Pivotal

STATISTICAL ANALYSIS PLAN

Study Code: ATL001-PI-CEP

Protocol Version: 4.0

Protocol Date: 13 - December - 2023

A Phase I, double-blind, randomized, placebo-controlled study to assess the safety and pharmacokinetics of ATL-001 (ciclopirox olamine) in healthy volunteers

Investigational Product	ATL-001
Indication Studied	N/A
NCT No	05647343
Phase of Study	Phase 1
Sponsor (company and address)	Atlas Molecular Pharma S.L., Parque Tecnológico de Bizkaia, Bld. 800

Pages Nº	32
SAP Version	2.1
SAP Date:	26-Abr-2024

Approvals

By signing this document, I acknowledge that I have read the Statistical Analysis Plan and approve of the planned statistical analysis described herein.

I agree that the planned statistical analyses are appropriate for the objective of the study and are consistent with the methodology described in the protocol, clinical development plan.

I also understand that any subsequent changes to the statistical analyses, as described herein, may have a regulatory impact and/or result in timeline adjustments. All changes to the planned analyses will be described in the clinical study report.

PIVOTAL

Developed by:

[redacted]

Signature / Date

Review by:

[redacted]

Signature / Date

WuXi Clinical

Approved by:

[redacted]

Signature / Date

ATLAS Molecular Pharma

Approved by:

[redacted]

Signature / Date

Table of Contents

STATISTICAL ANAYSIS PLAN VERSION

This section details the changes compared to previous version of the statistical analysis plan (SAP).

SAP Version	Date of SAP version	Author	Changes from the previous version
1.0	09-Aug-2023	[redacted]	NA
2.0	11-Mar-2024	[redacted]	Inclusion of additional cohort (new version of the protocol v4.0)
2.1	26-Apr-2024	[redacted]	Include the definition for calculating the derived variable Urea.

1. LIST OF ABBREVIATIONS

Abbreviation	Definition
AE	Adverse Event
AESI	Adverse Event of Special Interest
ANOVA	Analysis of Variance
ATC	Anatomical Therapeutic Chemical
$AUC_{(0-12)}$	Area Under the Curve from time 0 to 12 hours after drug administration
$AUC_{(0-24)}$	Area Under the Curve from time 0 to 24 hours after drug administration
$AUC_{(0-last)}$	Area Under the Curve from time 0 to the time of the last measured concentration
BLQ	Below the limit of quantitation
CEP	Congenital Erythropoietic Porphyria
CI	Confidence Interval
CL/F	Total body clearance
C_{max}	Maximum observed plasma drug concentration
COVID	Coronavirus Disease 2019
CRO	Contract Research Organization
CSP	Clinical Study Protocol
CSR	Clinical Study Report
DLT	Dose-Limiting Toxicity
DSMB	Data and Safety Monitoring Board
ECG	Electrocardiogram
eCRF	Electronic Case Report Form
eGFR	Estimated Glomerular Filtration Rate
FU-V	Follow-Up Visit
GCP	Good Clinical Practice
HIV	Human Immunodeficiency Virus
ICH	International Conference on Harmonization
LLoQ	Lower limit of quantitation
IMP	Investigational Medicinal Product
MedDRA	Medical Dictionary for Regulatory Activities
PCR	Polymerase Chain Reaction
PI	Principal Investigator
PK	Pharmacokinetics
QTcF	Corrected QT interval by Fredericia
RA	Regulatory Authority
RBMP	Risk-Based Monitoring Plan
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SOC	System Organ Class
SOP	Standard Operating Procedure
SUSAR	Suspected Unexpected Serious Adverse Reaction
$t_{1/2}$	Terminal half-life
TEAE	Treatment-Emergent Adverse Event

Abbreviation	Definition
t_{max}	Time to Maximum observed plasma drug concentration
UROIIIS	Uroporphyrinogen III Synthase
Vz/F	Volume of distribution
WHO	World Health Organization

2. INTRODUCTION

The Statistical Analysis Plan (SAP) has been written based on the Clinical Study Protocol (CSP) version 4.0 (13 December 2023).

The purpose of this document is to describe the procedures and the statistical methods to be applied in the data analysis and to ensure that the statistical methodologies that will be used are complete and appropriate to the study objectives.

The focus of this SAP is for the planned primary, secondary, and exploratory analysis at the final analysis in the study.

Pivotal and WuXi will perform the statistical analyses described in the protocol. Result of the analyses described in this SAP will be include in the Clinical Study Report (CSR).

2.1 Background & Rationale

Porphyria is a group of disorders resulting from deficient activity of specific enzymes in the heme biosynthetic pathway. Congenital erythropoietic porphyria (CEP), also known as Günther's disease, is a very rare inherited metabolic disorder resulting from a deficient activity of the enzyme uroporphyrinogen III synthase (UROIIIS), the fourth enzyme in the heme biosynthetic pathway.

ATL-001 is a new oral formulation of ciclopirox olamine, including alginate and tocophersolan, currently under development for the treatment of subjects with CEP. This new oral formulation was developed to improve the local tolerance of ciclopirox olamine, which otherwise induces dose dependent gastrointestinal toxicity.

The present randomized, placebo-controlled, dose escalation, Phase I study aims to investigate the safety, tolerability, and PK of ATL-001 at five different dose levels in healthy subjects.

The results of this study are intended to be used to identify appropriate and well tolerated doses of ATL-001 to be used in further studies.

2.2 Study Objectives

2.2.1 Primary Objective

- To investigate the safety and tolerability of ATL-001 in healthy subjects.

2.2.2 Secondary Objectives

- To determine the PK of ATL-001 in healthy subjects

2.3 Study Design

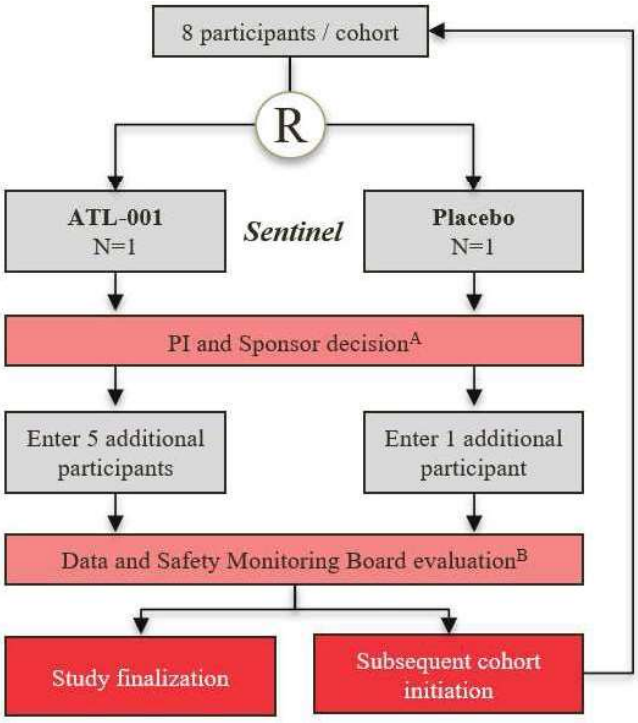
This is a Phase I, double-blind, randomized, parallel-design, placebo-controlled, and dose escalation study to investigate the safety and PK of ATL-001 (ciclopirox olamine) in healthy subjects.

The study will consist of five sequentially recruited cohorts of 8 subjects each, resulting in up to 40 randomized subjects. Subjects will not be allowed to participate in more than one cohort.

Five different ATL-001 doses will be tested along with their matching placebos: ATL-001 0.2 mg/kg, ATL-001 0.5 mg/kg, ATL-001 1 mg/kg, ATL-001 2 mg/kg and ATL-001 4 mg/kg. All of them are named as IMP in this plan. On the morning of Day 1 to Day 5, each subject will receive a single oral dose of ATL-001 or placebo (i.e., total of 5 administrations of IMP) administered under fasting conditions (overnight fast; at least 10 hours). Refer CSP Section 8.1 for more details.

The day of the first administration of the IMP will be considered Day 1 of the study.

Study Schema (per cohort):



2.3.1 Randomization

At each investigated cohort, subjects will be randomized in the ratio of 6 active treatments to 2 placebos (3:1). For safety reasons, the subjects of each cohort will be divided in 2 sub-cohorts with 2 sentinel subjects in the first sub-cohort and 6 additional subjects in the second sub-cohort. The randomization schedule will be designed so that in each sub cohort there will be 1 subject scheduled to receive placebo. In order to maintain the double-blind study, the administration of the IMP must occur individually and separately, and in the absence of the physician responsible for the study. This is because the reaction after its administration could be easily distinguishable between placebo and ATL-001.

2.3.2 Study treatment

The product name is referred to as ATL-001. ATL-001, ciclopirox olamine oral solution, 20 mg/ml is a new oral formulation of ciclopirox. On the morning of Day 1 to Day 5, each subject will receive a single oral dose of ATL-001 or placebo (i.e., total of 5 administrations of IMP) administered under fasting conditions (overnight fast; at least 10 hours) following the instructions below:

Cohort	Investigational medicinal product	Route	Administration Days
1	ATL-001 0.2 mg/kg / Placebo	Oral	1, 2, 3, 4, 5
2	ATL-001 0.5 mg/kg / Placebo	Oral	1, 2, 3, 4, 5
3	ATL-001 1 mg/kg / Placebo	Oral	1, 2, 3, 4, 5
4	ATL-001 2 mg/kg / Placebo	Oral	1, 2, 3, 4, 5
5	ATL-001 4 mg/kg / Placebo	Oral	1, 2, 3, 4, 5

2.3.3 Replacement of Subjects

In case of a withdrawn subject from the study, he/she may be replaced with another subject at the Sponsor's discretion. The replacement subject will be assigned to the same regimen dosage.

2.3.4 Follow up

The follow-up period will include 2 visits, one scheduled 3 days after the last IMP dose and one scheduled 30 days after the last IMP dose.

FU-V1 (3 days after the last IMP dose) and FU-V2 (30 days after the last IMP dose)

At both FU visits, the following activities and assessments will be performed:

- Assessment of changes in the medical history.
- Physical examination.
- Vital signs, including blood pressure, pulse rate and body temperature.
- Laboratory safety assessments, including hematology, biochemistry, and urinalysis.
- ECG.
- Concomitant medications.
- AEs.

2.3.5 Study Visits and Procedures

	Screening period	Treatment period												Follow-up period	
	Screening visit	Hospital Admission											Hospital discharge†	FU-V1	FU-V2
Day	Day -30 to Day -1	Baseline (Day 1)	Day 1 and Day 5										Day 2 and Day 6	3 days after the last IMP dose	30 days after the last IMP dose
Assessment time		Pre-dose (0 h)	15 min (±5 min)	30 min (±5 min)	1 h (±10 min)	1:30 h (±10 min)	2 h (±10 min)	2:30 h (±15 min)	3 h (±15 min)	4 h (±30 min)	6 h (±30 min)	8 h (±30 min)	12 h (±30 min)	24 h (±30 min)		
Informed consent	X															
Inclusion/Exclusion criteria	X	X[1]														
Randomization		X														
Demographic data	X															
Medical history	X	X[2]												X[2]	X[2]	
Physical examination	X	X					X							X	X	X
Vital signs[3]	X	X					X							X	X	X
Laboratory safety assessments[4]	X	X					X							X	X	X
Viral serology[5]	X															
Drug of abuse and alcohol screen	X	X														
Pregnancy test (females only)[6]	X	X[7]														
ECG	X	X					X							X	X	X
PK blood sampling		X[7]	X	X	X	X	X	X	X	X	X	X	X	X		
Concomitant medications	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Adverse events[8]	X[9]	X[9]	X	X	X	X	X	X	X	X	X	X	X	X	X	X

ECG, electrocardiogram; FU-V, Follow-up Visit; h, hour; IMP, investigational medicinal product; min, minutes; PK, pharmacokinetics

† Subjects will be discharged at Day 2 and Day 6 after completing their assessments.

[1] Assessment of changes in inclusion/exclusion criteria

[2] Assessment of changes in the medical history

[3] Vital signs include: blood pressure, pulse rate and body temperature

[4] Laboratory safety assessments include: hematology, biochemistry, and urinalysis

[5] Viral serology will comprise human immunodeficiency virus (HIV) I and II, hepatitis B virus surface antigen (HBsAg) and hepatitis C virus testing, and coronavirus disease (COVID)-19 testing by polymerase chain reaction (PCR)

[6] Pregnancy testing of female subjects only. At screening pregnancy test will be done with using the blood sample; urine pregnancy test will be done on admission.

[7] Also pre-dose at Day 5

[8] Adverse events will be daily recorded by the subject in the subject diary and will not involve visit to the hospital.

[9] Pre-treatment-emergent events.

2.4 Modifications made in the Statistical Analysis Plan

The following modifications have been made to the analyses specified in Protocol v4.0 (13 December 2023)

Screened population:

The definition of the Screened population has been added.

PK Analysis set:

Protocol definition:

PK analysis set are defined as all randomized subjects who received at least one dose of the study drug medication and have at least one valid PK measurement.

SAP definition:

PK analysis set are defined as all randomized subjects who received at least one dose of the study drug medication and have sufficient data to estimate at least one PK parameter. Subjects with major protocol deviations that might have impacted the PK data collection will be excluded from the population.

3. ANALYSIS POPULATIONS

The following analysis populations are described in the study protocol.

3.1 Safety Analysis Set

Safety analysis set are defined as all randomized subjects who receive at least one dose of the study medication regardless of if they have or have not completed the study.

Safety analyses will be based on Safety Analysis Set.

3.2 PK Analysis Set

PK analysis set are defined as all randomized subjects who received at least one dose of the study drug medication and have sufficient data to estimate at least one PK parameter. Subjects with major protocol deviations that might have impacted the PK data collection will be excluded from the population.

PK analyses will be based on PK analysis Set.

3.3 Other analysis sets

3.3.1 Screened population

The Screened population is defined as all screened subjects, including screening failures who are in the database.

Those subjects which did not pass the screening visit will not be randomized.

4. STUDY ENDPOINTS AND DERIVED VARIABLES DEFINITION

4.1 General considerations

The following time unit conversion factors will be applied to convert days into weeks, months, or years as needed.

Table 1 Time unit conversion factors

1 week	7 days
1 month	30.4375 days
1 year	365.25 days

Table 2 Units conversion factors

1 pounds	0.453597 kg
1 inch	2.54 cms

Periods:

Study day: The date of first dose of ATL001 administration is considered as Day 1 of the study.

For the events occurring on or after this day, the study day is calculated as the event date minus the date of the first administration of ATL001 plus 1 day.

For the events occurring before the start of treatment, study day is calculated as the event date minus the date of the first administration of ATL001.

The End of the study is defined as the date of the last study visit of a study subject ("last subject last visit"). The study will be completed after the last subject has performed the safety follow-up 30 days after the last IMP administration (FU-V2).

Baseline value: is defined as the last non-missing observation for each subject prior to the first administration of ATL001.

Difference from Baseline:

Change = Post baseline visit value – baseline value

If either the baseline or visit value is missing, the change from baseline is set to "missing".

Placebo:

All placebo receiving subjects will be combined into a single placebo group and presented as "Placebo" group.

4.2 Baseline and Demographics Characteristics

Age: The information of the variable age will be shown and analysed continuously and categorized according to the following distribution.

- < 18
- [18, 40)
- [40,65]
- >65

Weight: will be displayed in kilograms.

Height: will be displayed in centimetres.

Result of Urine drug screen:

- **Positive:** if result of any urine test is positive in any of the following substances, we will consider the test 'Positive'

Amphetamine/Barbiturates/Benzodiazepine/Ethanol/Cocaine/Opiate/Phencyclidine/Cannabinoids/Cotinine

- **Negative:** if the result of all urine tests is negative in the following substances, we will consider the test "Negative"

Amphetamine/Barbiturates/Benzodiazepine/Ethanol/Cocaine/Opiate/Phencyclidine/Cannabinoids/Cotinine

- **Not determined:** if the result is neither positive nor negative based on the above definitions (some or all of the tests have a ND result)

4.3 Prior and Concomitant medications

Prior medications: are defined as medications that started within 8 weeks prior to the Screening Visit (whether continuing or not).

Concomitant medications: are defined as any medications taken after the first administration of ATL001 through last study visit.

4.4 Efficacy Endpoints

This is a Phase I study in healthy subjects. The efficacy of the IMP will not be tested.

4.5 Safety Endpoints

4.5.1 Treatment exposure

Compliance (%): will be calculated as the percentage of actual doses the subject took with respect to the total doses planned.

4.5.2 Adverse events

Adverse events

- **Non-TEAE** is an AE that occurs prior to the initiation for the treatment.
- **TEAE** is an AE that occurs after administration of the first dose of ATL001 and through 30 days after the last dose of ATL001 administration. A TEAE also includes an AE present prior to administration of IMP which worsened after administration of IMP.

Note: *If an AE date of onset is incomplete, an imputation rule will be used to classify the AE as treatment-emergent or not. The algorithm for imputing date of onset is provided in the section Handling of Missing Data.*

Related to ATL-001:

- **Related:** TEAEs with relationship to study treatment (as recorded on the AE eCRF page), Relationship ("Definite" / "Probably" / "Possibly") reported by the investigator and those of missing or unknown relationship.
- **Not Related:** TEAEs with relationship to study treatment (as recorded on the AE eCRF page), Relationship ("Unlikely" / "Unrelated") reported by the investigator.

4.5.3 Laboratory results

4.5.3.1 Urea

Urea values will be calculated based on the standardised BUN values reported in the database.

The conversion factor to be applied for this calculation is:

$$\text{Urea (mmol/L)} = \text{BUN(mmol/L)} * 2.1428$$

4.6 Pharmacokinetic Endpoints

Blood samples are collected at pre-dose 0 hr and post-dose time points at 15 min, 30 min, 1 hr, 1.5 hr, 2 hr, 2.5 hr, 3 hr, 4 hr, 6 hr, 8 hr, 12 hr, and 24 hr on Day 1 and Day 5. To assess the PK profile of ATL001 after administration of multiple doses, the following PK parameters will be calculated using non-compartmental analysis (NCA) method with Phoenix® WinNonlin® software (Version 8.1 or higher, Certara, L.P.).

Parameter	Definition
Day 1 (Initial Dose)	
C_{max}	Maximum observed plasma ATL-001 concentration, determined directly from individual concentration-time data
t_{max}	Time until C_{max} is reached, determined directly from individual concentration-time data
$AUC_{0\text{-last}}$	Area under the plasma ATL-001 concentration-time curve from time 0 to the time of the last detectable concentration after the initial dose, before the next dose administration
$AUC_{0\text{-}12}$	Area under the plasma ATL-001 concentration-time curve from time 0 to 12 hours after the initial dose
$AUC_{0\text{-}24}$	Area under the plasma ATL-001 concentration-time curve from time 0 to 24 hours after the initial dose
Day 5 (Steady State after Multiple Doses)	
$C_{max\text{-}ss}$	C_{max} at steady state
$T_{max\text{-}ss}$	T_{max} at steady state
$C_{min\text{-}ss}$	C_{min} at steady state
AUC_{τ}	Area under the plasma ATL-001 concentration-time curve during one dosing interval at steady state
$AUC_{0\text{-last,ss}}$	Area under the plasma ATL-001 concentration-time curve from time 0 to the time of the last detectable concentration at steady state
$t_{1/2}$	Apparent terminal elimination half-life at steady state
CL_{ss}/F	Apparent systemic clearance at steady state
V_{ss}/F	Apparent volume of distribution at steady state
$R_{ac}(AUC)$	Accumulation ratio based on AUC, defined as $AUC_{\tau}/AUC_{0\text{-}24}$
$R_{ac}(C_{max})$	Accumulation ratio based on C_{max}, defined as $C_{max\text{-}ss}/C_{max}$

4.7 Other Derived Variables

In the case that more derived variables than the ones described in this document are required for the analysis, they will be defined and described in the statistical report.

5. GENERAL STATISTICAL METHODS

5.1 Sample Size

The study will consist of five sequentially recruited cohorts of 8 subjects each, resulting in up to 40 randomized subjects.

No technical sample size calculation was done. This sample size of 8 subjects per cohort with a randomization 3:1 for active/placebo treatment, is regarded sufficient to demonstrate the effects of the study drug under investigation and therefore to be sufficient to fulfil the objectives of the study.

In case of a withdrawn subject from the study during the treatment period and for other reasons than safety, he/she may be replaced with another subject, at the Sponsor's discretion.

5.2 Statistical Hypotheses

No formal statistical hypothesis of the safety or tolerability will be tested for this study.

5.3 Subject´s Disposition

For subject study status, the number and percentage of subjects for each one of the following categories will be presented.

- Screened subjects, including screened failures (number only).
- Screened-failure subjects (number only).
- Subjects enrolled.
- Subjects in the safety population.
- Subjects included in the PK population.
- Subjects completing treatment.
- Subjects who discontinued treatment by primary reason.
- Subjects completing the study per protocol.
- Subjects who discontinued study by primary reason.

For all categories of subjects, percentages will be calculated using the number of safety population subjects.

The subject disposition will be listed by subject (Screening population).

5.3.1 Protocol Deviations

All protocol deviations will be collected separately from the clinical database.

They will be recorded and categorized by the study monitors in Simple Trials. The PDs management will be done according to the Protocol Deviation Plan.

A listing of all patients with protocol deviations will be maintained by regional Clinical PM and periodically reviewed by the Global PM and Sponsor. The Sponsor may upgrade or downgrade the category of the protocol deviation, always based on the approved Protocol Deviation Plan.

The final list of protocol deviations will be reviewed before the database lock and provided to the biostatistician.

All protocol deviations will be listed by category and deviation type.

5.4 Summary of Statistical Methods

All subjects entered in the database will be included in subject data listings.

All relevant subject data will be included in listings.

The listings will be sorted by placebo/ dose-level cohort, subject, visit, and timepoint where applicable, unless specified otherwise.

All applicable data will be summarized overall. In addition, data will be summarized by visit and/or time-point when appropriate. Unscheduled or repeat assessments will not be included in summary tables but will be included in listings, if any.

Continuous variables will be summarized using the number of observations (n), missing, mean, standard deviation (SD), median, Q1, Q3, minimum, and maximum along with the total number of subjects contributing values.

Descriptive statistics for categorical/qualitative data will include frequency counts and percentages. The total number of subjects in the cohort (N) will be used as the denominator for percent calculations unless stated otherwise in the table footnote.

Results of statistical analysis will be interpreted descriptively by placebo/ dose-level cohort.

No adjustment for covariates is planned for this study.

No missing value imputation will be done (except for what is included in the missing data imputation section 5.4.8)

All analyses will be conducted on observed data.

5.4.1 Demographics, Other Characteristics, Prior, and Concomitant Medication

5.4.1.1 Demographic and Other Characteristics

The following variables for demographics characteristics will be summarized by placebo/ dose-level cohort and overall:

- Age (years) / Age range (< 65, [65,85], > 85).
- Gender (Male / Female / Unknown / Undifferentiated-Intersex).
- Race (Black or African American / American Indian or Alaska Native / Asian / Native Hawaiian or Other Pacific Islander / White /Other). If other, specify.
- Height (cm), weight (kg), Body Mass Index (kg / m^2).

This information will be extracted from the Screening visit.

The demographics and other characteristics will be listed by subject (Screening visit).

Summary of the substance use will be summarized by placebo /dose-level cohort, and overall:

- Smoking status (Yes / No)
- If yes, smoking conditions (Current smoker/Quit before 3 months prior to Screening/ Quit after 3 months prior to Screening/ Missing)
- Was the subject screened for urine Drug abuse? (Yes / No)
- If yes, result (Positive/ Negative / Not determine)
- Was the subject screened for alcohol use? (Yes / No)
- If yes, result (Positive / Negative)

The smoking status will be extracted from the Screening visit.

The urine Drug abuse, and alcohol abuse will be extracted from the Baseline visit if available, otherwise will be extracted from the Screening visit.

The substance use will be listed by subject including the information reported in the screening and baseline visit.

5.4.1.2 Medical History

Medical history will be coded to Medical Dictionary for Regulatory Activities (MedDRA) terms. The version used will be specified in the data display footnote.

The medical history will be listed including the MedDRA codes, but not summarized.

5.4.1.3 Prior and Concomitant Medications

Prior and concomitant medications will be listed but not summarized.

5.4.2 Efficacy Analyses

The efficacy analysis will not be performed in this study.

5.4.3 Safety Analyses

The population used for safety analyses will be the Safety Analysis Set. No formal statistical hypothesis of the safety or tolerability are to be tested for this study.

Safety variables include adverse events, clinical laboratory values, 12-lead ECG, vital signs, and Physical Examination.

5.4.3.1 Treatment Exposure

Exposure of the ATL001 will be summarized by placebo / dose-level cohort. The following variables will be summarized:

- Number of doses administered per Subject.
- Subjects with any delayed dose.
- Subjects with any modified dose.
- Subjects with at least one skipped dose.
- Subjects who do not received any full dose administration.
- Compliance.

A listing of ATL001 administration information will be provided.

5.4.3.2 Adverse events

All AEs (TEAEs and non TEAEs) will be coded to System Organ Class (SOC) and Preferred Term (PT) using the MedDRA dictionary v25.1. or later.

The NCI-CTCAE version 5.0 or higher when available will be used to assess the severity grade of TEAEs.

5.4.3.2.1 TEAEs – Overview

An overall summary table will be provided showing the number of AEs/TEAEs (events), the number of subjects, and proportion of subjects who presented:

- Any AE.
- Any TEAE.
- Any TEAE grade 3 or higher.
- Any TEAE grade 3.
- Any TEAE grade 4.
- Any TEAE grade 5.
- Any TEAE related to ATL001.
- Any TEAE related to ATL001 grade 3 or higher.
- Any TEAE related to ATL001 grade 3.
- Any TEAE related to ATL001 grade 4.
- Any TEAE related to ATL001 grade 5.
- Any TEAE leading to temporary interruption of ATL001.
- Any TEAE leading to treatment permanent discontinuation of ATL001.
- Any Serious TEAE.
- Any Serious TEAE related to ATL001.
- Any Serious TEAE with a fatal outcome.
- Any Serious TEAE related to study discontinuation.
- Any Serious TEAE related to ATL001 with fatal outcome.
- Any AESI.
- Any AESI related to ATL001.

This information will be presented by placebo/ dose-level cohort, and overall.

5.4.3.2.2 TEAEs.

All AE summaries will be restricted to TEAEs only.

Treatment Emergent Adverse Events will be summarized:

- By worst grade according to National Cancer Institute Common Terminology Criteria for Adverse Events (NCI-CTAE version 5.0), per subject, using the latest version of MedDRA as event category.

The following tables will be created:

TEAEs: Worst grade by subject (by placebo / dose-level cohort, and overall):

- Incidence of TEAEs will be tabulated by Primary System Organ Class (SOC), Preferred Term (PT) and maximum CTCAE grade, showing the total incidence of SOC and PT within SOC.

TEAEs: More than 5 % of incidence (by placebo / dose-level cohort, and overall):

- Incidence of TEAEs with more than 5% of incidence will be tabulated by Preferred Term (PT).

TEAEs: Worst grade by subject related to (by placebo / dose-level cohort, and overall):

- Incidence of TEAEs related to ATL001 will be tabulated by SOC, PT and maximum CTCAE grade, showing the total incidence of SOC and PT within SOC.

Serious Adverse events: Worst grade by subject (by placebo / dose-level cohort, and overall)

- Incidence of SAEs will be tabulated by SOC, PT and cohort, showing the total incidence of SOC and PT within SOC and maximum CTCAE grade, showing the total incidence of SOC and PT within SOC.

A listing of AEs (Non TEAEs /TEAEs) will be sorted by placebo/ dose-level cohort, subject, Start Date, System organ class, and Preferred term.

5.4.3.3 Deaths

In case of death during the study period, it would be reported in the reasons for end of treatment and/or end of study.

5.4.3.4 Clinical Laboratory Test

To evaluate the safety profile the following laboratory tests will be performed:

- **Hematology**

  Hemoglobin
  Erythrocytes
  Leukocytes
  Neutrophils
  Lymphocytes
  Monocytes
  Eosinophils
  Basophils
  Ferritin
  C reactive protein (CRP)

- **Biochemistry**

  Alkaline phosphatase
  AST / SGOT
  ALT / SGPT
  Gamma-glutamyl transferase
  Creatinine
  Glucose
  Total Bilirubin
  Lactate dehydrogenase
  Triglycerides
  Calcium
  Chloride
  Potassium
  Sodium
  TSH
  BUN

- **Urinalysis**
  - Quantitative
    - pH
    - Specific gravity
    - Microalbumin
    - Protein
  - Qualitative
    - Urine bilirubin (Positive / Negative /Not Done)
    - Ketones (Positive / Negative /Not Done)
    - Nitrite (Positive / Negative /Not Done)
    - Occult blood (Positive / Negative /Not Done)
    - Acid Urate Crystals (Positive / Negative /Not Done)
    - Urine glucose (Positive / Negative /Not Done)
  - Microscopic analysis quantitative (will be analyzed as qualitative)
    - RBC Casts (in range / above the range/ missing)
    - WBC Casts (in range / above the range/ missing)
  - Microscopic analysis qualitative
    - Urine Appearance (Normal / Abnormal /Not Done)
    - Urine Color (Normal / Abnormal /Not Done)

For each parameter, summaries will be presented for the Screening, Baseline, and each scheduled post-baseline visit.

For each quantitative laboratory parameter, the values, and changes from baseline will be summarized by placebo / dose-level cohorts (including number of observations, missing, mean, SD, median, Q1, Q3, minimum, and maximum) will be calculated for each visit.

The decimal precision for each parameter will be based on the maximum number of decimals to which the results were reported on the eCRF.

If a result for a parameter that is normally considered quantitative is reported as a range (i.e., the result for basophils is reported as '<0.01'), this value will be not considered in the summary table. All the values reported will be included in the respective listing.

Lab Original results with ND/ NA/ NE/ UNK/ NQ/ UN etc: will not be evaluated or used for the analysis purpose.

All biochemistry, hematology, and urinalysis data will be listed.

The listings will be sorted by placebo /dose-level cohort, subject, parameter, visit and timepoint (if applicable).

The results and normal ranges will be displayed to the same decimal precision in the listings.

The clinically significant abnormal values will be flagged.

Notes:

- Clinical laboratory data will be analysed using standardized values.
- Only evaluable data will be used, and missing data will not be imputed.
- The Microscopic analysis quantitative parameters will be analysed as categorical variable.
- Biochemistry: FSH and Glomerular Filtration Rate will be shown in a listing.

  Information on the derived variable Urea will be presented. The standardised values and changes from baseline will be summarised by placebo / dose level cohorts (including number of observations, missing, mean, SD, median, Q1, Q3, minimum and maximum) and will be calculated for each visit.

5.4.3.5 Vital Signs

Summary statistics for observed and change from baseline values of each vital sign measurements, including blood pressure (Systolic/Diastolic) (mmHg), Pulse Rate (beats/minute) and temperature (ºC) will be presented by placebo/ dose-level cohort and visit.

Vital signs will be summarized using the number of observations (n), Abnormal values (number of subjects with abnormal values), missing, mean, standard deviation (SD), median, Q1, Q3, minimum, and maximum along with the total number of subjects contributing values.

The decimal precision for each parameter will be based on the maximum number of decimals to which the results were reported on the eCRF.

All vital signs measurements will be listed. The listing will be sorted by placebo / dose-level cohort, subject, vital sign, visit and timepoint (if applicable).

The test status (Normal/Abnormal/ Not done) and the clinically significant abnormal values (Yes / No) will be shown.

5.4.3.6 12-Lead Electrocardiogram (ECG)

Summary statistics for observed and change from baseline values of each ECG parameter will be presented by placebo / dose-level cohort, and visit/timepoint. 12-lead ECG measurements will be conducted locally, including RR duration (sec), PR interval (ms), QRS duration (ms), QT duration (ms), QTcF (ms) and P wave duration (ms).

ECG will be summarized using the number of observations (n), Abnormal values (number of subjects with abnormal values), missing, mean, standard deviation (SD), median, Q1, Q3, minimum, and maximum along with the total number of subjects contributing values.

The decimal precision for each parameter will be based on the maximum number of decimals to which the results were reported on the eCRF.

All ECG measurements and overall interpretation results will be listed. The listing will be sorted by placebo/ dose-level cohort, Subject, parameter, visit and timepoint (if applicable).

5.4.3.7 Physical Examination

All subjects will undergo a standard physical examination. General physical examinations will include the following body systems:

Heent, dermatological, psychiatric, neurological, cardiovascular, respiratory system, renal, abdominal and gastrointestinal, genitourinary, musculoskeletal, endocrine and metabolic, hematological and lymphatic, allergies and drug sensitivities, others.

The examinations will be recorded and assessed as “normal” or “abnormal”. Abnormal findings will be assessed as “clinically significant” or “not clinically significant”.

Any clinically significant abnormalities will be recorded either as medical history or concomitant disease (if detected at screening) or as adverse event.

Data from physical examinations will be listed. The listing will be sorted by placebo /dose-level cohort, subject, parameter, visit, and timepoint (if applicable).

5.4.4 Pharmacokinetic Analyses

Missing pre-dose ATL-001 concentration values will be imputed as 0. Other missing PK concentration values will be treated as missing for summary tables and figures. PK concentration values that are missing or not reportable will be reported in listing as missing or "NR", respectively.

Plasma ATL001 concentrations that are below the lower limit of quantification (LLOQ) will be displayed as "<LLOQ" in data listings, and imputed as LLOQ/2 for summary tables and figures. LLOQ at pre-dose will be imputed as 0.

LLOQ values in the terminal phase and embedded LLOQ values (an LLOQ value between two quantifiable concentrations) will be set to missing for PK parameter calculation.

Plasma concentrations of ATL-001 will be summarized by ATL-001 dose group and nominal (scheduled) time point using descriptive statistics, including n, mean, SD, SE, minimum, maximum, median, coefficient of variation (CV%), geometric mean, and geometric CV%. Mean and individual concentration-time profiles will be presented graphically on linear and semi-log scales.

PK parameters will be summarized by ATL-001 dose group using descriptive statistics, including n, mean, SD, SE, minimum, maximum, median, coefficient of variation (CV%), geometric mean, and geometric CV%. Important PK parameters, e.g., $AUC_{0\text{-}24}$, AUC_{τ}, C_{max} and $C_{max\text{-}ss}$ will be graphically presented using box plot.

5.4.5 Subgroup analysis

No subgroup analysis is planned for this study.

5.4.6 Interim Analyses

A statistical Analysis has been conducted after completing Cohort 4 (2 mg/kg) and before enrolling subjects in Cohort 5 (4 mg/kg).

5.4.7 DSMB Analyses

DSMB will periodically review safety data from study subjects and will make recommendations concerning the continuation, modification, or termination of the trial. Only then the study may proceed with the next cohort for treatment administration after

This activity will be managed and documented by the Sponsor.

5.4.8 Handling of Missing Data

No missing value imputation will be done. All analyses will be conducted on observed data.

Only will be considered the imputation to classify the adverse events into TEAE or non TEAE. For this purpose, is strictly necessary the start date of the event so if it is missing, imputation will be performed following the next rules:

- If only the day is missing, 1st of the registered month will be interpreted.
- If both day and month are missing, it will be placed 1st of January of 2023

Although this option is being contemplated, it is not expected to occur.

Just in case the previous imputation generated an incoherence considering the rest of dates reported for any subject, the day imputed would be the corresponding day according to the rest of dates for the same subject.

Also, if there is any partial date and it has relation with the medication, it will be considered TEAE.

These data imputations are for analysis purpose only and will not be used in listings.

5.4.9 Reporting Conventions

Descriptive statistics will be reported to 2 decimals precision, unless otherwise specified in the sections.

Percentages should be rounded to 2 decimals precision.

Date and time display conventions: the following display conventions will be applied in all outputs where date and /or times are displayed:

Date: DDMMMYYYY
Date and time: DDMMMYYYY/HH:MM (24 hours)

If only partial information is available, unknown components of the date or time will be presented as 'UK' (Unknown), i.e., 'UKMAY2016'.

Listings will be presented and sorted by placebo / dose-level cohort, subject ID, when available, listings will also sort by visit and timepoint (If applicable).

5.4.10 Study Timelines

The anticipated study duration is about 11 months, with up to 66 days duration per subject.

For each cohort, after a 30-day Screening period to establish subjects' eligibility, the subjects will be treated for 5 days (the treatment period) and followed by 30 days of observation and assessment of treatment outcomes (the follow-up period).

5.4.11 Technical Details

The most updated study protocol has been used as a reference for this document.

SAS programs, SAS Logs and SAS outputs generated during the creation of the Statistical Report will be archived in the PIVOTAL's File System.

5.4.12 Software

The statistical analysis will be performed using the scientific software SAS® V9.4 or later releases and SAS® Enterprise Guide V8.3 or later releases.

6. REFERENCES

Study protocol version 4.0 (dated on 13 December 2023) has been used to prepare this document.

Study Case Report Forms version 4.0 (annotated and dated on 30-Jan-2024) has been referred during the development of this document.

7. TABLES, LISTINGS AND FIGURES

This information has been detailed in the following documents:

-ATL001 - Shells TFLs.

Included in this document are the TFLs generated from the data recorded in the eCRF.

-ATL001_PK_Shells TFLs.

Included in this document are the TFLs related to the PK Analysis.