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AD ST-07
Statistical Analysis Plan Approval Form

Sponsor:	Ocuphire Pharma, Inc.
Protocol:	OPI-NYXG-201
Protocol Title:	Randomized, Placebo-Controlled, Double-Masked Study of the Safety and Efficacy of Phentolamine Mesylate Ophthalmic Solution in Subjects with Open Angle Glaucoma or Ocular Hypertension
SAP Version:	Final V1.0
SAP Date:	02OCT2019

The statistical analysis plan has been reviewed and approved.

Sponsor:

[redacted]

Signature

[redacted]

Date

Consultant:

[redacted]

Signature

02 Oct 2019

Date

Author:

[redacted]

Signature

03 OCT 2019

Date

SUMMITANALYTICAL	CONFIDENTIAL

Ocuphire Pharma, Inc.

STATISTICAL ANALYSIS PLAN

Protocol Title: Randomized, Placebo-Controlled, Double-Masked Study of the Safety and Efficacy of Phentolamine Mesylate Ophthalmic Solution in Subjects with Open Angle Glaucoma or Ocular Hypertension

Study Number: OPI-NYXG-201 (ORION-1)

Phase: Phase 2

Sponsor: Ocuphire Pharma, Inc.

[redacted]

Author: [redacted]

SAP Date: 2019-10-02

Status: FINAL V1.0

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2. LIST OF ABBREVIATIONS AND DEFINITION OF TERMS

Only abbreviations and terms relevant to the SAP are repeated herein. The reader is referred to the protocol for the complete and comprehensive list of abbreviations and definitions of terms for the study.

Abbreviation/Term	Definition
ADaM	Analysis Data Model
AE	Adverse Event
AM	Ante Meridian – in the morning
ANCOVA	Analysis of Covariance
AR	All Randomized
ATC	Anatomical Therapeutic Chemical
BCDVA	Best Corrected Distance Visual Acuity
BP	Blood Pressure
CCLRU	Cornea and Contact Lens Research Unit
CDISC	Clinical Data Interchange Standards Consortium
CI	Confidence Interval
CRF	Case Report Form
CSR	Clinical Study Report
DCNVA	Distance Corrected Near Visual Acuity
DD	Drug Dictionary
ETDRS	Early Treatment Diabetic Retinopathy Study
FAS	Full Analysis Set
FDA	Food and Drug Administration
HR	Heart Rate
ICH	International Council for Harmonisation
IOP	Intraocular Pressure
IRB	Institutional Review Board
ITT	Intent-To-Treat
LSM	Least Squares Mean
LOCF	Last Observation Carried Forward
logMAR	logarithm of the Minimum Angle of Resolution
m	meter
MAR	Missing at Random
MCMC	Markov Chain Monte Carlo

Abbreviation/Term	Definition
mmHg	millimeters of mercury
MedDRA	Medical Dictionary for Regulatory Activities
Nyxol	Phentolamine Mesylate Ophthalmic Solution 1 % (Nyxol®)
OAG	Open Angle Glaucoma
OD	Right eye
OHT	Ocular Hypertension
OR	Odds Ratio
OS	Left eye
OU	Both eyes
PM	Post Meridian
PP	Per Protocol
PT	Preferred Term
QD	Once daily
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SDTM	Study Data Tabulation Model
SE	Standard Error
SOC	System Organ Class
SOP	Standard Operating Procedure
SP	Safety Population
TEAE	Treatment Emergent Adverse Event
TFL	Tables, Figures, and Listings
US	United States
VA	Visual Acuity
WHO	World Health Organization

3. INTRODUCTION

3.1. Preface

This document presents a statistical analysis plan (SAP) for Ocuphire Pharma, Inc. Protocol OPI-NYXG-201 (ORION-1) (*Randomized, Placebo-Controlled, Double-Masked Study of the Safety and Efficacy of Phentolamine Mesylate Ophthalmic Solution in Subjects with Open Angle Glaucoma or Ocular Hypertension*).

Reference materials for this statistical plan include the protocol OPI-NYXG-201 Amendment 1 (08APR2019) and Case Report Forms (Final Version 24APR2019).

The SAP described hereafter is an *a priori* plan. The SAP will be finalized and approved prior to unmasking of any study data.

For the reasons stated here the conduct of the study in the field is considered to be independent of any study outcome that might materialize upon enactment of the currently proposed statistical plan.

3.2. Purpose of Analyses

The purposes of the planned analyses described in this SAP are to evaluate the efficacy to lower intraocular pressure (IOP), to improve visual performance, and to evaluate the ocular and systemic safety of Nyxol Eye Drops - 1% Phentolamine Mesylate ophthalmic solution (Nyxol) compared to its vehicle, in Open Angle Glaucoma (OAG) and Ocular Hypertension (OHT). Results from the analyses completed will be included in the final clinical study report for OPI-NYXG-201, and may also be utilized for regulatory submissions, manuscripts, or other clinical development activities.

Post-hoc exploratory analyses not identified in this SAP may be performed to further examine the study data. These analyses will be clearly identified, where appropriate, in the final clinical study report. Additional analyses not prospectively identified in this SAP may also be completed for publications, or regulatory or funding inquiries. These analyses, if performed, may not be reported in the clinical study report, but will be fully documented in the document containing the additional analyses.

3.3. Summary of Statistical Analysis Changes to the Protocol

The analyses described in this analysis plan are consistent with the analyses described in the study protocol.

4. STUDY OBJECTIVES AND ENDPOINTS

Study objectives and endpoints defined in the protocol include safety and efficacy endpoints. Objectives and pre-specified endpoints are as follows:

4.1. Study Objectives

The objectives of this study are as follows:

- To evaluate the efficacy of Nyxol to lower IOP in OAG and OHT.
- To evaluate the ocular and systemic safety of Nyxol compared to its vehicle.
- To evaluate additional efficacy of Nyxol to improve visual performance.

4.2. Study Endpoints

4.2.1. Primary Endpoints

The primary efficacy endpoint from the protocol is the change from Baseline to Day 15 in mean diurnal IOP in the study eye. Mean diurnal IOP is the mean of the IOP measurements at all three timepoints (8AM, 10AM, 4PM).

The primary safety measures are:

- Objective biomicroscopic and ophthalmoscopic examination
- Subjective ocular tolerability
- Adverse events (AEs)

4.2.2. Secondary Endpoints

Secondary endpoints for efficacy and safety assessments include the following:

Efficacy:

Secondary efficacy endpoints will be analyzed by study eye, fellow eye and all eyes (the pooled data from the study eye and fellow eye) unless otherwise indicated, and will include:

- Change from Baseline to Day 15 in mean diurnal IOP in the fellow eye and all eyes.
- Change from Baseline to Day 8 in mean diurnal IOP.
- Mean IOP at each post-treatment timepoint (8AM, 10AM and 4PM; on Day 8 and Day 15).
- Change and percent change from Baseline to Day 8 and Day 15 in IOP at each timepoint (8AM, 10AM, 4PM), and Day 16 at 8AM.

- Percentage of subjects achieving reductions from Baseline to Day 8 and Day 15 in IOP at 8AM of greater than or equal to 10%, 15%, 20%, 25% and 30%.
- Percentage of subjects achieving Day 8, Day 15 and Day 16 IOP levels at 8AM of less than or equal to 16mmHg, 18mmHg, 20mmHg and 22mmHg.
- Change and percent change from Baseline Day 8, Day 15, and Day 16 in IOP at 8AM.
- Change and percent change to Days 8, 15, and 16 in pupil diameter at 8AM.
- Percentage of subjects achieving reductions from Baseline to Day 8, Day 15 and Day 16 in pupil diameter at 8AM, of greater than or equal to 10%, 15%, 20%, 25%, and 30%.
- Change from Baseline to Day 8, Day 15, and Day 16 in best corrected distance visual acuity (BCDVA) (Early Treatment Diabetic Retinopathy Study [ETDRS] high contrast) (photopic and mesopic) at 8AM.
- Change from Baseline to Day 8, Day 15, and Day 16 in distance corrected near visual acuity (DCNVA) (ETDRS high contrast) (photopic and mesopic) at 8AM.
- Percentage of subjects achieving improvements from Baseline to Day 8, Day 15, and Day 16 in BCDVA and DCNVA (photopic and mesopic) of greater than or equal to 1 line, 2 lines and 3 lines. For BCDVA, only the number of letters read is collected; this will be converted to lines as follows: 1 line = 3-7 letters read; 2 lines = 8-12 letters read; 3 lines = 13-17 letters read, etc. For DCNVA, only the logMAR value is collected; this will be converted to lines as follows: 1 line = 1.3 logMAR; 2 lines = 1.2 logMAR; 3 lines = 1.1 logMAR, etc. to 14 lines = 0.0 logMAR, 15 lines = -0.1 logMAR, 16 lines = -0.2 logMAR, and 17 lines = -0.3 logMAR.

All secondary endpoints related to IOP will be analyzed additionally in those subpopulations with Baseline IOP of <25mmHg and ≥25mmHg. Subjects will be categorized as "<25 mmHg" if their study eye Baseline IOP readings at all time points (8AM, 10AM, and 4PM) are <25 mmHg.

Safety and Tolerability:

- Vital signs (heart rate [HR] and blood pressure [BP])
- Urine pregnancy tests for females of childbearing potential

5. STUDY METHODS

5.1. General Study Design and Plan

As background for the statistical methods presented below, this section provides an overview of the study design and plan of study execution. The protocol is the definitive reference for all matters discussed in what follows.

Approximately 40 subjects with either OAG or OHT will be randomized, for a target of 36 completed subjects. Subjects will be randomized in a 1:1 ratio to receive 1% Nyxol or placebo once daily for 14 days beginning at 8PM to 10PM on Day 1 and continuing through Day 14. Treatment-study IOP evaluations will occur on Days 8 ± 1 Day and Day 15 ± 1 Day. There will be Follow-up Visits on Day 16 at 8AM ± 15 minutes and by phone on Day 22 (7 days after the last Treatment-study Visit).

ALL TREATMENTS WILL BE ADMINISTERED TO BOTH EYES AND IOP WILL BE MEASURED IN BOTH EYES.

The eye with the higher IOP at the Qualification/Baseline Visit at 8AM is designated as the study eye. In the case where both eyes have the same IOP, the study eye will be the RIGHT eye. Baseline IOP evaluations will occur before dosing on Day 1.

If the subject meets all of the inclusion criteria and none of the exclusion criteria, including all three timepoint IOP measurements, this qualification visit becomes the Baseline Visit, a subject number is assigned, and he/she is randomized into the study.

Site personnel will demonstrate the proper instillation technique to the subject at the Qualification/Baseline Visit (Day 1) and the subject will self-administer a dose of artificial tears at the study site, instilling 1 drop in each eye from the unit-dose bottle (Note: if a drop is not instilled into the eye, the subject should wait approximately 10-15 seconds and administer a second drop). After successful instillation of the drop in each eye, the subject should carefully empty any remaining contents as directed.

The subject is given their study medication dropper bottles, and with instructions when to administer the eye drop (8PM to 10PM), and when to return to the clinic.

Each subsequent evening of dosing, the subject will administer one drop to each eye from a single new unit-dose bottle, then empty the remaining contents and store the opened bottle in the baggie provided and place it in the medication box for return to the study site at the Day 8 Visit. Each subsequent evening of dosing (approximately 24 hours between doses), the subject will follow the same procedures. At the Day 8 visit the medication box, complete with opened bottles and any unopened study medication, will be returned to the study site where the baggies of opened medication will be removed, and the study medication box will be re-dispensed with the unopened medication. During the second week of treatment, subjects will continue to administer one drop of study medication to each eye every night

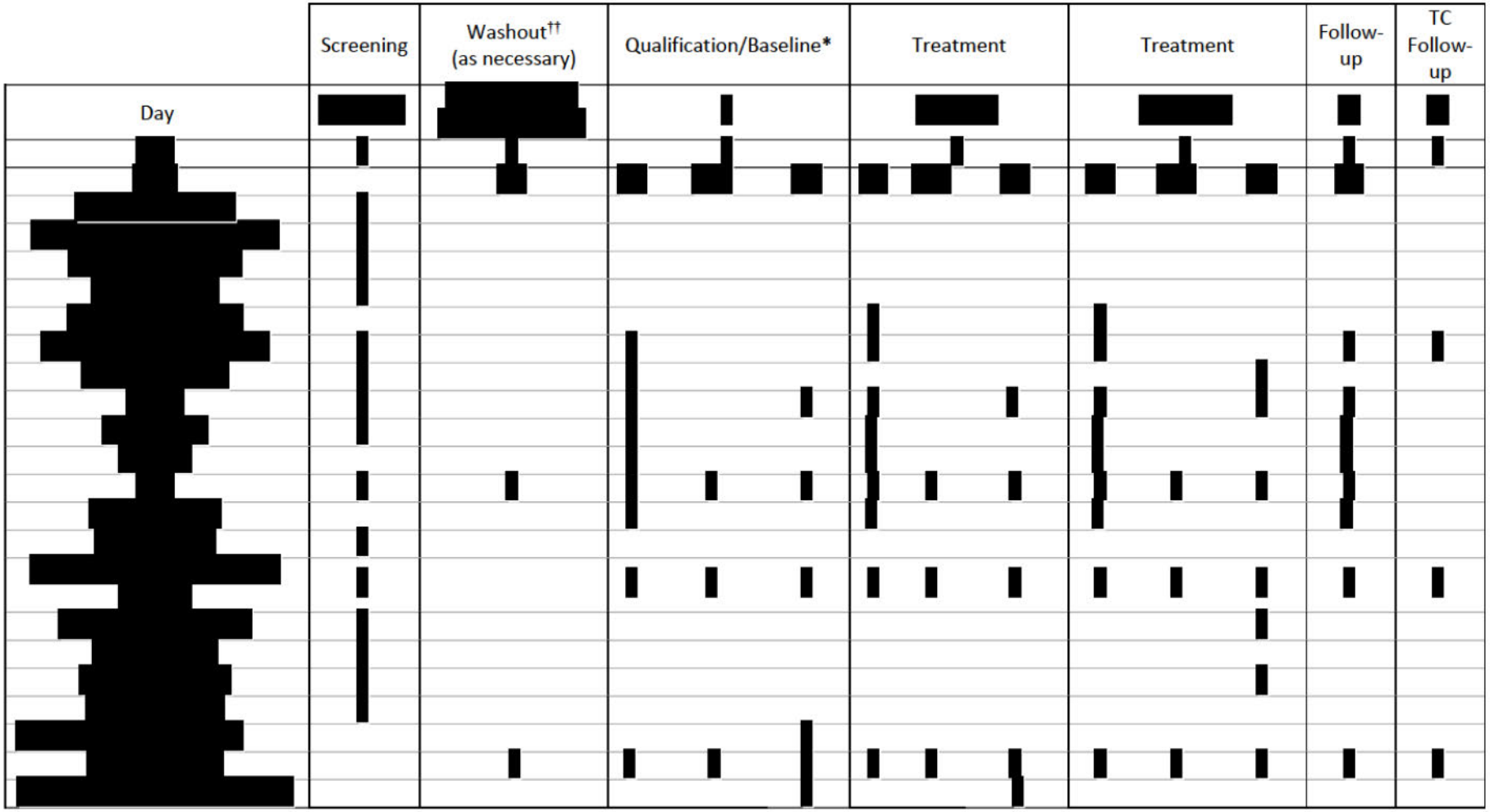
using a new bottle for each dose, then emptying the remaining contents of that bottle and storing the opened bottles in the baggies and placing them back in the box to return to the study site at the Day 15 Visit. The Day 15 visit will be the last day of study treatment; no further study medication will be dispensed at this visit.

IOP measurements at the Baseline Visit and both Treatment-study Visits will be done at 8AM, 10AM and 4PM. All IOP measurements have a window of ± 15 minutes with at least 2 hours between the 8AM and 10AM assessments. IOP will be measured twice in both eyes at all time points. The mean IOP at each timepoint (8AM, 10AM, 4PM) is the average of the 2 IOP measurements at that timepoint. If the difference in the two IOP measurements is > 5 mmHg, a third measurement will be obtained, and the three values averaged.

The lighting conditions will be kept the same from visit to visit. Every effort should be made to have the same clinician perform the IOP measurements at all timepoints and at all visits.

The schedule for assessments and timing of events is presented in Table 1.

Table 1 **Screening, Washout, Qualification, Treatment-study, and Follow-Ups Visits and Procedures**



[redacted]

5.2. Inclusion – Exclusion Criteria and General Study Population

The general study population will include approximately 40 subjects at least 18 years of age and with OAG or OHT. The inclusion and exclusion criteria defined in the protocol apply to all subjects and are not repeated herein the SAP. Reference is made to the final protocol for the specific inclusion and exclusion criteria for study subjects.

5.3. Randomization and Blinding

All eligible and consented subjects will be randomized in a 1:1 ratio to receive 1 % Nyxol or placebo. Once a subject is qualified for the study (Qualification/Baseline Visit) the subject is assigned a randomization number.

Rules for unblinding a subject for safety reasons are fully described in the protocol and not repeated herein this SAP.

5.4. Analysis Variables

Variables to be summarized include demographics and baseline characteristics, medical (non-ocular) and ocular history, concomitant medications, and study drug accountability.

Efficacy variables include:

- IOP
- DCNVA (i.e., Near VA)
- BCDVA (i.e., Distance VA)
- Pupil diameter

Safety variables include:

- Conjunctival hyperemia (eye redness) measured with [redacted]
  - [redacted]
  - [redacted]
  - [redacted]
  - [redacted]
- Biomicroscopy of anterior segment including evaluation of cornea, conjunctiva (other than redness) and anterior chamber. Fluorescein staining to be used.

- Ophthalmoscopy. Dilated fundus exam including optic nerve, macula, vessels and periphery.
- AEs
- Vital signs (HR and BP)
- Urine pregnancy tests for females of childbearing potential

6. SAMPLE SIZE

A sample size of 36 total completed subjects is needed for the study. Thirty-six completed subjects, who were originally randomized in a 1:1 ratio to the Nyxol 1% and the placebo groups, [redacted] dditionally, it is assumed that there will be approximately 10% drop-out between Baseline/Day 1 and Day 15. To account for this drop-out, a total of 40 subjects will be randomized into the study.

7. GENERAL CONSIDERATIONS

7.1. Analysis Populations

The following analysis populations will be defined for this study.

7.1.1. Full Analysis Set Population (FAS)

The FAS will include all randomized subjects who have received at least 11 doses of study medication without missing 2 consecutive doses and have both a Baseline and a Day 15 IOP measurement. The FAS will be used to analyze efficacy endpoints. Subjects included in the FAS will be analyzed as randomized.

7.1.2. Per Protocol Population (PP)

The PP population will include all subjects in the FAS who had no major protocol deviations. The PP population will be used in confirmatory efficacy analyses. Subjects included in the PP population will be analyzed as randomized.

7.1.3. All Randomized Population (AR)

The AR population will include all randomized subjects. This population is also known as the intent-to-treat (ITT) population. The AR population will be used in confirmatory efficacy analyses. Subjects included in the AR will be analyzed as randomized.

7.1.4. Safety Population (SP)

The Safety Population will include all randomized subjects who have received at least one dose of study medication. The Safety Population will be used to summarize safety variables. Subjects who are members of the Safety Population will be analyzed as treated.

7.2. Covariates and Subgroups

7.2.1. Planned Covariates

Planned covariates include baseline values for the given assessment.

7.2.2. Planned Subgroups

Subgroup analyses will be completed for all secondary efficacy endpoints related to IOP in those subpopulations with Baseline IOP in the study eye of <25mmHg and ≥25mmHg. Subjects will be categorized as "<25 mmHg" if their study eye Baseline IOP readings at all time points (8AM, 10AM, and 4PM) are <25 mmHg.

7.3. Management of Analysis Data

7.3.1. Data Handling

Data from unscheduled visits will not be included in the analysis of efficacy or safety, but will be listed.

7.3.2. Missing Data

The primary efficacy endpoint is the change from Baseline to Day 15 in mean diurnal IOP. For the analysis of the primary efficacy endpoint, observed case data will be used (no imputation will be performed for missing efficacy data) for the analysis using the FAS. However, confirmatory analyses will be performed using the AR population, with imputation performed for missing data as specified in Section 7.3.2.3.

Otherwise there will be no substitutions made to accommodate missing data points for efficacy data. All data recorded on the case report form will be included in data listings that will accompany the clinical study report.

Safety data will be imputed in limited situations. If the severity of an AE is missing, then the severity will remain missing. If relationship of the AE to study drug is missing, the relationship will remain missing. Missing or partial dates for AEs or concomitant medications will be imputed as described in Section 7.3.2.1. Otherwise, all summaries of safety endpoints will be completed using observed cases in the Safety population; no imputation will be completed.

7.3.2.1. Handling of Missing Date Values

Partial or Missing Dates

The following conventions will be used to impute missing portions of dates for adverse events and concomitant medications, if warranted. Note that the imputed values outlined here may not always provide the most conservative date. In those circumstances, the imputed value may be replaced by a date that will lead to a more conservative analysis.

A. Start Dates
   1) If the year is unknown, then the date will not be imputed and will be assigned a missing value.
   2) If the month is unknown, then:
      i) If the year matches the first dose date year, then impute the month and day of the first dose date.
      ii) Otherwise, assign 'January.'
   3) If the day is unknown, then:
      i) If the month and year match the first dose date month and year, then impute the day of the first dose date.
      ii) Otherwise, assign the first day of the month.

B. Stop Dates
   1) If the year is unknown, then the date will not be imputed and will be assigned a missing value.
   2) If the month is unknown, then assign 'December.'
   3) If the day is unknown, then assign the last day of the month.

7.3.2.2. Missing Baseline Data

Every effort will be made to ensure that accurate baseline information on the subjects is collected. In the event that a subject is missing baseline information the subject will be included in the safety population for assessment of safety, and excluded from the primary analyses. Each case of missing baseline data will be evaluated for potential inclusion in the exploratory endpoints. All baseline data will be observed cases, without imputation.

7.3.2.3. Imputation Methods

Imputation for efficacy data will only be performed for the confirmatory analysis of the primary efficacy endpoint, using the AR population. If 5% or fewer data are missing in all treatment groups, an analysis with last observation carried forward (LOCF) for missing data will be applied. If more than 5% of data in any treatment group are missing, multiple imputation will be employed to analyze incomplete data sets under the assumption that the mechanism responsible for the missing data is at worst characterized as missing at random (MAR).

Multiple imputation is a simulation-based approach where missing values are replaced by multiple Bayesian draws from the conditional distribution of missing data given the observed data and covariates, creating multiple completed data sets. These completed datasets can then be analyzed using standard analysis methods. Rubin (1987) presented rules for how to combine the multiple sets of estimates to produce overall estimates, confidence intervals, and tests that adequately incorporate missing data uncertainty.

Missing values for IOP will be imputed simultaneously based on an underlying joint normal distribution using a Markov Chain Monte Carlo (MCMC) method.

The imputations will be done separately for each treatment group and will include the following variables in the imputation model: IOP at Day 8 (8AM, 10AM, and 4PM) and Day 15 (8AM, 10AM, and 4PM). No imputation will be applied to the Baseline/Day 1 time points because those must be completed for the subject to meet the inclusion/exclusion criteria for study enrollment.

Example SAS code is provided below:

[redacted]

7.3.3. Handling of Early Termination Visit Information

In the event that a subject is terminated early from this study, the early termination visit data for safety variables will be assigned to the closest scheduled visit. If the closest visit has valid data, the early termination data will be assigned to the next available visit.

7.3.4. Pooling of Investigational Sites

The data from all study centers will be pooled together for all planned analyses.

7.3.5. Coding Conventions for Events and Medications

All adverse events, and medical history will be mapped to the Medical Dictionary for Regulatory Activities (MedDRA Version 22.0) system for reporting (preferred term and body system).

Prior and Concomitant medications will be coded using WHO-DD (Drug Dictionary) (Version January 2019).

7.3.6. Analysis Software

Data manipulation, tabulation of descriptive statistics, calculation of inferential statistics, and graphical representations will be performed primarily using SAS (release 9.4 or higher) for Windows. If the use of other software is warranted, the final clinical study report will detail what software was used and for what purposes.

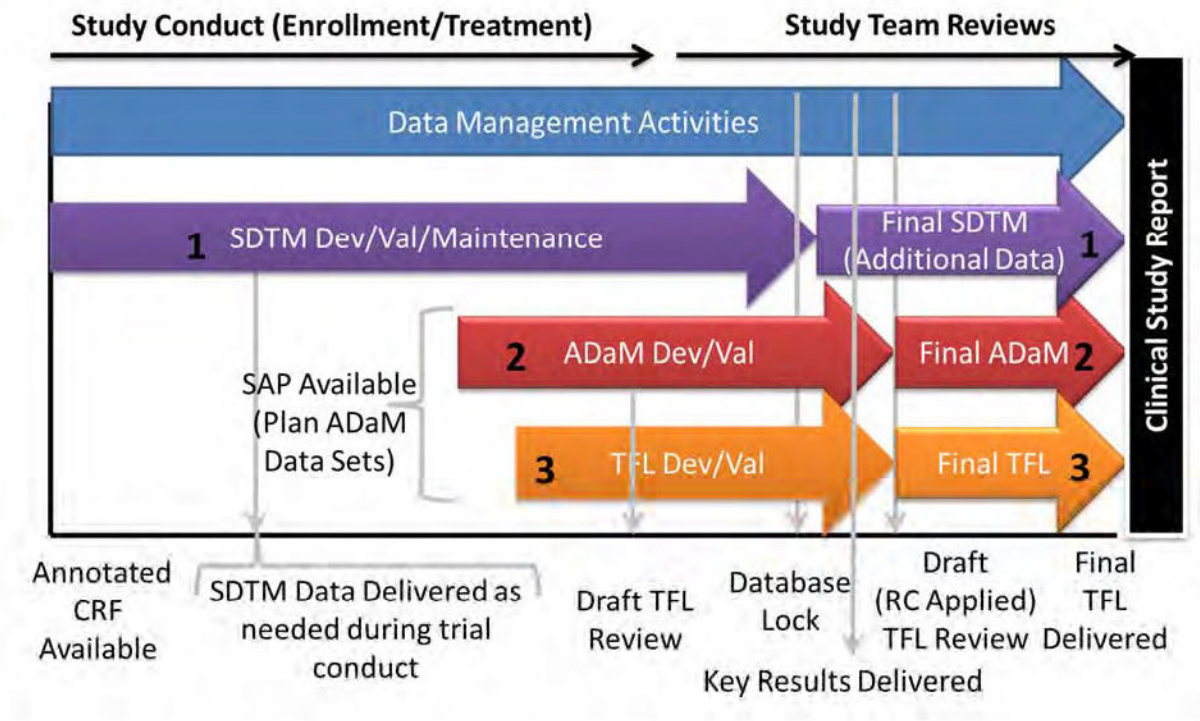
7.3.7. Study Data

Study data identified in the schedule for time and events (Table 1) are collected, and source verified, on paper CRFs; there is no electronic data capture tool.

All study data will be formulated into regulatory compliant data sets to provide transparency, traceability, and integrity of trial analysis results from the collection source. Observed study data will be mapped to the Clinical Data Interchange Standards Consortium (CDISC) Study Data Tabulation Model (SDTM) and serve as the source data from the trial. All study analyses will be completed using analysis data sets that are derived from the SDTM and follow the CDISC Analysis Data Model (ADaM) architecture.

The methods for programming the CDISC SDTM and ADaM data sets are described in Figure 1.

Figure 1 SDTM, ADaM, and TFL Development and Validation



Where:

1. Development, Validation, and Maintenance of SDTM Domains
2. Development and Validation of ADaM Data Sets, with input source the appropriate SDTM domains.
3. Development and Validation of Tables, Figures, and Listings (TFL), with input data source the SDTM domains and analysis specific ADaM data sets.

7.4. Planned Study Analyses

7.4.1. Statistical Summaries: Descriptive and Inferential

Categories for data presentation and analysis will consist of each treatment group separately: Nyxol or placebo.

All statistical tests will be two-sided and a difference resulting in a p-value of less than or equal to 0.05 will be considered statistically significant. All p-values will be rounded to and displayed in four decimals. If a p-value less than 0.0001 occurs it will be shown in tables as <0.0001.

Descriptive summaries of variables will be provided where appropriate. For continuous variables, the number of non-missing values (n), mean, standard deviation, median, minimum, and maximum will be tabulated by treatment. For categorical variables, the counts and proportions of each value will be tabulated by treatment. Expansion of

descriptive table categories within each treatment may occur if such elaborations are thought to be useful.

All study related data collected will be presented in listings. Study related data not subject to analysis according to this plan will not appear in any tables or graphs but will be included in the data listings.

7.4.2. Interim Analyses and Data Monitoring

No formal interim analysis or safety monitoring committee is planned for this study.

7.4.3. Final Analysis and Publication of Study Results

The final analysis will be completed after all subjects have completed the study.

7.5. Multiple Testing Procedures

There will be no adjustments for multiplicity and no formal multiple testing procedures are to be implemented with this analysis plan.

7.6. Baseline Values

Baseline values are the values obtained prior to the first study drug administration on Day 1 (scheduled between 8PM and 10PM in the evening). If the Day 1 value is missing, the value at Screening will be treated as the baseline.

8. SUMMARY OF STUDY DATA

8.1. Subject Disposition

A summary of the analysis sets includes the number and percentage of subjects by treatment group and overall for the following categories: subjects in the AR Population, subjects in the Safety Population, and subjects in the FAS Population. All percentages will be based on the number of subjects randomized.

End of trial information will also be summarized in this table, including the number of subjects completing the study, the number of subjects who prematurely discontinued the study with reasons for withdrawal, the number of subjects completing the study medication dosing, and the number of subjects who prematurely discontinued the study medication with reasons for study medication discontinuation.

A by-subject data listing of study completion information including the reason for premature study withdrawal, if applicable, will be presented.

8.2. Protocol Deviations

Major protocol deviations, as determined by a Sponsor blinded review of the data prior to database lock and unblinding of the study, may result in the removal of a subject's data from the PP Population. The Sponsor or designee will be responsible for producing the final deviation file; this file will include a description of the protocol deviation and clearly identify whether this violation warrants exclusion from the PP Population. This file will be finalized prior to database lock.

All protocol deviations will be presented in a by-subject data listing, with a flag to indicate if a deviation was considered major..

8.3. Demographics and Baseline Characteristics

Subject demographic data and baseline characteristics will be tabulated and summarized descriptively by treatment group and overall. The demographic data and baseline characteristics will be summarized for the Safety and FAS Populations.

The demographics and baseline characteristics consist of age (year), sex, race, ethnicity, and study eye (OD [right eye] or OS [left eye]), and baseline IOP category (<25 mmHg or ≥25 mmHg). A subject's age in years is calculated using the date of the informed consent and date of birth. Age will be summarized using descriptive statistics. The number and percentage of subjects by gender, race, ethnicity, study eye, and baseline IOP category will also be reported. Percentages will be based on the total number of subjects in the study population presentation.

The following baseline characteristics will be summarized for OD and OS, and for study eye, fellow eye, and all eyes, using descriptive statistics:

- Pupil diameter, photopic and mesopic
- BCDVA, photopic and mesopic, monocular only
- DCNVA, photopic and mesopic, monocular only
- IOP at 8am, 10am, 4pm, and mean diurnal

All demographic and baseline information will be listed by subject.

8.4. Medical History

The number and percent of subjects with individual medical histories will be summarized for all subjects by treatment group and overall. Non-ocular and ocular medical history will be summarized separately.

Medical history will be coded using the MedDRA Version 22.0. The number and percentage of subjects with any medical history will be summarized overall and for each system organ class (SOC) and preferred term (PT). Percentages will be calculated based on number of subjects in the Safety Population.

Subject medical history data including specific details will be presented in by-subject listings.

8.5. Prior and Concurrent Medications

The number and percentages of all concomitant medications will be summarized by treatment group, Anatomical Therapeutic Chemical (ATC) level 4, and PT. The total number of concomitant medications and the number and percentages of subjects with at least 1 concomitant medication will be summarized by treatment group. All summaries will be performed using the Safety Population.

A concomitant medication is defined as any medication taken on or after the day of first exposure to study drug.

Prior medications are defined as any medication that has a start and stop date prior to the day of first exposure to any study drug, collected from up to 30 days prior to Screening.

8.6. Treatment Compliance and Exposure

Continuous descriptive summaries for total exposure to study drug (in days) and percentage compliance will be presented for the Safety Population by treatment group. Exposure to study drug will be determined by counting the total number of used bottles returned.

Compliance will be determined by calculating the percent of used bottles returned to the total that should have been used during the treatment period.

9. EFFICACY ANALYSES

Unless otherwise noted, all efficacy analyses will be completed using the FAS, AR, and PP Populations. The FAS Population will be the primary analysis population for efficacy. All efficacy analyses will be completed using the planned dose that the subject received.

9.1. Clinical Efficacy

The evaluations of clinical efficacy will be performed using the FAS, AR, and PP Population, as specified below. Baseline values are generally taken at the Day 1 (Qualification/Baseline) Visit.

IOP, near and distance VA, and pupil diameter will be measured at the Screening Visit, the Qualification/Baseline Visit, the Treatment-study visits, and Follow-up Visit.

Table 2 Clinical Efficacy Assessments

Efficacy Assessment	Equipment Name	Measurement (Unit)	Procedure
IOP	[redacted]	mmHg	[redacted]
Distance VA	[redacted]	Letters	[redacted]
Near VA	[redacted]	Jaeger	[redacted]

	[redacted]		[redacted]
Pupil diameter	[redacted]	mm	[redacted]

*For this study photopic and mesopic light conditions are considered to be "with the lights on or with the lights off".

IOP measurements at the Baseline Visit and both Treatment-study Visits will be performed at 8AM, 10AM and 4PM. All IOP measurements have a window of ± 15 minutes with at least 2 hours between the 8AM and 10AM assessments. IOP will be measured twice in both eyes at each timepoint. The mean value at each timepoint for the study eye will be used in efficacy assessments. If the difference in the two IOP measurements is > 5mmHg, a third measurement will be obtained, and the three values averaged. Mean diurnal IOP is the mean of the IOP measurements at all three timepoints (8AM, 10AM, 4PM).

Near and distance VA and pupil diameter will be performed in photopic and mesopic conditions. Furthermore, near and distance VA will each be assessed using monocular (OD and OS separately) and binocular (OU [both eyes]) measurements.

Primary Efficacy Endpoint:

The primary efficacy endpoint will be analyzed using ANCOVA with change from Baseline to Day 15 in mean diurnal IOP in the study eye as the dependent variable; treatment as a factor; and Baseline mean diurnal IOP as the covariate. The ANCOVA will be performed using the FAS, with subjects included in their randomized treatment group regardless of the treatment they actually received. The least-squares mean (LSM) and standard error (SE) will be provided for both treatment groups, along with the placebo-corrected LSM (Nyxol – placebo), its 95% confidence interval (CI) and associated p-value.

A confirmatory analysis of the primary efficacy endpoint will be performed, using the AR population with missing Day 15 values imputed. See Section 7.3.2 for imputation methods.

Another confirmatory analysis of the primary efficacy endpoint will be performed using the PP population. No imputation of missing Day 15 values will be applied.

Secondary Efficacy Endpoints:

Secondary efficacy endpoints (for the study eye unless otherwise noted) are indicated in Section 4.2.2. Secondary efficacy endpoints will be analyzed by study eye, fellow eye and all eyes (unless otherwise indicated). "All eyes" refers to the pooled data from the study eye and fellow eye, so there will be 2 results per subject for any summary of all eyes if the endpoint was assessed for both the study eye and fellow eye. Secondary efficacy assessments not related to IOP will be analyzed only at 8AM. Binocular near and distance VA assessments will not be analyzed. All secondary efficacy assessment data, regardless of whether they are included in the analysis, will be presented in by-subject listings.

For each of the continuous secondary efficacy endpoints, the same ANCOVA for the primary efficacy endpoint will be used, with the respective Baseline included as the covariate. Each ANCOVA will be performed using the FAS with subjects included in their randomized treatment group regardless of the treatment they actually received. The output from each ANCOVA will include the LSM and SE for both treatment groups, along with the placebo-corrected LSM (Nyxol – placebo), its 95% CI and associated p-value.

For each of the secondary efficacy endpoints related to percentage of subjects (or percentage of eyes) meeting certain criteria, the analysis will be performed for the FAS using a logistic regression with treatment and Baseline included as independent factors. For each analysis, the percentage of subjects (or eyes) in each treatment group meeting the criteria, the odds ratio (OR) with 95% CI and p-value will be provided.

For the analysis of the secondary efficacy endpoints, only observed case data will be used.

10. SAFETY ANALYSES

All Safety analyses will be conducted using the Safety population. All safety analyses will be completed using the actual treatment a subject received. Observed case data will be used; no imputation will be performed for missing safety data except for the limited situations described in Section 7.3.2.

10.1. Adverse Events

Adverse events will be coded using MedDRA, Version 22.0.

Treatment-emergent adverse events (TEAEs) are defined as any adverse event that begins or worsens after initiation of the investigational product and through the subject's last study visit (study completion or early termination) or follow-up telephone call. Serious adverse events will be recorded from the date of informed consent, throughout the clinical trial.

If the onset of an AE is on or after the date of first dose of study medication, or increasing in severity after first dose of study medication, then the AE will be considered treatment-emergent.

Only TEAEs will be summarized; all AEs (TEAE, non-TEAE) will be included in a by-subject listing.

The number and percent of subjects with any TEAEs will be summarized by system organ class and preferred term by treatment group and overall. At each level of tabulation (ex. at the preferred term level) subjects will be counted only once if they had more than one such event reported during the AE collection period.

Note that in MedDRA, ocular events are coded to the SOC of "special senses". Thus, using SOC in the summaries will provide a separation of ocular and non-ocular adverse events.

The following summary tables will be presented for TEAE data:

- Overall summary of TEAEs
- Summary table of TEAEs by SOC and PT
- Summary table of TEAEs by highest relationship level to study drug by SOC and PT (not related, possibly related, related)
- Summary table of TEAEs by maximum severity by SOC and PT (mild, moderate, severe)
- Summary table of serious TEAEs by SOC and PT
- Summary table of TEAEs leading to withdrawal from the study by SOC and PT
- Summary table of TEAEs leading to study medication discontinuation by SOC and PT

10.2. Deaths, Serious Adverse Events and Other Significant Adverse Events

10.2.1. Deaths

The AE listing will include all AEs, including deaths, regardless of causality; one of the columns in the listing will specify whether the AE was fatal.

Clinical narratives for each death will be written to include important data and safety findings related to the individual death and included in the final clinical study report.

10.2.2. Serious Adverse Events

The AE listing will include all AEs, including serious adverse events (SAEs); one of the columns in the listing will specify whether the AE was an SAE.

Clinical narratives for each SAE observed will be written to include important data and safety findings related to the individual SAE and included in the final clinical study report.

10.2.3. Adverse Events Leading to Withdrawal from the Study

The AE listing will include all AEs, including AEs leading to withdrawal from the study; one of the columns in the listing will specify whether the AE led to withdrawal from the study.

10.2.4. Adverse Events Leading to Discontinuation of Study Medication

The AE listing will include all AEs, including AEs leading to discontinuation of study medication; one of the columns in the listing will specify whether the AE led to discontinuation of study medication.

10.3. Biomicroscopic and Ophthalmoscopic Examinations

Results from biomicroscopic examinations, including conjunctival redness, cornea, conjunctiva (other than redness), anterior chamber, and corneal fluorescein staining will be summarized descriptively for each treatment group and overall by time point for the observed value. Results from ophthalmoscopic examinations, including optic nerve, macula, vessels and periphery will be summarized similarly. Separate summaries will be created for the study eye and the fellow eye.

For conjunctival redness, a descriptive summary of the numeric values from the CCLRU card 4-point scale will also be summarized by treatment group and overall by time point.

All examination data will be presented in by-subject listings. Unscheduled examinations will not be summarized but will be included in the listings.

10.4. Vital Signs

Descriptive statistics of observed values will be presented for vital sign data by time point, including systolic BP (mmHg), diastolic BP (mmHg), and HR (bpm) by treatment group. Changes from baseline to each scheduled post-baseline visit and timepoint will be presented.

All vital sign data will be presented in a by-subject listing. Unscheduled visit or repeated results will not be summarized but will be included in the listings.

10.5. Other Safety Measures

Urine pregnancy tests for females of childbearing potential but will be presented in by-subject listings.

11. REFERENCES

[1] ICH E9 Expert Working Group. Statistical Principles for Clinical Trials: ICH Harmonized Tripartite Guideline, September 1998

12. APPENDICES

12.1. List of Planned Tables

The list of planned tables includes all of the *main* tables to be presented for the study.

Table	Description of Table	AR	Safety	FAS	PP
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]

Table	Description of Table	AR	Safety	FAS	PP
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]

12.2. List of Planned Listings

Listing	Description of Listing
[redacted]	[redacted]