn from the study every effort must be made to complete the follow-up assessments detailed in the SoA within 7-14 days of last dose and the Day 28 follow-up assessments.

8.4. Lost to Follow Up

A participant will be considered lost to follow-up if he or she repeatedly fails to return for scheduled visits and is unable to be contacted by the study site.

The following actions must be taken if a participant fails to return to the clinic for a required study visit:

- The site must attempt to contact the participant and reschedule the missed visit as soon as possible and counsel the participant on the importance of maintaining the assigned visit schedule and ascertain whether or not the participant wishes to and/or should continue in the study.
- Before a participant is deemed lost to follow up, the investigator or designee must make every effort to regain contact with the participant (where possible, 3 telephone calls and, if necessary, a certified letter to the participant's last known mailing address or local equivalent methods). These contact attempts should be documented in the participant's medical record.

- Should the participant continue to be unreachable, he/she will be considered to have withdrawn from the study with a primary reason of lost to follow-up.

9. STUDY ASSESSMENTS AND PROCEDURES

- Study procedures and their timing are summarized in the SoA (Section 2).
- Protocol waivers or exemptions are not allowed
- Immediate safety concerns should be discussed with the sponsor immediately upon occurrence or awareness to determine if the participant should continue or discontinue study treatment.
- Adherence to the study design requirements, including those specified in the SoA, is essential and required for study conduct.
- All screening evaluations must be completed and reviewed to confirm that potential participants meet all eligibility criteria. The investigator will maintain a screening log to record details of all participants screened and to confirm eligibility or record reasons for screening failure, as applicable.
- Procedures conducted as part of the participant's routine clinical management (eg, blood count) and obtained before signing of ICF may be utilized for screening or baseline purposes provided the procedure met the protocol-specified criteria and was performed within the time frame defined in the SoA.
    - The maximum amount of blood collected from each participant over the duration of the study, including any extra assessments that may be required, will not exceed 300mL.
    - Repeat or unscheduled samples may be taken for safety reasons or for technical issues with the samples.

9.1. Pharmacodynamics

9.1.1. Hemodynamics

Pulmonary arterial catheters will be placed in all participants enrolled. Participants will consent to have the catheter inserted for the purposes of the study. Catheter placement may be confirmed by chest X-ray or fluoroscopy, as clinically indicated. All effort will be made to minimize radiation exposure time.

Variables that will be recorded from the right heart catheterization, as data permit, include:

- right atrial pressure
- pulmonary artery systolic and diastolic pressure
- mean pulmonary artery pressure
- pulmonary wedge pressure
- cardiac output and index measured by thermodilution

- pulmonary vascular resistance (PVR)
- pulmonary artery oxygen saturation (PAsat)

The clinical sites will use their SOPs for the insertion, maintenance and removal of the catheter. The catheter will be removed following completion of the "4 hour post-dose" catheter measurements and sample collections. The participant will then be monitored for a minimum of 20 hours or until discharge.

9.2. Adverse Events

The definitions of an AE or SAE can be found in Appendix 4.

The investigator and any designees are responsible for detecting, documenting, and reporting events that meet the definition of an AE or SAE and remain responsible for following up AEs that are serious, considered related to the study treatment or the study, or that caused the participant to discontinue the study (see Section 8).

9.2.1. Time Period and Frequency for Collecting AE and SAE Information

- All SAEs will be collected from the signing of the ICF until the follow-up visit at the time points specified in the SoA (Section 2).
- All AEs will be collected from the start of treatment until the follow-up visit at the time points specified in the SoA (Section 2).
- Medical occurrences that begin before the start of study treatment but after obtaining informed consent will be recorded on the Medical History/Current Medical Conditions section of the CRF not the AE section.
- All SAEs will be recorded and reported to the sponsor or designee within 24 hours, as indicated in Appendix 4. The investigator will submit any updated SAE data to the sponsor within 24 hours of it being available.
- Investigators are not obligated to actively seek AEs or SAEs in former study participants. However, if the investigator learns of any SAE, including a death, at any time after a participant has been discharged from the study, and he/she considers the event to be reasonably related to the study treatment or study participation, the investigator must promptly notify the sponsor.
- The method of recording, evaluating, and assessing causality of AEs and SAEs and the procedures for completing and transmitting SAE reports are provided in Appendix 4.

9.2.2. Method of Detecting AEs and SAEs

Care will be taken not to introduce bias when detecting AE and/or SAE. Open-ended and non-leading verbal questioning of the participant is the preferred method to inquire about AE occurrence.

9.2.3. Follow-up of AEs and SAEs

After the initial AE/SAE report, the investigator is required to proactively follow each participant at subsequent visits/contacts. All SAEs, will be followed until the event is resolved, stabilized, otherwise explained, or the participant is lost to follow-up (as defined in Section 8.4). Further information on follow-up procedures is given in Appendix 4.

9.2.4. Regulatory Reporting Requirements for SAEs

- Prompt notification by the investigator to the sponsor of a SAE is essential so that legal obligations and ethical responsibilities towards the safety of participants and the safety of a study treatment under clinical investigation are met.
- The sponsor has a legal responsibility to notify both the local regulatory authority and other regulatory agencies about the safety of a study treatment under clinical investigation. The sponsor will comply with country-specific regulatory requirements relating to safety reporting to the regulatory authority, IRB/ IEC, and investigators.
- Investigator safety reports must be prepared for suspected unexpected serious adverse reactions (SUSAR) according to local regulatory requirements and sponsor policy and forwarded to investigators as necessary.
- An investigator who receives an investigator safety report describing a SAE or other specific safety information (eg, summary or listing of SAE) from the sponsor will review and then file it along with the IB and will notify the IRB/IEC, if appropriate according to local requirements.

9.2.5. Cardiovascular and Death Events

For any CV events and all deaths, whether or not they are considered SAEs, specific CV and Death sections of the CRF will be required to be completed. These sections include questions regarding CV (including sudden cardiac death) and non-CV death.

The CV CRFs are presented as queries in response to reporting of certain CV medical dictionary for regulatory activities (MedDRA) terms. The CV information should be recorded in the specific CV section of the CRF within one week of receipt of a CV Event data query prompting its completion.

The Death CRF is provided immediately after the occurrence or outcome of death is reported. Initial and follow-up reports regarding death must be completed within one week of when the death is reported.

9.2.6. Pregnancy

- Details of all pregnancies in female participants and, if indicated, female partners of male participants will be collected after the start of study treatment and until 1 week after dose.
- If a pregnancy is reported, the investigator should inform GSK within 24 hours of learning of the pregnancy and should follow the procedures outlined in Appendix 5.
- Abnormal pregnancy outcomes (eg, spontaneous abortion, fetal death, stillbirth, congenital anomalies, and ectopic pregnancy) are considered SAE.

9.3. Treatment of Overdose

For this study, any dose of GSK2586881 greater than 1.2mg/kg will be considered an overdose.

Sponsor does not recommend specific treatment for an overdose.

In the event of an overdose, the investigator/treating physician should:

1. Contact the Medical Monitor immediately.
2. Closely monitor the participant for AE/SAE and laboratory abnormalities until GSK2586881 can no longer be detected systemically (at least 3 days post last dose).
3. Obtain a plasma sample for PK analysis as described in the SoA (Section 2). PK assessments may be continued beyond 24 hours if requested by the Medical Monitor (determined on a case-by-case basis).
4. Document the quantity of the excess dose as well as the duration of the overdosing in the CRF.

Decisions regarding dose interruptions or modifications will be made by the investigator in consultation with the Medical Monitor based on the clinical evaluation of the participant.

9.4. Six-Minute Walk Test

Six minute walk (6MW) distance, as performed at screening or within 6 months prior to screening. The subject is asked to walk along a prescribed path (typically 30 meters long) as far as possible during a 6-minute interval. The subject may walk at whatever pace is comfortable, with the goal of walking the longest distance possible. The subject may rest as needed. The subject will need to walk a minimum distance of ≥ 100 meters and maximum distance of ≤500 meters to meet study inclusion criteria. The distance walked will be entered into the CRF.

9.5. Safety Assessments

Planned time points for all safety assessments are provided in the SoA (Section 2).

9.5.1. Physical Examinations

- A complete physical examination will include, at a minimum, assessments of the Skin, CV, Respiratory, Gastrointestinal and Neurological systems. Height and weight will also be measured and recorded (at screening only).
- A brief physical examination will include, at a minimum, assessments of the skin, lungs, CV system, and abdomen (liver and spleen).
- Investigators should pay special attention to clinical signs related to previous serious illnesses.

9.5.2. Vital Signs

- Pulse rate, respiratory rate, and BP will be assessed.
- Blood pressure and pulse measurements will be assessed (supine) with a completely automated device. Manual techniques will be used only if an automated device is not available.
- Blood pressure and pulse measurements should be preceded by at least 5 minutes of rest for the participant in a quiet setting without distractions (eg, television, cell phones). At screening there will be 1 pulse and 3 BP measurements (3 consecutive BP readings will be recorded at intervals of at least 1 minute). The average of the 3 BP readings will be recorded on the CRF.

9.5.3. Pulse Oximetry

- Oxygen saturation will be measured by pulse oximetry after the subject has been at rest for at least 5 minutes.

9.5.4. Electrocardiograms

- 12-lead ECG will be obtained using an ECG machine that automatically calculates the HR and measures PR, QRS, QT, and QTc intervals. Refer to Section 8.2 for QTc monitoring criteria and additional QTc readings that may be necessary.
- Triplicate ECG are required at screening and predose. At these time points, 3 individual ECG tracings should be obtained as closely as possible in succession, but no more than 2 minutes apart.

9.5.5. Telemetry

- Telemetry will be measured continuously as detailed in the SoA (Section 2).
- Only AE and SAE data related to telemetry will be databased.

9.5.6. Clinical Safety Laboratory Assessments

- Refer to Appendix 2 for the list of clinical laboratory tests to be performed and to the SoA (Section 2) for the timing and frequency.

- The investigator must review the laboratory report, document this review, and record any clinically relevant changes occurring during the study in the AE section of the CRF. The laboratory reports must be filed with the source documents. Clinically significant abnormal laboratory findings are those which are not associated with the underlying disease, unless judged by the investigator to be more severe than expected for the participant's condition.
- All laboratory tests with values considered clinically significantly abnormal during participation in the study or within 3 days after the last dose of study treatment should be repeated until the values return to normal or baseline or are no longer considered significantly abnormal by the investigator or medical monitor.
- If such values do not return to normal/baseline within a period of time judged reasonable by the investigator, the etiology should be identified and the sponsor notified.
- All protocol-required laboratory assessments, as defined in Appendix 2, must be conducted in accordance with the laboratory manual and the SoA.
- If laboratory values from non-protocol specified laboratory assessments performed at the institution's local laboratory require a change in participant management or are considered clinically significant by the investigator (eg, SAE or AE or dose modification), then the results must be recorded in the CRF.

9.6. Pharmacokinetics

Whole blood samples will be collected for measurement of plasma concentrations of GSK2586881 as specified in the SoA (Section 2). The actual date and time (24-hour clock time) of each sample will be recorded.

Additional sample time points may be added during the study if warranted and agreed upon between the investigator and the sponsor, however, the maximum allowed blood volume will not be exceeded.

Samples collected for analyses of GSK2586881 plasma concentration may also be used to evaluate safety or efficacy aspects related to concerns arising during or after the study.

Once the plasma has been analyzed for GSK2586881 any remaining plasma may be analyzed for other compound-related metabolites and the results reported under a separate protocol.

Details of blood sample collection (including volume to be collected), processing, storage and shipping procedures are provided in the lab manual.

Plasma analysis will be performed under the control of GSK, the details of which will be included in the lab manual.

9.7. Immunogenicity Assessments

Whole blood samples will be collected for measurement of immunogenicity assessments as specified in the SoA (Section 2).

Additional visits at 12, 24, and 36 weeks post-dose (and less frequently thereafter), to obtain immunogenicity samples may be required in the unlikely event that subjects develop a clinically relevant immunoglobulin response to the drug as described in the lab manual.

Details of immunogenicity blood sample collection (including volume to be collected), processing, storage and shipping procedures are provided in the lab manual.

9.8. Biomarkers

The following samples for biomarker research will be collected from all participants in this study as specified in the SoA (Section 2):

- RAS peptides Ang II, Ang (1-7) and Ang (1-5) and other vasoctive peptides in the RAS and bradykinin cascade, and Nitric Oxide may be analyzed from plasma as data permit.
- Biomarkers of disease activity: NT pro-BNP, and cardiac troponin I may be analyzed from serum as data permit.

Additional sample time points may be added during the study if warranted and agreed between the investigator and the sponsor, however the maximum allowed blood volume will not be exceeded.

In addition, with the participant's consent, samples will be stored and may be used to investigate additional biomarkers thought to play a role in PAH disease progression or to evaluate their association with observed clinical responses to GSK2586881.

Samples may also be used for research to develop methods or support identification of prognostic/diagnostic biomarkers associated with clinical outcomes in PAH and related diseases.

Details of blood sample collection (including volume to be collected), processing, storage and shipping procedures are provided in the lab manual.

9.9. Genetics

A 6 mL blood sample for deoxyribonucleic acid (DNA) isolation will be collected from participants who have consented to participate in the genetics analysis component of the study. Participation is optional. Participants who do not wish to participate in the genetic research may still participate in the study.

In the event of DNA extraction failure, a replacement genetic blood sample may be requested from the participant. Signed informed consent will be required to obtain a replacement sample unless it was included in the original consent.

Appendix 6 for Information regarding genetic research. Details on processes for collection and shipment and destruction of these samples can be found in SRM.

10. STATISTICAL CONSIDERATIONS

10.1. Sample Size Determination

The sample size chosen is not based on statistical considerations.

The design of the study is deemed acceptable under the anticipated safety profile, given previous clinical experience with GSK2586881.

If additional participants/cohorts are enrolled to allow for evaluation of additional dose levels the proposed analysis approach(s) would be modified accordingly.

10.2. Populations for Analyses

For purposes of analysis, the following populations are defined:

Population	Description
Enrolled	All participants who sign the ICF
Safety	All participants who take at least 1 dose of study treatment. Participants will be analyzed according to the treatment they actually received.
Evaluable	All participants who are in the Safety population who complete all Day 1 assessments (including up to 24 hours post dose) and were not deemed to have had major protocol deviations (as defined within the PDMP)
PK Population	Subjects in the 'Safety' population for whom a PK sample was obtained and analysed. PK population will be the population for reporting PK data.

10.3. Statistical Analyses

Limited subject level data/information from a GSK supported study (NCT01884051) is available at doses of 0.2 mg/kg and 0.4 mg/kg. This data may be used as supportive only in reference to basic comparisons of safety, PK and PD with the current study. This previous study data will not be incorporated into statistical analyses associated with the current study.

10.3.1. Pharmacodynamic and Safety Analyses

Endpoint	Statistical Analysis Methods
Primary	• Descriptive Summaries, Figures and Listings of Change from Baseline PVR, CO and mPAP by planned timepoints. • If data permit, linear regression of Dose vs Change from Baseline PVR, CO and/or mPAP at 4 hours (endpoints $\log_e$ transformed if necessary).
Secondary	• Descriptive Summaries, Figures and Listings. • Simple figures (e.g. scatterplots and/or scatterplot matrix plots) exploring correlation between key PD endpoints (e.g. RAS peptides) and the Primary endpoints (PVR, CO and mPAP). • All safety analyses will be performed on the Safety Population
Exploratory	Will be described in the RAP. Note that exploratory genetic analyses, should they occur will be detailed in a separate RAP.

10.3.2. Pharmacokinetic and Pharmacokinetic-Pharmacodynamic Analyses

Pharmacokinetics analysis will be performed by, or under direct auspices of the Clinical PK Modeling & Simulation department within GSK. Plasma GSK2586881 concentration-time data will be analyzed by non-compartmental methods with WinNonlin V6.3 or greater. Calculations will be based on the actual sampling times recorded during the study.

From the plasma concentration-time data, the following PK parameters will be determined, as data permit: maximum observed plasma concentration (Cmax), time to Cmax (tmax), area under the plasma concentration-time curve [AUC(0-t) and area under the concentration-time curve from time zero (pre-dose) extrapolated to infinite time [AUC(0-∞)], the last observed quantifiable concentration (Clast), time of the last quantifiable concentration (tlast), plasma clearance (CL), volume of distribution (V_D) and apparent terminal phase half-life (t1/2). Other PK parameters may also be determined.

Pharmacokinetic data will be presented in graphical and/or tabular form and will be summarized descriptively. All PK data will be stored in the Archives, GSK Pharmaceuticals, research and development (R&D).

A population PK analysis may be conducted. In addition the GSK2586881 plasma concentration-time data may be merged with historical data and analysed as part of a population PK meta-analysis. Further details will be provided in the RAP. If data permit, potential association between systemic exposure of GSK2586881 and identified biomarker (e.g. Ang II, Ang(1-7), Ang(1-5)), and/or clinical related endpoints (pulmonary vascular hemodynamic) may be studied. In addition the PK/PD data may be

merged with historical data and analysed as part of a population PK/PD (biomarker) meta-analysis. Further details will be provided in the RAP. Population PK and PK/PD exploratory analyses may be presented separately to the CSR.

10.3.3. Interim Analyses

Safety, PK and pulmonary hemodynamic data will be reviewed after 4 subjects have completed dosing in each cohort prior to dosing at the next level. A linear regression model may be fitted to the (log transformed, if necessary) Change from Baseline at 4h PVR, CO and/or mPAP data from the first three cohorts (Dose Vs baseline adjusted response); and the slope parameter estimated along with the (extrapolated) mean response for the planned 4th dose of 0.8mg/kg. This will be used in addition to safety, PK and other PD data, to aid the team in decision making with regards to additional subject recruitment at an optimum dose level(s).

The RAP will describe the planned interim analyses in greater detail.

11. REFERENCES

Cheng ZJ, Vapaatalo H, Mervaala E. Angiotensin II and vascular inflammation. Medical Science Monitor. 2005; 11:194–205.

Clarke NE and Turner AJ. Angiotensin-Converting Enzyme 2: The First Decade.International Journal of Hypertension. 2011; doi:10.1155/2012/307315

Crosswhite P, Sun Z. Molecular Mechanisms of Pulmonary Arterial Remodeling. *Molecular Medicine*.2014; 20(1): 191–201.

D'Alonzo E, et al. Survival in Patients with Primary Pulmonary Hypertension. *Annals of Internal Medicine*. 1991;15(5):343-349

Dai HL, Guo Y, Guang XF, Xiao ZC, Zhang M, Yin XL. The changes of serum angiotensin-converting enzyme 2 in patients with pulmonary arterial hypertension due to congenital heart disease. *Cardiology*. 2013;124(4):208-12.

De Man F, Tu, Handoko, et al. Dysregulated Renin-Angiotensin-Aldesterone System Contributes to Pulmonary Arterial Hypertension. American Journal of Respiratory and Critical Care Medicines. 2012;186(8):780-9

Galie N, et al. Guildines for the diagnosis and treatment of pulmonary hypertension. European Respiratory Journal. 2009; 34(6):1219-63

GlaxoSmithKline Document Number 2010N108777_03 Study ID GSK2586881.Investigator's Brochure for GSK2586881 (formerly known as APN01)– A Soluble Recombinant Human Angiotensin Converting Enzyme 2 (rhACE2). Report Date 18-Sep-2015

GlaxoSmithKline Document Number 2014N217963_00 Study ID ACE114622. A two part study to investigate the safety, tolerability, pharmacokinetics and pharmacodynamics of GSK2586881 in subjects with acute lung injury. Report Date 07-Oct-2015.

GlaxoSmithKline Document Number 2016N300457_00 Supplement to GSK2586881 Investigator's Brochure. Version 2. Report Date 07-Oct-2016.

Grobe JL, Mecca AP, Lingis M, Shenoy V, Bolton TA, Machado JM,et.al. Prevention of angiotensin induced cardiac remodeling by angiotensin-(1–7). American Journal of Physiology. *Heart and Circulatory Physiology*. 2007;292(2):H736–H742.

Haschke M, Schuster M, Poglitsch M, Loibner H, Salzberg M, Bruggisser M, et al. Pharmacokinetics and pharmacodynamics of recombinant human angiotensin-converting enzyme 2 in healthy human subjects. *Clinical Pharmacokinetics*. 2013 Sep;52(9):783-92.

Humbert 2004 Marc Humbert, M.D., Ph.D., Olivier Sitbon, M.D., and Gérald Simonneau, M.D. Treatment of Pulmonary Arterial Hypertension, *The New England Journal of Medicine*. 2004;351:1425-36.

Johnson JA, Hemnes AR, Perrien DS, et al. Cytoskeletal defects in Bmpr2-associated pulmonary arterial hypertension. *American Journal of Physiology-Lung Cellular and Molecular Physiology*. 2012;302:L474-84

Johnson JA, West J, Maynard KB, Hemnes AR. ACE2 improves right ventricular function in a pressure overload model. *PLoS One*. 2011;6(6):e20828

Lipworth BJ, Dagg KD. Vasoconstrictor effects of angiotensin II on the pulmonary vascular bed. *Chest*. 1994; 105(5):1360–1364.

Maron BA, Loscalzo J. Pulmonary hypertension: pathophysiology and signaling pathways. *Handbook of Experimental Pharmacology.* 2013;218:31-58.

Sampaio WO, Santos RA, Faria-Silva R, da Mata Machado LT, Schiffrin EL, Touyz RM. Angiotensin-(1–7) through receptor Mas mediates endothelial nitric oxide synthase activation via Akt-dependent pathways. *Hypertension* 2007;49(1):185–192.

Schiffrin EL, Touyz RM. Inflammation and vascular hypertrophy induced by angiotensin II: role of NADPH oxidase-derived reactive oxygen species independently of blood pressure elevation? *Arteriosclerosis, Thrombosis, and Vascular Biology*. 2003; 23:707–709.

Sitbon O, Humbert M, Nunes H, Parent F, Garcia G, Herve P, et.al. Long-Term Intravenous Epoprostenol Infusion in Primary Pulmonary Hypertension. *Journal of the American College of Cardiology.* 2002;40(4)

Takahashi Y, Haga S, Ishizaka Y, Mimori A. Autoantibodies to angiotensin-converting enzyme 2 in patients with connective tissue diseases. *Arthritis Research & Therapy.* 2010;12(3):R85

Tipnis SR, Hooper NM, Hyde R, Karran E, Christie G & Turner AJ. A human homolog of angiotensin-converting enzyme. Cloning and functional expression as a captopril-insensitive carboxypeptidase. The Journal of Biological Chemistry. 2000; 275, 33238–33243.

Vickers C, Hales P, Kaushik V, Dick L, Gavin J, Tang J et al. Hydrolysis of biological peptides by human angiotensin-converting enzyme-related carboxypeptidase. The Journal of Biological Chemistry. 2002; 277, 14838–14843.

Yamada K, Iyer SN, Chappell MC, Ganten D, Ferrario CM. Converting enzyme determines plasma clearance of angiotensin-(1–7). *Hypertension.*1998; 32(3):496–502.

Zhang F, Hu Y, Xu Q, Ye S. Different effects of angiotensin II and angiotensin-(1–7) on vascular smooth muscle cell proliferation and migration. *PLoS ONE.* 2010;5(8):e12323.

12. APPENDICES

12.1. Appendix 1: Abbreviations and Trademarks

ABBREVIATIONS

2D	Two-dimensional
6MW	Six minute walk
ACE2	Angiotensin converting enzyme type 2
AE	Adverse Event
ALT	Alanine aminotransferase (SGPT)
Ang II	Angiotensin II
ARDS	Acute respiratory distress syndrome
AST	Aspartate aminotransferase (SGOT)
aPTT	Activated prothrombin time
AUC(0-∞)	Area under the plasma concentration-time curve
AUC(0-t)	Area under the concentration-time curve from time zero (pre-dose) to last time of quantifiable concentration within a subject across all treatments
BID	Bi-Daily
BMI	Body mass index
BP	Blood pressure
BUN	Blood urea nitrogen
CIOMS	Council for International Organizations of Medical Sciences
Clast	Last observed quantifiable concentration
CL	Plasma clearance
Cmax	Maximum observed plasma concentration
CO	Cardiac output
CONSORT	Consolidated Standards of Reporting Trials
COPD	Chronic Obstructive Pulmonary Disease
CPK	Creatine phosphokinase
CRF	Case Report Form
CRM	Continual reassessment model
CSR	Clinical study report
CV	Cardiovascular
DLT	Dose limiting toxicity
DNA	Deoxyribonucleic acid
ECG	Electrocardiogram
eCRF	Electronic case report form
eGFR	Estimated glomerular-filtration-rate
FDA	Food and Drug Administration
FSH	Follicle Stimulating Hormone
FTIH	First time in humans
GCP	Good Clinical Practice
GFR	Glomerular-filtration-rate
GSK	GlaxoSmithKline

HB	hemoglobin
HBsAg	Hepatitis B surface antigen
hCG	Human chorionic gonadotropin
HIPAA	Health Insurance Portability and Accountability
HIV	Human Immunodeficiency Virus
HPAH	Heritable PAH
HPLC	High performance liquid chromatography
HR	Heart rate
HRT	Hormone replacement therapy
IB	Investigator's Brochure
ICH	International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use
ICF	Informed Consent Form
IEC	Independent Ethics Committee
IgG	Immunoglobulin G
IgM	Immunoglobulin M
IND	Investigational New Drug
INR	International normalized ratio
IP	Investigational Product
IPAH	Idiopathic PAH
IRB	Institutional Review Board
IUD	Intrauterine device
IUS	Intrauterine hormone-releasing system
IV	Intravenous
IWRS	Interactive Web Response System
LDH	Lactate dehydrogenase
MCH	Mean corpuscular hemoglobin
MCV	Mean corpuscular volume
MedDRA	Medical Dictionary for Regulatory Activities
mPAP	Mean pulmonary arterial pressure
MSDS	Material Safety Data Sheet
MTD	Maximal tolerated dose
NO	Nitric oxide
NSAIDS	Nonsteroidal anti-inflammatory drugs
NSVT	Non-sustained ventricular tachycardia
PAC	Pulmonary arterial catheterization
PAH	Pulmonary Arterial Hypertension
PAP	Pulmonary arterial pressure
PAsat	Pulmonary artery saturation
PCW	Pulmonary capillary wedge
PD	Pharmacodynamic
PDE	Phosphodiesterase
PDMP	Protocol Deviation Management Plan
PGx	Pharmacogenetics
PK	Pharmacokinetic
PT	Prothrombin time

PVC	Premature Ventricular Complexes
PVR	Pulmonary vascular resistance
QTcB	QT duration corrected for heart rate by Bazett's formula
QTcF	QT duration corrected for heart rate by Fridericia's formula
R&D	Research and development
RAP	Reporting and Analysis Plan
RAS	Renin-Angiotensin System
RBC	Red blood cells
rhACE2	Recombinant human angiotensin converting enzyme type 2
RNA	Ribonucleic acid
SAE	Serious adverse event(s)
SaO_2	Oxygen saturations
SGOT	Serum glutamic-oxaloacetic transaminase
SGPT	Serum glutamic pyruvic transaminase
SOA	Schedule of Activities
SOP	Standard Operating Procedure
SRM	Study Reference Manual
SUSAR	Suspected, Unexpected, Serious Adverse drug Reaction
t½	Terminal phase half-life
tlast	Time of last quantifiable concentration
tmax	Time of occurrence of Cmax
ULN	Upper limit of normal
V_D	Volume of distribution
WHO	World Health Organization
WOCBP	Women of Childbearing Potential

Trademark Information

Trademarks of the GlaxoSmithKline group of companies
NONE

Trademarks not owned by the GlaxoSmithKline group of companies
WinNonlin

12.2. Appendix 2: Clinical Laboratory Tests

- The tests detailed in Table 1 will be performed by the central laboratory.
- If a local sample is required, it is important that the sample for central analysis is obtained at the same time.
- Protocol-specific requirements for inclusion or exclusion of participants are detailed in Section 6 of the protocol.
- Additional tests may be performed at any time during the study as determined necessary by the investigator or required by local regulations.

Table 1 Protocol-Required Safety Laboratory Assessments

Laboratory Assessments	Parameters			
Hematology	Platelet Count RBC Count Hemoglobin Hematocrit		RBC Indices: MCV MCH %Reticulocytes	WBC count with Differential: Neutrophils Lymphocytes Monocytes Eosinophils Basophils
Clinical Chemistry[1]	BUN	Potassium	Aspartate Aminotransferase (AST)/ Serum Glutamic-Oxaloacetic Transaminase (SGOT)	Total and direct bilirubin
	Creatinine	Sodium	Alanine Aminotransferase (ALT)/ Serum Glutamic-Pyruvic Transaminase (SGPT)	Total Protein
	Glucose	Calcium	Alkaline phosphatase	International normalized ratio (INR) Activated Prothrombin time (PT) aPTT
Routine Urinalysis	• Specific gravity • pH, glucose, protein, blood, ketones, bilirubin, Microscopic examination (only if blood or protein is abnormal)			
Other Screening Tests	• Follicle-stimulating hormone and estradiol (as needed in women of non-childbearing potential only) • Serum or urine human chorionic gonadotropin (hCG) pregnancy test (as needed for women of childbearing potential)[2]			

Laboratory Assessments	Parameters
	• Serology (HIV antibody, hepatitis B surface antigen [HBsAg], hepatitis C RNA and hepatitis C virus antibody)

NOTES :

1. Details of liver chemistry stopping criteria and required actions and follow-up assessments after liver stopping or monitoring event are given in Section 8.1 and Appendix 7. All events of ALT ≥3 × upper limit of normal (ULN) and bilirubin ≥2 × ULN (>35% direct bilirubin) or ALT ≥3 × ULN and international normalized ratio (INR) >1.5, if INR measured, which may indicate severe liver injury (possible Hy's Law), must be reported as an SAE (excluding studies of hepatic impairment or cirrhosis).
2. Local urine testing will be standard for the protocol unless serum testing is required by local regulation or IRB/IEC.

12.3. Appendix 3: Study Governance Considerations

Regulatory and Ethical Considerations

- This study will be conducted in accordance with the protocol and with:
  - Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences (CIOMS) International Ethical Guidelines
  - Applicable ICH Good Clinical Practice (GCP) Guidelines
  - Applicable laws and regulations
- The protocol, protocol amendments, ICF, Investigator Brochure, and other relevant documents (e.g., advertisements) must be submitted to an IRB/IEC by the investigator and reviewed and approved by the IRB/IEC before the study is initiated.
- Any amendments to the protocol will require IEC/IRB approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study participants.
- The investigator will be responsible for the following:
  - Providing written summaries of the status of the study to the IRB/IEC annually or more frequently in accordance with the requirements, policies, and procedures established by the IRB/IEC
  - Notifying the IRB/IEC of SAE or other significant safety findings as required by IRB/IEC procedures
  - Providing oversight of the conduct of the study at the site and adherence to requirements of 21 CFR, ICH guidelines, the IRB/IEC, European regulation 536/2014 for clinical studies (if applicable), and all other applicable local regulations

Financial Disclosure

Investigators and sub-investigators will provide the sponsor with sufficient, accurate financial information as requested to allow the sponsor to submit complete and accurate financial certification or disclosure statements to the appropriate regulatory authorities. Investigators are responsible for providing information on financial interests during the course of the study and for 1 year after completion of the study.

Informed Consent Process

- The investigator or his/her representative will explain the nature of the study to the participant or his/her legally authorized representative and answer all questions regarding the study.

- Participants must be informed that their participation is voluntary. Participants or their legally authorized representative will be required to sign a statement of informed consent that meets the requirements of 21 CFR 50, local regulations, ICH guidelines, Health Insurance Portability and Accountability Act (HIPAA) requirements, where applicable, and the IRB/IEC or study center.
- The medical record must include a statement that written informed consent was obtained before the participant was enrolled in the study and the date the written consent was obtained. The authorized person obtaining the informed consent must also sign the ICF.
- Participants must be re-consented to the most current version of the ICF(s) during their participation in the study.
- A copy of the ICF(s) must be provided to the participant or the participant's legally authorized representative.
- Participants who are rescreened are required to sign a new ICF.

Data Protection

- Participants will be assigned a unique identifier by the sponsor. Any participant records or datasets that are transferred to the sponsor will contain the identifier only; participant names or any information which would make the participant identifiable will not be transferred.
- The participant must be informed that his/her personal study-related data will be used by the sponsor in accordance with local data protection law. The level of disclosure must also be explained to the participant.
- The participant must be informed that his/her medical records may be examined by Clinical Quality Assurance auditors or other authorized personnel appointed by the sponsor, by appropriate IRB/IEC members, and by inspectors from regulatory authorities.

Publication Policy

- The results of this study may be published or presented at scientific meetings. If this is foreseen, the investigator agrees to submit all manuscripts or abstracts to the sponsor before submission. This allows the sponsor to protect proprietary information and to provide comments.
- The sponsor will comply with the requirements for publication of study results. In accordance with standard editorial and ethical practice, the sponsor will generally support publication of multicenter studies only in their entirety and not as individual site data. In this case, a coordinating investigator will be designated by mutual agreement.
- Authorship will be determined by mutual agreement and in line with International Committee of Medical Journal Editors authorship requirements.

Dissemination of Clinical Study Data

- This study will be registered and study information from this protocol will be posted on publicly available clinical trial registers before enrolment of study participants begins.
- The results summary of this study will be posted to the GSK Clinical Study Register and other publicly available clinical trial registers within 8 months of the primary study completion date.
- A manuscript reporting the study results will be submitted to a peer reviewed journal within 18 months of the last participant's last visit.

Data Quality Assurance

- All participant data relating to the study will be recorded on printed or electronic CRF unless transmitted to the sponsor or designee electronically (eg, laboratory data). The investigator is responsible for verifying that data entries are accurate and correct by physically or electronically signing the CRF.
- The investigator must maintain accurate documentation (source data) that supports the information entered in the CRF.
- The investigator must permit study-related monitoring, audits, IRB/IEC review, and regulatory agency inspections and provide direct access to source data documents.
- The sponsor or designee is responsible for the data management of this study including quality checking of the data.
- Study monitors will perform ongoing source data verification to confirm that data entered into the CRF by authorized site personnel are accurate, complete, and verifiable from source documents; that the safety and rights of participants are being protected; and that the study is being conducted in accordance with the currently approved protocol and any other study agreements, ICH GCP, and all applicable regulatory requirements.
- Records and documents, including signed ICF, pertaining to the conduct of this study must be retained by the investigator for 25 years after study completion unless local regulations or institutional policies require a longer retention period. No records may be destroyed during the retention period without the written approval of the sponsor. No records may be transferred to another location or party without written notification to the sponsor.

Source Documents

- Source documents provide evidence for the existence of the participant and substantiate the integrity of the data collected. Source documents are filed at the investigator's site.

- Data reported on the CRF or entered in the eCRF that are transcribed from source documents must be consistent with the source documents or the discrepancies must be explained. The investigator may need to request previous medical records or transfer records, depending on the study. Also, current medical records must be available.

Study and Site Closure

GSK or its designee reserves the right to close the study site or terminate the study at any time for any reason at the sole discretion of GSK. Study sites will be closed upon study completion. A study site is considered closed when all required documents and study supplies have been collected and a study-site closure visit has been performed.

The investigator may initiate study-site closure at any time, provided there is reasonable cause and sufficient notice is given in advance of the intended termination.

Reasons for the early closure of a study site by the sponsor or investigator may include but are not limited to:

- Failure of the investigator to comply with the protocol, the requirements of the IRB/IEC or local health authorities, the sponsor's procedures, or GCP guidelines
- Inadequate recruitment of participants by the investigator
- Discontinuation of further study treatment development

12.4. Appendix 4: Adverse Events: Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting

Definition of AE

AE Definition
• An AE is any untoward medical occurrence in a clinical study participant, temporally associated with the use of a study treatment, whether or not considered related to the study treatment. • NOTE: An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of a study treatment.

Events <u>Meeting</u> the AE Definition
• Any abnormal laboratory test results (hematology, clinical chemistry, or urinalysis) or other safety assessments (eg, ECG, radiological scans, vital signs measurements), including those that worsen from baseline, considered clinically significant in the medical and scientific judgment of the investigator (ie, not related to progression of underlying disease). • Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition. • New conditions detected or diagnosed after study treatment administration even though it may have been present before the start of the study. • Signs, symptoms, or the clinical sequelae of a suspected drug-drug interaction. • Signs, symptoms, or the clinical sequelae of a suspected overdose of either study treatment or a concomitant medication. Overdose per se will not be reported as an AE/SAE unless it is an intentional overdose taken with possible suicidal/self-harming intent. Such overdoses should be reported regardless of sequelae.

Events <u>NOT</u> Meeting the AE Definition
• Any clinically significant abnormal laboratory findings or other abnormal safety assessments which are associated with the underlying disease, unless judged by the investigator to be more severe than expected for the participant's condition. • The disease/disorder being studied or expected progression, signs, or symptoms of the disease/disorder being studied, unless more severe than expected for the participant's condition. • Medical or surgical procedure (eg, endoscopy, appendectomy): the condition that leads to the procedure is the AE. • Situations in which an untoward medical occurrence did not occur (social and/or

convenience admission to a hospital).

- Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.

Definition of SAE

If an event is not an AE per definition above, then it cannot be an SAE even if serious conditions are met (eg, hospitalization for signs/symptoms of the disease under study, death due to progression of disease).

A SAE is defined as any untoward medical occurrence that, at any dose:

a. Results in death

b. Is life-threatening

The term 'life-threatening' in the definition of 'serious' refers to an event in which the participant was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe.

c. Requires inpatient hospitalization or prolongation of existing hospitalization

In general, hospitalization signifies that the participant has been detained (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalization are AE. If a complication prolongs hospitalization or fulfills any other serious criteria, the event is serious. When in doubt as to whether "hospitalization" occurred or was necessary, the AE should be considered serious.

Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline is not considered an AE.

d. Results in persistent disability/incapacity

- The term disability means a substantial disruption of a person's ability to conduct normal life functions.
- This definition is not intended to include experiences of relatively minor medical significance such as uncomplicated headache, nausea, vomiting, diarrhea, influenza, and accidental trauma (eg, sprained ankle) which may interfere with or prevent everyday life functions but do not constitute a substantial disruption.

e. Is a congenital anomaly/birth defect

f. Other situations:

- Medical or scientific judgment should be exercised in deciding whether SAE reporting is appropriate in other situations such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the participant or may require medical or surgical intervention to prevent

one of the other outcomes listed in the above definition. These events should usually be considered serious.

Examples of such events include invasive or malignant cancers, intensive treatment in an emergency room or at home for allergic bronchospasm, blood dyscrasias or convulsions that do not result in hospitalization, or development of drug dependency or drug abuse.

Definition of Cardiovascular Events

Cardiovascular Events (CV) Definition:

Investigators will be required to fill out the specific CV event page of the CRF for the following AEs and SAEs:

- Myocardial infarction/unstable angina
- Congestive heart failure
- Arrhythmias
- Valvulopathy
- Pulmonary hypertension
- Cerebrovascular events/stroke and transient ischemic attack
- Peripheral arterial thromboembolism
- Deep venous thrombosis/pulmonary embolism
- Revascularization

Recording AE and SAE

AE and SAE Recording

- When an AE/SAE occurs, it is the responsibility of the investigator to review all documentation (eg, hospital progress notes, laboratory, and diagnostics reports) related to the event.
- The investigator will then record all relevant AE/SAE information in the CRF.
- It is **not** acceptable for the investigator to send photocopies of the participant's medical records to GSK in lieu of completion of the GSK /AE/SAE CRF page.
- There may be instances when copies of medical records for certain cases are requested by GSK. In this case, all participant identifiers, with the exception of the participant number, will be redacted on the copies of the medical records before submission to GSK.
- The investigator will attempt to establish a diagnosis of the event based on signs,

symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE.

Assessment of Intensity

The investigator will make an assessment of intensity for each AE and SAE reported during the study and assign it to 1 of the following categories:

- Mild: An event that is easily tolerated by the participant, causing minimal discomfort and not interfering with everyday activities.
- Moderate: An event that causes sufficiently discomfort and interferes with normal everyday activities.
- Severe: An event that prevents normal everyday activities. An AE that is assessed as severe should not be confused with an SAE. Severe is a category utilized for rating the intensity of an event; and both AE and SAE can be assessed as severe.

  An event is defined as 'serious' when it meets at least 1 of the predefined outcomes as described in the definition of an SAE, NOT when it is rated as severe.

Assessment of Causality

- The investigator is obligated to assess the relationship between study treatment and each occurrence of each AE/SAE.
- A "reasonable possibility" of a relationship conveys that there are facts, evidence, and/or arguments to suggest a causal relationship, rather than a relationship cannot be ruled out.
- The investigator will use clinical judgment to determine the relationship.
- Alternative causes, such as underlying disease(s), concomitant therapy, and other risk factors, as well as the temporal relationship of the event to study treatment administration will be considered and investigated.
- The investigator will also consult the Investigator's Brochure (IB) and/or Product Information, for marketed products, in his/her assessment.
- For each AE/SAE, the investigator **must** document in the medical notes that he/she has reviewed the AE/SAE and has provided an assessment of causality.
- There may be situations in which an SAE has occurred and the investigator has minimal information to include in the initial report to GSK. However, **it is very important that the investigator always make an assessment of causality for every event before the initial transmission of the SAE data to** GSK**.**
- The investigator may change his/her opinion of causality in light of follow-up information and send an SAE follow-up report with the updated causality assessment.
- The causality assessment is one of the criteria used when determining regulatory reporting requirements.

Follow-up of AE and SAE
• The investigator is obligated to perform or arrange for the conduct of supplemental measurements and/or evaluations as medically indicated or as requested by GSK to elucidate the nature and/or causality of the AE or SAE as fully as possible. This may include additional laboratory tests or investigations, histopathological examinations, or consultation with other health care professionals. • If a participant dies during participation in the study or during a recognized follow-up period, the investigator will provide GSK with a copy of any post-mortem findings including histopathology • New or updated information will be recorded in the originally completed CRF. • The investigator will submit any updated SAE data to GSK within 24 hours of receipt of the information.

Reporting of SAE to GSK

SAE Reporting to GSK via Electronic Data Collection Tool
• The primary mechanism for reporting SAE to GSK will be the electronic data collection tool. • If the electronic system is unavailable for more than 24 hours, then the site will use the paper SAE data collection tool (see next section). • The site will enter the SAE data into the electronic system as soon as it becomes available. • After the study is completed at a given site, the electronic data collection tool will be taken off-line to prevent the entry of new data or changes to existing data. • If a site receives a report of a new SAE from a study participant or receives updated data on a previously reported SAE after the electronic data collection tool has been taken off-line, then the site can report this information on a paper SAE form (see next section) or to the medical monitor/SAE coordinator by telephone. • Contacts for SAE reporting can be found in SRM.

12.5. Appendix 5: Contraceptive Guidance and Collection of Pregnancy Information

Definitions

Woman of Childbearing Potential (WOCBP)

A woman is considered fertile following menarche and until becoming post-menopausal unless permanently sterile (see below)

Women in the following categories are not considered WOCBP

1. Premenarchal
2. Premenopausal female with ONE of the following:

- Documented hysterectomy
- Documented bilateral salpingectomy
- Documented bilateral oophorectomy

Note: Documentation can come from the site personnel's: review of participant's medical records, medical examination, or medical history interview.

3. Postmenopausal female

- A postmenopausal state is defined as no menses for 12 months without an alternative medical cause. A high follicle stimulating hormone (FSH) level in the postmenopausal range may be used to confirm a postmenopausal state in women not using hormonal contraception or hormonal replacement therapy (HRT). However, in the absence of 12 months of amenorrhea, a single FSH measurement is insufficient.
- Females on HRT and whose menopausal status is in doubt will be required to use one of the non-hormonal highly effective contraception methods if they wish to continue their HRT during the study. Otherwise, they must discontinue HRT to allow confirmation of postmenopausal status before study enrollment.

Contraception Guidance

Male participants

No specific contraception is required of males as the likelihood of study drug distribution in semen is low.

Female participants

Female participants of childbearing potential are eligible to participate if they agree to use a highly effective method of contraception consistently and correctly as described in Table 2 from screening up to 3 days following treatment with study drug.

Table 2 Highly Effective Contraceptive Methods

Highly Effective Contraceptive Methods That Are User Dependent [a] *Failure rate of <1% per year when used consistently and correctly.*
Combined (estrogen- and progestogen-containing) hormonal contraception associated with inhibition of ovulation • oral • intravaginal • transdermal
Progestogen-only hormonal contraception associated with inhibition of ovulation[b] • injectable
Highly Effective Methods That Are User Independent
• Implantable progestogen-only hormonal contraception associated with inhibition of ovulation • Intrauterine device (IUD) • Intrauterine hormone-releasing system (IUS) • bilateral tubal occlusion
Vasectomized partner *(A vasectomized partner is a highly effective contraception method provided that the partner is the sole male sexual partner of the WOCBP and the absence of sperm has been confirmed. If not, an additional highly effective method of contraception should be used.)*
Sexual abstinence (*Sexual abstinence is considered a highly effective method only if defined as refraining from heterosexual intercourse during the entire period of risk associated with the study drug. The reliability of sexual abstinence needs to be evaluated in relation to the duration of the study and the preferred and usual lifestyle of the participant.)*

NOTES:
a. Typical use failure rates may differ from those when used consistently and correctly. Use should be consistent with local regulations regarding the use of contraceptive methods for participants in clinical studies.

Pregnancy Testing

- WOCBP should only be included after a negative highly sensitive urine or serum pregnancy test
- Additional pregnancy testing should be performed as specified in the SoA during the treatment period.
- Pregnancy testing will be performed when pregnancy is otherwise suspected

- Pregnancy testing, with a high sensitivity test will be performed using the test kit in accordance with instructions provided in its package insert

Collection of Pregnancy Information

Female Participants who become pregnant

- Investigator will collect pregnancy information on any female participant, who becomes pregnant while participating in this study.
- Information will be recorded on the appropriate form and submitted to GSK within 24 hours of learning of a participant's pregnancy.
- Participant will be followed to determine the outcome of the pregnancy. The investigator will collect follow up information on participant and neonate, which will be forwarded to GSK Generally, follow-up will not be required for longer than 6 to 8 weeks beyond the estimated delivery date.
- Any termination of pregnancy will be reported, regardless of fetal status (presence or absence of anomalies) or indication for procedure.
- While pregnancy itself is not considered to be an AE or SAE, any pregnancy complication or elective termination of a pregnancy will be reported as an AE or SAE.
- A spontaneous abortion is always considered to be an SAE and will be reported as such.
- Any SAE occurring as a result of a post-study pregnancy which is considered reasonably related to the study treatment by the investigator, will be reported to GSK as described in Section 12.4. While the investigator is not obligated to actively seek this information in former study participants, he or she may learn of an SAE through spontaneous reporting.
- Any female participant who becomes pregnant while participating will be withdrawn from the study.

12.6. Appendix 6: Genetics

There are no reported genetic mutations in ACE2 that have been associated with the development of PAH. However, genetic studies of a common insertion/deletion (I/D) polymorphism within the *ACE* gene appears to explain much of the variance observed in ACE enzyme expression levels, with the D allele conferring higher ACE and Ang II levels in tissue and serum (Marshall, 2002). Numerous studies report genetic mutations in ACE and association with various forms of pulmonary hypertension. For example, in male subjects, but not in the female subjects, the ACE DD genotype was negatively associated with electrocardiographic evidence of right ventricular hypertrophy (van Suylen, 1999), and pulmonary hypertension evoked by exercise challenge in patients with Chronic Obstructive Pulmonary Disease (COPD) (Kanazawa, 2000) plus others.

USE/ANALYSIS OF DNA

- Genetic variation may impact a participant's response to therapy, susceptibility, severity and progression of disease. Variable response to therapy may be due to genetic determinants that impact drug absorption, distribution, metabolism, and excretion; mechanism of action of the drug; disease etiology; and/or molecular subtype of the disease being treated. Therefore, where local regulations and IRB/IEC allow, a blood sample will be collected for DNA analysis
- DNA samples may be used for research related to [GSK2586881] or [PAH] and related diseases. They may also be used to develop tests/assays including diagnostic tests) related to GSK2586881 and PAH and related diseases. Genetic research may consist of the analysis of the entire genome.
- DNA samples may be analyzed if it is hypothesized that this may help further understanding of the clinical data. Analysis could include, but not be limited to, evaluation of the impact of I/D polymorphisms in the Angiotensin Converting Enzyme (ACE) gene on RAS peptides, and responses to GSK2586881 administration.
- The samples may be analyzed as part of a multi-study assessment of genetic factors involved in the response to GSK2586881 or study treatments of this class. The results of genetic analyses may be reported in the clinical study report or in a separate study summary.
- The sponsor will store the DNA samples in a secure storage space with adequate measures to protect confidentiality.
- The samples will be retained while research on GSK2586881 (or study treatments of this class) or PAH continues but no longer than 15 years or other period as per local requirements.

12.7. Appendix 7: Liver Safety: Required Actions and Follow-up Assessments

There are no criteria for study discontinuation as this is a single dose study; however if any of the criteria for liver events (as described in the table below) are met, then the monitoring and follow-up assessments are required as described.

Phase II liver chemistry increased monitoring criteria have been designed to assure participant safety and evaluate liver event etiology

Phase II liver chemistry additional monitoring criteria and required follow up assessments

Liver Chemistry Additional Monitoring Criteria	
ALT-absolute	ALT ≥ 5xULN
ALT Increase	ALT ≥ 3xULN persists for ≥4 weeks
Bilirubin[1, 2]	ALT ≥ 3xULN **and** bilirubin ≥ 2xULN (>35% direct bilirubin)
INR[2]	ALT ≥ 3xULN **and** INR>1.5, if INR measured
Cannot Monitor	ALT ≥ 3xULN and cannot be monitored weekly for 4 weeks
Symptomatic[3]	ALT ≥ 3xULN associated with symptoms (new or worsening) believed to be related to liver injury or hypersensitivity

Required Actions and Follow up Assessments	
Actions	**Follow Up Assessments**
• Report the event to GSK **within 24 hours** • Complete the liver event CRF and complete an SAE data collection tool if the event also meets the criteria for an SAE[2] • Perform liver chemistry event follow up assessments • Monitor the participant until liver chemistries resolve, stabilize, or return to within baseline (see **MONITORING** below) **MONITORING:** **For bilirubin or INR criteria:** • Repeat liver chemistries (include ALT, AST, alkaline phosphatase, bilirubin) and perform	• Viral hepatitis serology[4] • Obtain INR and recheck with each liver chemistry assessment until the transaminases values show downward trend • Obtain blood sample for pharmacokinetic (PK) analysis (within 24 hours) after last dose • Serum creatine phosphokinase (CPK) and lactate dehydrogenase (LDH). • Fractionate bilirubin, if total bilirubin≥2xULN • Obtain complete blood count with

liver event follow up assessments within **24 hrs** • Monitor participants twice weekly until liver chemistries resolve, stabilize or return to within baseline • A specialist or hepatology consultation is recommended **For All other criteria:** • Repeat liver chemistries (include ALT, AST, alkaline phosphatase, bilirubin) and perform liver event follow up assessments within **24-72 hrs** • Monitor participants weekly until liver chemistries resolve, stabilize or return to within baseline	differential to assess eosinophilia • Record the appearance or worsening of clinical symptoms of liver injury, or hypersensitivity, on the AE report form • Record use of concomitant medications on the concomitant medications report form including acetaminophen, herbal remedies, other over the counter medications. • Record alcohol use on the liver event alcohol intake case report form (CRF) page **For bilirubin or INR criteria:** • Anti-nuclear antibody, anti-smooth muscle antibody, Type 1 anti-liver kidney microsomal antibodies, and quantitative total immunoglobulin G (IgG) or gamma globulins. • Serum acetaminophen adduct high performance liquid chromatography (HPLC) assay (quantifies potential acetaminophen contribution to liver injury in participants with definite or likely acetaminophen use in the preceding week (James, 2009). Liver imaging (ultrasound, magnetic resonance, or computerised tomography) and /or liver biopsy to evaluate liver disease; complete Liver Imaging and/or Liver Biopsy CRF pages.

1. Serum bilirubin fractionation should be performed if testing is available. If serum bilirubin fractionation is not immediately available, discontinue study treatment for that subject if ALT ≥ 3xULN **and** bilirubin ≥ 2xULN.. Additionally, if serum bilirubin fractionation testing is unavailable, **record presence of detectable urinary bilirubin on dipstick**, indicating direct bilirubin elevations and suggesting liver injury.
2. All events of ALT ≥ 3xULN **and** bilirubin ≥ 2xULN (>35% direct bilirubin) or ALT ≥ 3xULN **and** INR>1.5, if INR measured which may indicate severe liver injury (possible 'Hy's Law'), **must be reported as an SAE (excluding studies of hepatic impairment or cirrhosis)**; INR measurement is not required and the threshold value stated will not apply to participants receiving anticoagulants
3. New or worsening symptoms believed to be related to liver injury (such as fatigue, nausea, vomiting, right upper quadrant pain or tenderness, or jaundice) or believed to be related to hypersensitivity (such as fever, rash or eosinophilia)
4. Includes: Hepatitis A IgM antibody; Hepatitis B surface antigen (HbsAg) and Hepatitis B Core Antibody (IgM); Hepatitis C RNA; Cytomegalovirus IgM antibody; Epstein-Barr viral capsid antigen IgM antibody (or if unavailable, obtain heterophile antibody or monospot testing); Hepatitis E IgM antibody

Phase II liver chemistry increased monitoring criteria

Liver Chemistry Increased Monitoring Criteria – Liver Monitoring Event	
Criteria	**Actions**
ALT ≥3xULN and <5xULN **and** bilirubin <2xULN**, without** symptoms believed to be related to liver injury or hypersensitivity, **and** who can be monitored weekly for 4 weeks	• Notify the GSK medical monitor **within 24 hours** of learning of the abnormality to discuss participant safety. • Participant must return weekly for repeat liver chemistries (ALT, AST, alkaline phosphatase, bilirubin) until they resolve, stabilise or return to within baseline • If at any time participant meets the liver chemistry stopping criteria, proceed as described above • If, after 4 weeks of monitoring, ALT <3xULN and bilirubin <2xULN, monitor participants twice monthly until liver chemistries normalize or return to within baseline.

References

James LP, Letzig L, Simpson PM, Capparelli E, Roberts DW, Hinson JA, Davern TJ, Lee WM. Pharmacokinetics of Acetaminophen-Adduct in Adults with Acetaminophen Overdose and Acute Liver Failure. Drug Metab Dispos 2009; 37:1779-1784.

Kanazawa H, Okamoto T, Hirata K, Yoshikawa J. Deletion Polymorphisms in the Angiotensin Converting Enzyme Gene Are Associated with Pulmonary Hypertension Evoked by Exercise Challenge in Patients with Chronic Obstructive Pulmonary Disease. American Journal of Respiratory and Criticial Care Medicine. 2000; 162: 1235- 1238

Marshall RP, Webb S, Bellingan GJ et al. Angiotensin Converting Enzyme Insertion/Deletion Polymorphism Is Associated with Susceptibility and Outcome in Acute Respiratory Distress Syndrome. American Journal of Respiratory and Critical Care Medicine. 2002; 166: 646-650

van Suylen RJ, Wouters EF, Pennings HJ et al. The DD Genotype of the Angiotensin Converting Enzyme Gene Is Negatively Associated with Right Ventricular Hypertrophy In Male Patients with Chronic Obstructive Pulmonary Disease. American Journal of Respiratory and Critical Care Medicine. 1999; 156: 1791-1795

12.8. Appendix 8: Protocol Amendment History

The Protocol Amendment Summary of Changes Table for the current amendment is located directly before the Table of Contents (TOC).

Amendment 3 01-Dec-2017

Overall Rationale for the Amendment

This is a substantial amendment to enable characterisation of the response at a lower starting level of 0.1mg/kg GSK2586881.

Section # and Name	Description of Change	Brief Rationale
Throughout	Top (maximum) dose reduced from 1.2mg/kg to 0.8mg/kg.	1.2mg/kg dose removed to enable investigation of a 0.1mg/kg group without increasing the size of the study.
1. Synopsis and 7.2 Dose Modification	Planned doses changed from 0.2, 0.4, 0.8 and 1.2mg/kg to 0.1, 0.2, 0.4 and 0.8mg/kg.	To enable characterisation of the dose response at 0.1mg/kg.
1. Synopsis	Study design schematic.	Changed to reflect new planned dose cohorts.
1. Synopsis and 5.2 Number of Participants	Additional participants can be recruited into a cohort prior to completion of all dose escalation.	Changed to allow the total number of participants recruited per cohort to be determined through an ongoing review of the safety, tolerability, PK and PD data prior to dose escalation.

Amendment 2 07-Nov-2017

Overall Rationale for the Amendment:

This is a non-substantial amendment to clarify the target population to be enrolled, the changes to the inclusion and exclusion criteria are listed in the table below. In addition, an upper limit for the 6MW was added, the evaluable population was changed, and the proposed linear regression analysis was expanded.

Section # and Name	Description of Change	Brief Rationale
Throughout	Transpulmonary RAS biomarkers was changed to pulmonary wedge RAS.	Transpulmonary is the position the sample is taken however a more appropriate way of describing this sample is pulmonary wedge RAS.
2. Schedule of Activities	Notes were added for genetic consent and genetic sample	Notes were added to ensure the timing and consent process are clear for the genetic sample.
6.1 Inclusion Criteria	A maximum distance of 450 meters was added for the Six minute walk (6MW) distance.	To confine the participant population to WHO functional class I, II, and III subjects.
6.2 Exclusion Criteria	Added renin inhibitors as restricted concomitant medication.	Renin inhibitors may potentially reduce Ang I and subsequently Ang II levels which is the target of ACE2 so by excluding them patients have a better chance of having elevated Ang II.
10.2 Population for Analysis	Evaluable population was changed from participants that completed day 7 and 14 follow ups to include participants that complete day 1.	To ensure participants lost to follow up will still be included in the evaluable population.
10.3.1 and 10.3.3 Statistical Analysis	Proposed linear regression analysis expanded to include other primary endpoints CO and mPAP if data permit (Section 10.3.1 and Section 10.3.3).	Expanded to assess additional hemodynamic measurements (primary endpoints) in relation to dose of GSK2586881

Amendment 1 03-May-2017

Overall Rationale for the Amendment: The protocol was amended to include the risk of exposure to ionizing radiation that was previously omitted and update information on participant rescreening numbers.

Section # and Name	Description of Change	Brief Rationale
3.3.1 Risk Assessment	Risk of exposure to ionizing radiation was added	Some participants will be exposed to ionizing radiation as part of the right heart catheterization, therefore the risk was added.
9.1.1 Hemodynamics	The procedure of confirming the right heart catheter placement by chest X-ray or fluoroscopy was added.	Procedure was added to ensure clear understanding.
6.4 Screen Failures	Rescreened participants should be assigned a new participant number.	Due to a change in eCRF system rescreened participants will be assigned a new number.
12.4 Appendix 4	Added SAE Reporting to GSK via Paper CRF	Missing from original protocol