Statistical Analysis Plan: CY 6031

Title: A Phase 3, Multi-Center, Randomized, Double-blind, Placebo- controlled Trial to Evaluate the Efficacy and Safety of CK-3773274 in Adults with Symptomatic Hypertrophic Cardiomyopathy and Left Ventricular Outflow Tract Obstruction

NCT05186818

Approval Date: 2-Oct-2023

STATISTICAL ANALYSIS PLAN

VERSION: 2 Final

DATE OF PLAN:

October 2, 2023

STUDY DRUG:

CK-3773274 (aficamten)

PROTOCOL NUMBER:

CY 6031

STUDY TITLE:

A Phase 3, Multi-Center, Randomized, Double-blind, Placebo- controlled Trial to Evaluate the Efficacy and Safety of CK-3773274 in Adults with Symptomatic Hypertrophic Cardiomyopathy and Left Ventricular Outflow Tract Obstruction

BASED ON:

Protocol Amendment 3, 03 January 2023

SPONSOR:

Cytokinetics, Inc.
350 Oyster Point Blvd., South San Francisco, CA 94080
650-624-3000

This study is being conducted in compliance with good clinical practice, including the archiving of essential documents.

SIGNATURE PAGE

This document has been prepared and/or reviewed by*:

Amy Wohltman, ME, Director, Biostatistics Cytokinetics, Inc.	Signature	Date

This document has been reviewed and accepted by*:

[redacted], PhD, VP, Clinical Biometrics, Cytokinetics, Inc.	Signature	Date
[redacted], MD, VP, Clinical Research and Therapeutic Area Lead Cardiovascular, Cytokinetics, Inc.	Signature	Date
[redacted] MD, SVP, Chief Medical Officer, Cytokinetics, Inc.	Signature	Date
[redacted] SVP, Regulatory and Quality, Cytokinetics, Inc.	Signature	Date

*See electronic signatures at the end of the document.

TECHNICAL SUMMARY REPORT (TSR)

Name of Sponsor/Company Cytokinetics, Inc.	**Individual Study Table Referring to Part of the Dossier:** **Volume:**	***(For National Authority Use Only)*:**
Name of Finished Product: No generic or trade name assigned	**Page:**	
Name of Active Ingredient: Aficamten (CK-3773274)		
Title of Study: A Phase 3, Multi-Center, Randomized, Double-blind, Placebo- controlled Trial to Evaluate the Efficacy and Safety of CK-3773274 in Adults with Symptomatic Hypertrophic Cardiomyopathy and Left Ventricular Outflow Tract Obstruction		
Investigators: Study Center(s): Patients will be enrolled from approximately 105 sites worldwide.		
Studied period (years): 2022 to 2023	**Phase of development:** Phase 3	

Objectives and Endpoints:

Objectives	*Endpoint(s)*
Primary	
• To evaluate the effect of CK-3773274 on exercise capacity in patients with symptomatic oHCM	• Change in peak oxygen uptake (pVO2) by cardiopulmonary exercise testing (CPET) from baseline to Week 24
Secondary	
• To evaluate the effect of CK-3773274 on patient health status	• Change in Kansas City Cardiomyopathy Questionnaire – Clinical Summary Score (KCCQ-CSS) from baseline to Week 12 and Week 24
• To evaluate the effect of CK-3773274 on New York Heart Association (NYHA) Functional Classification	• Proportion of patients with ≥1 class improvement in NYHA Functional Class from baseline to Week 12 and Week 24
• To evaluate the effect of CK-3773274 on post-Valsalva left ventricular outflow tract gradients (LVOT-G)	• Change in post-Valsalva LVOT-G from baseline to Week 12 and Week 24 • Proportion of patients with post-Valsalva LVOT-G <30 mmHg at Week 12 and Week 24
• To evaluate the effect of CK-3773274 on duration of eligibility for septal reduction therapy	• Total duration of septal reduction therapy (SRT) eligible during the 24 Week treatment period in patients who were SRT eligible at baseline
• To evaluate the effect of CK-3773274 on exercise capacity	• Change in total workload during CPET from baseline to Week 24

Methodology:

This is a Phase 3 randomized, placebo-controlled, double-blind, multi-center trial in patients with symptomatic oHCM. Approximately 270 eligible patients will be randomized in a 1:1 ratio to receive CK-3773274 or placebo. Doses of 5, 10, 15, or 20 mg or matching placebo will be administered in an escalating manner using echocardiography to guide dose titration. Randomization will be stratified by use of beta-blockers and CPET exercise modality.

The trial will comprise three periods. The screening period will be up to 6 weeks in duration. The double-blind placebo-controlled treatment period will last 24 weeks. Following the final dose of investigational product (IP), there will be a 4-week safety follow-up period. IP will be administered orally once daily. During the initial six weeks of the treatment period, IP doses will be individually titrated at Weeks 2, 4, and 6 using echocardiography. Dose escalation at the Weeks 2, 4, and 6 visits will occur only if a patient has a post-Valsalva LVOT-G ≥30 mmHg and a biplane LVEF ≥55%. An echocardiogram will be performed at each subsequent visit during the trial and the dose down-titrated if necessary. The primary endpoint of pVO2 will be measured by CPET at screening and at end of treatment (Week 24).

Number of Subjects (planned and analyzed): Approximately 270 patients will be randomized to CK-3773274 or placebo at 1:1 ratio.

Diagnosis and main criteria for inclusion:

The key inclusion criteria are below. A full listing of eligibility criteria can be found in protocol Section 5.

- Males and females between 18 and 85 years of age, inclusive, at screening.
- Body mass index <35 kg/m^2.
- Diagnosed with HCM per the following criteria:
  − Has LV hypertrophy and non-dilated LV chamber in the absence of other cardiac disease and
  − Has an end-diastolic LV wall thickness as measured by the echocardiography core laboratory of:
    - ≥15 mm in one or more myocardial segments OR
    - ≥13 mm in one or more wall segments and a known-disease-causing gene mutation or positive family history of HCM
- Has resting LVOT-G ≥30 mmHg and post-Valsalva LVOT-G ≥50 mmHg during screening as determined by the echocardiography core laboratory.
- LVEF ≥60% at screening as determined by the echocardiography core laboratory.
- NYHA Functional Class II or III at screening.
- Hemoglobin ≥10g/dL at screening.
- Respiratory exchange ratio (RER) ≥1.05 and pVO_2 ≤90% predicted on the screening CPET per the core laboratory.
- Patients on beta-blockers, verapamil, diltiazem, or disopyramide should have been on stable doses for >6 weeks prior to randomization and anticipate remaining on the same medication regimen during the trial. Patients treated with disopyramide must also be concomitantly treated with a beta blocker and/or calcium channel blocker

Test product, dose and mode of administration:

Aficamten will be administered orally once daily with or without food. Patients receiving CK-3773274 start at a dose of 5 mg once daily and may escalate through doses of 10, 15, and 20 mg once daily during the initial six weeks of treatment if they continue to meet the escalation criteria (post-Valsalva LVOT-G ≥30 mmHg and a biplane LVEF ≥55%) or will stop at their current dose when escalation criteria are not met.

Duration of treatment:

After signing the informed consent form, patients will complete assessments to determine trial eligibility during a screening period of up to 6 weeks in duration. The double-blind placebo-controlled treatment period will last 24 weeks. Following the final dose of IP, there will be a 4-week safety follow-up period.

Reference therapy, dose and mode of administration:

Doses of 5, 10, 15, or 20 mg or matching placebo will be administered in an escalating manner using echocardiography to guide dose titration.

Criteria for evaluation (see protocol Section 3):

Efficacy:

The primary efficacy endpoint is change in peak oxygen uptake (pVO_2) by cardiopulmonary exercise testing (CPET) from baseline to Week 24.

The secondary endpoints are as follows:

- Change in Kansas City Cardiomyopathy Questionnaire – Clinical Summary Score (KCCQ-CSS) from baseline to Week 12 and Week 24
- Proportion of patients with ≥1 class improvement in NYHA Functional Class from baseline to Week 12 and Week 24
- Change in post-Valsalva LVOT-G from baseline to Week 12 and Week 24
- Proportion of patients with post-Valsalva LVOT-G <30 mmHg at Weeks 12 and 24
- Change in total workload during CPET from baseline to Week 24
- Total duration of SRT eligible during the 24 Week treatment period in patients who were SRT eligible at baseline

Safety:

- Incidence of reported major adverse cardiac events (cardiovascular [CV] death, cardiac arrest, non-fatal stroke, non-fatal myocardial infarction, CV hospitalization)
- Incidence of new onset persistent atrial fibrillation
- Incidence of appropriate implantable cardiac defibrillator (ICD) discharges and aborted sudden cardiac death
- Incidence of left ventricular ejection fraction (LVEF) <50%
- Incidence of treatment emergent adverse events

Statistical methods:

Unless specified otherwise, efficacy analyses will be performed on the full analysis set (FAS), which includes all randomized patients. The primary analysis will test the null hypothesis that there is no treatment difference in the primary endpoint between patients randomized to placebo and those randomized to CK-3773274 in the FAS. Change from baseline in pVO2 will be analyzed using an ANCOVA model with treatment group, randomization stratification factors, baseline pVO2 and baseline weight as covariates.

For preservation of the overall type I error rate at two-sided 0.05 for the primary and secondary endpoints, the primary and secondary endpoints will be tested in the following specified order using a closed testing procedure. The primary endpoint is tested first at two-sided 0.05. If the primary endpoint achieves statistical significance at two-sided $p<0.05$, then the secondary endpoints will be tested at two-sided 0.05, with their testing being in the sequential order of KCCQ-CSS change from baseline, proportion of patients with ≥1 NYHA functional class improvement, post-Valsalva LVOT-G change from baseline, and proportion of patients with post-Valsalva LVOT-G < 30 mmHg; duration of SRT eligibility for participants who are SRT eligible at baseline, for each after 24 weeks of treatment; then KCCQ-CSS change from baseline, proportion of patients with ≥1 NYHA functional class improvement, post-Valsalva LVOT-G change from baseline, and proportion of patients with post-Valsalva LVOT-G < 30 mmHg, for each after 12 weeks of treatment; and lastly change from baseline to Week 24 in total workload. SRT eligibility is defined as resting or post-Valsalva LVOT-G ≥ 50 mmHg AND NYHA Functional Class ≥ 3. See below table for illustration of the testing order.

Primary Endpoint		Significance Level
Step 1	pVO_2	0.05
Secondary Endpoints		↓
Step 2	KCCQ-CSS (24 wk)	0.05
		↓
Step 3	NYHA Class (24 wk)	0.05
		↓
Step 4	Valsalva Gradient (24 wk)	0.05
		↓
Step 5	%Valsalva Gradient (24 wk)	0.05
		↓
Step 6	Duration SRT Eligible (24 wk)	0.05
		↓
Step 7	KCCQ-CSS (12 wk)	0.05
		↓
Step 8	NYHA Class (12 wk)	0.05
		↓
Step 9	Valsalva Gradient (12 wk)	0.05
		↓
Step 10	%Valsalva Gradient (12 wk)	0.05
		↓
Step 11	Total workload (24 wk)	0.05

Multiplicity will be detailed in Section 7.6.

The proportion of responders in various exploratory endpoints in FAS will be analyzed using Cochran–Mantel–Haenszel (CMH) test stratified by randomization factors. The p-value and 95% confidence interval (CI) will be obtained using exact method. Other change from baseline endpoints will be

analyzed using mixed measures repeated model with treatment, visit, randomization stratification factors, treatment by visit, baseline by visit interaction as fixed effect and baseline assessment as covariate. Total of SRT eligible will be analyzed using an ANCOVA model with treatment group and randomization stratification factor beta blocker use/no use as fixed effects adjusting for significant baseline characteristics.

Safety analyses will be performed on the safety analysis set (SAS) which includes all patients who received at least one dose of IP. The pharmacokinetics analysis set (PKS) will consist of patients who have at least one evaluable plasma concentration of CK-3773274.

The number and percentage of patients reporting any treatment-emergent AEs will be coded using the MedDRA dictionary and be tabulated by system organ class and preferred term.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF ABBREVIATIONS

Abbreviation/Term	Explanation
AE	Adverse event
ALP	Alkaline phosphatase
ALT	Alanine aminotransferase
ANCOVA	Analysis of Covariance
AST	Aspartate aminotransferase
BSA	Baseline body surface area
CGI	Clinical Global Impression scale
CI	Confidence interval
CMH	Cochran–Mantel–Haenszel
CMR	Cardiac magnetic resonance
CPET	Cardiopulmonary exercise testing
CRF	Case report form
CSR	Clinical Study Report
CSS	Clinical Summary Score
cTTO	composite time trade-off
CV	Cardiovascular
CV%	Coefficient of Variation
ECG	Electrocardiogram
eCRF	Electronic Case Report Form
EQ-5D	EuroQol 5-dimension instrument
EQ-5D-5L	EuroQol 5-dimension 5-level instrument
EQ-VAS	EuroQol - Visual Analogue Scale
FAS	Full analysis set
HCM	Hypertrophic cardiomyopathy
hs-cTnI	High sensitivity cardiac troponin I
ICD	Implantable cardioverter defibrillators
ICF	Informed consent form
ICH	International Council for Harmonisation
ID	Identifier
IP	Investigational product
IWRS	Interactive web response system
KCCQ	Kansas City Cardiomyopathy Questionnaire
LLN	Lower Limit of Normal
LSM	Least Squares Mean
LV	Left ventricle(ular)
LVEDV	Left ventricular end-diastolic volume

Abbreviation/Term	Explanation
LVEF	Left ventricular ejection fraction
LVESV	Left ventricular end-systolic volume
LVOT	Left ventricular outflow tract
LVOT-G	Left ventricular outflow tract gradient
MAR	Missing At Random
MedDRA	Medical Dictionary for Regulatory Activities Terminology
MMRM	Mixed Model for Repeated Measures
MNAR	Missing Not At Random
NT-proBNP	n-terminal prohormone brain natriuretic peptide
NYHA	New York Heart Association
oHCM	Obstructive hypertrophic cardiomyopathy
PDCC	Protocol deviation classification committee
PGI-C	Patient Global Impression of Change scale
PK	Pharmacokinetics
PKS	Pharmacokinetics analysis set
PRO	Patient reported outcomes
PT	Preferred Term
pVO2	Peak oxygen uptake
QTcF	Fridericia corrected QT
RER	Respiratory exchange ratio
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SAQ-7	Seattle Angina Questionnaire -7
SAS	Statistical Analysis System (SAS®)
SD	Standard deviation
SOC	System Organ Class
SRT	Septal Reduction Therapy
TEAE	Treatment-Emergent Adverse Event
TESAE	Treatment-Emergent Serious Adverse Event
TSS	Total Symptom Score
ULN	Upper Limit of Normal
VAS	Visual Analogue Scale
VAT	Ventilatory anaerobic threshold
WHO	World Health Organization

SAP VERSION HISTORY

Version and Date	Revision	Rationale
Final 1.0	Not applicable: original version	Not applicable
Version 2.0	Synopsis and Section 7.6: updated testing hierarchy and removed the statement related to determination of the testing order of the duration of SRT eligibility and total workload.	The testing hierarchy was simplified from parallel gate keeping method to a closed testing procedure to allow the testing of the secondary endpoints including SRT eligibility at Week 24 at two-sided alpha level of 0.05 once the primary endpoint reaches statistical significance at a specified sequential testing order.
	Section 2.2.3: corrected typo in exploratory endpoints: number of patients with new or worsening ST depression during exercise at Week 12 and 24 by removing time point Week 12 added 1 exploratory efficacy endpoint	CPET is only performed at Week 24.
	Section 2.2.4: updated the definition of the endpoint of incidence of LVEF <50% with signs and symptoms of heart failure (concomitant adverse event of heart failure or dyspnea) and/or increase in NT-proBNP from baseline	Update was made to provide greater specificity to the increase in NT-proBNP (≥30% increase) in relation to worsening HF.
	Section 5.5.4: modified FAS definition by removing condition of requiring at least one post baseline efficacy measurement	Update was made to address FDA comments to the SAP.
	Section 7.2: added statement that if model assumptions are substantially violated, rank based analysis will be performed as supportive analysis.	Updates were made to address FDA comments to the SAP.
	Section 7.5: added additional analyses by IND Sites status	Updates were made to add additional analyses to have consistent scope of by IND sites status analyses
	Section 7.8: Remove COVID-19 sensitivity analyses from the secondary endpoints	The impact of COVID-19 reported on the study has been minimal.

Version and Date	Revision	Rationale
	Section 7.7.1: updated the reasons of invalid CPET	To use the same statements provided by the CPET laboratory used in the data transfer specification
	Section 7.7.4: updated to include criteria of condition when rank-based analysis will be performed and clarified that, if performed, rank-based analysis will be supportive analysis for the primary endpoint	Updates were made to address FDA comments to the SAP and provide specific criteria for the rank-based analysis to be used.
	Section 7.8.1: update the number of imputed datasets from 50 to 100	Updates were made per FDA comments to the SAP
	Section 7.8.1: clarified imputing missing Week 12 NYHA. Added sensitivity analysis for proportion of responders at Week 12 and 24 after treating patients with missing data as non-responder. Added statement to impute intermittent missing data use adjacent visits results in determining patients' SRT eligibility.	Updates were made per FDA comments to the SAP; added clarification statements on imputing intermittent missing data except Week 12 and Week 24 in determining patient's SRT eligibility
	Editorial updates made where applicable	To correct editorial issues

1. INTRODUCTION

The purpose of this statistical analysis plan (SAP) is to provide a technical elaboration of the planned analyses and detailed data displays to be included in the Clinical Study Report (CSR) for CY 6031 study.

This SAP was developed in accordance with International Council for Harmonisation (ICH) E9 and ICH E9 (R1) guideline. All decisions regarding final analysis, as defined in this SAP document, will be made prior to the study database lock. Further study information can be found in the protocol.

2. STUDY OBJECTIVES AND ENDPOINTS

2.1. Study Objectives

2.1.1. Primary Objective

To evaluate the effect of CK-3773274 on exercise capacity in patients with symptomatic obstructive hypertrophic cardiomyopathy (oHCM).

2.1.2. Secondary Objective

The secondary objectives of the study are listed as follows:

- To evaluate the effect of CK-3773274 on patient health status
- To evaluate the effect of CK-3773274 on New York Heart Association (NYHA) Functional Classification
- To evaluate the effect of CK-3773274 on post-Valsalva left ventricular outflow tract gradients (LVOT-G)
- To evaluate the effect of CK-3773274 on exercise capacity
- To evaluate the effect of CK-3773274 on duration of eligibility for SRT

2.2. Study Endpoints

2.2.1. Primary Endpoints

The primary endpoint of the study is Change in peak oxygen uptake (pVO2) by cardiopulmonary exercise testing (CPET) from baseline to Week 24.

2.2.2. Secondary Endpoints

- Change in Kansas City Cardiomyopathy Questionnaire – Clinical Summary Score (KCCQ-CSS) from baseline to Week 12 and Week 24
- Proportion of patients with ≥1 class improvement in NYHA Functional Class from baseline to Week 12 and Week 24
- Change in post-Valsalva LVOT-G from baseline to Week 12 and Week 24
- Proportion of patients with post-Valsalva LVOT-G <30 mmHg at Week 12 and Week 24
- Total duration of SRT eligibility during the 24-Week treatment period in patients who were SRT eligible at baseline
- Change in total workload during CPET from baseline to Week 24

2.2.3. Exploratory Endpoints

- Compared with baseline, proportion of patients at Week 24 achieving either:

  − Change from baseline of ≥1.5 mL/kg/min in pVO2 AND ≥1 class improvement in NYHA Functional Class

OR

− Change from baseline of ≥3.0 mL/kg/min in pVO2 AND no worsening of NYHA Functional Class

- Proportion of patients with >3 mL/kg/min improvement in pVO2 and ≥1 class improvement in NYHA Functional Class
- Proportion of patients with improvement of ≥5, 10, 15 and 20 points in KCCQ-CSS and KCCQ- Total Symptom Score (TSS) at Weeks 12 and 24
- Proportion of patients with resting LVOT-G <30 mmHg, post-Valsalva LVOT-G <50 mmHg, and NYHA Functional Class I at Week 12 and Week 24
- Proportion of patients with resting LVOT-G <30 mmHg, post-Valsalva LVOT-G <50 mmHg, and ≥1 class improvement in NYHA Functional Class at Week 12 and Week 24
- Change from baseline to Week 24 in CPET parameters of:
    - Ventilatory efficiency (VE/VCO2 slope)
    - Circulatory power (VO2 × systolic BP)
    - Ventilatory anaerobic threshold (VAT)
- Proportion of patients who remain SRT eligible at Week 24 in patients who were eligible for SRT at baseline. Proportion of patients who remain SRT eligible will also be evaluated at other scheduled visit weeks
- Time to first SRT ineligibility status in patients who were SRT eligible at baseline
- Change from baseline to Week 24 in individual responses to the EuroQol 5-dimension 5-level instrument (EQ-5D-5L)
- Change from baseline to Week 24 in total score and domain scores for the Seattle Angina Questionnaire -7 (SAQ-7)
- Change from baseline to Week 24 in echocardiographic measurements of cardiac structure and of systolic function including:
    - Left ventricular ejection fraction (LVEF)
    - Left ventricular global longitudinal strain (LV GLS)
    - Left ventricular end-systolic and end-diastolic volumes (LVESV and LVEDV)
    - Left atrial volume index
    - Left ventricular mass index
    - Maximal wall thickness
- Change from baseline values in n-terminal prohormone brain natriuretic peptide (NT-proBNP), high sensitivity cardiac troponin I (hs-cTnI) and other biomarkers through Week 24; proportional change (post randomization/baseline) will also be calculated and used in the analysis

- Change from baseline to Week 24 in patients enrolled in Cardiac magnetic resonance (CMR) substudy in CMR measurements of:
    - Left ventricular (LV) mass index
    - LVEF
    - Septal, free wall and maximal wall thickness
    - Left atrial volume index
    - LVESV
    - LVEDV
    - Extracellular volume proportion
    - Late gadolinium enhancement proportion
    - Mitral regurgitation severity
- Time to Maximal ST Segment Depression on CPET ECG
- Time to 1 mm ST depression below Baseline on CPET ECG
- Maximal ST segment depression on resting ECG in mm at Weeks 12 and 24
- Number of patients with new or worsening ST depression during exercise at Week 24
- Change from baseline values in all other summary KCCQ scores (Physical Limitation, Symptom Stability, Symptom Frequency, Symptom Burden, Total Symptom Score, Self-efficacy, Quality of Life, Social Limitation, Overall Summary Score) at Weeks 12 and 24.
- Proportion of patients with LVH with strain pattern (typical+ atypical) on Electrocardiogram (ECG) at Weeks 12 and 24
- Proportion of patients with all LVH (with or without strain) on ECG at Weeks 12 and 24

2.2.4. Safety Endpoints

- Incidence of reported major adverse cardiac events (cardiovascular [CV] death, cardiac arrest, non-fatal stroke, non-fatal myocardial infarction, CV hospitalization)
- Incidence of new onset persistent atrial fibrillation
- Incidence of appropriate implantable cardiac defibrillator (ICD) discharges and aborted sudden cardiac death
- Incidence of LVEF <50% with at least one of the following:
    - Signs and symptoms of heart failure (concomitant adverse event of heart failure or dyspnea)

      AND/OR
    - Increase in NT-proBNP (≥30% increase), relative to results from the most recent previous visit and above the upper limit of normal, at the time of LVEF assessment <50%

Note signs and symptoms refer to AEs with onset date within ± 7 days relative to the date when LVEF <50%.

- Incidence of LVEF <40%
- Incidence of LVEF <50%

Incidence of LVEF below 40% and 50% will be summarized for site read, core lab read and both.

- Incidence of treatment emergent adverse events (TEAEs)

2.2.5. PK Parameters

- $C_{post\ dose}$ and $C_{pre\text{-}dose}$

3. STUDY DESIGN

3.1. Summary of Study Design

This is a Phase 3, randomized, placebo-controlled, double-blind, multi-center trial in patients with symptomatic oHCM. Approximately 270 eligible patients will be randomized in a 1:1 ratio to receive CK-3773274 or placebo. Randomization will be stratified by use of beta-blockers (yes or no) and CPET exercise modality (treadmill or bicycle) and implemented in the Interactive Web Response System (IWRS). A cap on the number of patients taking beta-blockers and will not exceed approximately 70% of total enrollment. The number of patients taking disopyramide will be capped at approximately 10% of total enrollment. The number of patients with persistent atrial fibrillation at screening will also be capped at approximately 15%, and the number of patients using the bicycle CPET exercise modality will be capped at approximately 50% as well.

Investigational product (IP) will be administered orally once daily with or without food. During the initial six weeks of the treatment period, IP doses will be individually titrated at Weeks 2, 4, and 6 using echocardiography. Dose escalation at Weeks 2, 4, and 6 will occur only if a patient has a post-Valsalva LVOT-G ≥30 mmHg and a biplane LVEF ≥55%. Echocardiograms will be performed at each subsequent visit during the trial and the dose down titrated if necessary. The primary endpoint of pVO2 will be measured by CPET at screening and at end of treatment (Week 24). If applicable, patients will continue taking background HCM medications consistent with regional clinical practice guidelines during the trial.

3.2. Definition of Study Drugs

Table 1 describes any study drug: IP (ie, aficamten) or placebo intended to be administered to a trial patient according to the protocol.

Table 1: Investigational Products

	Active	Placebo
IP/Product Name	CK-3773274	Placebo
Type	Drug	Drug
Dose Formulation	Tablet	Tablet
Unit Dose Strength(s)	5 mg	Matching placebo
Dosage Level(s)	5mg, 10mg, 15mg, 20 mg	
Route of Administration	Oral	Oral
Use	Experimental	Placebo
IMP and NIMP	IMP	IMP
Packaging and Labeling	IP will be provided in blister packs which will be labeled as required per country requirement	IP will be provided in blister packs which will be labeled as required per country requirement

3.3. Sample Size Considerations

3.3.1. Sample Size Justifications

Assuming a difference in change from baseline in pVO2 of 1.5 mL/kg/min for CK-3773274 compared to placebo, a standard deviation (SD) of 3.5 mL/kg/min, accounting for limiting beta-blocker use (less than ~70%), limiting exercise modality of bicycle (less than ~50%) and 10% of patients missing change from baseline data of the primary endpoint, a sample size of 270 patients at randomization ratio of 1:1 (approximately 135 randomized to CK-3773274 and 135 randomized to placebo) provides more than 90% power to detect the difference in pVO2 change from baseline to Week 24 with a 2-sided type I error of 0.05.

During the study, Cytokinetics will periodically assess in a blinded fashion the aggregate pooled missing data rate and overall pooled SD for the change from baseline in pVO2 at Week 24. If the pooled SD is larger than expected, Cytokinetics may consider increasing the sample size once in order to maintain the intended power.

3.4. Randomization

All eligible patients will be centrally assigned to randomized IP using the IWRS. Randomization will be stratified by use of beta-blockers (yes or no) and CPET exercise modality (treadmill or bicycle) and implemented in the IWRS.

3.5. Clinical Assessments

3.5.1. Efficacy Assessments

Efficacy assessments include CPET, echocardiography, NYHA classification, patient-reported outcomes (KCCQ, EQ-5D-5L), Patient Global Impression of Change scale (PGI-C) and SAQ-7, and clinical global impression scale (CGI). CMR measurements will be assessed in patients included in CMR sub-study.

3.5.1.1. Echocardiography

Echocardiography will be done during screening, prior to dosing on Day 1, and 2 hours after dosing in the clinic on Weeks 2, 4, 6, 8, 12, 16, 20, 24, and 28.

Site read echocardiographic assessments include LVEF, resting and Valsalva LVOT at each visit. Unless otherwise specified, echocardiographic variables will be based on the core echocardiography laboratory assessments. Table 2 below lists the echocardiography parameters from core lab. A full list of echocardiography parameters is specified in the data transfer agreement from core lab.

Table 2: Echocardiographic Variables and Names

Endpoint Names
Variables Describing LV Structure
Left ventricular end diastolic diameter
Left ventricular end diastolic volume Index

Table 2: Echocardiographic Variables and Names (Continued)

Endpoint Names
Left Ventricular End Systolic Volume Index
Left ventricular end systolic diameter
Left Ventricular Posterior Wall Thickness, End-diastole
LV Mass indexed
Interventricular Septum Thickness, End-Diastole
Left ventricular Maximal wall thickness
Left ventricular relative wall thickness
Variable Describing LV Systolic Function
Left ventricular ejection fraction
Left ventricular fractional shortening
Left ventricular stroke volume Index
Left ventricular cardiac output Index
Left Ventricular Isovolumetric Contraction Time
Left Ventricular Isovolumetric Relaxation Time
Left Ventricular Ejection Time
Left Ventricular Myocardial Performance Index
Left Ventricular Outflow Tract Velocity Time Integral
Left Ventricular Global Longitudinal Strain
Left Ventricular Global Circumferential Strain
Variables Describing LV Diastolic Function
Peak E Wave Velocity
Peak A Wave Velocity
Mitral Lateral Annular Early Diastolic Velocity
Mitral Septal Annular Early Diastolic Velocity
Mitral E/A Wave Velocity Ratio
Mitral E Wave to Lateral Annular Early Diastolic Velocity Ratio
Mitral E Wave to Septal Annular Early Diastolic Velocity Ratio
LVOT Dynamic Gradient Assessment Variables
Peak Left Ventricular Outflow Tract Pressure Gradient at Rest
Peak Left Ventricular Outflow Tract Pressure Gradient during Valsalva Maneuver

Table 2: Echocardiographic Variables and Names (Continued)

Endpoint Names
Variables Describing LA Size and Function
Left Atrial Width
Left Atrial Volume Index
Variables Describing RV Size and Function
Right Ventricular Outflow Tract Velocity Time Integral
Right Ventricular Myocardial Performance Index
Tricuspid annular plane systolic excursion
Valvular Assessment Variables
Presence of Mitral Regurgitation
Mitral Regurgitation Jet Area to Left Atrial Area Ratio
Presence of Mitral Systolic Anterior Motion

3.5.1.2. Patient-Reported Outcomes

KCCQ and EQ-5D-5L will be assessed at Day 1, Weeks 2, 4, 6, 8, 12, 16, 20, 24 and 28 (4 weeks after last dose at end of the study). SAQ-7 will be assessed at Day 1, Weeks 4, 8, 12, 16, 20, 24 and 28. PGI-C will be assessed at the Week 24. Algorithms to derive the scores KCCQ, EQ-5D-5L and SAQ-7 are in Section 11.1.

EQ-5D-5L and EQ-VAS

The instrument EQ-5D is a standardized measure of health status for clinical and economic appraisal (EuroQol Group 1990), which consists of two parts: a short descriptive system questionnaire (EQ-5D-3L) and a visual analogue scale (EQ-VAS).

EQ-5D-5L is the 5-level version of EQ-5D introduced to improve the instrument's sensitivity and to reduce ceiling effects (EuroQol Group 2009). The descriptive system comprises 5 dimensions: mobility, self-care, usual activities, pain/discomfort and anxiety/depression. Each dimension has 5 levels indicating no problems, slight problems, moderate problems, severe problems or extreme problems. Five responses with a response from each of the 5 dimensions form a 5-digit number that defines a patient's health state profile. A health state can potentially be assigned a summary index score based on societal preference weights (societal perspective) for the health state. The health state preferences often represent national or regional values and can therefore differ between countries/regions. The health state index scores will be calculated using the composite time trade-off (cTTO) method based on the United States valuation of EQ-5D-5L (Pickard 2019) for patients from the United States and for the FAS. The health state index score ranges from less than 0 to 1 with higher scores indicating higher health utility; a score 0 represents death, negative values represent worse than death, and 1 represents full health.

EQ-VAS rates a patient's perceived health on a vertical visual analogue scale from 0 to 100, where 0 represents the worst imaginable health and 100 represents the best imaginable health.

The VAS can be used as a quantitative measure of health outcome that reflects the patient's own judgement.

3.5.1.3. CMR Assessments

A CMR imaging sub-study will assess the effects of administration of CK-3773274 dosing on cardiac morphology, function, and fibrosis in approximately 100 oHCM patients who are eligible and consent to participate. CMR will be performed during screening period and Week 24. CMR parameters are listed below in Table 3 and a final list of CMR parameters is specified in the data transfer agreement from core lab.

Table 3: CMR Parameters

LV Parameters
LVM - Left Ventricular Mass
LVMI Left Ventricular Mass Index
LVEDV - Left Ventricular End Diastolic Volume
LVEDVI - Left Ventricular End Diastolic Volume Index
LVESV - Left Ventricular End Systolic Volume
LVESVI - Left Ventricular End Systolic Volume Index
LVSV– Left Ventricular Stroke Volume=LVEDV-LVESV
LVSVI = Left Ventricular Stroke Volume Index=LVSV/Body Surface Area (mm2)
LVEF - Left ventricular Ejection Fraction
CO - Cardiac Output = Heart Rate x Stroke Volume
CI - Cardiac Index = Heart Rate x Stroke volume/Body Surface Area (m2)
LV maximal septal wall thickness
LV maximal lateral wall thickness
LV maximal wall thickness (each of 16 segments)
Overall LV maximal wall thickness (highest across all 16 segments)
LGE mass (g) (Global mass of LGE 6SD)
LGE mass % (as % of LV mass) (Global percent of LGE 6SD)
Global average ECVF

Table 3: CMR Parameters (Continued)

LV Parameters
LV Segmental assessment of ECVF
Global Native T1
Extracellular volume (ECV)
Global ECV mass
Global ECV mass index
ECVF normalized for height
HCM morphology (isolated basal septal hypertrophy, reverse septal curvature, apical, midcavity obstruction without apical aneurysm, midcavity obstruction with apical aneurysm, concentric, other
RV Parameters
RV end-diastolic volume (RVEDV)
RV end-diastolic volume index (RVEDVI)
RV end-systolic volume (RVESV)
RV end-systolic volume index (RVESVI)
RV stroke volume (RVSV)
RV stroke volume index (RVSVI)
RV ejection fraction (RVEF)
LA parameters
LA volume maximum
LA volume maximum index
LA reservoir amount
LA reservoir percent
LA contractile amount
LA contractile percent
LA total amount
LA total percent
LA global longitudinal strain
Mitral Valve Parameters
Exploratory Mitral Valve Regurgitation Measurements
Mitral regurgitation volume
Mitral regurgitation, regurgitation fraction

3.5.1.4. CPET Assessments

All patients will undergo CPET with gas-exchange analysis and the methodology will be standardized across all sites as specified in the CPET manual. Patients must use the same testing modality for all exercise tests during the trial. CPET are done at baseline and Week 24 post randomization. CPET parameters are listed below. A full list of CPET parameters is specified in the data transfer agreement from core lab.

- Workload
- Exercise Duration
- % of Predicted Oxygen Uptake
- Circulatory Power
- Predicted Oxygen Uptake
- Peak Oxygen Uptake per Kilogram
- Peak RER
- Oxygen Uptake Efficiency Slope
- Ventilatory Efficiency
- Anaerobic Threshold
- Aerobic Efficiency

3.5.2. Safety Assessments

Safety assessments include adverse events and serious adverse events (SAEs), ICD discharge, LVEF < 50% and <40%, electrocardiograms, laboratory assessments, physical examinations, and vital signs.

3.5.3. Pharmacokinetics Assessments

Blood samples will be collected to evaluate plasma concentrations of CK-3773274 at pre-dose and 2 hours post-dose at Day 1, Weeks 2, 4, 6, 8, 12, 16, 20 and 24.

4. PLANNED ANALYSES

4.1. Interim Analyses

No interim analysis is planned for this study.

4.2. Final Analyses

The final analysis will occur after all patients randomized in the study have completed the study including the 4-week safety follow up, all data has been entered into the clinical database, verified, and locked. Unblinding for the final analysis will occur after the database lock.

5. GENERAL CONSIDERATIONS FOR DATA ANALYSES AND HANDLING

5.1. General Summary Table and Individual Subject Data Listing Considerations

Descriptive statistics to be presented in a table include number of observations, mean, median, standard deviation, 1st and 3rd quartiles, minimum and maximum for continuous variables, and count of patients and the percentage for categorical variables. For variables that lognormal distribution assumptions may be appropriate geometric mean, and geometric coefficient of variation (CV%) will also be displayed. Geometric CV (%) will be derived as $100\% \times \text{sqrt}(\exp(s^2)-1)$, where s is the standard deviation of the natural logarithm (ln) transformed data.

For model-based analysis, least squares means (LSM), difference of least squares means between treatments, their standard errors and 95% confidence intervals (CI), and two-sided p-values for the statistical inferences will be presented.

Selected listings may be generated to include patient identifier (ID), demographics, randomized treatment group and other relevant items, and sorted by randomized treatment group, patient ID and date of assessment.

5.2. General Post Text Summary Table and Individual Subject Data Listing Format Considerations

Post text tables and individual subject data listings are prepared according to ICH Guideline E3. In general, summary and analysis tables will be presented by treatment groups and highest dose level administered.

5.3. Data Management

Data will be entered into the clinical database with programmed edit checks and manual data review to ensure integrity. The data will be reviewed and cleaned according to a data management plan. Clinical safety laboratory, ECG, Pharmacokinetics (PK) data, CMR, CPET and echocardiography will be provided per the pre-specified data transfer agreement from external laboratories.

5.4. Data Presentation Conventions

The following conventions will be applied to data presentations:

- For continuous variables, mean and median values are formatted to one more decimal place than the measured value. Standard deviation values are formatted to two more decimal places than the measured value. Minimum and maximum values are presented with the same number of decimal places as the measured value. For the statistical analyses results that are on the same scale as a measured value (e.g., change from baseline or treatment difference estimates), the LSM estimates and LSM estimate 95% CI boundary values will be formatted to one more decimal place than the measured value; Standard error of the mean (SEM) estimates will be formatted to two more decimal

places than the measured values. GLSM estimates for the proportional change from baseline, proportional change treatment ratios, odds ratios, and the corresponding 95% CIs will be presented with two decimal places.

- For categorical variables, the count and percentage of responses are presented in the form XX (XX.X%) where the percentage is in the parentheses.
- Date variables are formatted as YYYY-MM-DD for presentation. Time is formatted in military time as HH:MM for presentation.
- P-values, if applicable, will be presented to 3 decimal places. If the p-value is less than 0.0001 then it will be presented as <0.0001. If the rounded result is a value of 1.000, it will be displayed as >0.999.
- Unless otherwise stated, any statistical tests performed will use 2-sided tests at the 0.05 significance level.

The table and listing shells and table of contents provide the expected layout and titles of the tables, listings and figures. Any changes to format, layout, titles, numbering, or any other minor deviation will not necessitate a revision to the SAP, nor will it be considered a deviation from planned analyses. Only substantial deviation in the analysis methods will require an SAP revision or a change to planned analysis documented in the CSR.

5.5. Analysis Populations

5.5.1. All Screened Patients

All patients who signed the informed consent form (ICF) are included in the All Screened Patients Set. Patients who gave informed consent but are not randomized are considered screen failures. The following reasons for screen failures are collected: inclusion/exclusion criteria (including specific criteria not met), principal investigator decision, subject decision, lost to follow up, and other. For patients who are screen failures, the reasons for failing will be summarized.

5.5.2. All Randomized Set

All Randomized Set includes patients who are randomized to receive CK-3773274 or placebo.

5.5.3. Safety Analysis Set

Safety analyses will be performed on the safety analysis set (SAF), which includes all randomized patients who receive at least one dose of IP, CK-3773274 or placebo. Unless otherwise specified, for safety analyses, subjects will be grouped according to their randomized treatment group assignment with the following exception: if a subject receives treatment throughout the study that is different than the randomized treatment group assignment, then the subject will be grouped by the actual treatment group.

5.5.4. Full Analysis Set

Efficacy analyses will be performed on the full analysis set (FAS), which includes all randomized patients. Patients will be analyzed according to their randomized treatment group assignments.

5.5.5. Pharmacokinetics Analysis Set

All randomized patients who have at least one evaluable plasma concentration of CK-3773274, provided they have no major protocol deviations that could affect the PK of CK-3773274.

5.6. Baseline Definition

Unless otherwise specified, baseline is defined as the last available measurement taken prior to administration of the first dose of study drug. Baseline for KCCQ, EQ-5D-5L and SAQ-7 are assessments performed on Day 1. The assessments collected on the same date as the first dose of the IP that do not have assessment time are considered to have occurred prior to the first dose.

5.7. Derived and Transformed Data

5.7.1. Baseline Age

Age will be calculated as follows:

Age (years) = year of screening date – year of birth,

Patient age will be categorized as < 65 years or ≥ 65 years.

5.7.2. Body Measurements Variable Derivation

Baseline body surface area (BSA) will be calculated using the weight and height at screening using the DuBois and DuBois formula and rounded to two decimal points for the presentation of results:

BSA (m^2) = 0.007184 * (weight (kg) $^{0.425}$ * height (cm) $^{0.725}$).

5.7.3. Study Day

If the date of interest occurs on or after the first dose date, then study day will be calculated as (date of interest – date of first dose) + 1. If the date of interest occurs prior to the first dose date, then study day will be calculated as (date of interest – date of first dose). There is no study day 0.

5.7.4. Change from Baseline

Change from baseline is calculated as (post baseline value – baseline value).

Percent change from baseline is calculated as (change from baseline / baseline value) x 100%.

Proportional change from baseline is calculated as (post-baseline result/baseline value).

If either the baseline or the post-baseline value is missing, the change from baseline and percentage change from baseline will be set to missing.

5.7.5. Summary Scores for Patient Reported Outcomes (PRO)

Calculations of summary scores for KCCQ, SAQ-7 and EQ-5D-5L are specified in Section 11.1.

5.7.6. Analysis Windows

Since study visits do not always take place exactly as scheduled per protocol, it is necessary to assign the actual observation dates to analysis windows for analysis purposes.

For data collected at a scheduled post baseline visit, the analysis visit will be assigned based on the scheduled nominal visit as collected on the eCRF.

For unscheduled or early discontinuation post baseline visits, measurements taken on or after the first dose of study drug will be assigned to an analysis window using defined lower and upper bounds for each analysis window. Measurements assigned in an analysis window will have study day greater than or equal to the lower bound but no greater than the upper bound of the analysis window. The lower bound and the upper bound for the analysis windows are defined as the midpoints between the scheduled visits for all assessments (see Section 11.4).

Visits are identified as the nominal visits according to the eCRFs. Each visit will be identified with the visit descriptor (eg, “Week 24”).

5.7.7. Multiple Assessments

Once analysis windows are assigned, a patient’s individual analysis window could potentially contain more than one visit. Records from all visits, including scheduled, unscheduled and early discontinuation visits could be flagged as the “analyzed record” within the analysis window, although the records from scheduled visit will take priority.

In the event of multiple visits falling within an analysis window, the following rules will be used in sequence to determine the “analyzed record” for the analysis window:

- If a scheduled visit occurred during the analysis window, then the measurement taken from the scheduled visit will be used.
- If no scheduled visit occurred during the analysis window, the measurement taken closest to the scheduled day will be used as the “analyzed record.”
- If no scheduled visit occurred during the analysis window and there is a tie between unscheduled visits in the number of days before and after the scheduled day, measurements from the later visit will be used as the “analyzed record.”

For analyses by visit, only the “analyzed record” within each analysis window and the visit will be summarized in a table. Only protocol specified visits will be presented in the summary table. If there are other visit records within the analysis window, they will only be included in data listings.

5.7.8. Other Study Related Definitions

Actual Dose Group

Patients in the aficamten actual treatment group will be identified as 5 mg, 10 mg, 15 mg, 20mg or discontinuing IP prior to dose adjustment based on the dose assigned at Week 8. If a patient discontinues IP prior to the start of Week 8 (IWRS Week 8 dispensation), then the subject will be

identified as discontinued IP prior to achieve stable dose. The actual dose group may be used in selected displays.

Investigational Product Exposure Period

For subjects dosed with IP:
[(Last IP administration date – date of Study Day 1) + 1]/7 (in weeks)
Interruptions recorded in the eCRF page will be excluded from the expected dose calculation.

Treatment-emergent Adverse Event

For the purpose of reporting, an investigator-reported event starting on or after first dose of IP and up to and including 28 days after the last dose date of IP will be labeled as a treatment-emergent AE.

Last Titrated Dose

Last titrated dose is defined as the last titrated dose assigned to the patient during the 24 Week treatment period.

5.7.9. Derived Echocardiographic Parameters

The following BSA-indexed variables will be derived using the baseline BSA defined in Section 5.7.2:

- LVEDV-I (mL/m^2) = LVEDV/BSA
- LVESV-I (mL/m^2) = LVESV/BSA
- LVSVI (mL/m^2) = LVSV/BSA
- LAV-I (mL/m^2) = LAV/BSA
- LVCO-I ($mL/min/m^2$) = LVCO/BSA
- LVmass-I(g/m^2) = LVmas/BSA

For the presentation of results, the BSA-indexed variables will be rounded to the same number of decimal places as the corresponding non-indexed variables provided by the echocardiography core laboratory.

5.8. Handling of Missing Data

5.8.1. Missing Efficacy Endpoints

For the primary endpoint, missing data will be imputed under missing at random (MAR) assumption and all observed and imputed missing pVO_2 assessments will be included in the primary analysis of the primary endpoint. Missing secondary CPET endpoints will be handled the same as for the primary endpoint. Missing response for patient reported outcomes will be handled as described in Section 11.1.

5.8.2. Missing Start and Stop Dates for Prior and Concomitant Medication

To classify medications as baseline use or concomitant, missing start and stop dates of medications will be imputed as follows:

- If the medication start date day is missing, it will be imputed with the first of the month,
- If the medication start date day and month are missing, they will be imputed with 01 January,
- If the medication stop date day is missing, it will be imputed with the last day of the month or the date of the last contact with the patient, with the imputed date doesn't exceed the date of last contact,
- If the medication stop date day and month are missing, they will be imputed with 31 December or the date of the last contact with the patient, with the imputed date doesn't exceed the date of last contact,
- For the ongoing medications, the stop date will not be imputed.

5.8.3. Missing Start and Stop Dates for Adverse Events

For AEs with incomplete date information recorded in the eCRF, the imputation will follow the following algorithm:

For missing AE onset Date

- If an AE onset Day is missing and the Month of AE onset is known, then the first day of the month of AE onset will be imputed as the AE onset date. If the month and year of AE onset are the same as month and year of the first dosing, the missing day will be imputed as the first dosing date.
- If AE onset information is not available, then the first dosing date will be imputed as the AE onset date.
- If AE onset day and month are both missing, the missing month and day will be imputed as 01 January. If the year of AE onset is the same as the first dosing date, the AE onset will be imputed as the first dosing date.

For missing AE end Date:

- If the AE end Day is missing and it will be imputed with the last day of the month or the date of the last contact with the patient, with the imputed date doesn't exceed the date of last contact.
- If the AE end date day and month are missing, they will be imputed with 31 December or the date of the last contact with the patient, with the imputed date doesn't exceed the date of last contact.
- For the ongoing AEs, the stop date will not be imputed.

6. STUDY POPULATION

6.1. Subjects Disposition

Patient disposition will be summarized based on all randomized patients. The following will be summarized:

- The number and percentage of patients who completed the study and the number of patients who discontinued from the study early,
- For the patients who discontinued from the study early, reasons for early discontinuation,
- The number and percentage of patients who received at least one dose of the IP,
- For the patients who received at least one dose of the IP, the number and percentage of patients who completed study treatment and the number of patients who discontinued the study treatment early,
- For the patients who discontinued the study treatment early, reasons for early discontinuation.

The number and percentage of randomized patients included in each analysis set will be summarized. Reasons for exclusion from analysis sets will be listed.

6.2. Screen Failures

Screen failures will be listed and summarized by reasons of screening failure.

6.3. Protocol Deviations

Major protocol deviations are reviewed and confirmed by the protocol deviation classification committee (PDCC) during the protocol deviation reviews throughout the study prior to database lock. Major protocol deviations are a subset of protocol deviations that may significantly impact the completeness, accuracy, and/or reliability of the study data or that may significantly affect a subject's rights, safety, or well-being. E.g., of major protocol deviations are

- patients who entered the study even though they did not satisfy the entry criteria.
- patients who developed withdrawal criteria during the study but were not withdrawn.
- patients who received the wrong treatment or incorrect dose.
- patients who received an excluded concomitant treatment.

Number of patients with reported major protocol deviation will be summarized by treatment group for all randomized subjects. A summary of protocol deviations due to COVID-19 will be provided separately.

6.4. Demographic and Baseline Characteristics

Demographic and baseline characteristics, including age, age group [< 65, ≥ 65], sex, race, ethnicity, height, weight, BMI, BSA, randomization stratification variables and baseline disease characteristics will be summarized by randomized treatment group for the FAS using descriptive

statistics. Summary may be repeated for all randomized patients. All randomized patients will be included in the listing of demographic and baseline characteristics.

6.5. Listing of Subject Inclusion and Exclusion Criteria

A listing of randomized patients who did not meet the inclusion and exclusion criteria will be provided.

6.6. Medical History

Medical history will be summarized by treatment received for the Full Analysis Set. HCM-related medical history will be summarized, including time since initial diagnosis and number and percentage of patients meeting the oHCM criteria.

Select cardiovascular medical history and other medical history will be summarized by the Medical Dictionary for Regulatory Activities Terminology (MedDRA) system organ class (SOC) and preferred term (PT).

6.7. Baseline Medications Use

Medications will be coded using World Health Organization (WHO) Drug Dictionary. Baseline medication use is defined as medications that start before the first dose of IP and ends after the first dose of IP or ongoing. The count and percentage of patients with each medication history item will be presented by therapeutic class (Anatomical Therapeutic Chemical [ATC] Class 3) and preferred name. If ATC Class 3 is not available, ATC Class 2 will be used in the summary.

7. EFFICACY

7.1. General Considerations

Efficacy analyses will be performed in the FAS by the randomized treatment group. Unless otherwise specified, all hypothesis tests will be reported as 2-sided p-values. Exploratory endpoints and subgroup analyses will be assessed using a nominal alpha level of 0.05 and will not have multiplicity adjustments. In order to preserve an overall type I error rate for the primary and secondary endpoints testing of the primary and secondary endpoints will follow the testing procedures specified in Section 7.6.

7.2. Testing Statistical Assumptions Including Comparability at Baseline

The primary endpoint will be analyzed using an analysis of covariance (ANCOVA) model. Model assumption will be assessed by graphical examination of residuals. If assumptions are substantially violated, rank based analysis will be performed as supportive analysis. See Section 7.7.4 for more details.

7.3. Statement of the Null and Alternate Hypotheses

The null hypothesis for the primary endpoint is that the treatment difference (aficamten – placebo) of mean change from baseline of pVO_2 at Week 24 is 0 and the alternative hypothesis is that the treatment difference is > 0 (favors aficamten). The tests will be reported with two-sided p-values, but only the direction favoring aficamten direction will be considered success.

7.4. Planned Covariates

Baseline covariates include but are not limited to the stratification factors and baseline measurements. For CPET related endpoints, covariates age, sex, baseline weight will also be evaluated. See Section 7.7.4 for details evaluating covariates effect in the primary endpoint.

7.5. Subgroup Analyses

Subgroup analyses with relatively moderate sample size will be performed to examine the consistency of the observed treatment effect and to gain insight into the effectiveness of aficamten in subpopulations. Analyses of the primary endpoint will be conducted for the following subgroups:

- Sex (male, female)
- Age group (< 65, ≥ 65 years old)
- Baseline body mass index (<30 vs. ≥ 30)
- Baseline NYHA Class (II, III)
- Baseline KCCQ CSS (≤ median and > median)
- Baseline LVEF (≤ median and > median)
- Baseline N-terminal prohormone brain natriuretic (NT-proBNP) (≤ median and > median)

- CPET modality (treadmill, bicycle)
- Baseline pVO_2 (≤ median, > median)
- Beta Blocker (use, no use)
- Baseline resting LVOT (≤ median and > median)
- Sarcomeric gene mutation status (pathogenic or variant of uncertain significance, and non-disease causing or none)

Subgroup analysis will be performed by including the subgroup effect and subgroup by treatment interaction to the model. The subgroup analysis will be performed based on the imputed dataset generated for the primary analysis of the endpoint if the primary analysis is based on multiple imputation.

In addition, subgroup of IND site status (IND sites vs. non-IND sites) analysis will be performed for the primary estimand, proportion of patients with >1 NYHA functional class improvement, KCCQ CSS change from baseline, post-Valsalva LVOT-G change from baseline at Week 24 and overall adverse events summary to demonstrate compliance with relevant aspects of 21 CFR 312.120.

Analyses on the primary and secondary endpoints in patients who were SRT eligible at baseline may be explored.

7.6. Multiple Comparisons and Multiplicity

The null hypothesis for the primary and secondary efficacy variables in the FAS will be tested in the pre-specified order using a closed testing procedure.

For preservation of the overall type I error rate at two-sided 0.05 for the primary and secondary endpoints, the closed testing procedure will be used to address multiplicity. The primary endpoint is tested first at two-sided 0.05. If the primary endpoint achieves statistical significance at two-sided alpha level of 0.05, then the secondary endpoints will be tested at two-sided 0.05, with their testing being in the sequential order of KCCQ-CSS change from baseline, proportion of patients with ≥1 NYHA functional class improvement, post-Valsalva LVOT-G change from baseline, and proportion of patients with post-Valsalva LVOT-G < 30 mmHg in FAS at Week 24; duration of SRT eligibility up to 24 weeks of treatment for those who were SRT eligible at baseline; KCCQ-CSS change from baseline, proportion of patients with ≥1 NYHA functional class improvement, post-Valsalva LVOT-G change from baseline, and proportion of patients with post-Valsalva LVOT-G < 30 mmHg in FAS at Week 12; and change from baseline to Week 24 in total workload. The testing steps are described as follows in Table 4:

Table 4: Testing Steps

Step1	The null hypothesis for the primary endpoint is that there is no treatment difference in the change from baseline to Week 24 in pVO_2 between patients randomized to placebo and those randomized to aficamten in the FAS. The hypothesis will be tested at the two-sided significance level of 0.05. If this hypothesis is rejected, testing will proceed to Step 2; otherwise testing will stop.
Step 2	The null hypothesis for the first secondary endpoint is that there is no treatment difference in the change from baseline to Week 24 in KCCQ-CSS in the FAS. The hypotheses will be tested at the two-sided significance level of 0.05. If this hypothesis is rejected, testing will proceed to Step 3; otherwise testing will stop.
Step 3	The null hypothesis for the second secondary endpoint is that there is no treatment difference in proportion of patients with ≥1 NYHA functional class improvement at Week 24 in the FAS. The hypotheses will be tested at the two-sided significance level of 0.05. If this hypothesis is rejected, testing will proceed to Step 4; otherwise testing will stop.
Step 4	The null hypothesis for the third secondary endpoint is that there is no treatment difference in change from baseline to Week 24 in post-Valsalva LVOT-G in the FAS. The hypotheses will be tested at the two-sided significance level of 0.05. If this hypothesis is rejected, testing will proceed to Step 5; otherwise testing will stop.
Step 5	The null hypothesis for the fourth secondary endpoint is that there is no treatment difference in the proportion of patients with post-Valsalva LVOT-G < 30 mmHg at Week 24 in the FAS. The hypotheses will be tested at the two-sided significance level of 0.05. If this hypothesis is rejected, testing will proceed to Step 6; otherwise testing will stop.
Step 6	The null hypothesis for the fifth secondary endpoint is that there is no treatment difference in duration for SRT eligible during the 24-Week treatment period in patients who were SRT eligible at baseline in the FAS. The hypothesis will be tested at the two-sided significance level of 0.05. If this hypothesis is rejected, testing will proceed to Step 7; otherwise testing will stop.
Step 7	The null hypothesis for the sixth secondary endpoint is that there is no treatment difference in the change from baseline to Week 12 in KCCQ-CSS in the FAS. The hypothesis will be tested at the two-sided significance level of 0.05. If this hypothesis is rejected, testing will proceed to Step 8; otherwise testing will stop.
Step 8	The null hypothesis for the seventh secondary endpoint is that there is no treatment difference in proportion of patients with ≥1 NYHA functional class improvement at Week 12 in the FAS. The hypotheses will be tested at the two-sided significance level of 0.05. If this hypothesis is rejected, testing will proceed to Step 9; otherwise testing will stop.
Step 9	The null hypothesis for the eighth secondary endpoint is that there is no treatment difference in change from baseline to Week 12 in post-Valsalva LVOT-G in the FAS. The hypotheses will be tested at the two-sided significance level of 0.05. If this hypothesis is rejected, testing will proceed to Step 10; otherwise testing will stop.

Step 10	The null hypothesis for the ninth secondary endpoint is that there is no treatment difference in the proportion of patients with post-Valsalva LVOT-G < 30 mmHg at Week 12 in the FAS. The hypotheses will be tested at the two-sided significance level of 0.05. If this hypothesis is rejected, testing will proceed to Step 11; otherwise testing will stop.
Step 11	The null hypothesis for the tenth secondary endpoint is that there is no treatment difference in change from baseline to Week 24 in total workload in the FAS. The hypotheses will be tested at the two-sided significance level of 0.05.

7.7. Analysis of the Primary Efficacy Endpoint

7.7.1. Primary Efficacy Analysis

The primary endpoint is change in pVO_2 from baseline to Week 24. The **primary analysis** will be performed using an ANCOVA model that includes terms of treatment, randomization stratification factors (beta-blocker use status and CPET modality), baseline pVO_2 and baseline body weight as covariates in the FAS. Table 5 below displays details of the two estimands for the primary endpoint.

Table 5: Estimands for Primary Endpoint

Attributes	Primary Estimand	Secondary Estimand
Population	FAS, target population of potentially treatable aficamten subjects.	Hypothetical target population of potentially treatable aficamten subjects continue with treatment and are capable of completing the Week 24 assessment. Subjects with missing Week 24 pVO_2 due to intercurrent events or discontinuing treatment prior to Week 24 will be excluded.
Variable	Change from baseline to Week 24 in pVO_2. Data to be analyzed include all observed Week 24 pVO_2 values from subjects who complete 24 weeks of treatment, or from subjects who early terminate from the treatment but remain in the study and have Week 24 pVO_2 and imputed pVO_2 for subjects who don't have Week 24 pVO_2. Imputation details are provided below.	Change from baseline to Week 24 in pVO_2. Data to be analyzed include observed pVO_2 values from subjects who complete at least 24 weeks of treatment.
Measure of intervention effect	Mean treatment difference regardless of completing 24 weeks of treatment and experiencing intercurrent events.	Mean treatment difference among all subjects who remained on their randomized treatment for 24 weeks.

Subjects will be followed per the schedule of assessments from randomization through their final visit irrespective of whether the subject is continuing to receive study treatment. Reasons for not completing Week 24 CPET will be recorded on eCRF; categories of reasons include adverse

events, early termination, equipment failure, investigator decision, subject decision and other. The percentage of missing CPET data at Week 24 and the reasons for the missing data will be tabulated in the FAS. The following type of intercurrent events could preclude CPET at Week 24.

- Death
- Hospitalization
- CV AEs
- non-CV AEs (e.g., orthopedic injury)
- COVID-19 related intercurrent events e.g., subjects' decision to early terminate from the study due to the COVID-19 precautions, site closures, hospitalization due to COVID-19, or COVID-19 symptoms preventing subjects from coming to the Week 24 visit.

CPET data deemed to be invalid by the CPET core lab will be treated as missing CPET data in the analysis. CPET core lab flags the CPET results as invalid when there are:

- Technical equipment failure during CPET (i.e., air leak/lack of proper equipment utilization such as missing nose clip leading to lack of capture of gas exchange data)
- Transient non-cardiac issues that precluded conduct of the exercise study (as defined by inability to turn the pedals during the warm-up period for at least 3 min)
- CPET MOP-major CPET process deviation can impact one or more CPET variables.

Missing data as a result of an invalid assessment by the CPET core lab cab will be considered as missing at random.

Missing pVO2 at Week 24 regardless of type of intercurrent events will be imputed using multiple imputation methodology under the MAR assumption for the primary analysis of the primary estimand. Missing Week 24 CPET is expected to be low. Patients will be followed according to the schedule of activities in the protocol from randomization through the date of final visit irrespective of whether the patient is continuing to receive IP unless the patient has discontinued prematurely from the study or withdrawn consent. The protocol allows up to 4 weeks extension of Week 24 in the event that the subject is temporarily unable to exercise due to an AE e.g., ankle sprain, upper respiratory infection etc. or due to equipment malfunction to ensure post randomization CPET data collection. Missing Week 24 CPET due to intercurrent events i.e., non-CV AEs or COVID-19 related intercurrent events can be considered as MAR. Death event is expected to be very rare given the patient population and the duration of the treatment in this study. Assume patients' risk of death event or hospitalization due to HCM symptom and other CV AEs is balanced between treatment groups, missing data due to this type of intercurrent events for the primary estimand will be imputed as MAR.

The imputation model will use regression multiple imputation which includes treatment group, randomization stratification factors, baseline pVO_2, sex, age, baseline hemoglobin, baseline body weight, baseline KCCQ CSS, and baseline NYHA class and the last available post randomization NYHA functional class, resting and Valsalva LVOT. Categorical variables, i.e., treatment group, baseline NYHA functional class, and sex will be specified in the CLASS statement. One hundred

(100) imputed datasets will be generated. Change from baseline in pVO_2 will be calculated based on the observed and imputed data. Each of the imputed dataset will be analyzed using the primary analysis ANCOVA model. LSM estimate of treatment difference and the standard error will be combined using Rubin's rules (Rubin 1987) to produce a LSM estimate of the treatment difference, its 95% confidence interval, and p-value for the test of null hypothesis of no treatment effect. LSM, LSM difference and the corresponding standard error, 95% CI and p-value will be presented.

7.7.2. Sensitivity Analyses of the Primary Efficacy Endpoint

To evaluate the robustness of the primary analysis approach, sensitivity analyses, e.g., placebo-based imputation and tipping point analysis will be performed. In placebo-based imputation, missing pVO2 from subjects who discontinued from aficamten treatment or missing pVO2 from subjects from the placebo arm will be imputed based on the model that is constructed using observed pVO2 data from the placebo arm. Missing pVO2 from subjects who remained on aficamten treatment will be imputed based on the model that is constructed using observed pVO2 data from aficamten arm. Tipping point analysis will be performed by applying a range of negative shift to adjust the imputed value of missing pVO2 in aficamten group. If there are 10% or more missing data and/or 5% or more subjects missing data due to reason related to IP, the primary analysis will have the missing data imputed using placebo-based imputation.

The tipping point can be identified while the result is no longer statistically significant. Clinical judgment will be applied to evaluate the plausibility of the assumptions underlying this tipping point.

In addition, sensitivity analysis to evaluate COVID-19 impact will be performed by repeating the primary analysis after setting the Week 24 CPET to missing from subjects who are impacted by COVID pandemic.

Another sensitivity analysis is to fit a repeated measures mixed model to pVO_2 baseline and Week 24 data. The model includes stratification factors, visit, stratification by visit, and a numeric covariate which equals 0 for both treatment groups at baseline and equals 0 for placebo at Week 24 and equals 1 for aficamten group at Week 24. The primary treatment comparison is for the numeric covariate for treatments which corresponds to the treatment difference at week 24 in a specification for which there is no treatment difference at baseline.

7.7.3. Subgroup Analyses for the Primary Endpoint

Subgroup analyses will be performed by including the subgroup effect and subgroup by treatment interaction terms to the primary ANCOVA model for the primary endpoint. For subgroup analysis for the primary estimand, missing data imputed in the primary analysis will be used in the subgroup analyses. Only summary statistics will be presented for the subgroup level when the number of subjects in either treatment arm is ≤ 15 at this level. LSM estimate of the treatment difference, 95% confidence intervals for the mean treatment difference and nominal p-values will be provided for each subgroup level.

7.7.4. Supportive Analyses for the Primary Endpoint

To explain heterogeneity or identify treatment effect modifiers from the baseline characteristics, covariates used to define pre-specified subgroups and these covariates by treatment interaction

terms will be included in the ANCOVA model as supportive analysis. Global test of covariates by treatment interactions will be performed.

Stepwise model selection method will be used based on the stay or entry level of 0.05 to evaluate significant baseline covariates impact on the primary endpoint. ANCOVA model will be repeated by adjusting for the significant baseline covariates. Covariates measured as continuous will be introduced to the model as continuous variable. These analyses will be based on observed data.

The normality assumptions of the ANCOVA model will be investigated graphically. The scaled residuals will be examined. A supportive analysis will be performed after transforming pVO_2 data into ranks if greater than 5% of patients have an extreme change from baseline value at Week 24. Extreme values in the pooled data are defined as observations outside of Tukey's outer fences, i.e. observations that are less than the 25% quartile – 3 times the inter quartile range or greater than 75% quartile + 3 times the inter quartile range. Ranks will be applied to all changes from baseline data after the imputation step. Baseline data will be ranked separately.

The secondary estimand will be analyzed using the same primary analysis ANCOVA model for the primary estimand. Subgroup analysis for the secondary estimand will be performed using the same ANCOVA model for the subgroup analysis for the primary estimand.

7.8. Analysis of the Secondary Efficacy Endpoints

The secondary endpoint(s) of the trial are:

- Change in KCCQ-CSS from baseline to Week 12 and Week 24
- Proportion of patients with ≥1 class improvement in NYHA Functional Class from baseline to Week 12 and Week 24
- Change in post-Valsalva LVOT-G from baseline to Week 12 and Week 24
- Proportion of patients with post-Valsalva LVOT-G <30 mmHg at Weeks 12 and 24
- Total duration of SRT eligibility during the 24-Week treatment period in patients who were SRT eligible at baseline
- Change in total workload during CPET from baseline to Week 24

7.8.1. Analysis of the Secondary Efficacy Endpoints

1. Change in KCCQ-CSS from baseline to Week 12 and Week 24

The primary analysis for change in KCCQ-CSS from baseline to Week 12 and 24 will be performed using a MMRM model with baseline as covariate, randomization stratification factors, visit, treatment group, and interaction terms of treatment by visit and baseline by visit. An unstructured covariance matrix will be specified. All data observed up to Week 24 post randomization will be included in the model. Estimates for endpoints at Week 12 and Week 24 will be obtained from the LS Means estimate at visit of Week 12 and 24 from the model.

If there are >10% difference in baseline KCCQ CSS across regions, treatment by visit and region, baseline by visit and region will be included in the model. Treatment effect at each visit

week will be estimated from the interaction terms of treatment by visit by region, with coefficients for each region determined by proportion of patients evaluated in each region.

Sensitivity analysis based on multiple imputation with one hundred invocations will be performed. First the intermittent missing data will be imputed using the Markov Chain Monte Carlo (MCMC) method under MAR assumption. The imputation will be performed separately for each randomized treatment group and will include the following terms in the imputation model: region (for KCCQ only), endpoint observations from baseline up to Week 24. The monotone missing data will be imputed using the imputation model built from placebo group. 100 complete data sets will be generated and analyzed using the same model for the primary analysis of the change from baseline endpoint. The results from the 100 complete data sets will be combined using Rubin's combination rule for the inference.

Subgroup analysis will be performed using the primary analysis model MMRM model including additional effects of subgroup, subgroup by treatment and subgroup by treatment by visit interaction the model.

2. Change from baseline in post-Valsalva LVOT-G from baseline to Week 12 and Week 24

The analysis of this endpoint will follow the same specified above for change in KCCQ-CSS change from baseline to Week 12 and 24.

3. Proportion of patients with ≥1 class improvement in NYHA Functional Class at Weeks 12 and Week 24

For the proportion of patients with ≥1 class improvement in NYHA class at Week 12, Week 16 NYHA class will be used if Week 12 NYHA class is not available. Week 8 NYHA class will be used if Week 16 NYHA is not available. Patient will be considered as not achieving ≥1 improvement in NYHA class at Week 12 if no NYHA at Weeks 8, 12 and 16 is available. Similarly, Week 20 NYHA class will be used if Week 24 NYHA is not available. Patient will be considered as not achieving ≥1 improvement in NYHA class at Week 24 if Week 20 and Week 24 NYHA are not available. A sensitivity analysis will be performed by repeating Cochran–Mantel–Haenszel (CMH) test by assigning missing NYHA class at Weeks 12 and 24 as non-responders. Proportion of patients with ≥1 class improvement in NYHA class will be analyzed using CMH test stratified by randomization factors. The p-value and 95% CI will be obtained using exact method.

Subgroup analysis will be done by repeating the CMH test for each subgroup level without specifying stratifying by randomization stratification factors.

4. Proportion of patients with post-Valsalva LVOT-G <30 mmHg at Weeks 12 and 24

For the proportion of patients with post-Valsalva LVOT < 30 mmHg at Week 12, Week 16 post-Valsalva LVOT will be used if Week 12 visit is performed but post Valsalva LVOT is not available. Week 8 post-Valsalva LVOT will be used if Week 16 is not available. Similarly, Week 20 post Valsalva LVOT will be used if Week 24 is performed but post Valsalva LVOT is not available.

Proportion of patients with post-Valsalva LVOT-G <30 mmHg at Weeks 12 and 24 will be analyzed using Cochran– Mantel–Haenszel (CMH) test stratified by randomization factors. The p-value and 95% CI will be obtained using exact method.

Subgroup analysis will be done by repeating the CMH test for each subgroup level without specifying stratifying by randomization stratification factors.

5. Total duration of SRT eligibility during the 24-Week treatment period in patients who were SRT eligible at baseline

Patient SRT eligibility will be assigned after data handling in case there are missing NYHA class or LVOT assessments. Intermittent missing NYHA or LVOT will be imputed follows the same imputing method for missing Week 12 or Week 24; e.g., impute Week 16 NYHA if Week 20 NYHA is available or use Week 12 NYHA if Week 20 NYHA is not available. Patient will be treated as SRT eligible if SRT eligibility can't be determined due to the missing NYHA class or LVOT assessments or clinical visits not performed after patient early terminates from the study.

Total duration will be calculated as the number of days patients are SRT eligible, from the date of the randomization until Week 24. Total duration will be 24 weeks if a patient remains SRT eligible at all visits from baseline until Week 24 (regardless of whether a patient is SRT eligible at Week 24 or not). Because the SRT eligibility status of a patient may change during the study conduct, only intervals during which the patient is SRT eligible, will be summed. The start of the interval is the time of randomization (or resumption of SRT eligible status), and the end of the interval is the date prior to the visit when the patient becomes SRT not eligible.

SRT (Y/N) / Case #	Week 2	Week 4	Week 6	Week 8	Week 12	Week 16	Week 20	Week 24	Duration SRT Eligible (Weeks)
1	N	Y	Y	Y	N	N	Y	Y	12
2	Y	Y	N	N	Y	N	N	Y	10
3	Y	Y	N	N	N	N	N	N	6
4	Y	Y	N	N	Y	N	Y	N	14
5	Y	Y	N	N	Y	N	Y	Y	14

Total duration of SRT eligibility during the 24 Week of treatment period will be analyzed using an ANCOVA model includes treatment and randomization stratification factor beta blocker use/non-use as fixed effects and significant baseline characteristics as covariates. Stepwise model selection method will be used based on the default stay or entry level of 0.05 to evaluate significant baseline covariates. Sensitivity analysis of the SRT eligible endpoints includes repeating the same ANCOVA analysis the area under the curve calculated using the numeric value assigned to the SRT eligibility at each visit and treatment duration until Week 24, where SRT eligibility is assigned to value of 1 and SRT not eligible is assigned to value of 0.

6. Change in total workload during CPET from baseline to Week 24

The primary and sensitivity analysis for the total workload will follow the same analysis approach for the primary estimand of the primary endpoint.

Subgroup analysis will be performed similarly as specified for the primary endpoint.

Table 6 summarizes the primary and secondary efficacy endpoints and planned analysis method.

Table 6: Endpoint Summary Table

Endpoint	Primary Analysis Method	Sensitivity/Subgroup Analysis	COVID-19 Related Analyses
Primary Endpoint: Change in pVO2 on CPET from baseline to Week 24			
Primary estimand[1]	Missing data will be imputed using multiple imputation method (Section 7.7.1). Complete dataset will be analyzed using an ANCOVA model with fixed effects of treatment, randomization stratification factors baseline pVO_2 value and baseline body weight	Sensitivity analysis • ANCOVA model will be repeated with missing data from subjects who discontinued aficamten treatment as if the aficamten subjects were in the placebo arm • Tipping point analysis Supportive analysis • Mixed model with numeric covariate as 0 for baseline and placebo group at Week 24 and 1 for aficamten group at Week 24, visit, stratification factors and stratification factors by visit as fixed term. unscheduled covariance structure will be specified. • Multivariate ANCOVA model to evaluate treatment by covariates interaction; ANCOVA model with significant covariates per model selection Subgroup analyses for variables in Section 7.5	• Repeat the primary analysis after setting Week 24 pVO_2 to missing from subjects who were impacted by COVID pandemic • Summarize the number of subjects infected (positive COVID-19 test with or without symptoms) prior to Week 24 CPET.
Secondary estimand	ANCOVA model with fixed effects of treatment, randomization stratification factors baseline pVO2 value and baseline body weight.	Subgroup analyses for variables in Section 7.5	

Table 6: Endpoint Summary Table (Continued)

Endpoint	Primary Analysis Method	Sensitivity/Subgroup Analysis	COVID-19 Related Analyses
Secondary Endpoints			
Change in KCCQ-CSS from baseline to Week 12 and Week 24	MMRM model with baseline as covariate, randomization stratification factors, visit, treatment group, and interaction terms of treatment by visit and baseline by visit. An unstructured covariance matrix will be specified.	Sensitivity analysis: • Intermittent missing data be imputed using multiple imputation MCMC first and the monotone missing values will be imputed using the imputation model built from the placebo group. Complete data will be analyzed using ANCOVA model for each week separately. Subgroup analyses for variables in Section 7.5	
Change in post-Valsalva LVOT-G from baseline to Week 12 and Week 24	MMRM model with baseline as covariate, randomization stratification factors, visit, treatment group, and interaction terms of treatment by visit and baseline by visit. An unstructured covariance matrix will be specified.	Sensitivity analysis: • Intermittent missing data be imputed using multiple imputation MCMC first and the monotone missing values will be imputed using the imputation model built from the placebo group. Complete data will be analyzed using ANCOVA model for each week separately, Subgroup analyses for variables in Section 7.5	
Proportion of patients with ≥1 class improvement in NYHA Functional Class from baseline to Week 12 and Week 24	CMH test stratified by randomization factors	Treating patient as non-responder if NYHA is not performed and repeat CMH test. Subgroup analyses for variables in Section 7.5	

Table 6: Endpoint Summary Table (Continued)

Endpoint	Primary Analysis Method	Sensitivity/Subgroup Analysis	COVID-19 Related Analyses
Proportion of patients with post-Valsalva LVOT-G <30 mmHg	CMH test stratified by randomization factors	Subgroup analyses for variables in Section 7.5	
Total duration of SRT eligible during the 24-week of treatment period	Total duration of SRT eligible during the 24-week of treatment period will be analyzed using an ANCOVA model includes treatment and randomization stratification factor beta blocker use/non-use as fixed effects and significant baseline characteristics as covariates. Stepwise model selection method will be used based on the default stay or entry level of 0.05 to evaluate significant baseline covariates.	SRT eligible will be assigned value of 1 and SRT not eligible will be assigned value of 0. Area under the curve using the numeric value assigned and treatment duration until Week 24 will be calculated. AUC will be analyzed using the same ANCOVA model. Same ANCOVA model will be used to analyze the total time for SRT eligibility.	
Change in total workload during CPET from baseline to Week 24	Missing data will be imputed using multiple imputation method (Section 7.7.1). Complete dataset will be analyzed using an ANCOVA model with fixed effects of treatment, randomization stratification factors baseline total workload value and baseline body weight.	Sensitivity analysis • ANCOVA model will be repeated with missing data from subjects who discontinued aficamten treatment as if the aficamten subjects were in the placebo arm. • Tipping point analysis Subgroup analyses for variables in Section 7.5	

[1]See Table 5 for details on the two estimands for the primary endpoint.

7.9. Analysis of the Exploratory Efficacy Endpoints

Exploratory endpoints are specified in Section 2.2.3. Other echocardiography parameters and CPET parameters not listed as secondary endpoints or in Section 2.2.3 may also be analyzed as exploratory endpoints. All 10 summary scores will be derived for KCCQ. Change from baseline in each summary scores (except CSS) to Weeks 12 and 24 will be analyzed as exploratory endpoints. Patients with ≥5, 10 and 20 points improvement in KCCQ summary scores at Weeks 12 and 24 will also be summarized and analyzed. Domain scores and summary score of SAQ-7 will be derived. Health state for EQ-5D-5L and index value using US value set will be calculated. Analysis of change from baseline in index score and VAS score of EQ-5D-5L be performed. Proportional change in Valsalva and resting LVOT-G will be derived and analyzed as exploratory endpoints.

7.9.1. Analysis of the Exploratory Efficacy Endpoint

The same ANCOVA model specified for the primary endpoint will be used to analyze the endpoints from CPET and continuous CMR measurements that are only assessed once post randomization. Body weight will not be included in the model to analyze CMR parameters. The same MMRM model will be used to analyze the continuous endpoints measured at multiple visits post randomization. CMH test stratified by randomization factors will be used analyze endpoints evaluation proportion of responders or binary outcomes.

Proportion of responders at each week will be summarized. Responders that are defined based on NYHA classification or LVOTs will be assigned after data handling in case there are missing NYHA class or LVOT assessments. Missing NYHA classification or LVOT value will be imputed the same way as that for the secondary endpoints that are based on NYHA classification or LVOT value. Patient will be treated as non-responder if patient's response status can't be determined due to the missing NYHA classification or LVOT assessments (after the imputation above) or clinical visits not performed after the patient early terminates from the study. Proportion of responders will be analyzed using Cochran– Mantel–Haenszel (CMH) test stratified by randomization factors. The p-value and 95% CI will be obtained using exact method. Proportion of patients remaining SRT eligible at Week 24 will be analyzed using logistic model regression model stratified by beta blocker use/non-use adjusting for significant baseline characteristics. The model will include treatment as fixed effect and baseline characteristics as covariates. Covariates measured as continuous will be introduced to the model as continuous variable. Stepwise model selection method will be used based on the default stay or entry level of 0.05 to evaluate significant baseline covariates. Supportive analysis using CMH stratified by randomization factor (beta-blocker use/non-use) will be performed. Another supportive analysis of proportion of patients SRT eligible at each visit will be provided using a repeated measure logistic regression model with treatment, visit, treatment by visit and significant baseline characteristics from the model above. Time to first SRT ineligible in patients who are SRT eligible at baseline will be analyzed using Kaplan-Meier method.

For NT-proBNP and hs-cardiac-TnI, the log transformed proportional change will be analyzed using a MMRM model with log baseline as covariate, treatment group, randomization stratification factors, visit, log baseline by visit and treatment by visit interaction as fixed effects. Log transformed proportional change in Valsalva and resting LVOT will be analyzed using the

same model. Geometric LS Means estimate and ratio of proportional change in NT-proBNP and hs-cardiac-TnI between aficamten vs. placebo, 95% CI of ratio and p-value will be presented. Median and median difference of NT-pro-BNP and hs-cardiac- Tnl between treatment group and 95% confidence of the median difference will be presented at Week 12 and 24. Time to 1 mm ST depression at Week 24 will be analyzed using Cox regression model with treatment, randomization stratification factors as fixed effect. Patients didn't experience 1 mm ST depression will be censored at the end of CPET exercise. Logistic regression model will be fit to LVH strain pattern on ECG for Week 12 and 24, separately. The model will include baseline LVH pattern, stratification factors and treatment. Difference in proportion of patients with no LVH will be estimated and 95% CI for odds ratio (aficamten vs. placebo) and its corresponding p value will be obtained.

8. SAFETY AND TOLERABILITY

Safety and tolerability analyses will be based on the Safety Analysis Set. Safety data will be analyzed descriptively and tabulated by treatment groups.

8.1. Overall Summary of Tolerability

Overall summary of tolerability will include the following:

- Number of patients treated
- Number of patients with TEAEs
- Number of patients with treatment-emergent serious adverse events (TESAEs)
- Number of patients with TEAEs leading to premature treatment discontinuation
- Number of patients with at least one TEAE related to the study drug
- Patients with at least one moderate or severe TEAE
- Patients with at least one severe TEAE
- Number of Deaths

Summary of number and percent of patients with each AE category will be provided by treatment group and dose level at AE onset. Summaries of the number of events will also be provided.

8.2. Adverse Event Preferred Term and Body/Organ System Summary Tables

8.2.1. Summaries of Adverse Event Incidence Rates for All Subjects

All AE terms will be coded using MedDRA. TEAEs and TESAEs will be summarized by primary SOC and PT, and also by severity (mild, moderate and severe) and relationship to study drug (related and not related). For a TEAE reported more than once from a patient, the TEAE will be counted only once in the SOC or PT category using the most severe occurrence or closer relationship to the study drug. All AEs will be listed.

The following subsets of TEAEs will be summarized by SOC and PT:

- All TEAEs
- TEAEs related to study drug
- TEAEs leading to early discontinuation of study drug
- TESAEs

A summary of all TEAEs by PT will be provided. AE summaries will be sorted by descending order of SOC in aficamten group and descending order of preferred term within the SOC. TEAE and TESAE summary of ≥5% and ≥2%, respectively will be provided based on incidence rate in either aficamten or placebo group. Summary of TEAEs by maximum severity will display number and percentage of AEs with maximum severity being mild, moderate, or severe within

each SOC and PT. All TEAE summary will also be provided by dose level. Summary of number of events will be provided for all TEAE summary.

8.2.2. Summaries of Adverse Events of Special Interest

The following events are considered adverse events of special interest:

- Incidence of reported major adverse cardiac events (CV death, cardiac arrest, non-fatal stroke, non-fatal myocardial infarction, CV hospitalization)
- Incidence of new onset persistent atrial fibrillation
- Incidence of ventricular arrhythmias requiring treatment

Summary of patients counts and percentage by each event type will be provided by treatment group.

In addition, incidence of appropriate ICD discharges will be summarized for baseline and also post randomization. Incidence of aborted sudden cardiac death will be provided. Number of patients and number of incidences of LVEF <40% and 50% will be provided by treatment group. Number of patients and number of incidences of LVEF <50% and with signs and symptoms of heart failure (concomitant adverse event of heart failure or dyspnea) or experienced ≥30% increase in NT-proBNP, relative to results from the most recent previous visit and above the upper limit of normal, at the time of LVEF assessment will be provided by treatment group. Signs and symptoms and NT-proBNP increase referring to AEs with onset date or NT-proBNP assessment date within ± 7 days relative to the date when LVEF <50%.

8.3. Total Duration of Therapy, Final Daily Dose of Study Medication, and Compliance

8.3.1. Summary of IP Exposure and Overall Compliance

Total duration of treatment and total exposure of study drug will be summarized. IP compliance will be derived as:

IP compliance =100% * (number of tablets dispensed – number of tablets returned)/expected number of tablets administered.

Number of tablets dispensed and returned will be collected on the study drug accountability eCRF. For the IP kits not returned, the number of tablets returned will be set to 0 in this derivation, assuming all tablets were taken. Expected number of tablets administered will be derived as the number of daily tablets times the days in an IP dosing period, summed over all dosing periods. Days of dosing interruption will be excluded from the expected number of tablets calculation.

8.3.2. Summary of Dose Titration

IWRS-guided dose titration will be summarized showing the number and percentage of patients at each dose level by visit. Number and percentage of patients by last titrated dose will be provided.

8.4. Concomitant and Other Medications

Concomitant medications reported on the eCRF will be summarized. Medications with a start date that is 28 days after the last dose of the study drug will be excluded from the summary. The WHO Drug Dictionary will be used to classify medications by therapeutic class (ATC Class 3) and preferred name. If ATC Class 3 is not available, ATC Class 2 will be used in the summary. Coding will be performed using WHO Drug Dictionary.

8.5. Routine Laboratory Data

Clinical chemistry, hematology and urinalysis laboratory measurements and value changes from baseline at each laboratory blood sample collection time point will be summarized. Values below or above the quantifiable limits will be treated as equal to the limits in the summary. The count and percentage of patients who had normal or missing laboratory values at baseline and abnormal laboratory values post baseline will be presented. The lower limit of normal (LLN) and upper limit of normal (ULN) provided by the laboratories will be used as the criteria to determine abnormality. For each parameter, the denominator of the percentage will include patients with normal or missing assessments at baseline, and with at least one assessment post baseline. The numerator of the percentage will include patients who had at least one abnormal assessment post baseline among the patients that were counted in the denominator. Assessment collected at unscheduled visits, or the Follow-up Visit will be included in the summary.

Shift of clinical laboratory results from baseline severity to the maximum post baseline severity will be presented for selected laboratory parameters.

Liver function test results will be summarized as count and percentage of patients with normal baseline and abnormal post-baseline values in Alanine Aminotransferase (ALT), Aspartate Aminotransferase (AST), Alkaline Phosphatase (ALP) and bilirubin, with the following categories:

- ALT > 3xULN, > 5xULN, > 8xULN
- AST > 3xULN, > 5xULN, > 8xULN
- ALT and/or AST > 3xULN, > 5xULN, > 8xULN
- ALT and/or AST > 3xULN and total bilirubin > 2xULN and ALP < 2xULN
- ALT and/or AST > 3xULN and total bilirubin > 2xULN
- Bilirubin (total, indirect or direct) > 2xULN, >3xULN
- ALT or AST > 3 x ULN with symptoms including nausea, vomiting, anorexia, abdominal pain, fatigue, rash, dark-colored urine, light-colored bowel movements, jaundice, or fever

8.6. Vital Signs

Vital signs and changes from baseline will be summarized descriptively by treatment group over time. The changes from baseline at each post-baseline on-treatment visit will be additionally summarized by dose level at visit.

Patients will also be categorized into the following groups for each of the vital sign parameters if a post baseline value falls into a specific group. Unscheduled assessments will be included in the determination. The number of subjects in each group will be summarized for each dosing group.

Diastolic Blood Pressure

- ≤ 50 mmHg
- ≥ 100 mmHg

Systolic Blood Pressure

- ≤ 80 mmHg
- ≥ 160 mmHg

Heart Rate

- ≤ 50 beats/min
- ≥ 120 beats/min

Respiratory Rate

- > 18 breaths/min

8.7. Electrocardiogram

The baseline ECG is defined as the mean of all pre-dose assessments. PR, RR, QRS, QT, and Fridericia corrected QT (QTcF) intervals and their change from baseline will be summarized by treatment group and scheduled assessment. patients will be categorized into the following groups per their maximum change from baseline in QTcF. Unscheduled assessments will be included in the determination of the maximum change. The number and percentage of subjects in each group will be summarized.

- ≤30 msec
- >30 – 60 msec
- >60 msec

Patients will also be categorized into the following groups per their maximum post baseline QTcF. Unscheduled assessments will be included in the determination of the maximum post baseline value. The number of subjects in each group will be summarized for each dosing group.

- ≤450 msec
- >450 – 480 msec
- >480 – 500 msec
- >500 msec

ECG morphology analyses will be performed. New onset findings will be presented as the percentage of subjects with ‘new’ finding (ECG finding that were not present at any baseline ECG and became present on at least 1 ECG during the treatment) for the following variables

2:1 AV Block, AV Mobitz I,II, Complete heart block, first degree AV block, left atrial abnormality, left ventricular hypertrophy, right ventricular hypertrophy, incomplete left bundle branch block, incomplete right bundle branch block, intraventricular conduction defect, left anterior hemiblock, left bundle branch block, left posterior hemiblock, right bundle branch block, Wolff-Parkinson-White, Artificial pacemaker, atrial pacing, sinus bradycardia, sinus pauses, sinus tachycardia, atrial fibrillation, atrial flutter, atrial tachycardia, supraventricular tachycardia, prolonged QTC, ST depressed, ST elevated, T wave inverted, non-sustained ventricular tachycardia, ventricular fibrillation.

9. PHARMACOKINETICS

Plasma concentrations of CK-3773274 and its measured metabolites and PK parameters $C_{post\ dose}$ and $C_{pre\text{-}dose}$ will be summarized using descriptive statistics including arithmetic mean, standard deviation, coefficient of variation, geometric mean, geometric coefficient of variation, median, and range. Geometric mean concentrations over time will be graphically displayed.

10. REFERENCES

EuroQol Group. (1990). "Eq-5d-3l." EuroQol Research Foundation, from https://euroqol.org/publications/user-guides, Updated Date Accessed Date.

EuroQol Group. (2009). "Eq-5d-5l." EuroQol Research Foundation, from https://euroqol.org/publications/user-guides, Updated Date Accessed Date.

Pickard, A. S., Law, E. H., Jiang, R., Pullenayegum, E., Shaw, J. W., Xie, F., et al. (2019). "United states valuation of eq-5d-5l health states using an international protocol." Value in Health 22(8): 931-941.

Raghunathan, T., & Dong, Q. (2011). Analysis of variance from multiply imputed data sets. *Ann Arbor: University of Michigan*.

Rubin, D. B. (1987). Multiple imputation for nonresponse in surveys. New York, John Wiley & Sons, Inc.

11. APPENDIX

11.1. Patient-reported Outcome Scoring Algorithm

11.1.1. KCCQ

There are 10 summary scores within the KCCQ, which are calculated as follows:

1. Physical Limitation

- Code responses to each of Questions 1a-f as follows:

  Extremely limited = 1
  Quite a bit limited = 2
  Moderately limited = 3
  Slightly limited = 4
  Not at all limited = 5
  Limited for other reasons or did not do = <missing value>

- If at least three of Questions 1a-f are not missing, then compute
  Physical Limitation Score = 100*[(mean of Questions 1a-f actually answered) - 1]/4
  (see footnote at end of this appendix for explanation of meaning of "actually answered")

2. Symptom Stability

- Code the response to Question 2 as follows:

  Much worse = 1
  Slightly worse = 2
  Not changed = 3
  Slightly better = 4
  Much better = 5
  I've had no symptoms over the last 2 weeks = 3

- If Question 2 is not missing, then compute

  Symptom Stability Score = 100*[(Question 2) – 1]/4

3. Symptom Frequency

- Code responses to Questions 3, 5, 7 and 9 as follows:

  <u>Question 3</u>
  Every morning = 1
  3 or more times a week but not every day = 2
  1-2 times a week = 3
  Less than once a week = 4
  Never over the past 2 weeks = 5

  <u>Questions 5 and 7</u>
  All of the time = 1
  Several times a day = 2
  At least once a day = 3

3 or more times a week but not every day = 4
1-2 times a week = 5
Less than once a week = 6
Never over the past 2 weeks = 7

Question 9
Every night = 1
3 or more times a week but not every day = 2
1-2 times a week = 3
Less than once a week = 4
Never over the past 2 weeks = 5

- If at least two of Questions 3, 5, 7 and 9 are not missing, then compute:

S3 = [(Question 3) – 1]/4
S5 = [(Question 5) – 1]/6
S7 = [(Question 7) – 1]/6
S9 = [(Question 9) – 1]/4
Symptom Frequency Score = 100*(mean of S3, S5, S7 and S9)

4. Symptom Burden

- Code responses to each of Questions 4, 6 and 8 as follows:

Extremely bothersome = 1
Quite a bit bothersome = 2
Moderately bothersome = 3
Slightly bothersome = 4
Not at all bothersome = 5
I've had no swelling/fatigue/shortness of breath = 5

- If at least one of Questions 4, 6 and 8 is not missing, then compute

Symptom Burden Score = 100*[(mean of Questions 4, 6 and 8 actually answered) – 1]/4

5. Total Symptom Score

= mean of the following available summary scores:
Symptom Frequency Score
Symptom Burden Score

6. Self-efficacy

- Code responses to Questions 10 and 11 as follows:

Question 10
Not at all sure = 1
Not very sure = 2
Somewhat sure = 3
Mostly sure = 4
Completely sure = 5

Question 11
Do not understand at all = 1
Do not understand very well = 2
Somewhat understand = 3
Mostly understand = 4
Completely understand = 5

- If at least one of Questions 10 and 11 is not missing, then compute

  Self-Efficacy Score = 100*[(mean of Questions 10 and 11 actually answered) – 1]/4

7. Quality of Life

- Code responses to Questions 12, 13 and 14 as follows:

Question 12
It has extremely limited my enjoyment of life = 1
It has limited my enjoyment of life quite a bit = 2
It has moderately limited my enjoyment of life = 3
It has slightly limited my enjoyment of life = 4
It has not limited my enjoyment of life at all = 5

Question 13
Not at all satisfied = 1
Mostly dissatisfied = 2
Somewhat satisfied = 3
Mostly satisfied = 4
Completely satisfied = 5

Question 14
I felt that way all of the time = 1
I felt that way most of the time = 2
I occasionally felt that way = 3
I rarely felt that way = 4
I never felt that way = 5

- If at least one of Questions 12, 13 and 14 is not missing, then compute

  Quality of Life Score = 100*[(mean of Questions 12, 13 and 14 actually answered) – 1]/4

8. Social Limitation

- Code responses to each of Questions 15a-d as follows:

Severely limited = 1
Limited quite a bit = 2
Moderately limited = 3
Slightly limited = 4
Did not limit at all = 5
Does not apply or did not do for other reasons = <missing value>

- If at least two of Questions 15a-d are not missing, then compute

  Social Limitation Score = 100*[(mean of Questions 15a-d actually answered) – 1]/4

9. Overall Summary Score

= mean of the following available summary scores:
Physical Limitation Score
Total Symptom Score
Quality of Life Score
Social Limitation Score

10. Clinical Summary Score

= mean of the following available summary scores:
Physical Limitation Score
Total Symptom Score

Note: references to **"means of questions actually answered"** imply the following.

- If there are n questions in a scale, and the subject must answer m to score the scale, but the subject answers only n-i, where n-i >= m, calculate the **mean of those questions** as

  (sum of the responses to those n-i questions) / (n-i)
  not

(sum of the responses to those n-i questions) / n

11.1.2. SAQ-7

Three domain scores and one summary score are generated from the SAQ-7:

Physical Limitation Score (SAQ7-PL)

Angina Frequency Score (SAQ7-AF)

Quality of Life Score (SAQ7-QL)

Summary Score (SAQ7)

Scores are scaled 0-100, where 0 denotes the lowest reportable health status and 100 the highest.

Physical limitation Score

The physical Limitation score corresponds to Questions 1a, 1b and 1c. Responses are coded as follows:

Extremely limited	1
Quite a bit limited	2
Moderately limited	3
Slightly limited	4
Not at all limited	5
Limited for other reasons or did not do the activity	6

A response of 6 is treated as missing value for the purpose of scoring. If responses to two or more questions are missing, no score is computed. If the response to Question 1a or Question 1c is missing, it is assigned the responses from Question 1b. If the response to Question 1b is missing, it is assigned the average of responses to Questions 1a and 1c. The score is then calculated by taking the average of the three responses and rescaling to 0 – 100, as follows:

SAQ7-PL = 100*[(average of Questions 1a, 1b and 1c)-1]/4

Angina Frequency Score

The Angina Frequency score corresponds to Questions 2 and 3. Responses are coded as follows:

4 or more times per day	1
1 – 3 times per day	2
3 or more times per week but not every day	3
1 -2 times per week	4
Less than once a week	5
None over the past 4 weeks	6

If responses to both questions are missing, no score is computed. Otherwise, the score is calculated by taking the average of non-missing responses and rescale to 0-100 as follows:

SAQ7-AF = 100 *[(average of Questions 2 and 3) -1]/5

Quality of Life Score

The quality of life score corresponds to Questions 4 and 5. Responses are coded as follows:

Question 4	
It has extremely limited my enjoyment of life	1
It has limited my enjoyment of life quite a bit	2
It has moderately limited my enjoyment of life	3
It has slightly limited my enjoyment of life	4
It has not limited my enjoyment of life at all	5
Question 5	
Not satisfied at all	1
Mostly dissatisfied	2
Somewhat satisfied	3
Mostly satisfied	4
Completely satisfied	5

If responses to both questions are missing, no score is computed. Otherwise, the score is calculated by taking the average of the non-missing response and rescaling to 0 -100, as follows:

SAQ7-QL= 100*[(average of Question 4 and 5)-1]/4

Summary Score

The summary score represents an integration of the patients' physical limitation, angina symptom and quality of life. If all three domain scores are missing, no summary score is computed. Otherwise, the score is calculated as the average of the non-missing domain scores:

SAQ7 = average of SAQ- PL, SAQ- AF, and SAQ – QL

11.1.3. EQ-5D-5L

Five dimensions of the EQ-5D-5L include 'mobility', 'selfcare', 'activity', 'pain', and 'anxiety'. The US Pickard value set will be used to compute the EQ-5D-5L index values. The value set will be denoted as disut_mo for 'mobility', disut_sc for 'selfcare', disut_ua for 'activity', disut_pd for 'pain', and disut_ad for 'anxiety' in Table 7 below:

Table 7: EQ-5D-5L Value Set

		US value set
MOBILITY		disut_mo
I have no problems in walking about	1	0
I have slight problems in walking about	2	0.096
I have moderate problems in walking about	3	0.122
I have severe problems in walking about	4	0.237
I am unable to walk about	5	0.322
SELF-CARE		disut_sc
I have no problems washing or dressing myself	1	0
I have slight problems washing or dressing myself	2	0.089
I have moderate problems washing or dressing myself	3	0.107
I have severe problems washing or dressing myself	4	0.220
I am unable to wash or dress myself	5	0.261
USUAL ACTIVITIES		disut_ua
I have no problems doing my usual activities	1	0
I have slight problems doing my usual activities	2	0.068
I have moderate problems doing my usual activities	3	0.101
I have severe problems doing my usual activities	4	0.255
I am unable to do my usual activities	5	0.255
PAIN / DISCOMFORT		disut_pd
I have no pain or discomfort	1	0
I have slight pain or discomfort	2	0.060
I have moderate pain or discomfort	3	0.098
I have severe pain or discomfort	4	0.318
I have extreme pain or discomfort	5	0.414
ANXIETY / DEPRESSION		disut_ad
I am not anxious or depressed	1	0
I am slightly anxious or depressed	2	0.057
I am moderately anxious or depressed	3	0.123
I am severely anxious or depressed	4	0.299
I am extremely anxious or depressed	5	0.321
We would like to know how good or bad your health is TODAY	0 to 100	

disut_total=disut_mo+disut_sc+disut_ua+disut_pd+disut_ad;
The EQ-5D-5L index value (EQindex) =1-disut_total

The SAS code will be provided in Appendix Section 11.5

11.2. Table of Contents for Data Display Specifications

Table of contents for data display specifications will be provided in a separate document.

11.3. Data Display Specifications

Data display specifications will be provided in a separate document.

11.4. Analysis Windows

Measurements collected during the 24-week double-blind placebo-controlled period will be included only in the analysis windows up to Week 24. For data collected at a scheduled post randomization, the analysis visit will be the nominal visit as collected and visit window will not be applied.

For unscheduled or early discontinuation post randomization, analysis visit will be used according to Table 8 below when the scheduled visit is not available.

Table 8: Analysis Windows for Measurements

Visit	Scheduled Day	Lower Bound	Upper Bound
Screening	<1	<1	<1
Day 1	1	1	1
Week 2	15	2	21
Week 4	29	22	35
Week 6	43	36	49
Week 8	57	50	70
Week 12	85	71	98
Week 16	113	99	126
Week 20	141	127	154
Week 24	169	155	196
Week 28/Follow-up	last dose + 28 days	-	-

Note: Week 28/Follow-up should occur 4 weeks after last dose. The analysis visit of week 28/Follow-up will include the nominal visit End of Study (Week 28) and any unscheduled visits occurred after the week 24 window.

11.5. Sample SAS Codes

ANCOVA model for primary endpoint

```
proc mixed data=work;
        class <treatment arm (ref='0')> <Beta Blocker use> <Exercise Modality>;
        model chg=<base pvo2 > <base weight> <treatment arm> <beta blocker use> <Exercise
        Modality> /solution s Influence(EFFECT=usubjid) outp=out vciry;
        lsmeans <treatment arm>/pdiff cl;
        ods output solutionf=mixparms covB=mixcovb;
run;
```

This code assumes that the analysis involves 2 levels in treatment arm (e.g., placebo group is coded as 0 and aficamten is coded as1).

MMRM model

```
proc mixed data=work;
     class <Subject> <treatment arm (ref='0')> <Beta Blocker use> <Exercise Modality> <visit>;
model <chg> = <base> <treatment arm> <visit> <visit>*<treatment arm> <Beta Blocker use> <Exercise
Modality> <visit>*<base>/ddfm=kenwardroger;
repeated <visit> / type=un subject=<Subject>;
lsmeans <visit>*<treatment arm>/cl pdiff;
run;
```

This code assumes 2 level in treatment arm with placebo group is coded as 0. visit has level of nominal visit week where continuous measurements are assessed up to Week 24.

Imputation model for primary endpoint

```
proc mi data=work seed=&seed out=miout NIMPUTE=50;
        class <treatment arm> <Beta Blocker use> <Exercise Modality> <base NYHA> <randomization
        factors>;
        var <treatment arm> <randomization stratifications> <sex> <age> <base pVO2> <base
        hemoglobin> <base KCCQ CSS> <baseline NYHA> <last available post rand NYHA> <last
        available post rand resting LVOT> <last available post rand Valsalva LVOT> <W24 pVO2> ;
        monotone reg(W24 pVO2);
 run;
```

Placebo-based imputation for sensitivity analysis of the primary endpoint

Step 1:
generate input dataset for subjects in placebo group or subject who didn't complete treatment, and datasets for the rest of subjects in aficamten group.
Step2: impute missing data with separately with each input dataset using the imputation model for the primary endpoint.
Step 3. Combined the complete data from the imputations above as final imputed dataset.

Tipping point imputation for primary endpoint

```
proc mi data=work seed=&seed out=miout NIMPUTE=50;
        class <treatment arm> <Beta Blocker use> <Exercise Modality> <base NYHA> <>;
        var <treatment arm> <sex> <age> <base pVO2> <base hemoglobin> <base KCCQ CSS>
        <baseline NYHA> <last available post rand NYHA> <last available post rand resting LVOT>
        <last available post rand Valsalva LVOT> <W24 pVO2> ;
        monotone reg(W24 pVO2);
        mnar adjust (W24 pVO2/shift=&shift adjustobs=(Trt='1'));
run;
```

shift will be set so that the imputed value for aficamten group is 1 ml/min/kg worse as starting point with increment of 1 ml/min/kg worse each time until the result is no longer significant.

Multiple Imputation for sensitivity analysis for repeated measures endpoint

```
Step1:
proc mi data=work1 seed=&seed1 out=miout1 NIMPUTE=100;
        by <treatment arm>;
        mcmc IMPUTE=monotone ;
        var <base> <var week 2> <var week4> <var week6> <var week8> <var week12> <var week16>
<var week20> <var week24>;
run;
```

Step 2:

```
proc mi data=miout1 seed=&seed NIMPUTE=1 OUT=miout2;
 class <treatment arm>;
 by _IMPUTATION_;
 var <treatment arm> <base> <var week 2> <var week4> <var week6> <var week8> <var week12> <var
week16> <var week20> <var week24>;
 monotone reg (<base> <var week 2> <var week4> <var week6> <var week8> <var week12> <var
week16> <var week20>);
 mnar model (<base> <var week 2> <var week4> <var week6> <var week8> <var week12> <var
week16> <var week20> / modelobs = (trt01pn = '0'));
run;
```

Multiple Imputation for categorical endpoint

```
proc mi data=work seed=&seed out=outwork NIMPUTE=100;
         class <var at week8> < var at week 12> < var at week 16> < var at week 20> < var at week 24>
<treatment arm>;
  var <treatment arm> <var at week8> < var at week 12> < var at week 16> < var at week 20> < var at
week 24> ;
fcs logistic (<var at week8> < var at week 12> < var at week 16> < var at week 20> < var at week 24> =
<treatment arm> /link=glogit);
run;
```

SAS codes using proc minalyze to combine results from imputed datasets

The pooled estimates from the 50 imputed datasets are obtained from the following codes.

```
proc mianalyze data=est;
  modeleffects estimate;
  stderr stderr;
run;
```

Mixed model for CPET data

```
proc mixed data=work;
    class <subject ID> <visit> <stratification factors> ;
    model <chg in CPET>=<trtid> <stratification factors> <stratification factors>*<visit> /s;
    estimate 'active at Week 24' int 1 <trtid> 1 <stratification factors> &c1 &c2 stratification
    factors>*<visit> 0 0 &c1 & / e;
    estimate 'placebo at Week 24' int 1 <trtid> 0 <stratification factors> &c1 &c2 stratification
    factors>*<visit> 0 0 &c1 & / e;
    estimate 'active vs PBO at Week 24' trt 1;
    repeated <visit>/subject=<subject id> type=un;
run;
```

where trtid is assigned as 0 at baseline, 0 for placebo group at Week 24 and 1 for active group at Week 24. &c1 and &c2 are the proportion of patients evaluated in each stratification level among all the patients.

Subgroup analysis for the primary endpoint

```
proc mianalyze parms=mixparms covb(effectvar=rowcol)=mixcovb;
    class <treatment arm> <beta blocker use> <exercise modality> <subgroup> ;
    =<base pvo2 > <base weight> <treatment arm> <beta blocker use> <Exercise modality>
    modeleffects Intercept <base pvo2 > <base weight> <treatment arm> <beta blocker use>
    <Exercise Modality> <subgroup>*<treatment arm>;
    run;
```

SAS codes to evaluate normality assumption for the primary endpoint

The normality assumptions for the ANCOVA analysis will be assessed by residual illustration. The outpred option in the above code stores residuals which are used to test the assumption of normality. Examination of residuals can be done using the following codes.

```
proc univariate data=work normal;
var ScaledResid; QQPLOT ScaledResid;
ods output QQPlot=qqplot;
run;
```

SAS codes to perform CMH test

```
proc freq data=work;
 tables <Beta blocker use>*<CPET modality>*<treatment arm>*<response Y/N>/CMH
 exactcmh;
exact riskdiff (column=2) relrisk (column=2) commor.
Run;
where response is coded as Y=1, N=0.
```

SAS codes to rank observation

```
proc rank data=work out=rwork nplus1;
Var chg;
Ranks rchg;
Run;
```

SAS code for proportional hazard Cox regression model

```
Proc phreg data=work;
        Class <treatment arm>(ref=first) ;
        Model <time>*censor (1)= ;
        Strata <beta blocker use> <CPET modality> ;
Run;
```

SAS code for stratified logistics regression model

```
Proc phreg data=work;
        Class <treatment arm>;
        Model <Response> (event='Y')= <treatment arm> <baseline covariates>/selection=stepwise
details;
        Strata <beta blocker use> ;
Run;
```

SAS code for EQ-5D-5L Index Value

```
**********************************************************************
*SAS syntax code for the computation of index*
*values with the US TTO value set*
**********************************************************************

data WORK.CAT;
set WORK.CAT;

if mobility eq 1 then disut_mo=0;
else if mobility eq 2 then disut_mo=0.096;
else if mobility eq 3 then disut_mo=0.122;
else if mobility eq 4 then disut_mo=0.237;
else if mobility eq 5 then disut_mo=0.322;

if selfcare eq 1 then disut_sc=0;
else if selfcare eq 2 then disut_sc=0.089;
else if selfcare eq 3 then disut_sc=0.107;
else if selfcare eq 4 then disut_sc=0.220;
else if selfcare eq 5 then disut_sc=0.261;

if activity eq 1 then disut_ua=0;
else if activity eq 2 then disut_ua=0.068;
else if activity eq 3 then disut_ua=0.101;
else if activity eq 4 then disut_ua=0.255;
else if activity eq 5 then disut_ua=0.255;

if pain eq 1 then disut_pd=0;
else if pain eq 2 then disut_pd=0.060;
else if pain eq 3 then disut_pd=0.098;
else if pain eq 4 then disut_pd=0.318;
else if pain eq 5 then disut_pd=0.414;

if anxiety eq 1 then disut_ad=0;
else if anxiety eq 2 then disut_ad=0.057;
else if anxiety eq 3 then disut_ad=0.123;
else if anxiety eq 4 then disut_ad=0.299;
else if anxiety eq 5 then disut_ad=0.321;

disut_total=disut_mo+disut_sc+disut_ua+disut_pd+disut_ad;
EQindex=1-disut_total;
run;
```

Signature Manifest

Document Number: PRD-0381 Revision: 00

Title: CY 6031 - SAP Version 2 - A Phase 3, Multi-Center, Randomized, Double-blind, Placebo-controlled Trial to Evaluate the Efficacy and Safety of CK-3773274 in Adults with Symptomatic Hypertrophic Cardiomyopathy and Left Ventricular Outflow Tract Obstruction

Effective Date: 09 Oct 2023

All dates and times are in Pacific Time.

CY 6031 - SAP Version 2 - A Phase 3, Multi-Center, Randomized, Double-blind, Placebo- controlled Trial to Evaluate the Efficacy and Safety of CK-3773274 in Adults with Symptomatic Hypertrophic Cardiomyopathy and Left Ventricular Outflow Tract Obstruction

1: Electronic Approvals

Name/Signature	Title	Date	Meaning/Reason
[redacted]	Senior Director, Biostatistics	05 Oct 2023, 11:52:16 AM	Approved
[redacted]	SVP, Chief Medical Officer	05 Oct 2023, 11:54:20 AM	Approved
[redacted]	Vice President, Biometrics	05 Oct 2023, 06:10:52 PM	Approved
[redacted]	SVP, Regulatory Affairs & Quality	05 Oct 2023, 07:36:03 PM	Approved
[redacted]	VP, Clinical Research & Therapeutic Area Lead, Cardiovascular	09 Oct 2023, 01:39:31 PM	Approved