Statistical Analysis Plan

A Phase Ib/II, single center, placebo-controlled, randomized, blinded study in adult patients (> 18 years) with COVID-19 Respiratory Disease, to evaluate, Safety, Tolerability and mechanistic effect of Alvelestat on top of standard of care (COSTA)

1 STATISTICAL ANALYSIS PLAN APPROVAL

The signatures below indication review and approval of this Statistical Analysis Plan (SAP).

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2 LIST OF ABBREVIATIONS

Abbreviation	Definition
AE	Adverse Event
CI	Confidence Interval
COVID	Coronavirus Disease
CRF / eCRF	Case Report Form / Electronic Case Report Form
CSR	Clinical Study Report
ECMO	Extracorporeal Membrane Oxygenation
FDA	Food and Drug Administration
ICH	International Conference on Harmonization
ITT	Intention to Treat
MedDRA	Medical Dictionary for Regulatory Activities
Mg	Milligram
N	Number of patients/subjects
PCR	Polymerase Chain Reaction
PI	Principal Investigator
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SARS	Severe Acute Respiratory Syndrome
SD	Standard Deviation
SOC	Standard of Care
US	United States
WHO	World Health Organization

3 PREFACE

The SAP for "A Phase Ib/II, single center, placebo-controlled, randomized, blinded study in adult patients (≥ 18 years) with COVID-19 Respiratory Disease, to evaluate, Safety, Tolerability and mechanistic effect of Alvelestat on top of standard of care (COSTA)" describes and expands upon the statistical information presented in the protocol.

This document describes all planned analyses and provides reasons and justifications for these analyses. It also includes sample tables, listings, and figures planned for the final analyses.

Any deviation from this SAP will be described and justified in protocol amendments and/or in the clinical study report (CSR), as appropriate. The reader of this SAP is encouraged to also review the study protocol for details on conduct of the study and the operational aspects of clinical assessments.

4 INTRODUCTION

Neutrophils and neutrophil extracellular traps (NETs) have recently been established as a central feature of SARS-CoV-2 pathogenesis, and NET formation (NETosis) are associated with poor outcomes in patients hospitalized with COVID-19 (Zuo et al 2020; Barnes et al 2020). NETs are networks of extracellular DNA fibers, histones, myeloperoxidase (MPO) and neutrophil elastase (NE), released from neutrophils and involved physiologically in capturing bacteria. However, if uncontrolled, NETs are cytotoxic to endothelial and epithelial cells, acting as Damage Associated Molecular Pattern Molecules (DAMPs) and promoting cytokine release and thromboses (Kim et al 2019). Levels of NETs correlate with disease severity in ALI/ARDS (Li et al 2018) Neutrophil elastase is essential to NET formation (Papayannopoulos et al 2010) and remains active when decorating the NET fibers. The physiological inhibitor of NE is Alpha-1 antitrypsin (AAT), but studies in COVID-19 have shown this to be overwhelmed (McElvanay et al 2020). Providing NE inhibition therefore has potential to reduce both NET formation and damaging activity of existing NETs. In experimental models of ARDS, alvelestat, the NE inhibitor proposed for this clinical trial, protected the lung from inflammation and injury through NET inhibition (Li et al 2018). There is also evidence from clinical efficacy studies using NE inhibitors in ALI/ARDs. Sivelestat ('Elastpol') is a selective human neutrophil elastase inhibitor that is approved in Japan and South Korea for the treatment of ALI/ARDS accompanied by Systemic Inflammatory Response Syndrome. A meta-analysis of 6 randomised controlled trials in patients with all forms of ALI/ARDS, did not demonstrate an effect on mortality, Intensive Care Unit (ICU) or ventilator days. However, a sensitivity analysis detected a significant improvement in oxygen deficit measured by PaO2/FiO2 (SMD: 0.87; 95% CI: 0.39 to 1.35; $P < 0.001$, (Pu et al 2017) and oxygenation of Day 3 (Iwata et al 2010). Sivelestat appeared to have an effect on mortality in ARDS associated with disseminated intravascular coagulation (DIC) (Miyoshi et al 2014), a neutrophil-predominant disease, which is more closely aligned with COVID, and provides support for the NE mechanism.

Alvelestat (MPH966, AZD9668) is an oral, specific, potent human NE inhibitor (Stevens et al 2011). Alvelestat effectively inhibits neutrophil elastase activity in man, including during maximal neutrophil stimulation. It shows predictable systemic pharmacokinetic (PK) profile and therapeutically relevant concentrations in sputum (Gunawardena et al 2013). Clinical safety and efficacy experience has been gained in over 1000 healthy volunteers and patients with lung disease, including Chronic Obstructive Pulmonary Disease (COPD), cystic fibrosis and bronchiectasis (Kuna et al 2012; Elborn et al 2012; Stockley et al 2013). There has been evidence of mechanistic effect in clinical studies of neutrophil-driven diseases, with significant decrease in biomarkers of elastase activity in cystic fibrosis (Elborn et al 2012), and clinically significant improvement in lung function (Forced Expiratory Volume in 1 second [FEV1]) in bronchiectasis (Stockley et al 2013), with reduction in the pro-inflammatory biomarkers Interleukin (IL)-6 and RANTES in sputum and blood observed in both these studies. Alvelestat is currently being investigated in ongoing clinical trials in alpha-1 antitrypsin deficiency lung disease (Clinical

Trials.Gov NCT03636347; NCT03679598) and Bronchiolitis Obliterans Syndrome associated with chronic graft versus host disease (NCT02669251).

The scientific rationale for NE inhibition in SARS-CoV-2 infection, the evidence for an effect in other inflammatory lung diseases and the safety profile for alvelestat, supports investigating alvelestat in COVID-19. Here, we propose a pilot randomized, placebo-controlled trial assessing the safety and tolerability and exploring mechanistic effect on NETosis and clinical outcomes of alvelestat given on top of standard of care, compared to standard of care alone in patients hospitalized with COVID-19

4.1 Purpose of the Analyses

This SAP encompasses the final analysis of primary and secondary outcome measures. These analyses will be included in the CSR.
This is a pilot study which will expand our knowledge and understanding about how alvelestat can be used to facilitate recovery or prevent deterioration in hospitalized, COVID-19 patients.

5 INVESTIGATIONAL PLAN

5.1 Overall Study Design and Plan

This is a Phase Ib/II single-center, 1:1 randomized, blinded, placebo-controlled, parallel group, first in disease study. The purpose is to determine the safety, tolerability and PK and explore the mechanistic and clinical effect of alvelestat up to 240mg orally twice per day for 10 days added to standard of care in adult patients (≥18 years) with COVID-19 respiratory disease.

5.2 Objectives

The primary objective is to evaluate the safety and tolerability of alvelestat administered twice per day for 10 days in patients hospitalized with COVID-19.
Secondary objectives include the evaluation of:

- The effect of alvelestat on blood pharmacodynamic markers of Neutrophil Extracellular Trap (NET) activation, elastase, inflammatory and coagulopathy activity,
- The effect of alvelestat on alvelestat on oxygenation.
- The effect of alvelestat on disease progression
- The effect of alvelestat on clinical outcomes
- The effect of alvelestat on disease severity
- To explore pharmacokinetics (PK) in patients with COVID-19

5.3 Selection of Study Population

Male and non-pregnant female adults ≥18 to 80 years of age or older with COVID-19 and who meet all eligibility criteria will be enrolled at one site.

5.3.1 Inclusion Criteria

- Male or Female
- Age ≥18 years
- Proven SARS-Cov-2 infection (confirmed by PCR from a nasopharyngeal or lower respiratory tract sample)
- A score of Grade 3 to 5 on the WHO 9-point Ordinal Scale
- Male participants must agree to use a highly effective contraception
- Female participants are eligible to participate if not pregnant; not breastfeeding; and at least one of the following conditions is met:
    - Not a woman of childbearing potential
      OR
    - A woman of childbearing potential who agrees to follow the contraceptive

guidance during the treatment phase and for at least 4 days after the last dose of study medication

- Capable of giving signed informed consent which includes a commitment to comply with the requirements and restrictions listed in the informed consent form (ICF) and within this protocol.

5.3.2 Exclusion Criteria

- Patients who have previously had a score of 6 or 7 on the WHO 9-point Ordinal Scale
- Patients who require support with invasive mechanical ventilation at the time of inclusion, or expected to be required within 24 hours of randomization
- Alanine aminotransferase (ALT) OR aspartate aminotransferase (AST) >2 × the upper limit of normal (ULN) OR Total Bilirubin > ULN. In patients with a documented history of Gilbert's Syndrome AND baseline total bilirubin elevation consistent with an exacerbation of Gilbert's Syndrome (i.e. no other cause of total bilirubin elevation), subjects may enroll if total bilirubin is < 5x ULN.
- Diagnosis of liver cirrhosis, esophageal varices, ascites or hepatic encephalopathy
- Chronic liver diseases such as autoimmune hepatitis, primary biliary cholangitis, primary sclerosing cholangitis, Wilson's disease, haemochromatosis
- Significant renal disease or infection (as determined by the Investigator) including stage 4 chronic kidney disease or estimated glomerular filtration rate <60mL/min
- Absolute neutrophil count ≦ 1000/μL at screening
- Myocardial infarction, transient ischemic attack or stroke within 3 months prior to the first dose
- Current unstable angina or congestive heart failure (New York Heart Association III/IV)
- Screening 12-lead EKG with a measurable QTc interval according to Fridericia correction (QTcF) >450 ms
- Anticipated transfer to another hospital that is not the study center within 24 hours
- Allergy to study medication or excipients
- Inability to swallow tablets
- Other documented comorbidities or laboratory abnormalities that in the opinion of the Investigator could affect the outcome of the study assessments, participant safety, or ability of the participant to comply with the requirements of the protocol
- Any patient whose interests are not best served by study participation, as determined by the Investigator

Excluded Prior/Concomitant Therapy

- Requirement for medications mainly metabolized by CYP2C9 and with narrow therapeutic index (eg, warfarin, phenytoin) is prohibited unless therapeutic monitoring available for duration of alvelestat dosing
- Medicines that are potent CYP3A4 inhibitors including (but are not limited to) clarithromycin, diltiazem, erythromycin, itraconazole, ketoconazole, ritonavir, verapamil and potent inducers including but not limited to phenobarbital, phenytoin and rifampicin, will be exclusionary
- Requirement for medications substantially reliant on OATP1B1 for metabolism where discontinuation during study drug administration is not possible or where fluctuations in levels are considered clinically important (as per investigator judgement) and cannot be clinically monitored (e.g., statins, valsartan, olmesartan, enalapril, repaglinide)

Excluded Prior/Concurrent Clinical Study Experience

- Participation in any clinical investigation using investigational treatments within 4 weeks or 5 half-lives of the drug (whichever is longer) prior to the initial dosing (or longer if required by

local regulations) is prohibited. Use of remdesivir (Veklury) under the conditions of the authorization for emergency use in the US, and per manufacturer's instructions, is permitted.

5.4 Treatments Administered

Once eligibility is confirmed, patients will be randomized in a 1:1 ratio to one of two treatment arms listed in Table 1. Dosing will be performed twice daily approximately 12 hours apart in the morning and evening at approximately the same time each day.

Table 1. COSTA treatment arms.

Treatment Arm	n	Treatment
Treatment Arm 1	7-8	Blinded alvelestat (240 mg) tablets BID + SoC*
Treatment Arm 2	7-8	Blinded placebo tablets BID + SoC*
Abbreviations: SoC= standard of care, BID= twice daily, *SoC treatment for COVID-19 infection based on established practices within UAB		

5.5 Method of Assigning Patients to Treatment Groups (Randomization)

Randomization will take place prior to the first study medication administration in the order patients are enrolled and in accordance with a computer-generated randomization list supplied by the an independent statistician through the UAB Research Pharmacy. Available randomization numbers must be used sequentially for the next enrolled patient. Patients will be dosed with either blinded alvelestat or placebo according to the randomization scheme.

5.5.1 Selection and Timing of Dose for Each Patient

Each patient is randomly assigned 1:1 to a treatment group. Study medication dosing starts on Day 1 after randomization and continues twice daily, approximately every 12 hours, until discharge or Day 10, whichever is first.

5.5.2 Blinding

The UAB Research Pharmacy's unblinded pharmacist will label the study medication and perform accountability measures.

The blind will be broken only if specific emergency treatment would require knowing the treatment status of the patient or if the protocol review committee recommends unblinding data for safety review reasons. If the blind needs to be broken for an individual patient, the Investigator will contact the Sponsor as soon as feasible. The Investigator may unblind the study medication immediately if he/she feels it is necessary prior to contacting the Sponsor. However, the Investigator should promptly document and explain to the Sponsor any premature unblinding. Otherwise, all blinding will be maintained until all queries are resolved and the database is locked.

5.6 Prior and Concomitant Therapy

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

- UAB's standard treatment for COVID-19 infection is permitted, with the exception of prohibited medications listed below:
- Requirement for medications mainly metabolized by CYP2C9 and with narrow therapeutic index (eg, warfarin, phenytoin) is prohibited unless therapeutic monitoring available for duration of alvelestat dosing
- Medicines that are potent CYP3A4 inhibitors including (but are not limited to) clarithromycin, diltiazem, erythromycin, itraconazole, ketoconazole, ritonavir, verapamil

and potent inducers including but not limited to phenobarbital, phenytoin and rifampicin, will be exclusionary

- Requirement for medications substantially reliant on OATP1B1 for metabolism where discontinuation during study drug administration is not possible or where fluctuations in levels are considered clinically important (as per investigator judgement) and cannot be clinically monitored (e.g., statins, valsartan, olmesartan, enalapril, repaglinide)

5.7 Treatment Compliance

All days the study medication was received and the doses/day will be recorded on the appropriate eCRF. If study medication dose was missed, decreased or halted, the information will be recorded to track compliance.

6 DEFINITIONS

6.1 General

General clinical study definitions are listed in Table 2.

Table 2. COSTA clinical study general definitions.

Term	Definition for Study
Baseline	The most recent pre-dose assessment prior to first dose of study medication.
Clinical status assessment	Each assessment will be recorded as a number in the CRF based on the WHO Ordinal Scale status categories below. Where possible the timing of each assessment should be similar (i.e. morning or evening throughout). The Ordinal Scale is adapted from the WHO R&D Blueprint expert group which measures illness severity over time (https://www.who.int/blueprint/priority-diseases/key-action/COVID19_Treatment_Trial_Design_Master_Protocol_synopsis_Final_18022020.pdf according to the following criteria: 0. Uninfected, no clinical or virological evidence of infection 1. Ambulatory, no limitation of activities 2. Ambulatory, limitation of activities 3. Hospitalized – mild disease, no oxygen therapy* 4. Hospitalized – mild disease, oxygen by mask or nasal prongs* 5. Hospitalized – severe disease, noninvasive ventilation or high flow oxygen* 6. Hospitalized – severe disease, intubation and mechanical ventilation 7. Hospitalized – severe disease, ventilation and additional organ support – vasopressors, renal replacement therapy, extracorporeal membrane oxygenation 8. Death
COVID disease severity	At baseline/randomization, the categorization of COVID-19 disease severity will be in accordance with the FDA's document: COVID-19: Developing Drugs and Biological Products for Treatment or Prevention Guidance for Industry (May 2020). • Mild disease (meeting all inclusion and no exclusion criteria): Symptoms of mild illness that could include fever, cough, sore throat, malaise, headache, muscle pain, gastrointestinal symptoms, without shortness of breath or dyspnea and no clinical signs indicative of moderate, severe, or critical severity.

Term	Definition for Study
	• Moderate disease (meeting all inclusion and no exclusion criteria): could include any symptom of mild illness or shortness of breath with exertion. Clinical signs suggestive of moderate illness, such as respiratory rate ≥ 20 breaths per minute, saturation of oxygen (SpO2) > 93% on room air at sea level, heart rate ≥ 90 beats per minute and no clinical signs indicative of severe or critical severity. • Severe disease (meeting all inclusion and no exclusion criteria): could include any symptom of moderate illness or shortness of breath at rest, or respiratory distress. Clinical signs indicative of severe systemic illness, such as respiratory rate ≥ 30 per minute, heart rate ≥ 125 per minute, SpO2 ≤ 93% on room air at sea level or PaO2/FiO2 < 300 and no criteria of critical severity. • Critical disease: evidence of critical illness, defined by at least one of the following: 1) respiratory failure defined based on resource utilization requiring at least one of the following: endotracheal intubation and mechanical ventilation, oxygen delivered by high-flow nasal cannula (heated, humidified, oxygen delivered via reinforced nasal cannula at flow rates > 20 L/min with fraction of delivered oxygen ≥ 0.5), noninvasive positive pressure ventilation, ECMO, or clinical diagnosis of respiratory failure (i.e., clinical need for one of the preceding therapies, but preceding therapies not able to be administered in setting of resource limitation); 2) shock (defined by systolic blood pressure < 90 mm Hg, or diastolic blood pressure < 60 mm Hg or requiring vasopressors); or 3) multi-organ dysfunction/failure.
Day	Calendar day
Day 1	The day a patient received their first dose of study medication. • If a patient received their 1st dose of study medication the day following randomization, the study day number will be adjusted so that Day 1 = 1st dose of study medication.
Discharge	The patient is discharged and leaves the hospital alive. This does not include if a patient is moved to another unit. • The time to discharge = the elapsed time (in days) from Day 1 to the earliest day at which the patient is Discharged from hospital (= Day where the Ordinal Scale score is equal to 1 or 2).
Improvement in clinical status	A decrease from baseline of at least one point in the Ordinal Scale score (eg, 5 to 4).
Worsening clinical status	An increase from baseline of at least one point in the Ordinal Scale score (eg, 5 to 6).

6.2 Endpoints

Clinical study endpoints and associated definitions are listed in Table 3.

Table 3. COSTA clinical study endpoint definitions.

Endpoints		Definitions
Primary	To evaluate the safety and tolerability of alvelestat administered twice daily (bid) for 5 or 10 days	By treatment group (alvelestat/placebo): • Numbers and % of subjects who experience at least 1 treatment-emergent adverse event to Day 60 (Safety Outcome Assessment) • Adverse events of special interest (liver function abnormalities, corrected QT interval, infections, and neutropenia) and clinically significant safety monitoring labs tests to EoT • Number and % of subjects that discontinue alvelestat to Day 5 or 10 • Number and % of subjects requiring dose reduction to Day 5 or 10 • Number and % of subjects with AE of headache on prospective questioning and CTCAE severity Grade to Day 5 or 10 • Change in vital signs, EKG, safety laboratory measures to Day 5 or 10 or EOT
Secondary	To evaluate the effect of alvelestat on blood pharmacodynamic markers of Neutrophil Extracellular Trap (NET) activation, elastase, inflammatory and coagulopathy activity	By treatment group (alvelestat/placebo), change from baseline to Day 5 or 10 in: • Blood biomarkers of NETosis (cell-free DNA; citrullinated histone H3; Myeloperoxidase (MPO)-DNA complexes) • Blood biomarkers of inflammation (absolute neutrophil count; absolute lymphocyte count; neutrophil-lymphocyte ratio [NLR], C-Reactive protein, procalcitonin • Pro-inflammatory cytokines IL-1beta, IL-6, IL-8, TNF-alpha) • Coagulopathy as measured by D-Dimer • Plasma desmosine/isodesmosine
	To explore the effect of alvelestat on pulmonary function	By treatment group (alvelestat/placebo): Proportion of subjects alive and free of respiratory failure (without need for non-invasive or invasive mechanical ventilation, high flow oxygen, or Extracorporeal Membrane Oxygenation (ECMO) at Day 29
	The effect of alvelestat on clinical outcomes	By treatment group (alvelestat/placebo): • Mortality rate at Day 90 • Proportion of subjects alive and free of respiratory failure (without need for non-invasive or invasive mechanical

Endpoints		Definitions
		ventilation, high flow oxygen, or ECMO) at Days 5, 10 and 60 • Length of hospital stay • Length of ICU stay Time to room air (those on supplemental oxygen at Baseline)
	The effect of alvelestat on disease severity	By treatment group (alvelestat/placebo): • Percentage of patients deteriorating (>1 and >2 increase in WHO 9-point ordinal scale*) at Day 5, Day 10, Day, Day 29 and Day 60 Percentage of patients improving (>1 and > 2 decrease in WHO 9-point ordinal scale) at Day 5, Day 10, Day 29 and Day 60
	To explore Pharmacokinetics (PK) in patients with COVID-19	Plasma PK (sparse sampling taken pre or 1-2 hours post study drug dosing) in participants randomized to alvelestat.

7 GENERAL STATISTICAL CONSIDERATIONS

7.1 Sample Size

This is a pilot study and it is not powered based on any specific endpoint. Sample size has been set to minimize the number of patients exposed, but still large enough to give reliable estimates on the efficacy and safety of alvelestat in the target population.

7.2 General Principles

This is a double-blind, placebo controlled randomized pilot study which may inform on treatment effect for future studies.

Descriptive statistics

Continuous variables will be summarized using descriptive statistics including number of non-missing observations (n), arithmetic mean (Mean), median (Median), standard deviation (SD), minimum value (Min) and maximum value (Max).

For categorical variables, summaries will include the number of non-missing observations (n) or the number of patients in the population (N) as applicable, the counts of subjects and percentages. Percentages will be rounded to one decimal place.

Statistical Analyses

All analyses are considered exploratory and hypothesis-generating. All tests will be two-sided at 5% significance level. Hypothesis tested for the primary endpoint will be if addition of alvelestat to SoC for up to 10 days will be safe and well tolerated as compared to standard of care plus placebo.
Secondary endpoints are tested independently and there will be no adjustment for multiple tests.

7.3 Analysis Populations

Summaries and analysis of safety data will be presented for the Safety Analysis Population.
Summaries and analysis of efficacy data will be presented for the intent-to-treat (ITT) population.

- Intent to treat (ITT): Will consist of all randomized patients. Efficacy will be evaluated as randomized.
- Modified intention to treat (mITT): Will consist of randomized participants that have baseline and at least one follow-up assessment available.
- Per protocol population (PP): Will consist of all patients in ITT without any major protocol deviations (determined prior to database lock) deemed to have an effect on efficacy analyses.
- Safety set (SS): Will consist of all randomized and treated patients (at least one dose). Safety will be evaluated by actual treatment.

7.4 Missing Data

All attempts will be made to collect all data per protocol.

- The patients withdrawn from treatment will be followed up regarding their status according to the WHO Ordinal Scale at Day 29 as far as possible (exceptions for patients with withdrawn consent or lost to follow-up) and this data will be the primary source for handling missing data in the study.

Any data point that appears to be erroneous or inexplicable based on clinical judgment will be investigated as a possible outlier.

- If data points are identified as outliers, sensitivity analyses may be performed to examine the impact of including or excluding the outliers. Any substantive differences in these analyses will be reported.

For survival analyses, patients with death as outcome will be censored at Day 29 (Day 60), unless the required event was reached prior to death. Patients who are lost to follow-up or terminate the study prior to Day 29 (Day 60) for other reason than death and prior to observing/experiencing the event will be censored at the time of their last observed assessment. The only time to event analyses is the time to recovery.

For secondary endpoint analyses, the following imputation rules will be used for patients who are lost to follow-up, terminate early from the study, or do not have further outcome data available after discharge for any reason than death:

- Patients with no data collected at the actual time point of interest for other reason than death, the last assessment of their status prior to this time point will be used as a representative for that time point. Especially, if the patient's clinical status scale is 1 or 2 at the last observed assessment, then the patient will be considered to be recovered through Day 29.
- For endpoints assessing total time (oxygen or non-invasive ventilator care), the status for patients with no data collected for other reason than death, will be assumed unchanged from last assessment of the score up to last day in the definition of the endpoint (Day 29). Thus, no extra days will be added for patients without oxygen or patients without non-invasive ventilator care at last assessment.
- Patients that die in the study will be handled as worst case in all the analyses, that is they are considered to be on oxygen need, non-invasive and invasive mechanical ventilator need for all time points following the death.
- In analyses of oxygen need, patients in ventilator care (non-invasive or invasive mechanical) will be considered in oxygen need for the corresponding time period and patients with death as outcome will be considered in oxygen need for the remainder of the time period of evaluation.
- In analyses of non-invasive ventilator need, patients that require invasive mechanical ventilation will be considered also as having a non-invasive ventilator need. Patients with death as an outcome will be considered as having non-invasive ventilator need from time of death for the remainder of the time period of evaluation.

- In analyses of invasive mechanical ventilator need, patients with death as an outcome will be considered in invasive mechanical ventilator need for the remainder of the time period of evaluation
- The change from baseline in the WHO ordinal scale will use the baseline score and the score at the endpoint using imputation for missing data as stated above, that is the last assessment of their status prior to this time point will be used.
- If the patient is discharged and no further hospitalization data are available, then the patient will be assumed to not have been readmitted. Thus, no additional imputed days on hospitalization will be added to the number of days recorded on available assessments for these patients.

7.5 Interim Analyses
No interim analyses will be performed.

7.6 Safety Analysis
Time to first combined event.
Any patients that are lost to follow-up or terminated early prior to experiencing any of the events will be censored at the day of their last observed assessment.
Patients who complete follow-up but do not experience any of the events will be censored at the day of their Day 29 visit.

8 ANALYSIS
8.1 Descriptive Information
Table 4 lists the summaries for patient disposition, analysis populations and protocol deviations.

Table 4. COSTA analysis tables - Disposition.

Table #	Table Name	Table Contents
	Patient Dispositon.	The disposition of patients by treatment group and include study milestones • Total number screened, randomized, completed 5, 10 days of treatment, completed Day 29, Day 60 and Day 90 follow-ups and withdrawn patients in total and by withdrawal reason.
	Screen Failures.	A summary of the reasons that patients were screened but not enrolled.
	Major Protocol Deviations.	Major protocol deviations summarized by the reason for the deviation, the deviation category, treatment group, COVID disease severity and (separately) site for all patients.
	Analysis Sets	The composition of analysis populations, including reasons for patient exclusion by treatment group.

Table 5 summarizes patient demographics. Demographics include age/DOB, sex, height, weight, BMI (calculated), ethnicity, and race.

Table 5. COSTA analysis tables - Demographics.

Table #	Table Name	Table Contents
	Baseline Demographic Characteristics.	Overall, by treatment group, by COVID disease severity and by baseline clinical

		status assessment using the WHO ordinal scale for ITT, PP and SS (if different).

Summaries of baseline clinical characteristics are displayed in Table 6. Baseline clinical characteristics include brief physical exam findings, vital signs, clinical status assessment using the WHO ordinal scale score, COVID-19 symptom onset, admission date, COVID disease severity, existing co-morbidities, and COVID-19 standard medications and treatments.

Table 6. COSTA analysis tables - Baseline Clinical Characteristics.

Table #	Table Name	Table Contents
	Baseline clinical characteristics (Eg. WHO ordinal scale score, vital sign values, COVID disease severity, co-morbid conditions - obesity, hypertension, diabetes, asthma, and COPD).	Overall, by treatment group and COVID disease severity for ITT, PP and SS (if different).
	Proportion patients with WHO ordinal scale score of 3, 4 and 5, ITT.	Overall and by treatment group.
	Proportion patients with COVID disease severity of mild, moderate, severe and critical, ITT.	Overall and by treatment group.
	Disease onset (days with COVID symptoms, hospital admission date, COVID disease severity and oxygen need at randomization), ITT.	Overall and by treatment group.
	Number and proportion taking concomitant dexamethasone or remdesivir, ITT.	Overall, by treatment group and COVID disease severity.

8.2 Study Medication Compliance

Summaries of study non-medication compliance are listed in Table 7.

Table 7. COSTA analysis tables - Study Medication Compliance.

Table #	Table Name	Table Contents
	Study Medication Compliance (number of patients who had study medication halted/slowed and the number of patients with missed doses)	The number of patients with discontinued or missed doses (except due to discharge) by treatment group for ITT and PP.
	Patient study medication discontinuation by study day.	By treatment group for ITT and PP.

8.3 Primary Endpoint Analysis

The primary endpoint is safety and tolerability of alvelestat. Safety endpoints include SAEs and AEs. Adverse events will be analyzed using quantitative and qualitative measures.
Treatment-emergent adverse events will be summarized by treatment group for all AEs, related AEs, serious adverse events, deaths, adverse events leading to discontinuation of study medication or to withdrawal from study, adverse events of different severity and adverse events of different chronicity.
Treatment-emergent adverse events will be coded using MedDRA and summarized by system organ class and preferred term for each treatment group.

The time to the composite AE endpoint (death, SAE, severe AE or ET/W of treatment during first 10 days) will be compared between treatment groups using the log-rank test and difference between treatment arms will be expressed as a difference in proportions as measured by the Cochran Mantel-Haenzel (stratum)-weighted estimator based on the fitted logistic regression model

- If '0' events, proportions will be presented.

Summaries of the results of the safety analysis are displayed in **Table 8**.

Table 8. COSTA analysis tables - Safety Analyses.

Table #	Table Name
	Time to first composite AE (Safety Analysis Set)
	Number/Proportion Treatment emergent adverse events summary (Safety Analysis Set)
	Number/Proportion TEAEs leading to discontinuation of study treatment by system organ class and preferred term (Safety Analysis Set)
	Number/Proportion TEAEs with an outcome of death by system organ class and preferred term (Safety Analysis Set)

Deaths, Serious Adverse Events and other Significant Adverse Events
Narratives will be provided for deaths and serious adverse events.

Pregnancies.
For any patients in the Safety population who become pregnant during the study, every attempt will be made to follow these patients to completion of pregnancy to document the outcome, including information regarding any complications with pregnancy and/or delivery.

8.4 Secondary Endpoint Analysis

The biological effect of alvelestat will be assessed through measurement of pharmacodynamic markers of NET activation, elastase activity, inflammation, and coagulopathy as detailed in Table 9.

Table 9. COSTA analysis tables - Biomarker Analyses.

Table #	Table Name	Table Contents
	Descriptive Summary over time and Change from baseline in the amount of NETosis: amount of cell-free DNA, MPO-DNA complexes, and mount of citrullinated histone H3	Overall and by Treatment group Day 1 – Day 5/10/EOT
	Descriptive Summary over time and Change from baseline in blood biomarkers of inflammation (absolute neutrophil count; absolute lymphocyte count; neutrophil-lymphocyte ratio [NLR], C-Reactive protein)	Overall and by Treatment group Day 1 – Day 5/10/EOT
	Descriptive Summary over time and Change from baseline in blood pro-inflammatory cytokines IL-1beta, IL-6, IL-8, TNF-alpha	Overall and by Treatment group Day 1 – Day 5/10/EOT
	Descriptive Summary over time and Change from baseline in d-dimer	Overall and by Treatment group Day 1 – Day 5/10/EOT

Table #	Table Name	Table Contents
	Descriptive Summary over time and Change from baseline in plasma desmosine/isodesmosine	Overall and by Treatment group Day 1 – Day 5/10/EOT

- Change from baseline in markers of NETosis will be compared between treatments using ANCOVA models with treatment allocation as a factor and baseline marker of NETosis as a covariate. Estimated treatment difference with 95% confidence intervals and associated 2-sided p-value will be given.
- Change from baseline in markers of inflammation (e.g. CRP) will be compared between treatments using ANCOVA models with treatment allocation as a factor and baseline inflammatory marker as a covariate. Estimated treatment difference with 95% confidence intervals and associated 2-sided p-value will be given.
- Change from baseline in cytokines will be compared between treatments using ANCOVA models with treatment allocation as a factor and baseline cytokine value as a covariate. Estimated treatment difference with 95% confidence intervals and associated 2-sided p-value will be given.
- Change from baseline in D-dimer will be compared between treatments using ANCOVA models with treatment allocation as a factor and baseline D-dimer as a covariate. Estimated treatment difference with 95% confidence intervals and associated 2-sided p-value will be given.
- Change from baseline in desmosine/isodesmosine will be compared between treatments using ANCOVA models with treatment allocation as a factor and baseline desmosine/isodesmosine as a covariate. Estimated treatment difference with 95% confidence intervals and associated 2-sided p-value will be given.
- Biomarker values that are below the lower limit of quantitation of assay will be reported as the lower limit of quantitation (LLQ)/2.
- Biomarker values may be log transformed if appropriate.

The proportion of patients with recovery at Day 29 will be compared between treatment groups using a logistic regression model adjusting for treatment and COVID disease severity. The difference between treatment arms will be expressed as a difference in proportions as measured by the Cochran Mantel-Haenzel (stratum)-weighted estimator based on the fitted logistic regression model. The delta method will be used to calculate the standard error for the difference and the associated confidence interval [Ge, 2011]. In addition, the raw proportions per treatment group will be given. If 0 events in any of the groups only raw proportions will be given.

Summaries of the results of the secondary analysis for recovery are displayed in Table 10.

Table 10. COSTA analysis tables – Secondary Analysis (Recovery)

Table #	Table Name	Table Contents
	Proportion of Patients with Recovery at 29 Days, ITT	Overall and by Treatment Group • By Age Group • By Sex • By COVID Disease Severity
	Proportion of Patients with Recovery at 29 Days, PP	Overall and by Treatment Group

Patients that die in the study will be handled as worst case in all analyses.
To have an evaluation using all patients we will count an event as the type of need or worse (e.g., patients with oxygen need; non-invasive ventilator need; ventilator need; or with death at time of evaluation will be counted as having data supporting oxygen need), and included in both denominator and numerator.

8.4.1 Time to a recovery state (Day 1 - Day 29; measured in days)
The time to recovery from Day 1 to Day 29 will be compared between treatment groups using a log-rank test; patients not in recovery at end-of study/withdrawal will be censored at last observed time point in the treatment period or at last time status was assessed if collected post treatment-withdrawal. Patients with death as outcome will be censored at the scheduled end-of-study Day 29. Data will be illustrated using Kaplan-Meier plots and the median time to recovery estimated if appropriate.

8.4.2 Proportion of patients endpoint analyses
Secondary endpoints assessing proportion of patients fulfilling an event will be compared between treatments using a logistic regression model adjusting for treatment. The difference between treatment arms will be expressed as a difference in proportions as measured by the Cochran Mantel-Haenzel (stratum)-weighted estimator based on the fitted logistic regression model. The delta method will be used to calculate the standard error for the difference and the associated confidence interval. For endpoints with a low number of events, the baseline disease state will be dropped from the model. In addition, the raw proportions per treatment group will be given. If 0 events in any of the groups only raw proportions will be given.

- Proportion of patients who are in a recovery state (Days 29 and 60).
- Proportion of patients with improvement of one category using the WHO ordinal scale (Days 10 and 29).
- Proportion of patients with improvement of two categories using the WHO ordinal scale (Days 10 and 29).
- Proportion of patients in use of oxygen (Days 10 and 29).
- Proportion of patients alive and not in respiratory failure (invasive mechanical ventilation or ECMO (Days 10 and 29).
- Proportion of patients receiving non-invasive ventilation (Days 10 and 29).
- All-cause mortality

8.4.2.1 Sub-sets based on post-randomization outcome
Descriptive summaries of the proportion of patients as well as the duration of the worsening following initiation of study medication will be provided for these endpoints:

- Proportion of patients needing re-hospitalization (post-dose Day 1 – Day 60).
    - Applies to the sub-set of patients that were randomized, treated with study medication and discharged from the hospital alive then re-admitted to the hospital for COVID-19 related treatment.
- Proportion of patients and duration of new oxygen use (post-dose Day 1 - Day 29; measured in days).
    - Applies to the sub-set of patients that were randomized and treated with study medication but then had a new need for supplemental oxygen (score worsened to 4, 5 or 6).
    - Or those which improved during hospitalization (to 3) and then their clinical status worsened again (to 4, 5 or 6).
- Proportion of patients and duration of new non-invasive ventilation or high flow oxygen use (post-dose Day 1 - Day 29; measured in days).

- Applies to the sub-set of alive patients that were randomized and treated with study medication but then had a new need for non-invasive ventilation or high-flow oxygen use (score worsened to 5 or 6).
- Or those which improved during hospitalization (to 3 or 4) and then their clinical status worsened again (to 5 or 6)

The total number of days with recorded use will be the sum of all reported days, regardless of whether the days occur consecutively or in disjoint intervals.

8.4.3 Change from baseline in WHO ordinal scale (Days 10 and 29).
Change from baseline in WHO ordinal scale will be compared between treatments using ANCOVA models with treatment allocation as a factor and baseline WHO score as a covariate. Estimated treatment difference with 95% confidence intervals and associated 2-sided p-value will be given.

8.4.4 Total time on supplemental oxygen (Day 1 - Day 29; measured in days).
The total number of days with recorded use will be the sum of all reported days, regardless of whether the days occur consecutively or in disjoint intervals. Results will be summarized by descriptive statistics including medians and quartiles by treatment arm and compared between treatment groups using Wilcoxon rank sum test. Patients that do not reach the specified score on any day will be counted as 0 days.

8.4.5 Total time on non-invasive ventilator use (Day 1 - Day 29; measured in days).
The total number of days with recorded use will be the sum of all reported days, regardless of whether the days occur consecutively or in disjoint intervals. Results will be summarized by descriptive statistics including medians and quartiles by treatment arm and compared between treatment groups using Wilcoxon rank sum test. Patients that do not reach the specified score on any day will be counted as 0 days.

8.4.6 Duration of hospitalization (Day 1 - Day 29; measured in days)
The total number of days with recorded use will be the sum of all reported days, regardless of whether the days occur consecutively or in disjoint intervals. Results will be summarized by descriptive statistics including medians and quartiles by treatment arm and compared between treatment groups using Wilcoxon rank sum test.

Summaries of the results of the secondary analyses are displayed in Table 11.

Table 11. COSTA analysis tables - Secondary Analyses.

Table #	Table Name	Table Contents
	Time to recovery, Day 1 - Day 29	Overall and by Treatment group
	Proportion of patients in recovery at Day 60, ITT	Overall and by Treatment group Day 1 – Day 10 Day 1 – Day 60
	Proportion of patients with improvement of one category using the WHO ordinal scale from Day 1, ITT	Overall and by Treatment group Day 1 – Day 10 Day 1 – Day 29
	Proportion of patients with improvement of two categories using the WHO ordinal scale from Day 1, ITT	Overall and by Treatment group Day 1 – Day 10 Day 1 – Day 29
	All-cause mortality, ITT	Overall and by Treatment group At Day 29 At Day 60

Table #	Table Name	Table Contents
		At Day 90
	Proportion of patients alive and not in respiratory failure (invasive mechanical ventilation or extracorporeal membrane oxygenation (ECMO) at Day 29, ITT	Overall and by Treatment group Day 1 – Day 10 Day 1 – Day 29
	Summary of the proportion of patients re-hospitalized following initiation of study medication and subsequent discharge (over 60 days), ITT	Overall and by Treatment group
	Duration of hospitalization from Day 1 (Day 1 - Day 29; measured in days), ITT	Overall and by Treatment group
	Change from baseline WHO ordinal scale at Days 10 and 29, ITT	Overall and by Treatment group Baseline – Day 10 Baseline – Day 29
	Total time on supplemental oxygen (Day 1 - Day 29; measured in days), ITT	Overall and by Treatment group
	Total time of non-invasive ventilation or high flow oxygen use (post-dose Day 1 - Day 29; measured in days).	Overall and by Treatment group
	Summary of the proportion of patients with oxygen use at Days 10 and 29, ITT	Overall and by Treatment group
	Summary of the proportion of patients with non-invasive ventilation or high flow oxygen use at Days 10 and 29), ITT	Overall and by Treatment group
	Proportion of patients receiving invasive mechanical ventilation or extracorporeal membrane oxygenation (ECMO, WHO ordinal scale criteria) at Days 10 and 29, ITT	Overall and by Treatment group Day 1 – Day 10 Day 1 – Day 29

8.5 Data Listings

Relevant subject data, including those derived, will be presented in individual subject data listings. All listings will be sorted by treatment regimen, subject number, date/time and visit. Listings will be based on all patients randomized

8.6 Figures

The study specific figures are displayed in Table 12.

Table 12. COSTA Figures.

Figure #	Figure Name
1	Box and whisker plot for each biomarker by treatment group (e.g. marker of NETosis) at day 1, Day 5and Day 10
2	Kaplan–Meier plot of time to Recovery, by treatment group ITT
3	Histogram of ordinal scores at Day 10 and 29 by treatment group and baseline ordinal scale, ITT.

9 REPORTING CONVENTIONS

P-values will be reported to three decimal places; p-values less than 0.0005 will be reported as "<0.001" and p-values greater than 0.9995 will be reported as ">0.999".
The mean, standard deviation and other statistics will be reported to1 decimal place greater than the original data. The minimum and maximum will use the same number of decimal places as the original data.
Proportions will be presented as 2 decimal places; values greater than zero but <0.005 will be presented as "<0.01".
Estimated parameters, not on the same scale as raw observations (e.g. regression coefficients) will be reported to 3 significant figures.

10 TECHNICAL DETAILS

Additional summaries/data points may be included in the final version of a table, figure, or listing. Additional tables, figures, and listings may be generated to supplement the planned output.

11 REFERENCES

1 Ge M, Durham LK, Meyer RD, Xie W, Thomas N. Covariate-adjusted Difference in Proportions from Clinical Trials Using Logistic Regression and Weighted Risk Differences. Drug Information Journal, Vol. 45, pp. 481–493, 2011