An Observational Study to Characterize Patient Global Impression Questions for Activity-induced Symptoms in Patients with Pulmonary Arterial Hypertension

IND Number 138,142

Protocol RIN-PRN-201

CONFIDENTIAL

UNITED THERAPEUTICS CORPORATION

Original Protocol Date: 31 January 2019

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INVESTIGATOR'S AGREEMENT

I have read the attached protocol entitled "An Observational Study to Characterize Patient Global Impression Questions for Activity-induced Symptoms in Patients with Pulmonary Arterial Hypertension" dated 31 January 2019 and agree to abide by all provisions set forth therein.

I agree to comply with the International Council for Harmonization (ICH) Guideline for Good Clinical Practice and applicable Food and Drug Administration regulations/guidelines set forth in 21 Code of Federal Regulations Parts 50, 54, 56, and 312 and any local regulations per country.

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

This protocol has been received for information only and must not be implemented before all necessary regulatory agency and Ethics Committee (EC)/Institutional Review Board (IRB) approval documents have been obtained.

Signature of Principal Investigator	Date
Printed Name of Principal Investigator	

PROTOCOL SYNOPSIS

Title	An Observational Study to Characterize Patient Global Impression Questions for Activity-induced Symptoms in Patients with Pulmonary Arterial Hypertension
Study Phase	2
Indication	Pro Re Nata (PRN) "as needed" prevention and/or treatment of episodic/activity-induced symptoms in patients with pulmonary arterial hypertension (PAH)
Primary Objective	• To characterize the psychometric properties and performance of Patient Global Impression of Severity (PGI-S) questions, designed to measure severity, character, and duration of patients' self-reported symptoms of PAH following effort.
Secondary Objectives	• To characterize the relationship of PGI-S questions with: o Borg dyspnea score o Oxygen saturation o Heart rate (HR) o HR recovery at 1 minute after the ISWT • To characterize the recovery profile for activity-induced symptoms
Safety Objectives	• To evaluate safety based on: o Adverse events (AEs) o Vital sign measurements
Study Design	This is an observational, multicenter, single-day, Phase 2 study. This study will include a 14-day Screening Period and Study Day 1 clinic visit. Subjects will be required to perform an activity to induce symptoms of PAH, and subjects' severity of self-reported symptoms of PAH will be measured from pre-activity, immediately after the activity, and through the 30-minute recovery. Subjects will be asked about their PAH symptoms using 3 PGI-S questions that address their overall PAH symptoms, shortness of breath, and physical fatigue. PAH symptoms will be induced via the Incremental Shuttle Walk Test (ISWT). The total number of shuttles completed by a subject during the Screening ISWT will be the maximum targeted for that subject during the remaining ISWTs in the study.

After Screening, subjects will be assigned to 1 of 2 cohorts based on PAH medications as prescribed by their physician: Cohort A will include subjects who are currently prescribed and using inhaled treprostinil for the treatment of PAH and Cohort B will include subjects who are taking other PAH medications (instead of inhaled treprostinil).

Subjects in Cohort A will be assigned to 1 of 2 sequences:

Sequence 1:

- Period 1: The ISWT will be initiated 3 to 4 hours after a subject's previous dose of inhaled treprostinil (expected trough level). Activity will be timed to allow all procedures, including recovery, to be completed prior to Period 2.

- Period 2: The ISWT will be initiated within approximately 30 minutes of the previous inhaled treprostinil dose (expected peak level). Following recovery and the Investigator's confirmation of return to baseline status, the subjects will be discharged from the study.

Sequence 2:

- Period 1: The ISWT will be initiated within approximately 30 minutes of the previous inhaled treprostinil dose (expected peak level). Activity will be timed to allow all procedures, including recovery, to be completed prior to Period 2.

- Period 2: The ISWT will be initiated 3 to 4 hours after a subject's previous dose of inhaled treprostinil (expected trough level). Following recovery and Investigator's confirmation of return to baseline status, the subjects will be discharged from the study.

Subjects in Cohort B will also undergo 2 activity periods:

- Period 1: The ISWT will be initiated approximately 4 hours after the morning dose of PAH medication.

- Period 2: The ISWT will be initiated at least 1 hour following completion of the previous ISWT. Following recovery and the Investigator's confirmation of return to

	baseline status, the subjects will be discharged from the study. The baseline PGI-S and Borg dyspnea score assessments will be conducted prior to (-15 and 0 minutes; pre-ISWT) activity that is typically expected to induce symptoms of PAH. The subject will initiate activity (ISWT), and these same assessments will be performed immediately at the end of the ISWT (within 1 minute of completing the activity). The PGI-S assessments will also be performed throughout recovery following completion of the ISWT. Continuous pulse oximetry will be performed at each clinic visit starting at each pre-ISWT and through recovery. Vital sign assessments will be performed at the beginning and at the end of each clinic visit. On Day 1, subjects will be provided rest between the 2 ISWTs (at least 1 hour). The total study duration for a subject, including the Screening Period, will be up to 15 days.
Sample Size	Approximately 40 subjects will be enrolled in this study to ensure at least 36 evaluable subjects complete the study.
Summary of Subject Eligibility Criteria	**Inclusion criteria:** Subjects who meet the following criteria may be included in the study: 1. Subject voluntarily gives informed consent to participate in the study. 2. Males and females aged 18 years and above at the time of informed consent. 3. Established primary diagnosis of PAH that is either idiopathic or familial PAH (World Health Organization [WHO] Group 1), collagen vascular disease associated PAH, PAH associated with human immunodeficiency virus (HIV) infection, PAH induced by anorexigens/toxins, or PAH associated with repaired congenital systemic-to-pulmonary shunts (repaired ≥1 year). 4. Subject is deemed WHO Functional Class I, II, or III. 5. Subject has shortness of breath upon exertion (exhibits a ≥1-point change in Borg dyspnea score) as assessed by the ISWT and a minimum completion of 3 shuttles (30 meters) of the ISWT. Subject may have other symptoms as well.

6. Subject is on stable dose of all prescribed FDA-approved PAH treatments (exceptions are anticoagulants and diuretics) for at least 60 days prior to Screening.
7. In the opinion of the Investigator, the subject can communicate effectively with study personnel and is considered reliable, willing, and likely to be cooperative with protocol requirements.

Exclusion Criteria:

The following will exclude potential subjects from the study:

1. The subject is known to be pregnant or nursing.
2. The subject has PAH related to any condition not covered under inclusion criteria, including, but not limited to, pulmonary venous hypertension, pulmonary veno-occlusive disease, pulmonary capillary hemangiomatosis, chronic thromboembolic pulmonary hypertension, or other conditions under WHO Group 2, 3, 4, and 5 classifications.
3. The subject has evidence of clinically significant left-sided heart disease (including, but not limited to, left ventricular ejection fraction <40%, left ventricular hypertrophy) or clinically significant cardiologic conditions, such as congestive heart failure, coronary artery disease, or valvular heart disease.
4. The subject has any form of congenital heart disease (repaired or unrepaired; other than a patent foramen ovale).
5. The subject has any ambulatory or orthopedic limitations that would interfere with the ability to perform the activity.
6. The subject has been hospitalized within 30 days of Screening.
7. Current use of prostacyclin analogs/agonists, except inhaled treprostinil, for the treatment of PAH.
8. Use of any other investigational drug/device, or participation in any investigational study with therapeutic intent within 30 days of Screening (concurrent participation in registry studies is allowed).
9. Any other clinically significant illness that, in the opinion of the Investigator, might put the subject at risk of harm during the study or might adversely affect the interpretation of the study data.

Drug Dosage and Formulation	Not applicable.
Control Group	None.
Route of Administration	Not applicable.
Procedures	The procedures are as follows: • PGI-S question assessments performed at each clinic visit at Pre-ISWT (-15 and at 0 minutes prior to inducement of symptoms), at the end of the ISWTs (within 1 minute of completing the activity), every minute for the first 5 minutes of recovery followed by every 5 minutes for the remainder of the 30-minute recovery. • Pulse oximetry to be performed continuously. Pulse oximetry will include the collection of SpO_2 and HR. Pulse oximetry will start at the beginning of pre ISWT (15 minutes prior to the ISWT) and will be continuous through the end of recovery of each individual period. The SpO_2 and HR measures obtained at -15 and at 0 minutes (prior to activity), every minute during the ISWT, every minute for the first 5 minutes of the recovery, followed by every 5 minutes for the remainder of the 30-minute recovery will be recorded in the subject's source documents and eCRF. • Borg dyspnea score performed at each clinic visit at pre-ISWT (-15 and at 0 minutes prior to inducement of symptoms) and immediately at the end of the ISWTs (within 1 minute of completing the activity). • Vital signs (systolic blood pressure, diastolic blood pressure) will be assessed at Screening (prior to any activity and after recovery) and on Day 1 (prior to initiating Period 1 assessments and prior to discharge). • Following completion of all study procedures, and the Investigator deems that the subject has returned to baseline status, per pulse oximetry readings, the subject will be discontinued from the clinic.
Statistical Considerations	**Sample size:** Approximately 40 male and female subjects will be enrolled, with the plan to have at least 36 evaluable subjects complete the study.

	Data for all measurements will be tabulated overall and by cohort using descriptive statistics appropriate to the data. No formal statistical analyses are planned.
Sponsor	United Therapeutics Corp. 55 TW Alexander Drive Research Triangle Park, NC 27709 United States of America
Study Conducted by	Lung Biotechnology PBC 1040 Spring Street Silver Spring, MD 20910 United States of America

LIST OF ABBREVIATIONS

AE	Adverse event
DBP	Diastolic blood pressure
eCRF	Electronic Case Report Form
FDA	Food and Drug Administration
GCP	Good Clinical Practice
HR	Heart rate
ICF	Informed Consent Form
ICH	International Council for Harmonisation
IEC	Independent Ethics Committee
IRB	Institutional Review Board
ISWT	Incremental Shuttle Walk Test
MedDRA	Medical Dictionary for Regulatory Activities
PAH	Pulmonary arterial hypertension
PGI-S	Patient Global Impression of Severity
PRN	Pro Re Nata
SAE	Serious adverse event
SAP	Statistical Analysis Plan
SBP	Systolic blood pressure
SpO_2	Saturation peripheral capillary oxygenation
UTC	United Therapeutics Corporation
WHO	World Health Organization
WHODrug	World Health Organization Drug Dictionary

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1 BACKGROUND AND RATIONALE

1.1 DEFINITION OF CLINICAL PROBLEM

Pulmonary arterial hypertension (PAH), defined as an elevation in pulmonary arterial pressure and pulmonary vascular resistance, is a severe hemodynamic abnormality common to a variety of diseases and syndromes. Elevation in pulmonary arterial pressure causes an increase in right ventricular afterload, impairing right ventricular function and ultimately leading to heart failure and death (McLaughlin 2009, Rubin 1997). PAH can present with shortness of breath, chest tightness, exercise intolerance, dizziness, syncope, elevated respiratory and/or heart rate, decreased appetite, fatigue, chest and abdominal pain, lower extremity edema, and cyanosis.

In a Food and Drug Administration (FDA)-supported forum of PAH patients (summarized in The Voice of the Patient-PAH meeting summary report, December 2014) participants reported that despite the availability of current PAH medications, therapies specifically focused on treating debilitating and functionally limiting symptoms represent an area where patient needs are currently not met. Patients reported shortness of breath and fatigue, followed by dizziness and swelling of the legs and ankles, as having the greatest adverse impact on their daily life. Due to the episodic nature of the appearance of these symptoms, patients avoid physical activities that trigger these symptoms, such as household chores, playing with children, or simply walking around. Therefore, a therapeutic option that provides patients with greater control over treatment of their disease symptoms on an "as needed" basis would be beneficial.

Pro Re Nata (PRN) use of pharmacological agents has a long history in the treatment of various conditions. Examples where PRN treatment options have empowered patients to proactively effect a positive change in their medical condition and daily activity include symptomatic treatment of several pulmonary diseases, eg, asthma (Ventolin® HFA [albuterol sulfate; GlaxoSmithKline], Proventil® HFA [albuterol sulfate; Merck & Co. Inc.]) and chronic obstructive pulmonary disease (Proair® HFA [albuterol sulfate; Teva Respiratory, LLC]), as well as nonpulmonary diseases, such as patient-controlled pain management (Actiq® [fentanyl citrate; Cephalon, Inc.).

Episodic symptoms can be evaluated through the use of Patient Global Impression (PGI) questions. A published review of US package inserts for drug approvals occurring between 2006 to 2010 identified 20 approvals based on symptom-related primary endpoints (Gnanasakthy 2012). Specifically, the drugs Savella® (approved 2009 [milnacipran HCl; Allergan Pharmaceuticals International Limited) and Simponi® (approved 2009 [golimumab; Janssen Pharmaceutical Companies]) were approved based on PGI measures. Measurement of symptoms associated with PAH (eg shortness of breath and fatigue) should be shown as feasible.

1.2 CLINICAL HYPOTHESIS

PGI questions can be used as endpoints in clinical studies to evaluate episodic symptoms of PAH. Subjects will be asked about their PAH symptoms using 3 Patient Global Impression of Severity (PGI-S) questions that address their overall PAH symptoms, shortness of breath, and physical fatigue (Appendix 15.1). The hypothesis is that the PGI-S questions will be sensitive to detect changes in activity-induced symptoms of PAH. The Incremental Shuttle Walk Test (ISWT; Singh 1992) will be the activity used to induce symptoms in patients. This ISWT requires the subject to walk back and forth on a 10-meter course (Appendix 15.2). The speed at which the subject walks is dictated by an audio signal.

2 OBJECTIVES

2.1 PRIMARY OBJECTIVES

The primary objective of this study is to characterize the psychometric properties and performance of PGI-S questions, designed to measure the severity, character, and duration of patients' self-reported symptoms of PAH following effort.

2.2 SECONDARY OBJECTIVES

The secondary objectives of this study are the following:

- To characterize the relationship of PGI-S questions with:
  - Borg dyspnea score
  - Oxygen saturation
  - Heart rate (HR)
  - HR recovery at 1 minute after the ISWT

- To characterize the recovery profile for activity-induced symptoms.

2.3 SAFETY OBJECTIVES

Safety will be evaluated based on:

1. Adverse events (AEs)
2. Vital sign measurements

3 EXPERIMENTAL PLAN

3.1 STUDY DESIGN

This is an observational, multicenter, single-day, Phase 2 study. This study will include a 14-day Screening Period and a Study Day 1 clinic visit. The ISWT (Appendix 15.2) will be conducted to induce PAH symptoms and to measure PGI-S (Appendix 15.1). Subjects will be asked about their PAH symptoms using 3 PGI-S questions that address their overall PAH symptoms, shortness of breath, and physical fatigue.

The total number of shuttles completed by a subject during the Screening ISWT will be the maximum targeted for that subject during the remaining ISWTs in the study.

After Screening, subjects will be assigned to 1 of 2 cohorts based on PAH medications as prescribed by their physician; Cohort A will include subjects who are currently prescribed and using inhaled treprostinil for the treatment of PAH and Cohort B will include subjects who are taking other PAH medications (instead of inhaled treprostinil).

Subjects in Cohort A will be assigned to 1 of 2 sequences:

Sequence 1:

- Period 1: The ISWT will be initiated 3 to 4 hours after a subject's previous dose of inhaled treprostinil (expected trough level). Activity will be timed to allow all procedures, including recovery, to be completed prior to Period 2.
- Period 2: The ISWT will be initiated within approximately 30 minutes of the previous inhaled treprostinil dose (expected peak level). Following recovery and the Investigator's confirmation of return to baseline status, the subjects will be discharged from the study.

Sequence 2:

- Period 1: The ISWT will be initiated within approximately 30 minutes of the previous inhaled treprostinil dose (expected peak level). Activity will be timed to allow all procedures, including recovery, to be completed prior to Period 2.
- Period 2: The ISWT will be initiated 3 to 4 hours after a subject's previous dose of inhaled treprostinil (expected trough level). Following recovery and the Investigator's confirmation of return to baseline status, the subjects will be discharged from the study.

Subjects in Cohort B will also undergo 2 activity periods:

- Period 1: The ISWT will be initiated approximately 4 hours after the subject's morning dose of PAH medication.
- Period 2: The ISWT will be initiated at least 1 hour following the completion of the previous ISWT. Following recovery and the Investigator's confirmation of return to baseline status, the subjects will be discharged from the study.

The baseline PGI-S and Borg dyspnea score assessments will be conducted prior to (-15 and 0 minutes; pre-ISWT) an activity that is typically expected to induce symptoms of PAH. The subject will initiate activity (ISWT; Appendix 15.2), and these same assessments will be performed immediately at the end of the ISWT (within 1 minute of completing the activity). The PGI-S assessments will also be performed throughout recovery following completion of the ISWT. Continuous pulse oximetry will be performed at each clinic visit starting at each Baseline and through recovery.

Vital sign assessments will be performed at the beginning and at the end of each clinic visit. On Day 1, subjects will be provided rest (at least 1 hour) between the 2 ISWTs. Vital sign assessments will be performed at the beginning and at the end of each clinic visit.

All AEs will be documented from the time of informed consent until the time screen failure is documented, the subject discontinues, or the subject completes the study.

The study will consist of the following periods:

Screening: During the Screening Period, subjects who sign the Informed Consent Form (ICF) will first undergo all Screening assessments to evaluate eligibility as listed in the

inclusion/exclusion criteria. A window of up to 14 days is permitted to enroll a subject after Screening. When feasible, Screening assessments should be completed on the same day. Rescreening will be allowed with the Sponsor's Medical Monitor approval.

During Screening, the ISWT will be performed to determine eligibility and the ability to induce symptoms. Completion of a minimum of 3 shuttles (30 meters) of the test will be required for study eligibility. Pre-ISWT, ISWT conduct, and ISWT recovery will be performed and specific assessments will be conducted as outlined below.

Pre-ISWT: Baseline PGI-S (Appendix 15.1) and Borg dyspnea score (Appendix 15.3) assessments completed at -15 and 0 minutes prior to activity inducement of PAH symptoms will be performed. Continuous pulse oximetry, for the assessment of oxygen saturation and HR, will be initiated at the beginning of pre-ISWT and performed until the end of recovery from the ISWT. Vital signs (systolic blood pressure [SBP], diastolic blood pressure [DBP]) will be assessed at the beginning of pre-ISWT.

ISWT: PGI-S and Borg dyspnea scores will be assessed immediately upon completion of the ISWT. Continuous pulse oximetry, for the assessment of oxygen saturation and HR, will be assessed continuously during the activity.

ISWT Recovery: After completion of the ISWT, PGI-S assessments will be performed every minute during the first 5 minutes followed by every 5 minutes for the remainder of the 30-minute recovery. Continuous pulse oximetry, for the assessment of oxygen saturation and HR, will be assessed during recovery.

Vital sign assessments will be performed at the end of this clinic visit.

Day 1: The same assessments mentioned above will be performed during 2 ISWTs. PGI-S assessments will be performed as outlined below. At least a 1-hour rest period between the conduct of the 2 ISWTs will be required.

During enrollment, subjects meeting eligibility criteria will be enrolled into the study and assigned to 1 of 2 cohorts.

For subjects who are currently taking inhaled treprostinil (Cohort A), the ISWT will be initiated either 3 to 4 hours (expected trough level) or within 30 minutes (expected peak level) of a subject's previous dose in a cross-over manner. Baseline PGI-S and Borg dyspnea score assessments, completed at -15 and 0 minutes prior to activity inducement of PAH symptoms, will be performed. Continuous pulse oximetry will be performed from pre-ISWT until the end of recovery. Vital signs (SBP and DBP) will be assessed at the beginning of the Day 1 Visit.

For subjects who are taking other PAH medications (instead of inhaled treprostinil) (Cohort B), the ISWT will be initiated approximately 4 hours after the subject's morning dose of PAH medication.

Discharge: Following completion of all study procedures, recovery from study symptom-induced activities, and when the Investigator is satisfied that the subject has returned to the baseline status, per pulse oximetry readings, the subject will be discharged from the clinic.

3.2 OVERALL SCHEDULE OF TIMES AND EVENTS

Table 3-1 Schedule of Time and Events

Study Procedures	Screening Period[m] (Days -14 to -1)			Day 1							
				Period 1			Rest[h]	Period 2			Discharge[j]
	Pre-ISWT	ISWT[a]	Recovery (30 minutes)	Pre-ISWT	ISWT[a]	Recovery (30 minutes)		Pre-ISWT	ISWT[a]	Recovery (30 minutes)	
Informed Consent	X[l]										
Demographics	X[l]										
Medical History[i]	X[l]			X							
Vital Signs[b]	X[l]		X	X							X
Subject Eligibility	X[l]			X							
Enrollment[c]				X							
Borg Dyspnea Score[d]	X	X[k]		X	X[k]			X	X[k]		
PGI-S Questions[e]	X	X[k]	X	X	X[k]	X		X	X[k]	X	
Pulse Oximetry[f]	X	X	X	X	X	X		X	X	X	
Adverse Events[g]	X[l]	X	X	X	X	X	X	X	X	X	X
Concomitant Medications	X[l]	X	X	X	X	X	X	X	X	X	X

Abbreviations: AE, adverse event; eCRF, electronic Case Report Form; HR, heart rate; ISWT, Incremental Shuttle Walk Test; PAH, pulmonary arterial hypertension; PGI-S, Patient Global Impression of Severity; SpO_2, saturation of peripheral capillary oxygenation; WHO, World Health Organization

[a] ISWTs will be performed during Screening and on Day 1 of the study. Subjects receiving supplemental oxygen during the Screening assessment must continue to receive the same flow rate at all subsequent ISWTs. The supplemental oxygen flow rate must be recorded for each activity, as applicable. Supplemental oxygen therapy must not be introduced for subjects after the Screening assessment, unless there is an urgent medical need in the clinical judgement of the Investigator. For Cohort A, the ISWT will be initiated either 3 to 4 hours after a subject's previous dose of inhaled treprostinil (expected trough level) or within approximately 30 minutes of the last previous inhaled treprostinil dose (expected peak level). For Cohort B, the ISWT will be initiated approximately 4 hours after the subject's morning dose of PAH medication and then again at least 1 hour after the previous ISWT.

[b] Vital signs (systolic blood pressure, diastolic blood pressure) will be assessed at Screening (prior to any activity and after recovery) and on Day 1 (prior to initiating Period 1 assessments and prior to discharge).

[c] Subjects will be enrolled to 1 of 2 cohorts. Cohort A: Subjects are currently prescribed and using inhaled treprostinil for the treatment of PAH; Cohort B: Subjects are taking other PAH medication (instead of inhaled treprostinil).

[d] See Appendix 15.3. The subject should be asked to rate his/her maximal breathlessness at pre-ISWT (-15 and 0 minutes prior to activity inducement of PAH symptoms) and immediately at the end of the ISWT (within 1 minute of completing the activity). Assessments will be recorded in the subject's source documents and eCRF.

[e] Assessed at pre-ISWT (-15 and 0 minutes prior to activity inducement of PAH symptoms), at the end of the ISWT (within 1 minute of completing the activity), every minute for the first 5 minutes of the recovery, followed by every 5 minutes for the remainder of the 30-minute recovery. Assessments will be recorded in the subject's source documents and eCRF. Assessments will be performed in the following order: 1) PGI-S global PAH question; 2) PGI-S shortness of breath question; 3) PGI-S physical fatigue question. PGI-S assessments are to be conducted prior to the Borg dyspnea score assessments.

[f] Pulse oximetry to be performed continuously. Pulse oximetry will include the collection of SpO_2 and HR. Pulse oximetry will start at the beginning of pre-ISWT (15 minutes prior to the ISWT) and will be continuous through the end of recovery of each individual period. The SpO_2 and HR measures obtained at -15 and at 0 minutes (prior to activity), every minute during the ISWT, every minute for the first 5 minutes of the recovery, followed by every 5 minutes for the remainder of the 30-minute recovery will be recorded in the subject's source documents and eCRF.

[g] All AEs will be documented from time of informed consent until the subject discontinues or subject completes the study.

[h] Subjects will be provided at least a 1-hour period for rest between ISWTs (until subject feels they are rested enough to perform again at their baseline level) prior to Period 2 assessments. The start and end times for this period should be documented in subject's source documents and eCRF.

[i] Includes PAH history and WHO Functional Class. Any significant changes to the subject's medical condition must be documented throughout the course of the study.

[j] Following completion of all study procedures and once the Investigator deems that the subject has returned to their baseline status, per pulse oximetry readings, the subject will be discharged from the clinic. If the subject permanently discontinues the study prior to completion of all study assessments, for any reason, the following assessments, which are also performed prior to discharge, should be conducted, if possible: vital signs, AEs, and concomitant medications.

[k] Assessments will be performed immediately (within 1 minute) after the end of the ISWT.

[l] Assessments to be performed prior to initiating other Screening pre-ISWT assessments.

[m] A window of up to 14 days is permitted to enroll a subject after Screening. When feasible, Screening assessments should be completed on the same day. Rescreening will be allowed with the Sponsor's Medical Monitor approval.

3.3 CLINICAL ASSESSMENTS

3.3.1 Efficacy

There are no efficacy measures since this is an observational study.

3.3.2 Safety

Overall safety during the conduct of the study will be assessed on the following parameters: AEs, vital sign measurements, concomitant medications, and oxygenation.

3.3.2.1 Medical History and Demographics

A complete medical history (inclusive of PAH and World Health Organization [WHO] Functional Class) and demographics will be collected during Screening. Any known changes to the medical history prior to enrollment should be recorded. Significant past or present illnesses, current prescription or nonprescription medications (including vitamins and herbal products), and history of allergies or idiosyncratic responses to drugs should be recorded. Any significant changes to the subject's medical condition must be documented throughout the course of the study.

3.3.2.2 Vital Signs

Vital signs will include SBP and DBP. HR will be collected via continuous pulse oximetry (Section 3.3.2.3). Vital signs will be collected at the beginning and at the end of each clinic visit.

3.3.2.3 Oxygenation (Pulse Oximetry)

Pulse oximetry will be performed from the beginning of pre-ISWT (15 minutes prior to the ISWT) and will be continuous through the end of recovery. Pulse oximetry will include the collection of saturation of peripheral capillary oxygenation (SpO_2) and HR. The SpO_2 and HR measurements obtained at -15 and 0 minutes (prior to activity), every minute during the ISWT, every minute for the first 5 minutes of the recovery, followed by every 5 minutes for the remainder of the 30-minute recovery, will be recorded in the subject's source documents and electronic Case Report Form (eCRF).

A specific pulse oximeter model will be recommended by the Sponsor and the site will purchase and provide the Sponsor an invoice for reimbursement. In the event a pulse oximeter cannot be used (ie, subject has known issues with obtaining accurate readings from a finger probe, etc), an alternative device may be used with prior Sponsor approval, so long as the same device is used throughout the study.

3.3.2.4 Adverse Events

Adverse events will be recorded throughout the course of the study from the time of informed consent until the time a subject discontinues or subject completes the study. Each subject will be questioned for AEs at each study visit. Subjects will also be instructed to voluntarily report all AEs throughout the study.

All AEs should be followed until resolution (or return to normal or baseline values), until they are judged by the Investigator to no longer be clinically significant, or for at least 7 days if the AE extends beyond the end of study assessments. All AEs meeting criteria for a serious adverse event (SAE) should be followed until resolution, death, or until the subject is lost to follow-up, even if they are ongoing more than 7 days after study completion. All AEs/SAEs that occur while the subject is in the study will be recorded as instructed in this protocol.

Section 9 and Appendix 15.4 provide definitions and guidelines for recording AEs, respectively.

3.3.3 Concomitant Medications

All concomitant medications taken throughout the course of the study, including those taken for AEs, should be recorded in the subject's source documents and captured in the eCRF as required.

3.3.4 Assessment of PGI-S Questions and Borg Dyspnea Scores

3.3.4.1 Incremental Shuttle Walk Test

The ISWT is the activity that will be used to induce symptoms of PAH during Screening and on Day 1 of the study. All activities will be conducted in a designated area which meets the requirements described in Appendix 15.2.

Subjects receiving supplemental oxygen during the Screening assessment must continue to receive the same flow rate at all subsequent ISWTs. The supplemental oxygen flow rate must be recorded for each activity, as applicable. Supplemental oxygen therapy must not be introduced for subjects after the Screening assessment, unless there is an urgent medical need in the clinical judgement of the Investigator.

The total number of shuttles completed by a subject during the Screening ISWT will be the maximum targeted for that subject during the remaining ISWTs in the study.

3.3.4.2 PGI-S Questions

Three PGI-S questions will be administered at pre-ISWT (-15 and 0 minutes prior to activity inducement of PAH symptoms), immediately at the end of the ISWTs (within 1 minute of completing the activity), every minute for the first 5 minutes of recovery followed by every 5 minutes for the remainder of the 30-minute recovery. Assessments will be recorded in the subject's source documents and eCRF.

PGI-S assessments will be performed in the following order prior to the Borg dyspnea score assessment:

[redacted]

3.3.4.3 Borg Dyspnea Score

The Borg dyspnea score (Appendix 15.3) is a 10-point scale rating the level of dyspnea experienced during the study-defined activity and is administered at each clinic visit at pre-ISWT (-15 and 0 minutes prior to activity inducement of PAH symptoms) and immediately at the end of the ISWTs (within 1 minute of completing the activity). Scores range from 0 (no shortness of breath) to 10 (maximal shortness of breath). Assessments will be recorded in the subject's source documents and eCRF.

3.4 NUMBER OF CENTERS

Up to 20 US centers that treat PAH subjects will be identified and recruited for this study.

3.5 NUMBER OF SUBJECTS

The total sample size will be approximately 40 subjects to ensure at least 36 evaluable subjects complete the study.

3.6 ESTIMATED STUDY DURATION

The total study duration for a subject, including the Screening Period, will be up to 15 days.

4 SUBJECT ELIGIBILITY

4.1 INCLUSION CRITERIA

Subjects who meet the following criteria may be included in the study:

1. Subject voluntarily gives informed consent to participate in the study.
2. Males and females aged 18 years and above at the time of informed consent.
3. Established primary diagnosis of PAH that is either idiopathic or familial PAH (WHO Group 1), collagen vascular disease associated PAH, PAH associated with human immunodeficiency virus (HIV) infection, PAH induced by anorexigens/toxins, or PAH associated with repaired congenital systemic-to-pulmonary shunts (repaired ≥1 years).
4. Subject is deemed WHO Functional Class I, II, or III.
5. Subject has shortness of breath upon exertion (exhibits a ≥1-point change in Borg dyspnea score) as assessed by the ISWT and a minimum completion of 3 shuttles (30 meters) of the ISWT. Subject may have other symptoms as well.
6. Subject is on stable dose of all FDA-approved PAH treatments (exceptions are anticoagulants and diuretics) for at least 60 days prior to Screening.
7. In the opinion of the Investigator, the subject can communicate effectively with study personnel, and is considered reliable, willing, and likely to be cooperative with protocol requirements.

4.2 EXCLUSION CRITERIA

The following will exclude subjects from the study:

1. The subject is known to be pregnant or nursing.
2. The subject has PAH related to any condition not covered under inclusion criteria, including, but not limited to, pulmonary venous hypertension, pulmonary veno-occlusive disease, pulmonary capillary hemangiomatosis, chronic thromboembolic pulmonary hypertension, or other conditions under WHO Group 2, 3, 4, and 5 classifications.
3. The subject has evidence of clinically significant left-sided heart disease (including, but not limited to, left ventricular ejection fraction <40%, left ventricular hypertrophy)

or clinically significant cardiologic conditions, such as congestive heart failure, coronary artery disease, or valvular heart disease.

4. The subject has any form of congenital heart disease (repaired or unrepaired; other than a patent foramen ovale).
5. The subject has any ambulatory or orthopedic limitations that would interfere with the ability to perform the activity.
6. The subject has been hospitalized within 30 days of Screening.
7. Current use of prostacyclin analogs/agonists, except inhaled treprostinil, for the treatment of PAH.
8. Use of any other investigational drug/device, or participation in any investigational study with therapeutic intent within 30 days of Screening (concurrent participation in registry studies is allowed).
9. Any other clinically significant illness that, in the opinion of the Investigator, might put the subject at risk of harm during the study or might adversely affect the interpretation of the study data.

4.3 PRESCRIBED THERAPY

Subjects must not be receiving any prostacyclin analogs/agonists other than inhaled treprostinil for the treatment of PAH. All concomitant medications taken during the conduct of the study, including those taken for AEs, should be recorded in the subject's source documents and transcribed into the eCRF.

5 SUBJECT ENROLLMENT

5.1 TREATMENT ASSIGNMENT

This is an observational study; therefore, no treatment will be provided in the study.

5.2 ENROLLMENT

The same number of subjects will be enrolled into Cohort A (subjects taking inhaled treprostinil therapy) and Cohort B (subjects who are taking other PAH medications [instead of inhaled treprostinil]). Subjects will be assigned to Cohort A or B based on the use of inhaled treprostinil as determined by the Investigator.

Subjects meeting eligibility criteria will be enrolled into the study and assigned a sequential subject number within their cohort.

5.3 SEQUENCE ASSIGNMENT FOR COHORT A

Subjects in Cohort A will be assigned to either Sequence 1 or 2 by the Sponsor.

5.4 BLINDING

Not applicable. This is an observational study without an investigational medicinal product.

6 DRUGS AND DOSING (OR TREATMENT PROCEDURES)

6.1 DRUG DOSAGE, ADMINISTRATION AND SCHEDULE

Not applicable. This is an observational study and no investigational drug will be administered.

6.2 ACCESS TO BLINDED TREATMENT ASSIGNMENT

This is an observational study of an assessment tool and involves no blinded treatments.

6.3 COMPLIANCE

This is not applicable since this is an observational study.

7 EXPERIMENTAL PROCEDURES

Procedures in this study will be performed at Screening and on Day 1. Refer to Section 3.2 for the Overall Schedule of Times and Events.

7.1 SCREENING PERIOD (STUDY DAYS -14 TO -1)

A window of up to 14 days is permitted to enroll a subject after Screening. When feasible, Screening assessments should be completed on the same day. Rescreening will be allowed with the Sponsor's Medical Monitor approval. The following assessments for Screening are required prior to initiating the Screening pre-ISWT assessments:

- Informed consent
- Demographics
- Medical history, including PAH history and WHO Functional Class
- AEs (documented from time of informed consent)
- Concomitant medications
- Vital signs (SBP and DBP)
- Assess subject eligibility per inclusion/exclusion criteria

If eligible after the above assessments, the pre-ISWT, ISWT, and ISWT recovery assessments will be conducted. Any significant changes to the subject's medical condition must be documented throughout the course of the study.

7.1.1 Pre-ISWT

AEs and concomitant medications will be assessed throughout the study.

The following assessments are required at -15 and 0 minutes prior to activity inducement of PAH symptoms:

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will be started 15 minutes prior to initiating the ISWT and will be performed continuously through the end of recovery during Period 1. The SpO_2 and HR measurements obtained at -15 and 0 minutes (prior to activity) will be recorded in the subject's source documents and eCRF. Subjects receiving supplemental oxygen during the Screening assessment must continue to receive the same flow rate at all subsequent ISWTs. The supplemental oxygen flow rate must be recorded for each activity, as applicable. Supplemental oxygen therapy must not be introduced for subjects after the Screening assessment, unless there is an urgent medical need in the clinical judgement of the Investigator.

7.1.2 ISWT

AEs and concomitant medications will be assessed throughout the study.

The ISWT will be initiated and the following assessments will be performed immediately at the end of the test (within 1 minute of completing):

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will be performed continuously through the ISWT. The SpO_2 and HR measurements obtained every minute during the ISWT will be recorded in the subject's source documents and eCRF.

For subjects who meet the minimum level of activity (3 shuttles; ie 30 meters) per inclusion criterion #5, site personnel will schedule their appointment time for the Day 1 Visit based on their assigned cohort and sequence (for subjects on inhaled treprostinil).

7.1.3 ISWT Recovery

AEs and concomitant medications will be assessed throughout the study.

The following assessments are required every minute for the first 5 minutes of recovery followed by every 5 minutes for the remainder of the 30-minute recovery:

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Pulse oximetry (SpO_2 and HR measurements)

Vital signs (SBP and DBP) will be measured prior to subject leaving the clinical site.

7.2 STUDY DAY 1

AEs and concomitant medications will be assessed throughout the study.

The following will be confirmed or updated prior to initiating the ISWT:

- Eligibility per inclusion/exclusion criteria
- Medical history, including PAH history and WHO Functional Class
- Enrollment

Subjects assigned to Cohort A and Sequence 1 will follow procedures presented in Sections 7.2.1.1 and 7.2.1.2. Subjects assigned to Cohort A and Sequence 2 will follow procedures presented in Sections 7.2.1.3 and 7.2.1.4.

Subjects assigned to Cohort B will follow procedures presented in Sections 7.2.2.1 and 7.2.2.2.

7.2.1 Cohort A

Cohort A will include subjects who are currently prescribed and using inhaled treprostinil for the treatment of PAH. Subjects will be assigned to 1 of 2 sequences (Section 3.1).

7.2.1.1 Sequence 1/Period 1

The ISWT will be initiated 3 to 4 hours after a subject's previous dose of inhaled treprostinil (expected trough level).

Pre-ISWT

AEs and concomitant medications will be assessed throughout the study.

The following assessments are required (-15 and 0 minutes prior to activity inducement of PAH symptoms):

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will start 15 minutes prior to initiating the ISWT and will be performed continuously through the end of recovery. The SpO_2 and HR measurements obtained at -15 and 0 minutes (prior to activity) will be recorded in the subject's source documents and eCRF.

ISWT

AEs and concomitant medications will be assessed throughout the study.

The ISWT will be initiated and the following assessments will be performed immediately at the end of the test (within 1 minute of completing):

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will be performed continuously throughout. The SpO_2 and HR measurements obtained every minute during the ISWT will be recorded in the subject's source documents and eCRF.

ISWT Recovery

AEs and concomitant medications will be assessed throughout the study.

The following assessments are required every minute for the first 5 minutes of recovery followed by every 5 minutes for the remainder of the 30-minute recovery:

- PGI-S questions measurements
- Pulse oximetry (SpO_2 and HR measurements)

Rest Period

Subjects will be provided at least a 1-hour period for rest between ISWTs (until subject feels they are rested enough to perform again at their baseline level) prior to Period 2 assessments. The start and end times for this period should be documented in the subject's source documents and eCRF. AEs and concomitant medications will be assessed throughout this period.

7.2.1.2 Sequence 1/Period 2

The ISWT will be initiated within approximately 30 minutes of the previous inhaled treprostinil dose (expected peak level).

Pre-ISWT

AEs and concomitant medications will be assessed throughout the study.

The following assessments are required (-15 and 0 minutes prior to activity inducement of PAH symptoms):

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will start 15 minutes prior to initiating the ISWT and will be performed continuously through the end of recovery. The SpO_2 and HR measurements obtained at -15 and 0 minutes (prior to activity) will be recorded in the subject's source documents and eCRF.

ISWT

AEs and concomitant medications will be assessed throughout the study.

The ISWT will be initiated and the following assessments will be performed immediately at the end of the test (within 1 minute of completing):

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will be performed continuously throughout. The SpO_2 and HR measurements obtained every minute during the ISWT will be recorded in the subject's source documents and eCRF.

ISWT Recovery

AEs and concomitant medications will be assessed throughout the study.

The following assessments are required every minute for the first 5 minutes of recovery followed by every 5 minutes for the remainder of the 30-minute recovery:

- PGI-S questions measurements
- Pulse oximetry (SpO_2 and HR measurements)

7.2.1.3 Sequence 2/Period 1

The ISWT will be initiated within approximately 30 minutes of the previous inhaled treprostinil dose (expected peak level).

Pre-ISWT

AEs and concomitant medications will be assessed throughout the study.

The following assessments are required (-15 and 0 minutes prior to activity inducement of PAH symptoms):

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will start 15 minutes prior to initiating the ISWT and will be performed continuously through the end of recovery. The SpO_2 and HR measurements obtained at -15 and 0 minutes (prior to activity) will be recorded in the subject's source documents and eCRF.

ISWT

AEs and concomitant medications will be assessed throughout the study.

The ISWT will be initiated and the following assessments will be performed immediately at the end of the test (within 1 minute of completing):

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will be performed continuously throughout. The SpO_2 and HR measurements obtained every minute during the ISWT will be recorded in the subject's source documents and eCRF.

ISWT Recovery

AEs and concomitant medications will be assessed throughout the study.

The following assessments are required every minute for the first 5 minutes of recovery followed by every 5 minutes for the remainder of the 30-minute recovery:

- PGI-S questions measurements
- Pulse oximetry (SpO_2 and HR measurements)

Rest Period

Subjects will be provided at least a 1-hour period for rest between ISWTs (until subject feels they are rested enough to perform again at their baseline level) prior to Period 2 assessments. The start and end times for this period should be documented in the subject's source documents and eCRF. AEs and concomitant medications will be assessed throughout this period.

7.2.1.4 Sequence 2/Period 2

The ISWT will be initiated 3 to 4 hours after a subject's previous dose of inhaled treprostinil (expected trough level).

Pre-ISWT

AEs and concomitant medications will be assessed throughout the study.

The following assessments are required (-15 and 0 minutes prior to activity inducement of PAH symptoms):

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will start 15 minutes prior to initiating the ISWT and will be performed continuously through the end of recovery. The SpO_2 and HR measurements obtained at -15 and 0 minutes (prior to activity) will be recorded in the subject's source documents and eCRF.

ISWT

AEs and concomitant medications will be assessed throughout the study.

The ISWT will be initiated and the following assessments will be performed immediately at the end of the test (within 1 minute of completing):

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will be performed continuously throughout. The SpO_2 and HR measurements obtained every minute during the ISWT will be recorded in the subject's source documents and eCRF.

ISWT Recovery

AEs and concomitant medications will be assessed throughout the study.

The following assessments are required every minute for the first 5 minutes of recovery followed by every 5 minutes for the remainder of the 30-minute recovery:

- PGI-S questions measurements
- Pulse oximetry (SpO_2 and HR measurements)

7.2.2 Cohort B

7.2.2.1 Period 1

The ISWT will be initiated approximately 4 hours after the subject's morning dose of PAH medication.

Pre-ISWT

AEs and concomitant medications will continue to be assessed throughout the study.

The following assessments are required (-15 and 0 minutes prior to activity inducement of PAH symptoms):

- PGI-S question measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will start 15 minutes prior to initiating the ISWT and will be performed continuously through the end of recovery. The SpO_2 and HR measurements obtained at -15 and 0 minutes (prior to activity) will be recorded in the subject's source documents and eCRF.

ISWT

AEs and concomitant medications will be assessed throughout the study.

The ISWT will be initiated and the following assessments will be performed immediately at the end of the test (within 1 minute of completing):

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)

- Borg dyspnea score measurement

Pulse oximetry will be performed continuously throughout. The SpO_2 and HR measurements obtained every minute during the ISWT will be recorded in the subject's source documents and eCRF.

ISWT Recovery

AEs and concomitant medications will be assessed throughout the study.

The following assessments are required every minute for the first 5 minutes of recovery followed by every 5 minutes for the remainder of the 30-minute recovery:

- PGI-S questions measurements
- Pulse oximetry (SpO_2 and HR measurements)

Rest Period

Subjects will be provided at least a 1-hour period for rest between ISWTs (until subject feels they are rested enough to perform again at their baseline level) prior to Period 2 assessments. The start and end times for this period should be documented in the subject's source documents and eCRF. AEs and concomitant medications will be assessed throughout this period.

7.2.2.2 Period 2

The ISWT will be initiated at least 1 hour after completion of the previous ISWT.

Pre-ISWT

AEs and concomitant medications will continue to be assessed throughout the study.

The following assessments are required (-15 and 0 minutes prior to activity inducement of PAH symptoms):

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will start 15 minutes prior to initiating the ISWT and will be performed continuously through the end of recovery. The SpO_2 and HR measurements obtained at -15 and 0 minutes (prior to activity) will be recorded in the subject's source documents and eCRF.

ISWT

AEs and concomitant medications will continue to be assessed throughout the study.

The ISWT will be initiated and the following assessments will be performed immediately at the end of the test (within 1 minute of completing):

- PGI-S questions measurements (to be conducted prior to Borg dyspnea score assessment)
- Borg dyspnea score measurement

Pulse oximetry will be performed continuously throughout. The SpO_2 and HR measurements obtained every minute during the ISWT will be recorded in the subject's source documents and eCRF.

ISWT Recovery

AEs and concomitant medications will continue to be assessed throughout the study.

The following assessments are required every minute for the first 5 minutes of recovery followed by every 5 minutes for the remainder of the 30-minute recovery:

- PGI-S questions measurements
- Pulse oximetry (SpO_2 and HR measurements)

7.2.3 Discharge

If the subject permanently discontinues the study prior to completion of all study assessments, for any reason, the following assessments, which are also performed prior to discharge, should be conducted, if possible:

- Vital signs (SBP and DBP)
- AEs
- Concomitant medications

Following completion of all study procedures and once the Investigator deems that the subject has returned to their baseline status, per pulse oximetry readings, the subject will be discharged from the clinic.

8 STUDY TERMINATION

8.1 CRITERIA FOR SUBJECT WITHDRAWAL

A subject may voluntarily withdraw from the study at any time for any reason. A withdrawal of consent would preclude data collection regarding that subject after the date of withdrawal.

In the event a subject discontinues prematurely due to an AE, the subject will be followed until either the Investigator determines that the AE has resolved, it is no longer considered clinically significant, the subject is lost to further follow-up, or for 7 days if the AE extends beyond the final visit. A Subject Discontinuation Form must be completed for all subjects discontinued from the study.

8.2 CRITERIA FOR TERMINATING THE STUDY

The study may be stopped at any time if, in the opinion of the Sponsor, continuation of the study represents a serious medical risk to the subjects. This may include, but is not limited to, the presence of serious, life-threatening, or fatal AEs or AEs that are unacceptable in nature, severity, or frequency. The Sponsor reserves the right to discontinue the study for any reason at any time.

8.3 CRITERIA FOR DISCONTINUING A SITE

The study may also be terminated at a given site if any of the following occur:

- The Principal Investigator elects to discontinue the study
- The Sponsor elects to discontinue the study at the site
- US FDA regulations are not observed
- Concerns with protocol violations
- Changes in personnel or facilities adversely affect performance of the study

9 ADVERSE EVENT REPORTING

All AEs/SAEs that occur while the subject is participating in the study will be recorded as instructed in Section 9.2.

9.1 DEFINITIONS

9.1.1 Adverse Event

An AE is any untoward or unfavorable medical event experienced by a subject during a clinical study. An AE may include an intercurrent illness, injury, or any other concomitant impairment of the subject's health if deemed to have clinical significance. Adverse events may also include worsening of pre-existing symptoms or conditions that occur as a result of protocol-mandated procedures.

9.1.2 Serious Adverse Event

An SAE is an AE that results in any of the following outcomes:

- Death
- A life-threatening AE
- Inpatient hospitalization or prolongation of existing hospitalization
- A persistent or significant disability/incapacity
- A congenital anomaly/birth defect

In addition, important medical events that may not result in death, be life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they jeopardize the subject and require medical/surgical intervention to prevent any of the outcomes listed above.

Life-threatening means that the subject was at immediate risk of death from the event as it occurred. It does not mean that the event, had it occurred in a more severe form, might have caused death.

9.2 DOCUMENTATION OF ADVERSE EVENTS

All AEs will be documented from time of informed consent until the subject discontinues or subject completes the study. An AE or SAE occurring during the study must be documented in the subject's source documents and on the appropriate eCRF page(s). Information relating to the AE, such as onset and cessation date and times, intensity, seriousness, and outcome, is also to be documented in the eCRF (see Appendix 15.4 for definitions). Where possible, AEs should be recorded using standard medical terminology. If several signs or symptoms are

clearly related to a medically defined diagnosis or syndrome, the diagnosis or syndrome should be recorded on the eCRF page, not the individual signs and symptoms.

9.3 FOLLOW UP OF ADVERSE EVENTS

In the event a subject discontinues prematurely due to an AE, the subject will be followed until either the Investigator determines that the AE has resolved, it is no longer considered clinically significant, the subject is lost to further follow-up, or for 7 days if the AE extends beyond the final visit.

Follow-up of AEs and SAEs considered related (either possible or probable) to inhaled treprostinil will be processed as a post-marketing case.

9.4 REPORTING RESPONSIBILITIES OF THE INVESTIGATOR

All SAEs, as defined in Appendix 15.4, must be reported using the SAE report form via e-mail or fax within 24 hours of the investigator's knowledge of the SAE occurrence:

- Email to drugsafety@unither.com (primary method)
- Fax to (919) 313-1297 (back-up method)

The Investigator or Sponsor (if appropriate) must also notify their Institutional Review Board (IRB), Independent Ethics Committee (IEC), and/or other local equivalent agency of the reported SAE, including any follow-up information. Copies of each report and documentation of IEC/IRB/local equivalent body notification and receipt will be kept in the Investigator's study file.

9.5 SAFETY REPORTS

Any SAE that is possibly or reasonably attributable to concomitant PAH treatment (inhaled treprostinil) will be processed as a post-marketing case and requires immediate notification via email to drugsafety@unither.com (primary) or via fax at (919) 313-1297 (back-up) using the SAE report form.

10 STATISTICAL CONSIDERATIONS

This section briefly describes the planned statistical analyses. A complete description of the methodology will be specified in the Statistical Analysis Plan (SAP), which will be finalized

prior to database lock. Any changes in the statistical methods described in this protocol that occur prior to database lock will be documented in the SAP and will not require a protocol amendment.

10.1 DATA PROCESSING

The results of assessments will be transcribed into an eCRF for each subject who signs an ICF until study completion, or study discontinuation for any reason. A representative from the Sponsor will verify eCRF data fields against source documentation. Data clarifications will be generated, and the database will be edited as appropriate. The eCRFs will be reviewed by the Investigator for completeness and accuracy. The Investigator must electronically sign each subject's eCRF to signify his/her approval of the data. The Investigator will be required to re-sign an eCRF if changes are made to a subject's eCRF by the site after the Investigator initially signs the eCRF. The database will be final when all data management and quality assurance procedures are complete.

10.2 SAMPLE SIZE

Approximately 40 subjects will be enrolled to ensure at least 36 evaluable subjects complete the study. A sample size of 36 is expected to provide sufficient power in the context of this study to evaluate change in the PGI-S items. Assuming a change from baseline endpoint with values that may range between -4 and 4, a common standard deviation as the 50th percentile of the range (~2 units) for each PGI-S item, a correlation between post-activity and baseline measurements of 0.75, and a standard deviation of the difference scores as ~1.3, a 2-sided 95% confidence interval for a paired mean difference, based on the large sample z statistic, will extend approximately 0.52 units. With a confidence interval length at the mid-point of PGI-S response ratings, reasonable confidence in the point estimates observed for the PGI-S items should be achieved.

10.3 ANALYSIS PLAN

The data collected in this study will be presented in listings and tables. No inferential statistical analyses are planned. Presentations will be appropriate to the data analyzed. Summary statistics may include the mean, N, standard deviation, median, minimum, maximum, the 25th and 75th percentile values for continuous variables, and frequencies and

percentages for categorical variables. Confidence intervals may be included for informal purposes.

Medical history and prior/concomitant medications will be coded using the World Health Organization Drug Dictionary (WHODrug), and AEs will be coded using the Medical Dictionary for Regulatory Affairs (MedDRA). Coding dictionary versions will be specified in the SAP.

All statistical analyses will be performed by the Sponsor's biostatistics department personnel (or appropriate designees) using SAS®, Version 9.3 or higher, or other validated software.

10.3.1 Efficacy Endpoint(s)

There are no efficacy measures since this is an observational study.

10.3.2 PGI-S Questions Assessment

The primary objective is to characterize the psychometric properties and performance of 3 PGI-S questions (Appendix 15.1) in PAH subjects during pre-ISWT, at the end of activity, and through recovery.

The focus of the analyses will be to describe PGI-S measurement sensitivity of the subjects' self-reported symptoms of PAH at the end of the activity and during recovery from activity. The Sponsor will tabulate symptom scores and compute changes from baseline (pre-activity) scores for each question. Data may be displayed graphically and will be summarized overall and by cohort and sequence/period (as appropriate).

Additionally, for Cohort A, the sensitivity to a known inhaled treprostinil drug effect will be assessed tabulating, across time, the PGI-S measurements with inhaled treprostinil to PGI-S measurements without inhaled treprostinil. Test-retest assessments for PGI-S measurements will be conducted for Cohort B.

Correlations between the 3 PGI-S questions and with Borg dyspnea scores, oxygen saturation, and HR will be assessed. Data for HR recovery at 1 minute after the ISWT will be examined in an exploratory fashion overall and as related to PGI-S measurements.

The onset and recovery profile for activity-induced symptoms will also be characterized.

10.3.3 Safety Analyses

Safety will be evaluated on AEs, vital signs, and oxygenation. All AEs as recorded by the Investigators will be assigned a MedDRA Preferred Term and System Organ Class by the Sponsor for reporting purposes. The summary of AEs will include the number and percentage of subjects, as well as the number of events reported for each Preferred Term. No inferential analyses are planned for the AEs.

Data collected at pre-ISWT will serve as baseline values for the evaluation of data collected during ISWT and recovery. Summary statistics will be calculated for measured values and changes from baseline values. No inferential analyses are planned on these safety endpoints.

10.4 INTERIM ANALYSIS

There is no planned interim analysis for the study. However, given the open-label nature of the study, results may be tabulated by the Sponsor during the course of the study.

10.5 OTHER ANALYSES

Other analyses may be conducted based on available study data. Details for any other planned statistical analyses will be specified in the SAP.

10.6 DATA LISTINGS AND SUMMARIES

All scientifically relevant data gathered in this study will be presented in summary tables and listings in the clinical study report.

11 PACKAGING AND FORMULATION

11.1 CONTENTS OF STUDY DRUG

Not applicable. This is an observational study and no investigational drug will be administered.

11.2 LABELING

Not applicable. This is an observational study and no investigational drug will be administered.

11.3 STORAGE AND HANDLING OF CLINICAL STUDY MATERIAL

Not applicable. This is an observational study and no investigational drug will be administered.

11.4 SUPPLY AND RETURN OF CLINICAL STUDY MATERIAL

Not applicable. This is an observational study and no investigational drug will be administered.

11.5 DRUG ACCOUNTABILITY

Not applicable. This is an observational study and no investigational drug will be administered.

12 REGULATORY AND ETHICAL OBLIGATION

12.1 US FDA OR APPLICABLE REGULATORY REQUIREMENTS

The study will be conducted in accordance with International Council for Harmonisation (ICH), Good Clinical Practices (GCP) guidelines, and all applicable national regulations. The Sponsor will obtain the required approval from the regulatory authority to conduct the study. During the conduct of the study, an annual safety report will be compiled by the Sponsor for submission to those regulatory authorities and IRBs/IECs that require it. Any additional national reporting requirements specified by the applicable regulations, regulatory authorities, or IRB/IEC will also be fulfilled during the conduct of the study.

12.2 INFORMED CONSENT REQUIREMENTS

Before a subject is enrolled in the study, the Investigator or his/her designees must explain the purpose and nature of the study, including potential benefits and risks and all study procedures to the subject. The subject must sign and date an IRB/IEC-approved ICF prior to the conduct of any study-related activities. A copy of the signed consent form will be given to the subject, and the original will be retained in the study site's records.

12.3 INDEPENDENT ETHICS COMMITTEE/INSTITUTIONAL REVIEW BOARD

Prior to study initiation at each site, the Investigator will obtain approval for the study from an appropriate IRB/IEC and provide the Sponsor or designee with a copy of the approval letter. The IRB/IEC must also review and approve the study site's ICF and any other written

information provided to the subject prior to enrollment, as well as any advertising materials used for subject recruitment. Copies of the site-specific ICF and advertising materials must be forwarded to the Sponsor or designee for review before submission to the IRB/IEC and prior to the site starting the study.

If, during the study, it is necessary to amend either the protocol or the ICF, the Investigator is responsible for obtaining IRB/IEC approval of these amended documents prior to implementation. Copies of the IRB/IEC correspondence and approval letters must be sent to the Sponsor or designee.

During the conduct of the study, an annual progress report will be compiled by the Sponsor for submission to those IRBs/IECs that require it.

A written summary of the study will be provided by the Investigator to the IRB/EC following study completion or termination according to the IRB/IEC standard procedures. Additional updates will also be provided in accordance with the IRB/IEC's standard procedures.

12.4 PRESTUDY DOCUMENTATION REQUIREMENTS

Before the commencement of the clinical study, the following documents will be provided to the site: protocol, ICF, budget agreement, and eCRF.

The site will be required to provide the following documents to United Therapeutics Corporation's (UTC's) designee (Lung Biotechnology PBC) prior to study start: signature page of the protocol, Form FDA 1572, Financial Disclosure Form, IRB/IEC Composition and Roster, IRB/IEC protocol and informed consent approval letters, and Curriculum Vitae of study staff listed on the 1572.

12.5 SUBJECT CONFIDENTIALITY

Every effort will be made to keep medical information confidential. UTC, Lung Biotechnology PBC, the FDA or other regulatory bodies, and the IRB/IEC governing this study may inspect the medical records of any subject involved in this study. The Investigator may release the subject's medical records to employees or agents of the Sponsor, the IRB/IEC, or the FDA or appropriate local regulatory agencies for purposes of checking the

accuracy of the data. A number will be assigned to all subjects, and any report published will not identify the subject's name.

13 ADMINISTRATIVE AND LEGAL OBLIGATIONS

13.1 PROTOCOL AMENDMENTS AND STUDY TERMINATION

Protocol amendments that could potentially affect the safety of participating subjects or that alter the scope of the investigation, the scientific quality of the study, the experimental design, duration of therapy, assessment variables, the number of subjects treated, or subject selection criteria may be made only after consultation between UTC, its designee (Lung Biotechnology PBC), and the Investigator.

All protocol amendments must be submitted to and approved by the appropriate regulatory authorities and IRB/IEC prior to implementation.

A report documenting study termination must also be submitted to and acknowledged by the appropriate IRB/IEC for each study site.

At the end of the study, where applicable, a final report will be provided to the local regulatory agencies.

13.2 STUDY DOCUMENTATION AND STORAGE

In accordance with federal/national regulations, ICH, and GCP guidelines, the Investigator must retain study records for at least 2 years after the last approval of a marketing application in an ICH region and until there are no pending or contemplated marketing applications in an ICH region, or at least 2 years have elapsed since the formal discontinuation of clinical development of the investigational product. The Investigator must notify UTC or its designee, Lung Biotechnology PBC, before any disposal or change in location of study records.

13.3 STUDY MONITORING AND DATA COLLECTION

In accordance with federal/national regulations, ICH, and GCP guidelines, monitors for UTC or its designee, Lung Biotechnology PBC, will periodically contact the site and conduct on-site visits. During these visits, the monitor will, at a minimum, confirm ethical treatment

of subjects, assess study progress, review data collected, conduct source document verification, verify drug accountability periodically, and identify any issues requiring resolution.

The Investigator agrees to allow the monitor direct access to all relevant documents and to allocate his/her time and his/her staff to the monitor to discuss any findings or any relevant issues.

13.4 QUALITY ASSURANCE

The Sponsor is responsible for ensuring that the study is conducted and data are generated, documented, and reported in compliance with the protocol, GCP guidelines, and applicable regulatory requirements.

The Sponsor or a contracted representative of the Sponsor may conduct a quality assurance audit of this study. If such an audit occurs, the Investigator agrees to allow the auditor direct access to all relevant study documents and source data and to allocate time to discuss findings and any relevant issues. In addition, this study is subject to an audit by the relevant Regulatory Authorities. If such a regulatory inspection occurs, the Investigator agrees to allow the inspector direct access to all relevant study documents and source data.

14 REFERENCES

Center for Drug Evaluation and Research. The voice of the patient. A series of reports from the U.S. Food and Drug Administration's (FDA's) patient-focused drug development initiative. Pulmonary arterial hypertension. Silver Spring, MD: Center for Drug Evaluation and Research, U.S. Food and Drug Administration; December 2014.

Gnanasakthy A, Mordin M, Clark M, et al. A review of patient-reported outcome labels in the United States: 2006 to 2010. Value Health. 2012;15(3):437-442.

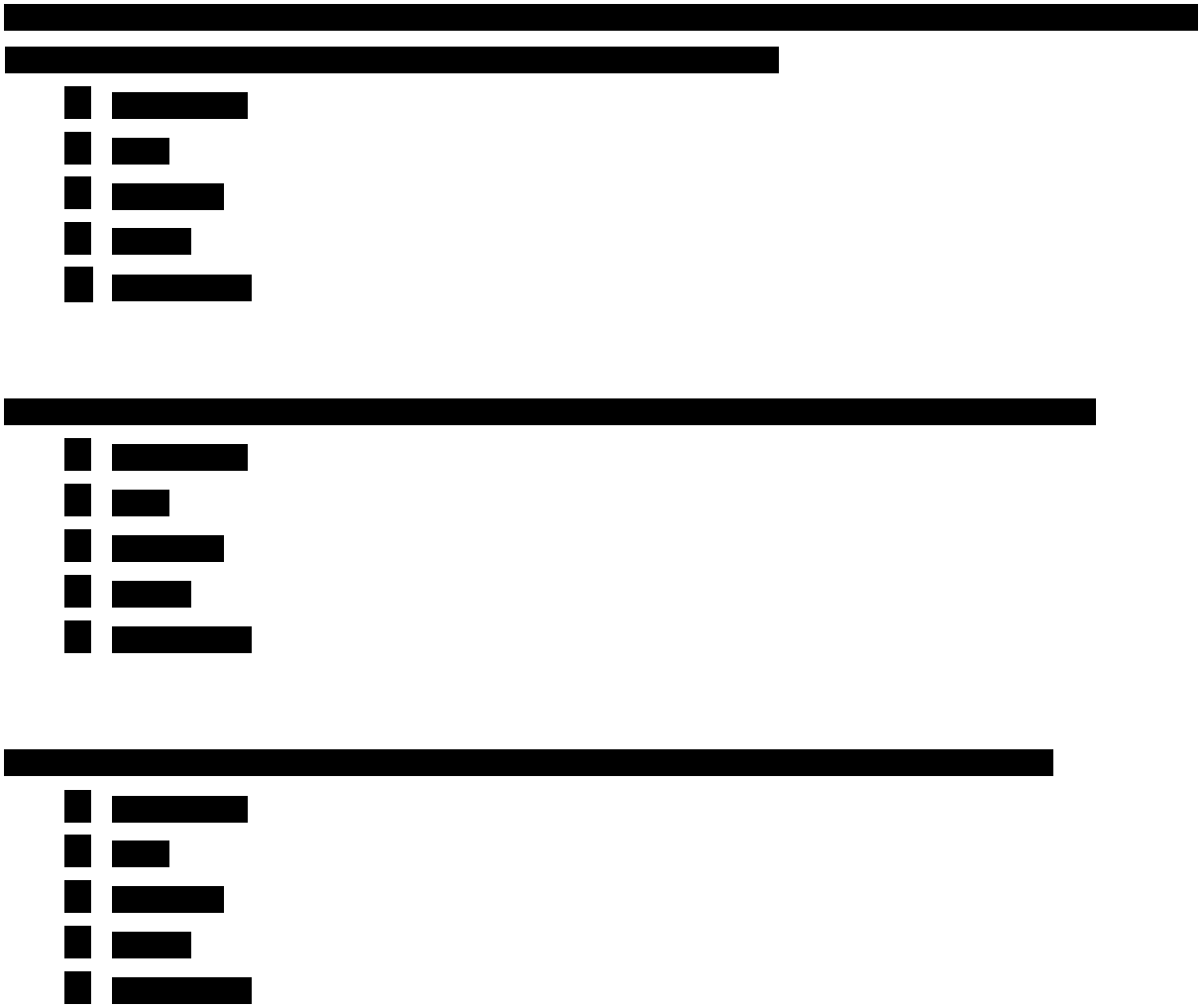
McLaughlin VV, Archer SL, Badesch DB, et al. ACCF/AHA 2009 expert consensus document on pulmonary hypertension: a report of the American College of Cardiology Foundation Task Force on Expert Consensus Documents and the American Heart Association: developed in collaboration with the American College of Chest Physicians, American Thoracic Society, Inc., and the Pulmonary Hypertension Association. Circulation. 2009;119(16):2250-2294.

Rubin LJ. Primary pulmonary hypertension. N Engl J Med. 1997;336(2):111-117.

Singh SJ, Morgan MD, Scott S, et al. Development of a shuttle walking test of disability in patients with chronic airways obstruction. Thorax. 1992;47(12):1019-1024.

15 APPENDICES

15.1 PATIENT GLOBAL IMPRESSION OF SEVERITY (PGI-S) QUESTIONS



15.2 INCREMENTAL SHUTTLE WALK TEST

Standardization

Standardization of the ISWT is very important for obtaining meaningful outcomes. It is advisable to time the performance as an additional measure, to confirm manual recording of the number of shuttles completed.

Please note that:

- If a repeat test is performed on the same day, at least a 1-hour rest should be allowed between tests.
- Only standardized instructions from the provided media should be used.
- No encouragement should be given throughout the ISWT.
- A comfortable ambient temperature and humidity should be maintained for all tests.
- The walking track must be the same for all tests for a patient:
  - Cones are placed 9 meters apart.
  - The distance walked around the cones is 10 meters.

The total number of shuttles completed by a subject during the Screening ISWT will be the maximum targeted for that subject during the remaining ISWTs in the study.

Before the ISWT

- Instruct the patient to dress comfortably and to wear appropriate footwear.

During the ISWT

Follow the instructions:

The objective of the ISWT is to walk as long as possible, there and back, along the 10-meter course (identified for the patient by 2 cones with an insert of 0.5 meters from either end), keeping to the speed indicated by the beeps on the audio recording.

Instructions for the subject: You will hear these beeps at regular intervals. You should walk at a steady pace, aiming to turn around the cone at 1 end of the course when you hear the first beep, and at the other end when you hear the next. At first, your walking speed will be very slow, but you will need to speed up at the end of each minute. Your aim should be to follow

the set rhythm for as long as you can. Each single beep signals the end of a shuttle and each triple beep signals an increase in walking speed. You should stop walking only when you become too breathless to maintain the required speed or can no longer keep up with the set pace.

The test is maximal and progressive. In other words, it is easier at the start and harder at the end. The walking speed for the first minute is very slow. You have 20 seconds to complete each 10-meter shuttle, so don't go too fast. The test will start in 15 seconds, so get ready at the start now. Level 1 starts with a triple beep after the 4-second countdown.

Ending the ISWT

The ISWT ends if any 1 of the following occur:

- The patient is **more than** 0.5 meters away from the cone when the beep sounds (allow 1 lap to catch up to the pace of the audio recording).
- The patient reports that they are too breathless to continue.

At the End of the ISWT

- Seat the patient or, if the patient prefers, allow the patient to stand.
- Record 1 of the following reasons for terminating the test (the patient can be asked, "What do you think stopped you from keeping up with the beeps?"):
  - Shortness of breath
  - Physical fatigue
  - Chest pain
  - Other

The patient should remain in a clinic area for at least 15 minutes following an uncomplicated test.

Modified from https://pulmonaryrehab.com.au/patient-assessment/assessing-exercise-capacity/incremental-shuttle-walking-test/

15.3 MODIFIED BORG DYSPNEA SCALE

The person administering the test will obtain a rating of dyspnea using the Borg dyspnea scale. The person will use the following dialogue:

"I would like to use the following scale to indicate your shortness of breath at this moment (indicate the Borg dyspnea scale). If there is no shortness of breath at all you will point to 0; if the shortness of breath is not very great you will choose from 0.5 to 2; if you are somewhat more short of breath you will select 3; and if the breathing is getting very difficult, you will choose 4 to 9, depending on just how hard it is; 10 represents the greatest shortness of breath you have ever experienced in your life. If one of the numbers does not exactly represent how short of breath you are at this moment, then you can choose a fraction in between. For example, if you have shortness of breath somewhere between 4 and 5, you can choose 4.5."

Perceived Breathlessness (Borg Scale)

0 NOTHING AT ALL

0.5 VERY VERY SLIGHT (just noticeable)

1 VERY SLIGHT

2 SLIGHT

3 MODERATE

4 SOMEWHAT SEVERE

5 SEVERE

6

7 VERY SEVERE

8

9 VERY VERY SEVERE (almost maximum)

10 MAXIMUM

15.4 GUIDELINES AND DEFINITIONS FOR RECORDING ADVERSE EVENTS

The Investigator or a designated member of his/her staff will probe each subject for any AEs that may have occurred. The Investigator should always ask the same question when conducting the verbal probe in order to ensure uniformity between subjects. The Investigator should ask:

"How are you doing (feeling)?"

Based on the subject's response to this question, the Investigator should ask additional questions relevant to the specific complaint such as:

"How severe is/was the symptom?"
"How often did the symptom occur?"
"How long did the symptom last?"

It is the Investigator's responsibility to review the results of all diagnostic and laboratory tests as they become available and ascertain if there is a clinically significant change from baseline. If the results are determined to be a clinically significant change from baseline, this should be reported as an AE. The Investigator may repeat the diagnostic procedure or laboratory test or request additional tests to verify the results of the original tests. When possible, a diagnosis associated with the abnormality should be used as the reported AE.

Using provided definitions, the Investigator will then:

(1) rate the intensity and seriousness of the AE and (2) note actions taken to counteract the AE.

Definitions of Intensity, Seriousness, and Outcome

INTENSITY

An assessment of the relative intensity (severity) of an AE is based on the Investigator's clinical judgment. The maximum intensity encountered during the evaluation period should be checked. The assessment of intensity should be independent of the assessment of the seriousness of the AE.

SERIOUSNESS

An SAE is one that represents an actual or potential significant hazard. This includes, but is not limited to, an event that is fatal, life-threatening, permanently or severely disabling, requires or prolongs inpatient hospitalization[a], is a congenital abnormality (offspring of subject) or is medically significant (important medical events that may not result in death, be life-threatening, or require hospitalization may be considered an SAE when, based upon appropriate medical judgment, they may jeopardize the subject and may require medical or surgical intervention to prevent 1 of the outcomes listed in this definition).

[a] Hospitalizations that would not be considered SAEs include those for:

- Routine treatment or monitoring of the study indication not associated with any deterioration in condition (eg, hospitalization for a routine right heart catheterization).
- Treatment which was elective or pre-planned, for a pre-existing condition not associated with any deterioration in condition (eg, pre-planned operation which does not lead to further complications etc).
- Treatment of an emergency, in an outpatient setting for an event not fulfilling any of the definitions of serious as given above and not resulting in hospital admission.

OUTCOME

- Fatal – The study subject died.
- Not Recovered/Not Resolved – The AE was ongoing at the time of death or at the time the subject was lost to follow up.
- Recovered/Resolved – The AE resolved.
- Recovered/Resolved with Sequelae – The AE is considered resolved; however, there is residual sequelae. Some events do not return to baseline, such as metastasis or progression of disease; however, once these events are determined by the Investigator to be stable or chronic, the Investigator may consider the event to be resolved or resolved with sequelae.
- Recovering/Resolving – The AE is improving but is not yet completely recovered/resolved.
- Unknown – The outcome of the AE cannot be determined.