Beta-blockers in Pulmonary Arterial Hypertension – A Phase 2 Double-Blind, Placebo-Controlled, Crossover Study Evaluating the Efficacy, and Safety of Carvedilol for Right Ventricular Dysfunction in Pulmonary Arterial Hypertension

Regulatory Sponsor:

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Study Product: Carvedilol

IND Number:

Trial Registration:

Version / Date: **10.12.17**

Statement of Compliance

This document is a protocol for a human research study. This study will be conducted according to US and International standards of Good Clinical Practice, applicable government regulations and Institutional research policies and procedures.

All individuals responsible for the design and conduct of this study have completed Human Subjects Protection Training and are qualified to be conducting this research prior to the enrollment of any subjects.

As Principal Investigator, I agree to conduct this clinical study in accordance with the design and specific provisions of this protocol. Modifications to the study are acceptable only with an approved protocol amendment. I agree to obtain approval from the IRB and/or regulatory bodies of competent jurisdiction, for the protocol and informed consent before initiating the study, to obtain consent from subjects prior to their enrollment in the study, to collect and record data as required by this protocol and case report forms, to prepare adverse event and study reports as required by this protocol and to maintain study documentation for the period of time required.

Thenappan Thenappan 06/05/2015

Print Name/Title of Principal Investigator Date

As a Sub-Investigator, I agree to conduct this clinical study in accordance with the design and specific provisions of this protocol. Modifications to the study are acceptable only with an approved protocol amendment. I agree to obtain approval from the IRB and/or regulatory bodies of competent jurisdiction, for the protocol and informed consent before initiating the study, to obtain consent from subjects prior to their enrollment in the study, to collect and record data as required by this protocol and case report forms, to prepare adverse event and study reports as required by this protocol and to maintain study documentation for the period of time required.

Marc R Pritzker 06/05/2015

Print Name/Title of Sub- Investigator Date
Print Location if multi-site protocol

Rebecca Cogswell 06/05/2015

Print Name/Title of Sub- Investigator Date
Print Location if multi-site protocol

Version History

Version #	Approval Date	Significant Changes from Previous Version
Version 1	08.12.2015	Original Protocol Version
Version 2	10.08.2015	• +/- 5 days window for all clinic visits • phone call at 4 weeks after completing the 13-month visit to assess clinical safety
Version 3		• Remove open-label access post study completion • Clarification of screening window

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Study Summary

Title	**A Phase 2 Double-Blind, Placebo-Controlled, Crossover Study Evaluating the Efficacy, and Safety of Carvedilol for Right Ventricular Dysfunction in Pulmonary Arterial Hypertension**
Short Title	Carvedilol PAH
Protocol Numbers	Protocol identifiers
Study Sponsor	List the regulatory sponsor and/or IND holder
Phase	II
Principal Investigator	Thenappan Thenappan, MD
Study Design	**Phase 2 Double-Blind, Placebo-Controlled, Crossover Study**
Study Duration	1 year
Study Center(s)	University of Minnesota, 420 Delaware Street SE, MMC 508 Minneapolis, MN 55435
Objectives	Estimate the efficacy of chronic beta-adrenergic receptor blockade with carvedilol on RV function in patients with PAH. Assess the safety and tolerability of chronic carvedilol therapy in patients with PAH
Number of Subjects	26
Main Inclusion Criteria	1. Age > 18 years 2. WHO category 1 pulmonary arterial hypertension (Dana Point 2008) 3. WHO functional class II-III 4. RVEF by cardiac MRI < 45% 5. Stable on PAH-specific therapy as defined by no change in PAH-specific treatment and functional class in the past 3 months. Patient can be on either mono or combination PAH-specific therapy
Study Product, Dose , Route, Regimen	Carvedilol (generic) Dose: 3.125 mg bid to 12.5 mg bid as tolerated Route: Oral
Duration of Administration	13 months
Reference Therapy	Placebo
Endpoints	The primary efficacy outcome is the mean change in RVEF as measured by cardiac MRI before and after 6 months of carvedilol treatment compared to placebo

	The primary safety outcome is the absence of adverse events associated with carvedilol
Statistical Methods	We will compare RVEF before and after therapy using paired t-tests or Wilcoxon signed-rank tests. The null hypothesis is that treatment with carvedilol has no effect on RVEF in patients with PAH with a two-sided alternative that carvedilol changes RVEF. Using a similar approach, we will also compare echo parameters of RV function, right heart hemodynamics, 6-minute walk distance, plasma NT-ProBNP, plasma catecholamine level, and MLHF questionnaire before and after study drug therapy.

List of Abbreviations:

AE	Adverse Event
CFR	Code of Federal Regulations
CRF	Case Report Form
CMRI	Cardiac Magnetic Resonance Imaging
DSMB	Data and Safety Monitoring Board
DSMP	Data and Safety Monitoring Plan
EF	Ejection fraction
FDA	Food and Drug Administration
GCP	Good Clinical Practice
HIPAA	Health Insurance Portability and Accountability Act of 1996
ICH	International Conference on Harmonization
IDS	Investigational Drug Services
IRB	Institutional Review Board
PAH	Pulmonary arterial hypertension
PHI	Protected Health Information
RV	Right Ventricle
SAE	Serious Adverse Event

1.0 Study Contact Information

1.1 Principal Investigator Contact Information (Coordinating Center)

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1.3 Key Study Personnel

Clinical Coordinator: Gretchen Peichel, RN
Study Monitor: CTSI
Medical Monitor: Thenappan Thenappan, MD
Statistician: Sue Duval, Ph.D.

2.0 Introduction / Background and Rationale

Safety and Efficacy of Beta-blockers in Pulmonary Arterial Hypertension: Pulmonary arterial hypertension (PAH) is characterized by progressive narrowing of the resistance pulmonary arteries, ultimately leading to right ventricular (RV) failure and death. It is a debilitating disease, associated with significant morbidity and a 1-year mortality rate of 15-20%. Moreover, the economic burden of PAH is high; estimated PAH-related healthcare costs in the United States are ~ $ 29 billion dollars per year. As the disease progresses, the RV dilates due to maladaptive remodeling and eventually fails. Multiple studies have consistently shown that long-term outcomes in PAH are largely determined by RV function. Despite this, all approved therapies for PAH target the pulmonary vasculature and not the RV. Neurohormonal activation has been observed in isolated RV failure secondary to PAH. Although neurohormonal modulation with pharmacological blockade of the β-adrenergic receptors is one of the main treatment strategies in left ventricular systolic dysfunction, the efficacy and safety of this treatment approach in isolated RV failure secondary to PAH has not been determined. In fact, beta-blockers are considered not safe in PAH, as these patients are dependent on the heart rate to maintain cardiac output. However, increasing evidence from animal studies and early human experience suggest that chronic β-adrenergic receptor blockade indeed improves RV function in PAH. Thus, β-adrenergic receptor blockers, if proven safe and efficacious in humans, may offer an inexpensive therapy for improving RV function, the main determinant of long-term PAH outcomes.

The **long-term goal** of our research is to develop novel therapies that improve RV function and thereby improve outcomes in PAH. **In this proposal**, we will evaluate pharmacological blockade of the β-adrenergic receptor as a potential therapeutic target for the treatment of RV failure resulting from PAH. Carvedilol is a non-selective α1/β1/β2-adrenergic receptor antagonist that causes reverse remodeling of the left ventricle and improves survival in patients with left ventricular systolic dysfunction. In animal models of PAH, carvedilol reverses RV remodeling and restores RV β-receptor signaling pathways, ultimately leading to improved RV function. Moreover, results from a retrospective, propensity score analysis published by our group and a recent small open label trial show that β-adrenergic receptor antagonists are safe in PAH patients**.** Based on these preliminary data, *we **hypothesize** that by inhibiting neurohormonal activation, treatment with carvedilol will improve RV function as measured by cardiac magnetic resonance imaging (MRI), and will be safely tolerated, in PAH patients.*

Specific Aim: Determine the efficacy (improvement in RV function) and safety of long-term β-adrenergic receptor blockade with carvedilol in patients with PAH. We will conduct a, randomized, phase 2, placebo-controlled, double-blinded, crossover trial of carvedilol in 26 PAH patients with World Health Organization functional class II or III symptoms and RV ejection fraction (EF) < 45% for 6 months.

Protocol: Adult PAH patients on a stable dose of an approved PAH medication will undergo the following baseline assessments: cardiac magnetic resonance imaging (MRI), right heart catheterization (RHC), echocardiogram, 6-minute walk test (6-MWT), plasma NT-ProBNP (biomarkers of RV function) and plasma catecholamine (measure of sympathetic activation), and quality of life. Patients will be randomized to carvedilol (3.25 mg bid and escalated to 12.5 mg bid, as tolerated, over 3 months) or placebo in a 1:1 fashion. After 6 months, testing is repeated and patients are crossed over to the alternate treatment. Testing is repeated at the end of the study (month 13). Outcome measures will include the following:

1.1 Primary Efficacy Outcome: 6-month change in RVEF as measured by cardiac MRI. We hypothesize that carvedilol will increase RVEF. A 5-unit increase in RVEF will be considered meaningful, as this has been associated with improvement in survival.

1.2 Secondary Efficacy Outcomes: 6-month change in RV function (as measured by echocardiogram and RHC), exercise endurance (6-MWT), serum NT-ProBNP, plasma catecholamine levels, and quality of life (MLHF quality of life survey).

2. Primary Safety Outcome: Absence of hypotension (<90 mm Hg), bradycardia (<50 bpm or advanced atrioventricular nodal block), bronchospam, or acute decompensated right heart failure requiring hospitalization. We hypothesize that carvedilol will be safely tolerated by PAH patients.

These pilot data will enable us to implement a large phase III, placebo-controlled, randomized clinical trial. Carvedilol has great potential as a safe and effective therapy to improve long-term outcomes in PAH. Implementation and completion of this study will substantially enhance the applicant's expertise in conducting investigator initiated clinical trials while providing important preliminary data for an R01 application to further the goal of identifying targeted efficacious interventions for RV dysfunction in PAH.

A. Significance

Significance of RV dysfunction in PAH: Despite progress in understanding the pathogenesis and treatment of PAH, it remains a fatal illness[1]. The Food and Drug Administration (FDA) has approved 11 PAH-specific therapies. Most act on the pulmonary vasculature by inhibiting pulmonary vasoconstriction, while potentially decreasing pulmonary vascular smooth muscle proliferation and endothelial cellular proliferation. None have a proven direct effect on the RV except for phosphodiesterase-5-inhibitors, which may have a RV inotropic effect but this has not been confirmed in humans[2]. These therapies produce a modest improvement in functional capacity, and overall survival data are equivocal[3,4].

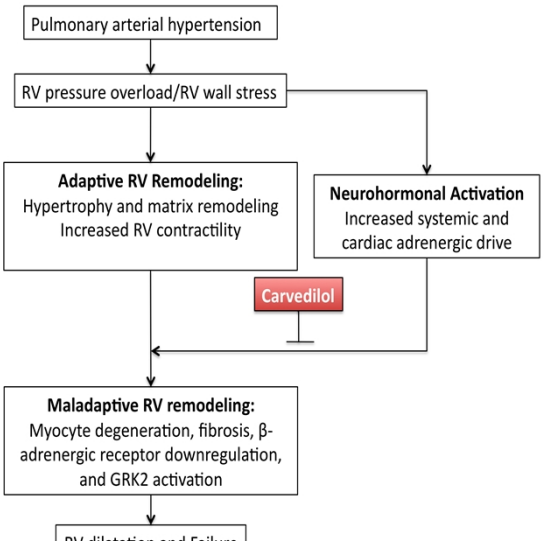
Initial RV hypertrophy in PAH is an adaptive response to the increased afterload with preserved contractile function. As the disease progresses, the RV dilates due to maladaptive remodeling and eventually fails [5]. The mechanisms that switch the adaptive, compensatory hypertrophy of the RV to maladaptive RV dilatation and ultimately RV failure are under investigation[6] [7]. Observational PAH registries and clinical trial data have consistently shown that baseline RV function, including RV ejection fraction, cardiac output, and right atrial pressure, is the major predictor of long-term mortality in patients with PAH [1,8-11]. A recent metaanalysis of prognostic factors in PAH reported that right atrial pressure was the most commonly reported parameters to be associated with mortality in patients with PAH. In fact, the RV response to therapy rather than the change in pulmonary vascular resistance (PVR) is the key determinant of clinical outcomes in PAH [12]. Despite frequency of PAH patient death from RV failure, RV function has not been investigated as a potential additional treatment target. ***Thus, there is an unmet need for novel cardiac-specific therapies preventing or reversing RV dysfunction in PAH***[5].

Neurohormonal Activation and RV Dysfunction in PAH: Neurohormonal activation has been proposed as a mechanism for the switch from adaptive to maladaptive RV remodeling in PAH (Figure

1). Similar to left ventricular systolic dysfunction, neurohormonal activation has been observed in isolated RV failure due to PAH in both experimental animal models and human PAH[13 6,14-17]. Over time, this high catecholamine-state leads to significant down-regulation and desensitization of the β-adrenergic receptors in the failing RV in PAH through activation of G-receptor protein kinase 2 (GRK2) [6,18]. Eventually, this leads to many adverse consequences, including a reduction in RV contractility and reduced responsiveness to exogenous catecholamine stimuli [6].

Pharmacological blockade of the β-adrenergic receptors is one of the main treatment strategies in left ventricular systolic dysfunction. Multiple, large, randomized, double-blind controlled trials have demonstrated that long-term beta-blockade therapy in left ventricular systolic dysfunction improves cardiac function and reduces maladaptive ventricular remodeling and sudden arrhythmias, ultimately leading to improved quality of life and survival[19-22]. By analogy, a β-adrenergic receptor inhibitor may have therapeutic benefit in RV failure due to PAH.

Figure 1: Neurohormal Activation in PAH



Inhibition of Neurohormonal Activation in Animal Models of PAH: Pre-clinical animal experiments demonstrate that inhibition of neurohormonal activation by chronic β-adrenergic receptor blockade improves RV function, reverses RV remodeling, and restores RV β-receptor signaling pathways [6,23-26]. Chronic treatment with arotinolol (a pure α and β adrenergic receptor blocker) in a rat model of monocrotaline (MCT)-induced PAH prevented development of PAH and RV hypertrophy[25]. Galein, a small molecule inhibitor of GRK2, decreased GRK2 activation and improved RV function in two experimental models of PAH with RV failure[6]. Bisoprolol (a $ß_1$-cardioselective blocker) improved RV function in MCT-induced PAH; this improvement in RV function was associated with reductions in RV fibrosis and inflammation, and restoration of RV β-adrenergic signaling[24]. In Sugen/hypoxia and MCT rat models of PAH, treatment with carvedilol (a non-selective $\alpha_1/\beta_1/\beta_2$-adrenergic receptor antagonist) reduced RV hypertrophy, improved RV function, and reversed RV remodeling, resulting in improved exercise endurance and survival[23]. The improvement in survival with carvedilol treatment was impressive: 50% of the vehicle-treated rats died, whereas all rats in the carvedilol-treated group survived. The improvement in RV function was associated with reduction in RV fibrosis and capillary rarefaction, increased reactivation of fetal genes with a pronounced switch from α to β myocyte heavy chain, and normalization of calcium handling protein Serca2a. The improvement in RV function with carvedilol in this model was due to a direct effect of carvedilol on the RV as there was no change in the severity of the angioproliferative pulmonary vasculopathy. Metoprolol, a cardio-selective β-blocker with no α-blocking properties, had similar but less pronounced effects in the SU5416 and hypoxia model. This finding, along with the significant improvement in survival with carvedilol treatment, suggests that the α-blocking and the anti-oxidant properties of carvedilol may have an additional beneficial role.

Based on the beneficial effects observed in preclinical animal experiments, we hypothesize that long-term pharmacological blockade of the β-adrenergic receptors with carvedilol will reverse RV remodeling and improve RV function in patients with PAH and RV dysfunction. By improving RV function, chronic treatment with carvedilol will improve functional capacity and survival in patients with PAH. ***To test this hypothesis, we propose a*** randomized, phase 2, placebo-controlled, double-blinded, crossover trial of carvedilol ***in 26 PAH patients with World Health Organization functional class II or III symptoms and RVEF < 45% for 6 months***.

B. Innovation

Currently β-blockers are contraindicated in patients with PAH because of a theoretical concern that their negative inotropic properties will lead to worsening right ventricular function and ultimately failure. However, this is based on a small study demonstrating improved exercise capacity and hemodynamics after withdrawal of propranolol in 10 patients with porto-pulmonary hypertension[27]. In addition, this study lacked a sensitivity analysis with a long-term comparison between those who continued beta-blocker use and those who discontinued it. Moreover, 8 of the 10 patients in this study were receiving the first generation, non-selective beta-blocker propranolol that perhaps has a greater bronchial and myocardial depressive effect than the currently available selective beta-blockers. ***Chronic* β-adrenergic receptor blockade with carvedilol *would provide a novel strategy for treatment of RV dysfunction in PAH patients, the important determinant of long-term outcomes, and thereby fills an important deficit in the management of PAH.*** Carvedilol has been available in the market for treatment of left ventricular systolic dysfunction for the last two decades and it has been well tolerated. Should the proposed phase-2 study confirm safety and suggest efficacy in PAH, carvedilol has the potential to move quickly to a Phase 3 clinical trial. Such a therapy would address a major unmet need for this orphan disease: an inexpensive oral therapy, which improves the RV.

One Phase 1/2 trial of β-blockers in patients with PAH was recently completed in the Netherlands (NCT01246037-PI Dr. A. Vonk Noordegraaf)[28]. This trial tested bisoprolol, a cardioselective β1-blocker. Although bisoprolol was safely tolerated, there was no significant improvement in RV function. Compared to bisoprolol, carvedilol has a superior profile with additional vasodilator and anti-oxidant properties[29,30]. Furthermore, carvedilol by acting as a “biased” ligand stimulates β-arrestin signaling in addition to antagonizing G-protein mediated signaling[31]. β-arrestin-dependent signaling is cardioprotective in the presence of chronic catecholamine stimulation[32]. Consistent with this, the beneficial effects observed with carvedilol in PAH animal experiments were impressive with a 50% reduction in mortality[23]. Furthermore, the Netherlands trial included patients with both normal and reduced RVEF. However, similar to left ventricular systolic dysfunction, it possible that beta-blockers are effective specifically in patients with reduced RVEF. Thus, we chose to study carvedilol in PAH patients with RVEF < 45%. Currently, there is another clinical trial studying the role of carvedilol in PAH (NCT01586156-PI Dr. Samara Farha). Our study differs from theirs critically in ours is a crossover design restricted to only patients with WHO category I PAH. This is important, as there is significant heterogeneity among patients with various WHO categories of pulmonary hypertension, which could potentially influence the results. We also have different end points (RVEF by cardiac MRI and invasive hemodynamics) and test different biomarkers (plasma catecholamines). These trials are complementary.

C. Preliminary Work

C.1. Beta-blocker therapy is not associated with adverse outcomes in PAH: a propensity score analysis: We recently showed that beta-blocker therapy is not associated with increased long-term outcomes in PAH using propensity score-matching analysis[33]. We studied patients in the Pulmonary Hypertension Connection (PHC) registry to assess the safety of β-blockers in patients with isolated RV failure due to PAH[33]. PHC, which was initiated in March 2004, enrolled all patients evaluated at a single United States practice over time at 3 different university hospitals between 1982 and 2014.

Methods: Over the study period, 4 physicians acquired all the clinical data.

Data were collected by chart review and entered with an Internet-based electronic data capture system. Patients were entered retrospectively from 1982 to February 2004 and prospectively from March 2004 through 2013. From the PHC registry, we identified all adult patients (> 18 years of age) with PAH (n = 564). Of the 564 patients, 71 patients with PAH were on chronic β-blocker therapy at the time of presentation. We compared baseline clinical, echocardiographic, and hemodynamic characteristics, and long-term survival in PAH patients with (n=71) and without beta-blocker (n=493) use at baseline. Propensity score-matching was used to match pairs of PAH patients with and without beta-blocker use (matched cohort). We compared all-cause mortality between the groups in the total cohort and the matched cohort using bootstrap validation, Kaplan-Meier, and Cox proportional hazard analyses.

Table 1: Baseline Characteristics by Beta-blocker Use

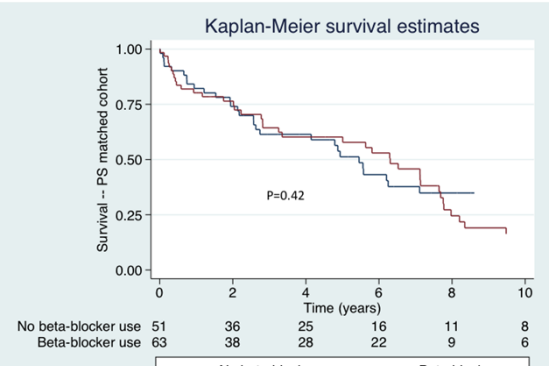
Characteristics	Total Cohort			Matched Cohort		
	Beta-Blocker			Beta-blocker		
	Yes	No	P-value	Yes	No	P-value
	(N=71)	(N=493)		(N=63)	(N=51)	
Age	59 ± 14	55 ± 14	0.02	58 ± 14	59 ± 15	0.63
Female	54 (76.1)	400 (81)	0.34	48 (76)	39 (77)	0.427
Etiology			0.01			0.91
Idiopathic	23 (33)	227 (46)		21 (33)	15 (29)	
CTD	24 (34)	155 (31)		23 (37)	17 (33)	
CHD	6 (9)	36 (7)		5 (8)	7 (14)	
Portopulmonary	15 (21)	37 (8)		11 (17.5)	9 (18)	
Anorexigen	1 (1)	20 (4)		1 (1.5)	0 (0)	
Familial	1 (1)	16 (3)		1 (1.5)	2 (4)	
Others	1 (1)	2 (1)		1 (1.5)	1 (2)	
WHO FC III/IV	58 (81.7)	428 (86.8)	0.24	53 (84.1)	43 (84.3)	0.97
Hypertension	48 (67.6)	141 (28.6)	<0.01	40 (63.5)	30 (58.8)	0.61
Diabetes	7 (9.9)	36 (7.3)	0.45	7 (11.1)	7 (13.7)	0.67
History of Obesity	27 (38.0)	87 (17.7)	<0.01	21 (33.3)	14 (27.5)	0.50
CAD	9 (12.7)	25 (5.1)	0.01	7 (11.1)	6 (11.8)	0.91
Medications						

Results: Comparison of Clinical, Echocardiographic, and Hemodynamic Characteristics (Table 1): PAH patients on chronic β-blocker therapy were older; more often had diabetes, coronary artery disease, and obesity; had worse renal function; were more often on diuretics, digoxin, and angiotensin converting enzyme inhibitors; and more frequently had left ventricular and RV hypertrophy than patients not taking β-blockers. Baseline heart rate was lower in patients taking β-blockers. The severity of pulmonary hypertension and RV dysfunction was not different between those with and without beta-blocker use. After propensity matching, 63 patients with beta-blocker use were compared to 51 patients without beta-blocker use.

Comparison of Long-term Outcomes: During a median follow-up time of 4.8 years, there were 339 (60%) deaths in the total cohort and 70 deaths (61%) in the matched cohort. There was no difference in absolute mortality between those with and without beta-blockers (P=0.71). Beta-blocker use was not associated with increased all cause-mortality in the total cohort after adjusting for propensity score (adjusted HR 1.0, 95% CI 0.7-1.5) and in the matched cohort (HR 1.2, 95% CI 0.8-2.0) (Figure 2).

Conclusion: There was no statistically significant difference in long-term mortality between propensity score-matched pairs of PAH patients with and without beta-blocker use.

Figure 2: Comparison of Survival by Beta-blocker Use in the Propensity Score (PS) Matched Cohort



C.2. Safety and feasibility of carvedilol in PAH patients with RV dysfunction: In a recent small, open label trial of 6 patients with PAH, carvedilol was safely tolerated without major side effects[34]. The median dose tolerated was 18.75 mg bid with three patients reaching the maximum dose of 25 mg bid. None of the patients had to down titrate or discontinue carvedilol. In addition, carvedilol improved RVEF and RV stroke volume, due to a reduction in RV end-systolic volume and without changes in RV end-diastolic volume. There was also a trend towards improvement in 6-MWD.

We currently have an ongoing phase I/2 open label trial to assess the safety and feasibility of carvedilol therapy in patients with PAH and RV dysfunction (NCT02120339 –PI: Thenappan Thenappan, MD). This trial was initiated in August 2014 and we have enrolled 5 patients. DSBM reviewed the interim analysis of the first five patients and recommended enrolling more patients to see a definite signal for efficacy and safety of carvedilol for RV dysfunction in PAH.

C.3. Summary: These preliminary data suggest that beta-adrenergic blockade with carvedilol is safe and feasible in patients with PAH. This is also a testament for the feasibility of an investigator-initiated phase 1/2 trial by our team at University of Minnesota.

3.0 Investigational Agent(s)

3.1 Description / Indications

Carvedilol, a non-selective α1/β1/β2-adrenergic receptor antagonist, will be used in this trial to evaluate the effect of chronic beta-adrenergic receptor blockade in patients with PAH. Carvedilol is approved by the FDA, and it is currently used in the treatment of left ventricular systolic dysfunction and hypertension. It is well tolerated and has minimal side effects. Carvedilol will be used in this study in patients with right ventricular failure due to PAH, which is not a FDA approved indication. Investigational new drug application has been filled and permission has been already. Patients with PAH routinely take carvedilol for other indications and it is well tolerated.

3.2 Placebo / Active Control Agent

This is a placebo controlled crossover trial. Hence, subjects will be randomized to carvedilol vs. placebo in a 1: 1 fashion. After 6 months, testing is repeated and patients are crossed over to the alternate treatment.

3.3 Blinding of Study Drug

The subjects, investigator team, and the treating physicians will be blinded to the study drug (carvedilol vs. placebo) until the analysis of the month 12 (primary endpoint) data is complete for all subjects. The investigational drug pharmacy at University of Minnesota will be employed to manage the conduct of the study.

3.4 Drug Accountability

The investigation drug service (IDS) pharmacy will be used to maintain a central log documenting screening, implement stratification and randomization, assess current inventories of the study drug, initiate any necessary resupply of study drugs, and to document discontinuation of the study drug.

The investigational pharmacy at University of Minnesota will provide adequate doses of the study drug in a double-blinded fashion (carvedilol vs. placebo) during each study visit. The study drug supply will be replenished at the baseline, 4 week, 8 week, 12 week, month 4 and month 5 study visits. The drug will be stored at room temperature, away from excess heat and moisture. The investigational pharmacy will assign bottle numbers and provide instructions for dispensing of blinded study drug.

Study drug accountability: Accountability of the study drug is the responsibility of the principal investigator. The investigator will ensure that the study drug is used in accordance with the protocol. The investigator may choose to assign some of the drug accountability responsibilities to a pharmacist or other appropriate individual. Drug accountability records indicating the drug's

delivery date, use by each subject and end of study destruction and disposal of the drug will be maintained. These records will adequately document that the subjects were provided the doses as specified in the protocol.

4.0 Study Objectives

The goal of this study is to evaluate the effect of long-term inhibition of neurohormonal activation with carvedilol, a nonselective α1/β1/β2-adrenergic receptor antagonist, on RV reverse remodeling and function. In addition, we will determine the safety and tolerability of long-term carvedilol therapy in PAH patients.

General Hypothesis and Outcome Measures

By inhibiting neurohormonal activation, carvedilol, will cause reverse remodeling of the RV and improve its function as measured by Cardiac MRI in PAH patients.

Specific Aim: Estimate the efficacy and safety of long-term β-adrenergic receptor blockade using carvedilol on RV function in PAH patients

4.1. Primary Objective:

1. The primary efficacy outcome is the mean change in RVEF as measured by cardiac MRI before and after 6 months of treatment. An improvement of 5% will be considered to be clinically significant. Assessment of the RV is challenging due to its complex geometry. Cardiac MRI offers the ability to acquire 3-dimensional datasets that do not require geometric modeling. In addition to being highly reproducible[35], RVEF measured by cardiac MRI can be used to identify PAH patients that are likely to have clinical worsening[8]. The prognostic ability of cardiac MRI-measurements of RVEF is similar to that as mean pulmonary artery pressure and exercise capacity.
2. The primary safety outcome is the absence of adverse events associated with carvedilol including hypotension (<90 mm Hg), bradycardia (<50 bpm or advanced atrioventricular nodal block), bronchospam, or acute decompensated right heart failure requiring hospitalization.

4.2. Secondary Objective:
The following exploratory, secondary efficacy outcomes will be studied.

1. Mean change in tricuspid annular plane systolic excursion (TAPSE), a measure of RV function by transthoracic echocardiography[36]
2. Mean change in cardiac output and right-sided filling pressures measured by RHC
3. Mean change in exercise capacity assessed by 6-MWT
4. Mean change in plasma NT-ProBNP level, a biomarker of RV function in PAH[37,38]
5. Mean change in plasma catecholamine levels
6. Improvement in quality of life assessed by MLHF quality of life survey, which has been validated in PAH[40,41]

5.0 Study Design

5.1 Overview of Study Design

Study Type: Intervention
Study Design: Controlled,
Allocation: Randomized
End Point Classification: Safety/Efficacy study
Intervention Model: Study drug vs. placebo
Masking: Double-blinded
Primary purpose: Treatment
Intervention: The study drug (carvedilol vs. placebo) will be started in an escalating dose regimen as tolerated. Patients will be randomized to carvedilol (3.25 mg bid and escalated to 12.5 mg bid, as tolerated, over 3 months) or placebo in a 1:1 fashion. Carvedilol will be started at 3.125 mg twice a day. The dose will be increased as tolerated to 6.25 mg twice a day at week 4, 9.375 mg twice a day at week 8, and 12.5 mg twice a day at week 12 or the highest dose as tolerated by the patient. After 6 months, testing is repeated, study drug is weaned gradually over 2 weeks and patients are crossed over to the alternate treatment after an additional wash out period of 2 weeks. Testing is repeated at the end of the study (month 13).

Estimated number of patients: 26

5.2 Anticipated Duration of the Clinical Investigation

The anticipated duration of the study from enrollment of the first subject to completion of the final report to the IRB is approximately 3 years. The anticipated accrual rate is approximately 1 patient a month.

5.3 Evaluation Criteria

5.3.1. Primary endpoint:

1. The primary efficacy outcome is the mean change in RVEF as measured by cardiac MRI before and after 6 months of treatment. An improvement of 5% will be considered to be clinically significant. Assessment of the RV is challenging due to its complex geometry. Cardiac MRI offers the ability to acquire 3-dimensional datasets that do not require geometric modeling. In addition to being highly reproducible[35], RVEF measured by cardiac MRI can be used to identify PAH patients that are likely to have clinical worsening[8]. The prognostic ability of cardiac MRI-measurements of RVEF is similar to that as mean pulmonary artery pressure and exercise capacity.
2. The primary safety outcome is the absence of adverse events associated with carvedilol including hypotension (<90 mm Hg), bradycardia (<50 bpm or advanced atrioventricular nodal block), bronchospam, or acute decompensated right heart failure requiring hospitalization.

5.3.2. Secondary endpoints:

The following exploratory, secondary efficacy outcomes will be studied.

1. Mean change in tricuspid annular plane systolic excursion (TAPSE), a measure of RV function by transthoracic echocardiography[36]
2. Mean change in cardiac output and right-sided filling pressures measured by RHC
3. Mean change in exercise capacity assessed by 6-MWT
4. Mean change in plasma NT-ProBNP level, a biomarker of RV function in PAH[37,38]
5. Mean change in plasma catecholamine levels
6. Improvement in quality of life assessed by Minnesota living with Heart Failure Questionnaire (MLHFQ), which has been previously used and validated in PAH[40,41]

5.4 Study Population

The study will enroll only patients with WHO category I PAH who meets the inclusion and exclusion criteria described below.

5.4.1 Sample Size

The effect of carvedilol therapy on RV function in patients with PAH is not well known. To date, no prior clinical trials have reported the effects of chronic β-blocker therapy on RV function secondary to PAH. Addetia et al showed that to detect a 5% difference in RVEF by cardiac MRI for 80% power and significance $\alpha<0.05$ in PAH patients, the number of subjects needed is 23[35]. Assuming a 10% drop out rate, we are planning to enroll 26 subjects to obtain at least 23 completed studies. This is a proof-of-concept trial, thus the goal of this study is to obtain data to estimate the variability of the outcome measures and obtain preliminary estimates of treatment effects in order to plan an appropriately powered, larger, phase 3, randomized, placebo-controlled trial. We will use the findings from the pilot study to refine our sample size calculation. We expect that a sample size of 26 will be sufficient to detect trends in echo parameters of RV function, hemodynamics, 6MWT, NT-ProBNP, plasma catecholamine, and MLHF score.

5.4.2 Subject Recruitment

Subjects will be recruited from the pulmonary hypertension clinic at the University of Minnesota who fulfill the inclusion and exclusion criteria.

5.4.3 Subject Screening

The study consists of a screening visit and a treatment period. Potential participants will be screened using data collected at their routine outpatient appointment at the pulmonary hypertension clinic at University of Minnesota. Eligible participants will complete the screening assessment including written consent, physical examination, medical history, electrocardiogram, blood to assess routine hematology, biochemistry, NT-ProBNP, 6 minute walk test 28 days prior to baseline, and transthoracic echocardiogram, cardiac MRI to assess baseline right ventricular function, and right heart catheterization (if none available within previous 3 months). Potential drug interactions with carvedilol will also be assessed. After completion of screening

assessment and obtaining written consent, patients will proceed to the baseline (week 0) study visit.

5.4.4 Prior and Concomitant Therapy

At screening visit and subsequent visits concomitant medications will be assessed for potential drug interactions with carvedilol. Subjects have to be on stable PAH-specific therapies for at least 3 months before enrolling in this clinical trial. Subjects will continue to take PAH-specific therapies during this clinical trial. No increase in current background PAH medication dose (oral, inhaled, Intravenous, subcutaneous) or addition of a new PAH medication will be allowed during the study duration of 12 months. If a subject needs up titration of PAH specific therapy due to progression of underlying pulmonary vascular disease, subjects will exit the study.

5.4.5 Inclusion Criteria

1. Subjects will be eligible to participate in the study if all of the following conditions exist:
2. Age > 18 years
3. WHO category 1 pulmonary arterial hypertension (Nice 2013)
4. WHO functional class II-III
5. RVEF by cardiac MRI < 45%
6. Stable on PAH-specific therapy as defined by no change in PAH-specific treatment and functional class in the past 3 months. Patient can be on either mono or combination PAH-specific therapy

5.4.6 Exclusion Criteria

Subjects will be excluded from participation in the study if any of the following conditions exist:

1. Significant persistent bradycardia (resting heart rate < 60 bpm) without a permanent pacemaker
2. Second or third degree AV block without a permanent pacemaker
3. Significant sinus tachycardia (resting heart rate > 110 bpm)
4. Use of anti-arrhythmic drugs
5. Hypotension defined as systolic blood pressure < 100 mmHg at the time of enrollment
6. Significant illness in the past 30 days requiring hospitalization
7. Acute decompensated right heart failure within past 30 days
8. Known allergy or intolerance to carvedilol or other β blockers
9. Cardiac index < 2 l/min/m2 or right atrial pressure > 15 mm Hg by right heart catheterization within last 3 months
10. Clinically significant severe asthma
11. Positive pregnancy test in patients of child bearing-potential

5.4.7 Exit/Discontinuation Criteria

Subjects will be discontinued from the study for the following reasons:

1. The subject no longer wishes to participate
2. Development of intolerable side-effects related to carvedilol

3. Development of significant hypotension (reduction in blood pressure associated with dizziness or syncope or SBP less than 100 mm Hg despite dose reduction)
4. Development of significant bradycardia despite dose reduction (HR < 50 bpm without a permanent pacemaker or reduction in heart rate associated with dizziness or syncope)
5. Development of decompensated right heart failure requiring hospitalization
6. Subject is a female of child-bearing potential who becomes pregnant
7. Subject death
8. Subject completes the protocol.
9. Subject is non-compliant with the protocol (See Section 6.8 for definition of non-compliance)
10. Subject's well-being, in the opinion of the Investigator would be compromised by study continuation.
11. DSMB and/or IRB recommendation

Patients who discontinue or wish to drop out early from the study will undergo final assessment at the time discontinuation.

6.0 Study Procedures

6.1 Consent Process

All subjects for this study will be provided a consent form describing this study and providing sufficient information for subjects to make an informed decision about their participation in this study. The consent form will be submitted with the protocol for review and approval by the IRB for the study. The formal consent of a subject, using the IRB-approved consent form, must be obtained before that subject is submitted to any study procedure. The subject or legally acceptable surrogate must sign this consent form, and the investigator-designated research professional obtaining the consent. A blank copy of the IRB-approved form must be kept on-site and by the sponsor-investigator.

6.2 Randomization Scheme

Subjects will complete the randomization visit within 4 weeks of the screening visit. After review of the inclusion and the exclusion criteria to confirm continued eligibility, subjects will be randomized to study drug in a 1:1 fashion to placebo vs. carvedilol administered twice daily.

Randomization will be done by the investigational pharmacy at University of Minnesota either in a sealed envelope or using block randomization scheme. The IDS pharmacy will maintain the randomization code until the completion of the study.

6.3 Blinding

6.3.1 Blinding procedure

The subjects, investigator team, and the treating physicians will be blinded to the study drug (carvedilol vs. placebo) until the analysis of the month 12 (primary endpoint) data is complete for all subjects. The investigational drug pharmacy at University of Minnesota will be employed to manage the conduct of the study.

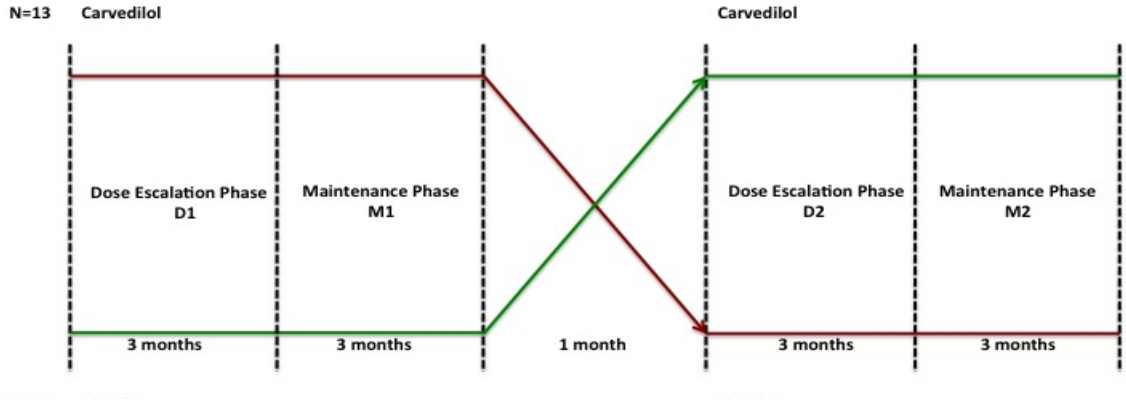
6.3.2 Unblinding procedure

In the event of a medical emergency where breaking the blind is required to provide medical care to the subjects, the investigators may obtain the treatment assignment directly from the investigational drug service pharmacy for that subject. Treatment assignment should remain blinded unless that knowledge is necessary to determine subject emergency medical care. The rationale for unblinding must be clearly explained in source documentation and on case report forms along with the data on which the treatment assignment was obtained.

6.4 Clinical Procedures

We propose a randomized, phase 2, placebo-controlled, double-blinded, crossover study to determine the safety and efficacy of carvedilol in in stable PAH patients with WHO functional class II or III symptoms and RVEF < 45% (Figure 3). Potential patients will be identified from our pulmonary hypertension clinic at the University of Minnesota.

Figure 3: Study Design



- Screening and Randomization: Following initial evaluation and consent, 26 patients meeting inclusion and exclusion criteria will be randomized to carvedilol or placebo in a 1: 1 ratio as the initial therapy in this 6-month crossover trial.

- Dose escalation phase (D1 and D2): Study drug (carvedilol or placebo) dose will be gradually escalated as tolerated every 4 weeks over a 3-month period. Carvedilol will be started on day 0 at 3.125 mg twice a day. The dose will be increased as tolerated to 6.25 mg twice a day at week 4, 9.375 mg twice a day at week 8, and 12.5 mg twice a day at week 12 as tolerated. Dose escalation will be stopped and the dose decreased to the previous dose (50% reduction) if the patient develops: 1. Systolic blood pressure < 100 mm Hg, 2. Heart rate < 55 bpm, or 3. Fluid retention requiring intravenous diuretics.

- Maintenance phase (M1 and M2): Patients will be continued on a stable dose of the study drug (maximum tolerated dose of carvedilol or placebo) for additional 3 months.

- Washout and Cross-over: After 6 months of treatment, the study drug will be tapered down over two weeks and after additional two weeks of washout period, patients will be crossed over to the alternative treatment for 6 months.

- Completion phase: The study drug dose (carvedilol or placebo) will be tapered down slowly over a 2-week period. All patients will have a follow up phone call at 4 weeks after completing the study to assess clinical stability.

Visit and assessment schedule:

Screening

Screening should be performed no more than 28 days prior to the baseline visit (TTE, cMRI and RHC within the previous 3 months). After going through the consent process and obtaining written consent, participant eligibility will be determined using data collected during their routine outpatient visit. These include the following:

- Obtain written consent
- Demographics
- Medical history
- Physical examination
- Concomitant medications
- Vital signs
- EKG
- WHO functional class
- 6 minute walk test
- Transthoracic echocardiogram
- Blood samples for routine hematology, blood chemistry and NT-ProBNP
- Cardiac MRI for assessment of right ventricular function (right ventricular size, volumes, and ejection fraction)
- Right heart catheterization

Baseline visit (Week 0)

The following will be obtained during baseline visit.

- Medical history
- Physical examination
- Concomitant medications
- Vital signs
- EKG
- Plasma catecholamine levels
- Quality of life assessment with MLHF questionnaire
- Pregnancy test if subject is a female of child-bearing potential

After obtaining above procedures subject will be randomized 1: 1 to placebo vs. carvedilol 3.125 mg orally twice a day.

Week 2

The following assessments will be done as an outpatient.

- Medical history
- Physical examination
- Concomitant medications
- Vital signs
- EKG
- WHO functional class

Week 4

The following assessments will be done as an outpatient.

- Medical history
- Physical examination
- Concomitant medications
- Vital signs
- EKG
- WHO functional class
- If patient is tolerating the study drug, dose will be increased to 2 pills (placebo or carvedilol 3.125 mg) twice a day

Week 6

The following assessments will be done as an outpatient.

- Medical history
- Physical examination
- Concomitant medications
- Vital signs
- EKG
- WHO functional class

Week 8

The following assessments will be done as an outpatient.

- Medical history
- Physical examination
- Concomitant medications
- Vital signs
- EKG
- WHO functional class

- If patient is tolerating the study drug, dose will be increased to 3 pills (placebo or carvedilol 3.125 mg) twice a day

Week 10

The following assessments will be done as an outpatient.

- Medical history
- Physical examination
- Concomitant medications
- Vital signs
- EKG
- WHO functional class

Week 12

The following assessments will be done as an outpatient.

- Medical history
- Physical examination
- Concomitant medications
- Vital signs
- EKG
- WHO functional class
- 6 minute walk test
- Blood samples for routine hematology, blood chemistry and NT-ProBNP
- If patient is tolerating the study drug, dose will be increased to 1 pill (placebo or carvedilol 12.5 mg) twice a day

Month 4

The following assessments will be done as an outpatient.

- Medical history
- Physical examination
- Concomitant medications
- Vital signs
- EKG
- WHO functional class

Month 5

The following assessments will be done as an outpatient.

- Medical history
- Physical examination

- Concomitant medications
- Vital signs
- EKG
- WHO functional class

Month 6

The following will be obtained during baseline visit.

- Medical history
- Physical examination
- Concomitant medications
- Vital signs
- EKG
- WHO functional class
- 6 minute walk test
- Transthoracic echocardiogram
- Blood samples for routine hematology, blood chemistry and NT-ProBNP
- Cardiac MRI for assessment of right ventricular function (right ventricular size, volumes, and ejection fraction)
- Right heart catheterization
- Quality of life assessment with MLHF questionnaire
- Plasma catecholamine levels

6 months of treatment, the study drug will be tapered down over two weeks and after additional two weeks of washout period, patients will be crossed over to the alternative treatment for 6 months.

All study clinic visits will be scheduled within ± 5 days of the actual expected calendar date as per the protocol.

6.4 Follow-Up Procedures

Study Assessments:

Physical examination and medical history:

Physical examination will be performed to ensure suitability according to the inclusion and exclusion criteria at screening and to document the health status at the time points specified in the schedule of events. The physical examination comprises measurement of height and body weight and a routine medical examination.

A detailed medical history will be recorded at screening and brief history will be taken at other visits. The medical history will elicit information concerning existing medical conditions, major illnesses, and related surgical procedures. Any prescribed or over the counter medications that the subject received within the past 30 days should be recorded. Subjects will be instructed to notify the study doctor before beginning new prescribed or over the counter medications.

EKG: Electrocardiogram will be formed during each study visit to exclude bradyarrythmias

RV Function by Cardiac MRI: Cardiac MRI will be performed on a Siemens 1.5 T Sonata scanner (Siemens Medical Solutions, Erlangen, Germany) with simultaneous EKG recording according to a protocol described previously. Short-axis cine images of the heart from apex to base will be acquired, covering the whole RV. The endo- and epicardial contours of the RV will be delineated manually by a blinded observer and processed using specialized software to obtain RV mass, end-systolic, and end-diastolic volume. Three measurements of RV wall thickness will be obtained for the RV free wall at the mid-ventricular level and the mean value will be calculated. Stroke volume will be determined from the flow in the main pulmonary artery in an image plane positioned perpendicular to the main pulmonary artery using a two-dimensional, spoiled gradient-echo pulse sequence, and one-dimensional velocity-encoding signal parallel to the flow in the pulmonary artery. Cardiac output will be determined by multiplying stroke volume by heart rate. Parameters are indexed by correcting for body surface area. RV ejection fraction will be determined by dividing stroke volume by RV end-diastolic volume.

RV Function by Echocardiography: Echocardiograms will be performed according to standard American Society of Echocardiography criteria using commercially available ultrasound machines. Cardiac chamber sizes will be assessed from multiple views. RV outflow tract flow will be assessed with the sample volume positioned immediately below the pulmonic valve in the parasternal short-axis view with pulse wave and continuous wave Doppler. Analysis of the pulsed wave Doppler signal in the RV outflow tract will allow for measurement of the velocity time integral (surrogate of stroke volume), time to peak velocity (acceleration time) and for categorization of the basic morphology of the Doppler signal (no notch, late systolic notch, mid-systolic notch). Color Doppler will be used to assess the degree of tricuspid regurgitation in multiple views. The highest right atrial to RV gradient from continuous wave Doppler will be used as an estimate of RV systolic pressure. Tricuspid annular plane systolic excursion will be measured using either M-mode imaging or two-dimensional imaging from the apical four-chamber view. Tissue Doppler imaging will be used for assessment of RV systolic and diastolic performance.

Right heart catheterization (RHC): Patients will undergo RHC using standard techniques. With the patient supine, the pressure transducers will be leveled at the mid-thorax. Mild sedation will be administered per patient request. The RHC will utilize a balloon flow-directed thermodilution catheter that will be placed in the PA. The catheter will be advanced to the pulmonary wedge position and validated by hemodynamic tracings, fluoroscopy, and blood sampling for oxygen saturation if needed. Measurements of mean right atrial pressure, pulmonary artery systolic, diastolic and mean pressure, and pulmonary wedge pressure will be recorded on paper for 5 seconds and measured at end-expiration. Thermodilution cardiac output will be measured in triplicate and averaged. O2 saturation will be measured from the pulmonary artery with a co-oximeter. Systemic blood pressure will be determined by cuff manometry and systemic oxygen saturation by pulse oximetry.

6MWT: The 6MWT will be performed according to American Thoracic society guidelines[42]. Briefly, the 6MWT is performed in an enclosed, level, measured corridor. Subjects will be instructed to walk at their own pace but to cover as much ground as possible in 6 minutes. The supervising study personnel accompanies the subject and encourages them with the standardized statements, "You are doing well" or "Keep up the good work," but avoids other phrases of encouragement.

Plasma Catecholamine levels: plasma catecholamine levels will be measured on blood samples as a send out test at ARUP Reference Labs in Utah.

Plasma NT-ProBNP: Plasma NT-proBNP testing will be performed at the University of Minnesota Medical Center laboratory.

Quality of Life: We will use the Minnesota Living with Heart Failure Questionnaire for assessing quality of life. Minnesota Living with Heart Failure survey has been used in previous PAH clinical trials and has also been validated as a reliable measure of quality of life in PAH patients[40,41].

6.7 Study Timetable / Schedule of Events

Assessment	Baseline	Week 2	Week 4	Week 6	Week 8	Week 10	Week 12	4-month	5-month	6-month		Week 2	Week 4	Week 6	Week 8	Week 10	Week 12	4-month	5-month	6-month
Clinical assessment	X	x	x	x	x	x	x	x	x	X		x	x	x	x	x	x	x	x	X
EKG	X	x	x	x	x	x	x	x	x	X		x	x	x	x	x	x	x	x	X
Echo	X									X										X
6MWT	X									X										X
cardiac MRI	X									X	crossover									X
RHC	X									X										X
Plasma NT-proBNP	X									X										X
Serum catecholamine	X									X										X
CAMPHOR questionaire	X									X										X

6.8 Study Protocol Compliance / Treatment Adherence

We will do pill counts at each clinic visit as a measure of compliance. If a subject is non-compliant with taking his study drug for more than 2 weeks continuously, that subject will be excluded from the study. The subject will undergo final assessment including cardiac MRI and right heart catheterization at the time of exclusion from the study.

6.9 Deviations from the Clinical Protocol

When a deviation from the protocol is necessary for an individual subject, the investigator must complete a description of the deviation from the protocol and justification on the Protocol Deviation Form.

6.10 Protocol Amendments After Study Initiation

Should changes in the study plan or protocol become necessary in the course of the clinical trial, those specific changes will be discussed and agreed upon by the Sponsor, its acting representative if appropriate, Investigator, and appropriate IRB approval obtained before the changes are implemented. All changes must be documented as protocol amendments. For studies conducted under an IND, FDA approval and/or notification may be required in addition to the IRB approval.

6.11 Subject Withdrawal

6.11.1 How to Withdraw Subjects

Subjects will be discontinued from the study for the following reasons:

1. The subject no longer wishes to participate
2. Development of intolerable side-effects related to carvedilol
3. Development of significant hypotension despite dose reduction (reduction in blood pressure associated with dizziness or syncope or SBP less than 90 mm Hg)
4. Development of significant bradycardia despite dose reduction (HR < 50 bpm without a permanent pacemaker or reduction in heart rate associated with dizziness or syncope)
5. Development of decompensated right heart failure requiring intravenous diuretics or hospitalization

Patients who discontinue or wish to drop out early from the study will undergo final assessment at the time discontinuation. Carvedilol will be gradually tapered of over 4 weeks period unless the patient is having a serious adverse event.

6.11.2 Data Collection and Follow Up for Withdrawn Subjects

Patients who discontinue or wish to drop out early from the study will undergo final assessment at the time discontinuation including cardiac MRI and right heart catheterization. A subject will be considered lost to follow up if we are unable to reach the patient despite 5 phone calls to the subject or to the next-of-kin and 3 certified letters.

6.12 Subject Compensation

Subjects will receive a $25 gift card at the completion of the 6 month and 13-month visit.

7.0 Data Collection and Analysis

7.1 Subject Population(s) for Analysis

All-treated population: Any subject randomized into the study that received at least one exposure to study drug will be subjected to the study analysis – both for the primary analysis and any applicable secondary analyses.

7.2 Statistical Methods

The primary objective of this study is to determine the effect of carvedilol therapy on RV function in PAH patients. We will measure the mean change in RVEF before and after carvedilol therapy. We will compare this variable before and after carvedilol therapy using paired t-tests or Wilcoxon

signed-rank tests. The null hypothesis is that treatment with carvedilol has no effect on RVEF in patients with PAH with a two-sided alternative that carvedilol changes RVEF. Using a similar approach, we will also compare echo parameters of RV function, right heart hemodynamics, 6-minute walk distance, plasma NT-ProBNP, plasma catecholamine level, and MLHF questionnaire before and after carvedilol therapy.

8.0 Safety and Adverse Events

8.1 Definitions

Adverse Event (AE)

An adverse event (AE) is any symptom, sign, illness or experience that develops or worsens in severity during the course of the study. Intercurrent illnesses or injuries will be regarded as adverse events. Abnormal results of laboratory or diagnostic procedures are considered to be adverse events if the abnormality:

- Results in study withdrawal
- Is associated with a serious adverse event
- Is associated with clinical signs or symptoms
- Leads to additional treatment or to further diagnostic tests
- Is considered by the Investigator to be of clinical significance.

Adverse Reaction

An adverse reaction is any adverse event caused by a drug. Adverse reactions are a subset of suspected adverse reactions.

Suspected Adverse Reaction

A suspected adverse reaction is an adverse event for which there is a reasonable possibility that the drug caused the adverse event.

Serious Adverse Event (SAE)

A serious adverse event (SAE) is any adverse event that is:

- Fatal
- Life-threatening
- Requires or prolongs a hospital stay
- Results in persistent or significant disability or incapacity
- A congenital anomaly or birth defect

Important medical events are events that may not be immediately life threatening, but are clearly of major clinical significance and may be SAEs. They may jeopardize the subject, and may require intervention to prevent one or the other serious outcomes noted above.

Hospitalization

Hospitalization shall include any initial admission (even if less than 24 hours) to a healthcare facility as a result of a precipitating clinical adverse effect; to include transfer within the hospital to an intensive care unit. Hospitalization or prolongation of hospitalization in the absence of a precipitating, clinical adverse effect (e.g., for a preexisting condition not associated with a new adverse effect or with a worsening of the preexisting condition; admission for a protocol-specified procedure) is not, in itself, a serious adverse effect.

Expected Adverse Event
Expected adverse events are those that are known to be associated with or have the potential to arise as a consequence of participation in the study.

Unexpected Adverse Event
An adverse event or suspected adverse reaction is considered unexpected if it is not listed in the Investigator Brochure or Protocol at the specificity or severity that has been observed.

Unanticipated Problems Involving Risk To Subjects or Others (UPIRTSO)
An adverse event that in the opinion of the Principal Investigator is unexpected, related to the drug, and serious.

8.2 Recording of Adverse Events

At each contact with the subject, the investigator must seek information on adverse events by specific questioning and, as appropriate, by examination. Information on all adverse events should be recorded immediately in the source document, and also in the appropriate adverse event module of the case report form (CRF). All clearly related signs, symptoms, and abnormal diagnostic procedures results should be recorded in the source document, though should be grouped under one diagnosis.

All adverse events occurring during the study period must be recorded. The clinical course of each event should be followed until resolution, stabilization, or until it has been determined that the study treatment or participation is not the cause. Serious adverse events that are still ongoing at the end of the study period must be followed up to determine the final outcome. Any serious adverse event that occurs after the study period and is considered to be possibly related to the study treatment or study participation should be recorded and reported immediately.

8.3 Reporting of Serious Adverse Events

8.3.1 IRB Notification by Investigators

Reports of all serious adverse events (including follow-up information) must be submitted to the IRB within 5 working days if it falls under the UPIRTSO guidelines. Copies of each report and documentation of IRB notification and receipt will be kept in the Clinical Investigator's binder.

8.3.2 UPIRTSO Events

Investigators are required to submit a report of UPIRTSO events to the IRB within 5 working days of first learning of the event.

8.4 Safety Monitoring Plan

Subjects will be evaluated at the University of Minnesota PH clinic every 2 weeks initially for 3 months and then monthly for additional 3 months in each of the 6 month segments One of the PH team providers (Drs. Pritzker, Thenappan, Rebecca Cogswell) will evaluate subject for signs of right heart failure, bradycardia, and hypotension. Patients will also get electrocardiogram during each clinic visit to rule out bradyarrythmias including sinus bradycardia, atrioventricular nodal block, and new bundle branch block.

8.4.1 Anticipated Risks / Risk Mitigation

Risk associated with study procedures: Most of the procedures performed in this study are mostly part of the routine patient care. There is no risk associated with EKG, echocardiogram, cardiac MRI, and six-minute walk distance. The risk associated with right heart catheterization includes pain, infection, bleeding, pneumothorax, arrhythmias, pulmonary artery rupture, and death. Based on prospective and retrospective analyses, the rate of overall serious adverse events is around 1.1%. To minimize the risk of adverse events, an experience heart failure cardiologist or interventional cardiologist in the catheterization laboratory will perform the procedure using ultrasound and fluoroscopy guidance. The risk associated with phlebotomy includes discomfort, bleeding, or rarely, infection.

Risk associated with Carvedilol: The most serious adverse effects of carvedilol are hypotension, significant bradyarrythmias including advanced atrioventricular block, and worsening right heart failure due to negative inotropic effect. To mitigate these risks, we will exclude patients with resting heart rate < 60 beats per minute, second or third degree atrioventricular block, systolic blood pressure < 100 mm Hg, sinus tachycardia with a resting hear rate > 100 bpm, or if they have NYHA class IV symptoms or decompensated right heart failure. During the dose escalation and maintenance phase, subjects will be evaluated at the University of Minnesota PH clinic by one of the PH team providers (Drs. Pritzker, Thenappan, Cogswell) every 2 weeks initially for 3 months and then monthly for additional 3 months. We will decrease the carvedilol dose to the next lowest dose (50% reduction) if the heart rate is < 55 or systolic blood pressure is < 100 mm Hg. Subjects will be withdrawn if they develop advanced atriovenricular block or worsening right heart failure requiring hospitalization or intravenous diuretics. Subjects will be screened during each clinic visit with an EKG to detect asymptomatic bradyarrythmias. We will check serum NT-ProBNP, a marker of heart failure, every 3 months to screen for worsening right ventricular failure.

There is also the risk of loss of privacy, which will be minimized as follows. All the paper documents, including consent forms, will be stored in a locked drawer in a securely locked room. The database will be password protected and access will be granted only to approved investigators. For the analysis phase, the data will be de-identified prior to exporting it.

Written consent will be obtained in the patient's native language and the risks, benefits, and alternatives to participating in the study will be reviewed. Patients will be required to demonstrate adequate reasoning faculties and an understanding of the facts before consent is obtained.

Study Procedure	Anticipated Risks	Risk Mitigation
RHC	Pain, infection, bleeding, pneumothorax, arrhythmias, pulmonary artery rupture, and death	Experienced heart failure cardiologist will perform the procedure under ultrasound guidance
Phlebotomy	Discomfort, bleeding, or rarely, infection	Experienced phlebotomist will do the blood draw

8.4.2 Medical Monitoring for Participant Safety

The Principal Investigator will oversee the safety of the study, including careful assessment and appropriate reporting of adverse events as outlined in Section 7.4. Medical monitoring will include a regular assessment of the number and type of serious adverse events.

8.4.3 Study Stopping Rules

The study will be stopped if one of the following endpoints are reached:

A statistically significant treatment effect is demonstrated with regard to the primary outcome for any treatment arm relative to the others.

A statistically significant treatment effect cannot be achieved with regard to the primary outcome for the treatment arm relative to the others (analysis for futility).

Voluntary subject withdrawal in any arm of the study is significantly greater than expected, such that the statistical validity of the comparisons between arms is compromised.

The rate of severe adverse events in any arm of the study is such that continued participation would compromise subject safety.

8.4.4 Anticipated Adverse Events

The anticipated adverse effects of carvedilol are listed below:

Common adverse effects:

- Cardiovascular: Bradyarrhythmia (3%-10%), Hypotension (1.8% to 20.2%), Peripheral edema (1% to 7%).
- Endocrine metabolic: Abnormal weight gain (10% to 12%), Hyperglycemia (5% to 12%). Gastrointestinal: Diarrhea (2% to 12%).
- Neurologic: Dizziness (6% to 33%)

Serious Adverse Effects:

- Cardiovascular: Atrioventricular block (1% to 3%)
- Dermatologic: Erythema multiforme, Stevens-Johnson syndrome, Toxic epidermal necrolysis
- Hematologic: Aplastic anemia
- Respiratory: Asthma with status asthmaticus (rare)

8.5 Data Safety Monitoring Board

A data safety monitoring board consisting of Drs. Gary S Francis, Stephen L. Archer, Harm Boggard and Stuart Rich will review all serious events that occur during the course of the study, regardless of the treatment received by the subject and relationship to the drug. This board will review all data and safety. An interim analysis after 5, 10, and 15 patients have been enrolled will be performed and reports submitted per IRB safety monitoring guidelines.

A serious adverse event will be reported to the DSMB by the investigator by telephone, email, or fax within 24 hours of the event. A Serious Adverse Event (SAE) form must be completed by the investigator and either emailed or faxed to the DSMB within 24 hours. The investigator will

keep a copy of this SAE form on file at the study site. Report serious adverse events by phone and facsimile to:

Thenappan Thenappan, MD

Assistant Professor of Medicine

Cardiology Division

University of Minnesota

420 Delaware Street, SE MMC 508

Minneapolis, MN 55455

Phone: 612 624 8970

Fax: 612 626 4411

At the time of the initial report, the following information should be provided:

- Study identifier
- Subject number
- A description of the event
- Date of onset
- Current status
- Whether study treatment was discontinued
- The reason why the event is classified as serious
- Investigator assessment of the association between the event and study treatment

Within the following 48 hours, the investigator must provide further information on the serious adverse event in the form of a written narrative. This should include a copy of the completed Serious Adverse Event form, and any other diagnostic information that will assist the understanding of the event. Significant new information on ongoing serious adverse events will be provided promptly to the IRB and DSMB.

9.0 Data Handling and Record Keeping

9.1 Confidentiality

Information about study subjects will be kept confidential and managed according to the requirements of the Health Insurance Portability and Accountability Act of 1996 (HIPAA). Those regulations require a signed subject authorization informing the subject of the following:

- What protected health information (PHI) will be collected from subjects in this study
- Who will have access to that information and why
- Who will use or disclose that information
- The rights of a research subject to revoke their authorization for use of their PHI.

In the event that a subject revokes authorization to collect or use PHI, the investigator, by regulation, retains the ability to use all information collected prior to the revocation of subject authorization. For subjects that have revoked authorization to collect or use PHI, attempts should be made to obtain permission to collect at least vital status (i.e. that the subject is alive) at the end of their scheduled study period.

9.2 Source Documents

Source Data are the clinical findings and observations, laboratory and test data, and other information contained in *Source Documents*. *Source Documents* are the original records (and certified copies of original records); including, but not limited to, hospital medical records, physician or office charts, physician or nursing notes, subject diaries or evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, x-rays, etc. When applicable, information recorded on the CRF shall match the *Source Data* recorded on the *Source Documents*.

9.3 Case Report Forms

The study case report form (CRF) is the primary data collection instrument for the study. All data requested on the CRF must be recorded. All missing data must be explained. If a space on the CRF is left blank because the procedure was not done or the question was not asked, write “N/D”. If the item is not applicable to the individual case, write “N/A”. All entries should be printed legibly in black ink. If any entry error has been made, to correct such an error, draw a single straight line through the incorrect entry and enter the correct data above it. All such changes must be initialed and dated. DO NOT ERASE OR WHITE OUT ERRORS. For clarification of illegible or uncertain entries, print the clarification above the item, then initial and date it.

A Case Report Form will be completed for each subject enrolled into the clinical study. The investigator will review, approve and sign/date each completed CRF; the investigator’s signature serving as attestation of the investigator’s responsibility for ensuring that all clinical and laboratory data entered on the CRF are complete, accurate and authentic.

9.4 Clinical Reports

An annual progress report will be submitted to the IRB. Investigators will submit a final report of the clinical study to the reviewing IRB within 3 months of termination or completion of the clinical study or the Investigator’s part of the clinical study.

9.5 Records Retention

The investigator will retain the specified records and reports for up to 2 years from completion of IRB related work and 6 years for HIPAA.

10.0 Study Monitoring, Auditing, and Inspecting

This study will be monitored according to FDA/GCP guidelines. The Investigator will allocate adequate time for such monitoring activities. The Investigator will also ensure that the monitor or other compliance or quality assurance reviewer is given access to all study-related documents and study related facilities (e.g. pharmacy, diagnostic laboratory, etc.), and has adequate space to conduct the monitoring visit.

10.1 Study Monitoring Plan

Independent monitoring of the clinical study for clinical protocol and IDE application compliance will be conducted periodically (i.e., at a minimum of annually) by qualified staff of the University of Minnesota's Clinical and Translational Science Institute (CTSI) in accordance with the attached monitoring plan.

10.2 Quality Assurance Procedures

Weekly meetings of the study's principal investigator and staff will be held to discuss matters related to the safety of protocol participants, validity and integrity of the data, enrollment rate, retention of participants, adherence to protocol, and data completeness.

10.3 Auditing and Inspection

The investigator will permit study-related monitoring, audits, and inspections by the IRB, the sponsor, government regulatory bodies, and University compliance and quality assurance groups of all study related documents (e.g. source documents, regulatory documents, data collection instruments, study data etc.). The investigator will ensure the capability for inspections of applicable study-related facilities (e.g. pharmacy, diagnostic laboratory, etc.).

Participation as an investigator in this study implies acceptance of potential inspection by government regulatory authorities and applicable University compliance and quality assurance offices.

11.0 Administrative Study Information

11.1 Pre-Study Site Qualification

This is an investigator-initiated single center study.

11.2 Materials Provided by Sponsor / Coordinating Center

This is an investigator-initiated single center study.

12.0 Ethical Considerations

This study is to be conducted according to US and international standards of Good Clinical Practice (21 CFR 312 and International Conference on Harmonization guidelines), applicable government regulations and Institutional research policies and procedures.

This protocol and any amendments will be submitted to a properly constituted independent Institutional Review Board (IRB), in agreement with local legal prescriptions, for formal approval of the study conduct. The decision of the IRB concerning the conduct of the study will be made in writing to the investigator and a copy of this decision will be provided to the sponsor before commencement of this study.

13.0 Study Finances

13.1 Funding Source

This study is financed through American Heart Association Scientist Development Grant: 15SDG25560048 awarded to the PI – Dr. Thenappan Thenappan

13.2 Conflicts of Interest

Any investigator who has a conflict of interest with this study (patent ownership, royalties, or financial gain greater than the minimum allowable by their institution, etc.) must refer to the Regents Policies on Individual Conflict of Interest Policy or Institutional Conflict of Interest Policy. These policies require University Faculty and staff to report external professional activities and business and significant financial interests related to his or her University activities by submitting a REPA (Report of External Professional Activities) at least once per year. Faculty and staff should also file a REPA when substantial changes in business or financial interests occur, when an activity that presents a potential conflict of interest is anticipated, or when submitting an application for research support or technology transfer, submitting research protocols to the IRB, or receiving financial contributions. All University of Minnesota investigators will follow the University conflict of interest policy.

14.0 Publications Plan

The principal Investigator, Dr. Thenappan, holds the primary responsibility for publication of any results of the study. Neither the complete nor any part of the results of the study carried out under this protocol, nor any of the information provided by the principal investigator for the purposes of performing the study, will be published or passed on to any third party without the consent of the principal investigator. Any investigator involved with this study is obligated to provide the principal investigator with complete test results and all data derived from the study.

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