SUMMITANALYTICAL	**CONFIDENTIAL**

Ocuphire Pharma, Inc.

STATISTICAL ANALYSIS PLAN

Protocol Title:	Randomized, Placebo-Controlled, Double-Masked Study of the Safety and Efficacy of Nyxol (0.75% Phentolamine Ophthalmic Solution) in Subjects with Dim Light Vision Disturbances
Study Number:	OPI-NYXDLD-301 (LYNX-1)
Phase:	Phase 3
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SAP Date:	2022-03-28
Status:	FINAL V1.0

SUMMIT ANALYTICAL	**AD ST-07** **Statistical Analysis Plan Approval Form**

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SAP Version: Final V1.0
SAP Date: 28MAR2022

The statistical analysis plan has been reviewed and approved.

Sponsor: *Ocuphire Pharma, Inc.*

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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
ANCOVA	analysis of covariance
ARP	All Randomized Population
ATC	Anatomical Therapeutic Chemical
BAT	Brightness Acuity Tester
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
DLD	dim light vision disturbances
EDC	electronic data capture
ETDRS	Early Treatment Diabetic Retinopathy Study
HR	heart rate
IOP	Intraocular pressure
ITT	Intention-To-Treat
LOCF	last observation carried forward
logMAR	logarithm of the minimum angle of resolution
LSM	least squares mean
MAR	missing at random
MCMC	Markov Chain Monte Carlo
MedDRA	Medical Dictionary for Regulatory Activities
mHCVA	mesopic high-contrast visual acuity
mITT	Modified Intention-To-Treat
Nyxol	Phentolamine Mesylate Ophthalmic Solution 1% (Nyxol®)

Abbreviation/Term	Definition
OD	right eye
OPD	optical path difference
OR	odds ratio
OS	left eye
OU	both eyes
PD	pupil diameter
pLCVA	photopic low-contrast visual acuity
PP	Per Protocol
PT	preferred term
QD	once daily
RMS	root-mean square
SAE	serious adverse event
SAP	Statistical Analysis Plan
SDTM	Study Data Tabulation Model
SE	standard error
SOC	system organ class
SP	Safety Population
TEAE	treatment-emergent adverse event
TFL	tables, figures, and listings
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-NYXDLD-301 (LYNX-1) (*Randomized, Placebo-Controlled, Double-Masked Study of the Safety and Efficacy of Nyxol (0.75% Phentolamine Ophthalmic Solution) in Subjects with Dim Light Vision Disturbances*).

Reference materials for this statistical plan include the protocol OPI-NYXDLD-301 amendment 3 (29JUN2021) and Case Report Forms (CRFs) Version 3.0 (02AUG2021).

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 LYNX-1 study is a randomized, parallel arm, double-masked, placebo-controlled study of the safety and efficacy of Nyxol (0.75% Phentolamine Ophthalmic Solution) to evaluate the efficacy of Nyxol to improve mesopic low-contrast visual acuity (mLCVA) in subjects with dim light vision disturbances (DLD).

The Sponsor intends to use this first Phase 3 registration study to evaluate Nyxol for the [redacted]

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 (CSR). 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 CSR but will be fully detailed in the document containing the additional analyses.

3.3. Summary of Statistical Analysis Changes to the Protocol

A hierarchical analysis was added to formally test a family of endpoints beyond the primary efficacy endpoint. Otherwise, the study protocol includes the subject questionnaire as both an efficacy and safety endpoint; in this analysis plan, all subject questionnaire endpoints are treated as efficacy variables. Also, two secondary efficacy endpoints were added: [redacted] Otherwise, the analyses described in this analysis plan are consistent with the analyses described in the 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 improve mLCVA in subjects with DLD
- To evaluate the efficacy of Nyxol to improve visual performance
- To evaluate the safety of Nyxol
- To evaluate any additional benefits of Nyxol in treating DLD subjects

4.2. Study Endpoints

4.2.1. Primary Endpoints

The primary efficacy endpoint is the percent of subjects with ≥ 15 Early Treatment Diabetic Retinopathy Study (ETDRS) letters (≥ 3 lines) of improvement in the study eye compared to baseline in monocular mLCVA at Day 8.

4.2.2. Secondary and Exploratory Endpoints

Secondary endpoints for efficacy and safety assessments include the following:

Efficacy:

Secondary efficacy endpoints (for the study eye, the non-study eye, the best of either eye, and binocular) include:

- [redacted]

- [redacted]

Exploratory efficacy endpoints (for the study eye, the non-study eye, and the best of either eye) are collected and analyzed for the sites that have an optical path difference (OPD) Scanner and include but are not limited to:

- [redacted]

In addition, [redacted]

Efficacy Measurements:

- PD is measured with a [redacted]
- mLCVA is measured under [redacted] (letters recorded, converted to logMAR).
- pLCVA is measured under [redacted] (letters recorded, converted to logMAR).
- DCNVA is measured under mesopic conditions by a high-contrast Near Visual [redacted]
- Best-corrected distance visual acuity (BCDVA) is measured under mesopic conditions by a [redacted] at [redacted] (letters recorded, converted to logMAR).

- [redacted]
- Subject questionnaire is a brief symptom survey.

Safety and Tolerability:

- Conjunctival hyperemia
- Adverse events (AEs)
- Vital signs (heart rate [HR] and blood pressure [BP])
- Intraocular pressure (IOP)
- Biomicroscopy
- Ophthalmoscopy
- Dry eye measurements
- Urine pregnancy tests for females of childbearing potential

Safety Measurements:

- Conjunctival hyperemia is assessed visually with a grading scale (0-3) using images from the Cornea and Contact Lens Research Unit (CCLRU).
- IOP is measured with the Tono-Pen.
- BP and HR is measured manually or with available digital devices.

5. STUDY METHODS

5.1. General Study Design and Plan

A sample size of approximately 160 DLD subjects ≥ 18 years of age were randomized in a 1:1 ratio to Nyxol or placebo, with the expectation that approximately 136 subjects were evaluable for efficacy, where evaluable means receiving at least 1 dose of study medication and having a Baseline and a Day 8 mLCVA measurement. [redacted]. The subjects were recruited from approximately 15 investigational sites.

Following the successful completion of screening, [redacted] The Screening Visit should occur the same day as the Baseline Visit.

[redacted] The study eye and non-study eye, as well as binocular, was evaluated at all appropriate assessments.

The subject instilled one drop once daily (QD) at or near bedtime (8PM to 10PM) in both eyes (OU) for 14 days (+ 1 day). Each subject received sufficient study medication for the duration of the trial. A study medication box was dispensed at Visit 1 (Screening/ Baseline; Day 1). The first dose of study medication was taken on the evening of the Baseline Visit (Day 1).

All opened bottles as well as any unopened study medication were returned at Visit 2 (Treatment; Day 8) and Visit 3 (Treatment; Day 15). At Visit 2, the box was re-dispensed to the subject following study drug accountability and removal of opened study medication materials. Study medication accountability was conducted at Visit 3, but the box was not re-dispensed.

Up to 3 days after Visit 3, a follow-up phone call (Visit 4) was completed. Subjects were asked if they had any problems with their eyes from the last visit, including whether their eyes are red, and if there have been any changes in their medical condition or concomitant medications since their last visit. There was no treatment administered at Visit 4.

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

Table 1 Screening, Baseline, Treatment, and Follow-Up Visits and Procedures Schedule

Day	[redacted]
Visit	
Informed consent	
Subject ID number assigned	
Medical/ophthalmic history [b]	
Demographics	
Drug accountability and review of subject diary	
Concomitant medications [c]	
Urine pregnancy test [d]	
Heart rate/Blood pressure [e]	
[redacted]	
[redacted]	
[redacted]	
[redacted]	
[redacted]	
[redacted]	
[redacted]	
[redacted]	
[redacted]	
[redacted]	
[redacted]	
[redacted]	
[redacted]	
Adverse events	
Randomization number assigned	
Medications dispensed/Re-dispensed	
Diary dispensed/Re-dispensed	

Abbreviations: BAT, Brightness Acuity Tester; BCDVA, best-corrected distance visual acuity; CCLRU, Cornea and Contact Lens Research Unit; DCNVA, distance-corrected near visual acuity; mLCVA, mesopic low-contrast visual acuity

[redacted]

5.2. Inclusion – Exclusion Criteria and General Study Population

The study population will be approximately 160 DLD subjects ≥18 years of age, with approximately 136 evaluable subjects. Written informed consent will be obtained from each subject.

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

A randomization code for allocating subjects to treatment will be prepared by a masked biostatistician not connected with the study. At the initiation of study-related procedures, every subject who is screened is assigned a subject identification number in numerical order within site. Once a subject is qualified for the study, the subject is assigned a randomization number in the order provided by the biostatistician, and randomized into the study.

Treatment randomization will be 1:1, Nyxol or Placebo (vehicle), [redacted] It is assumed that there will be approximately 15% drop-out between Day 1 and Day 15.

The study medication will be masked to investigators, study staff, study subjects, the clinical research organization, and Ocuphire. Only in case of medical emergency or occurrence of serious adverse events (SAEs) will the randomization code be unmasked by the study pharmacist and made available to the Investigator, Ocuphire, and/or other personnel involved in the monitoring or conduct of this study. Rules for unmasking 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:

- Mesopic low-contrast BCDVA (mLCVA)
- Photopic low-contrast BCDVA (pLCVA)
- Mesopic high-contrast BCDVA (mHCVA)
- Mesopic high-contrast DCNVA
- Photopic pupil diameter

- Mesopic pupil diameter
- Total RMS error (score) and higher-order RMS (spherical, coma, trefoil) error
- Subject questionnaire

Safety variables include:

- Conjunctival hyperemia (eye redness) measured with a CCLRU card 4-point scale:
    - None (0) = Normal Appears white with a small number of conjunctival blood vessel easily observed.
    - Mild (+1) = Prominent, pinkish-red color of both the bulbar and palpebral conjunctiva.
    - Moderate (+2) = Bright, scarlet red color of the bulbar and palpebral conjunctiva.
    - Severe (+3) = Beefy red with petechiae, dark red bulbar and palpebral conjunctiva with evidence of subconjunctival hemorrhage.
- AEs
- Vital signs (HR and BP)
- IOP
- Biomicroscopy
- Ophthalmoscopy
- Urine pregnancy tests for females of childbearing potential

6. SAMPLE SIZE

A sample size of approximately 136 subjects that are evaluable for efficacy (68 per treatment group) is needed for the study.

It is assumed that there will be approximately 15% drop-out between Day 1 and Day 15. To account for this drop-out, a total of approximately 160 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. Modified Intention-to-Treat (mITT)

The mITT will include all randomized subjects who received at least 1 dose of study medication and have a Baseline and any Day 8 efficacy measurement. The mITT will be used for the primary endpoint analysis and to analyze efficacy endpoints, with subjects included in their randomized treatment regardless of the treatment they actually received.

7.1.2. Per Protocol Population (PP)

The PP population includes all subjects in the mITT who received at least 5 doses of study medication during the first 7 days of dosing including the day prior to Day 8, have a Baseline and a Day 8 mLCVA measurement, and have no major protocol deviations considered to have significant impact on treatment outcome. The PP population will be used to analyze efficacy endpoints, with subjects included in their randomized treatment regardless of the treatment they actually received.

7.1.3. All Randomized Population (ARP)

The ARP will include all randomized subjects. This population is also known as the Intention-To-Treat (ITT) population. The ARP will be used in confirmatory efficacy analyses, with subjects included in their randomized treatment regardless of the treatment they actually received.

7.1.4. Safety Population (SP)

The SP will include all randomized subjects who have received at least one dose of study medication. The SP will be used to summarize safety variables, using the actual treatment a subject received.

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 by [redacted]

[redacted]
select efficacy endpoints. Other possible subgroups include sex and race.

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 percent of subjects with ≥ 15 ETDRS letters (≥ 3 lines) of improvement in the study eye compared to baseline in monocular mLCVA at Day 8. For the analysis of the primary efficacy endpoint, imputation will be performed for missing efficacy data as specified in Section 7.3.2.3 for the analysis using the mITT. Confirmatory analyses will be performed using the ARP, also using imputation for missing data.

Otherwise, there will be no substitutions made to accommodate missing data points for efficacy data. All data recorded on the CRF will be included in data listings that will accompany the CSR.

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 SP; 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 AEs 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 SP 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

[redacted]

The imputations will be done separately for each treatment group and will include the following variables in the imputation model: mLCVA at baseline and Day 8.

The number of imputations [redacted] The change from baseline will be calculated from these imputed datasets. The observed outcome ([redacted]) for each imputed dataset will be [redacted] See Section 9.1 for details on these models. The estimates

and standard errors (SEs) of the odds ratios based on the [redacted]

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 data for safety and efficacy variables will be assigned to the closest visit (Day 8 or Day 15). 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 AEs and medical history will be mapped to the Medical Dictionary for Regulatory Activities (MedDRA Version 23.1) system for reporting (preferred term and body system).

Prior and concomitant medications will be coded using WHO-DD (World Health Organization Drug Dictionary) (Global Version 2020-09-01).

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 CSR 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 the electronic data capture (EDC) MedNet version 1.212.2.

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

[redacted]

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 (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 group. For categorical variables, the counts and proportions of each value will be tabulated by treatment group. 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 any drug administration on the evening of the Baseline Visit (Day 1). If the Day 1 value is missing, any value collected prior to treatment administration 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 ARP, subjects in the SP, subjects in the mITT Population, and subjects in the PP Population. All percentages will be based on the number of subjects in the ARP.

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, , 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 considered to have significant impact on treatment outcome, 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 mITT Population, PP Population, SP, and ARP. If the mITT population is equivalent to any of the other populations, then only the mITT version will be generated rather than repeating equivalent summaries.

The demographics consist of age (year), sex, race, ethnicity, and study eye, iris color (light blue, dark blue, blue with peripupillary brown, uniform green, green with brown iris ring, central brown and peripheral green, brown with some peripheral green, or brown), irides type (light or dark), and eyeglasses-wearing status (yes or no; distance vision or near vision). 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 sex, race, ethnicity, study eye, iris color, irides type, and distance vision/near vision correction status, 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 study eye and non-study eye, and for binocular for visual acuity assessments, using descriptive statistics:

- mLCVA
- pLCVA
- mHCVA
- DCNVA
- Pupil diameter (photopic and mesopic)
- IOP
- Total RMS error (score)
- Total wavefront higher-order error (score)
- Higher-order RMS (spherical, coma, trefoil) error

All demographic and baseline information will be presented in by-subject listings.

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 23.1. 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 SP.

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 SP.

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. The total number of prior medications and the number and percentages of subjects with at least 1 prior medication will be summarized by treatment group.

8.6. Treatment Exposure and Compliance

9. EFFICACY ANALYSES

Unless otherwise noted, efficacy will be assessed using the mITT and PP populations, with subjects included in their randomized treatment regardless of the treatment they actually received. For the analysis of the primary efficacy endpoint, imputation will be performed for missing data as described in Section 7.3.2.3. If the analysis using the mITT Population shows a positive effect for Nyxol at the 0.05 level of significance, the primary endpoint will be considered met.

Confirmatory analysis of the primary efficacy endpoint will be performed using the ARP, also using imputation for missing data. For the analysis of the secondary efficacy endpoints, only observed case data will be used.

To formally test the significance of endpoints of interest beyond primary efficacy, endpoints will be tested in a predefined sequence using observed data only, each at the significance level 0.05, until the first nonsignificant test. The primary efficacy endpoint will be first in this sequence. The endpoints in the sequence are for the mITT population and study eye, and either Day 8 or Day 15. The sequence is specified in Section 12.3.

All efficacy assessment data, regardless of whether they are included in the analysis, will be presented in by-subject listings.

9.1. Clinical Efficacy

All efficacy data will be summarized by treatment group (Nyxol or placebo) and visit (Day 1, Day 8, and Day 15).

9.1.1. Primary Efficacy Analysis

[redacted]

By including [redacted] primary efficacy analysis model, the model efficiency as well as a change in the treatment effect will be increased. Including this factor in the model will also make the results more generalizable to other studies in which the sample characteristics may differ from the current study [2]. Additionally, a sensitivity analysis will be applied to the primary efficacy endpoint for the mITT and PP populations,

which uses a logistic regression model with only treatment as a [redacted]).

For the analysis of the PP Population, subjects who did not dose the night before, or the morning of, their Day 15 visit will be excluded from the Day 15 visit summary.

In addition, the primary efficacy endpoint [redacted] using the same model indicated above [redacted]. For these subgroup analyses, observed case data only will be used; that is, missing values will not be imputed.

9.1.2. Secondary and Exploratory Efficacy Analysis

Secondary and exploratory efficacy endpoints are indicated in Section 4.2.2. Secondary and exploratory efficacy endpoints will be analyzed by study eye, non-study eye, the best of either eye, and binocular, unless otherwise indicated.

Visual acuity assessments (mLCVA, pLCVA, mHCVA, and DCNVA) will be summarized using letters. For mLCVA, pLCVA, and mHCVA, the assessments will also be summarized using logMAR units. Only letters will be recorded in the CRF. Letters will be converted to logMAR using the following formula: logMAR = 0.02*(S - # Letters), where S = 55. The values of S are the number of letters read equivalent to a Snellen Acuity of 20/20. For example, for mLCVA, if the # letters = 55, then logMAR = 0.02*(55 - 55) = 0.00; if # letters > 55, then logMAR is negative.

Each of the continuous secondary efficacy endpoints will be analyzed using analysis of covariance (ANCOVA), with change from baseline as the dependent variable, treatment, and [redacted] and the respective baseline value included as the covariate.

Each ANCOVA will be performed using the mITT and PP populations. The output from each ANCOVA will include the LSM and SE for both treatment groups, along with the placebo-corrected LSM, its 95% CI and associated p-value.

For each of the secondary endpoints related to percent of subjects achieving certain criteria, the analysis will be performed using a logistic regression model with treatment [redacted] s, and the respective baseline as a covariate. For each analysis, the percentage of subjects in each treatment group meeting the criteria, the OR with 95% CI and p-value will be provided.

In addition, each secondary efficacy endpoint will be analyzed by [redacted] the same model indicated above but without irides as a factor.

For Day 8 and Day 15, the time since last dose of study treatment will be calculated as follows: date/time of visit minus date/time of the most recent dose of study medication in the subject diary. [redacted] For each subject, the category could be different for Day 8 and Day 15. Summaries of for improvement in mLCVA, and change and percent change in pupil diameter, will be presented based on these categorizations.

Questions from the subject questionnaire, including those questions for which it is not appropriate to be analyzed using the methods above, will be summarized categorically for each treatment group and visit, as well as by [redacted]

10. SAFETY ANALYSES

All safety analyses will be conducted using the SP. 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.

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

10.1. Adverse Events

AEs will be coded using MedDRA, Version 23.1.

Treatment-emergent adverse events (TEAEs) are defined as any AE that begins or worsens after initiation of the investigational product and through the subject's last study visit (study completion/follow-up phone call, or early termination).

If the onset of an AE is on or after the date of first dose of study medication or is 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 SOC and PT by treatment group and overall. At each level of tabulation (e.g., at the PT 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 "Eye Disorders". 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 SOC, PT, and by greatest relationship level to study drug (not related, unlikely related, possibly related, probably related, definitely related, or unknown)
- Summary table of TEAEs by SOC, PT, and maximum severity (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.

10.2.2. Serious Adverse Events

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

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. Conjunctival Hyperemia

Results from the conjunctival hyperemia assessment, measured with a CCLRU card 4-point scale, will be summarized descriptively using counts and percentages for each treatment group at each visit (Day 1, Day 8, and Day 15) for the observed value and change from baseline (Day 1). Separate summaries will be created for the study eye, the non-study eye, and the best of either eye.

Additionally, conjunctival hyperemia will be summarized as a continuous variable. Treatments will be compared using the same ANCOVA model proposed for the continuous secondary efficacy endpoints.

10.4. Vital Signs

Descriptive statistics of observed values will be presented for vital sign data at each visit (Day 1, Day 8, and Day 15), including systolic BP (mmHg), diastolic BP (mmHg), and HR (bpm) by treatment group and overall. Changes from baseline (Day 1) to each scheduled post-baseline time point will be presented.

10.5. IOP

Observed values and change from baseline in IOP will be summarized descriptively using counts and percentages for each treatment group at each visit (Day 1, Day 8, and Day 15) for the observed value and change from baseline (Day 1). Separate summaries will be created for the study eye, the non-study eye, and the best of either eye. Treatments will be compared using the same ANCOVA model proposed for the continuous secondary efficacy endpoints.

10.6. Biomicroscopy and Ophthalmoscopy

Results from biomicroscopic and ophthalmoscopic examinations at Day 1 and Day 15 will be summarized by treatment group.

10.7. Other Safety Measures

Urine pregnancy tests for females of childbearing potential will be presented in by-subject listings. Dry eye tests (TearLabs osmolarity test, tear break-up time and cornea fluorescein staining), performed only at Day 1, will also be listed.

11. REFERENCES

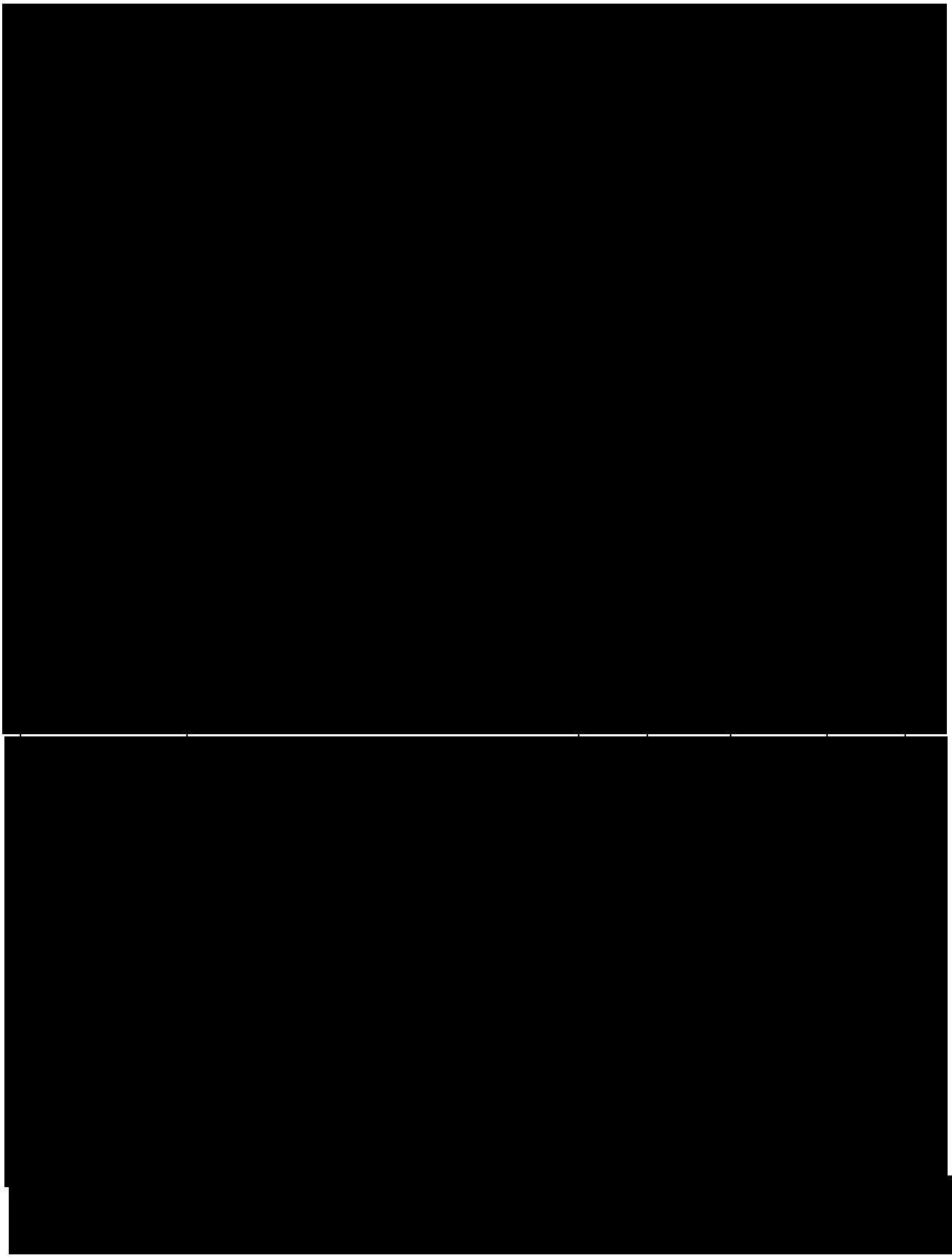
[1] ICH E9 Expert Working Group. Statistical Principles for Clinical Trials: ICH Harmonized Tripartite Guideline, September 1998

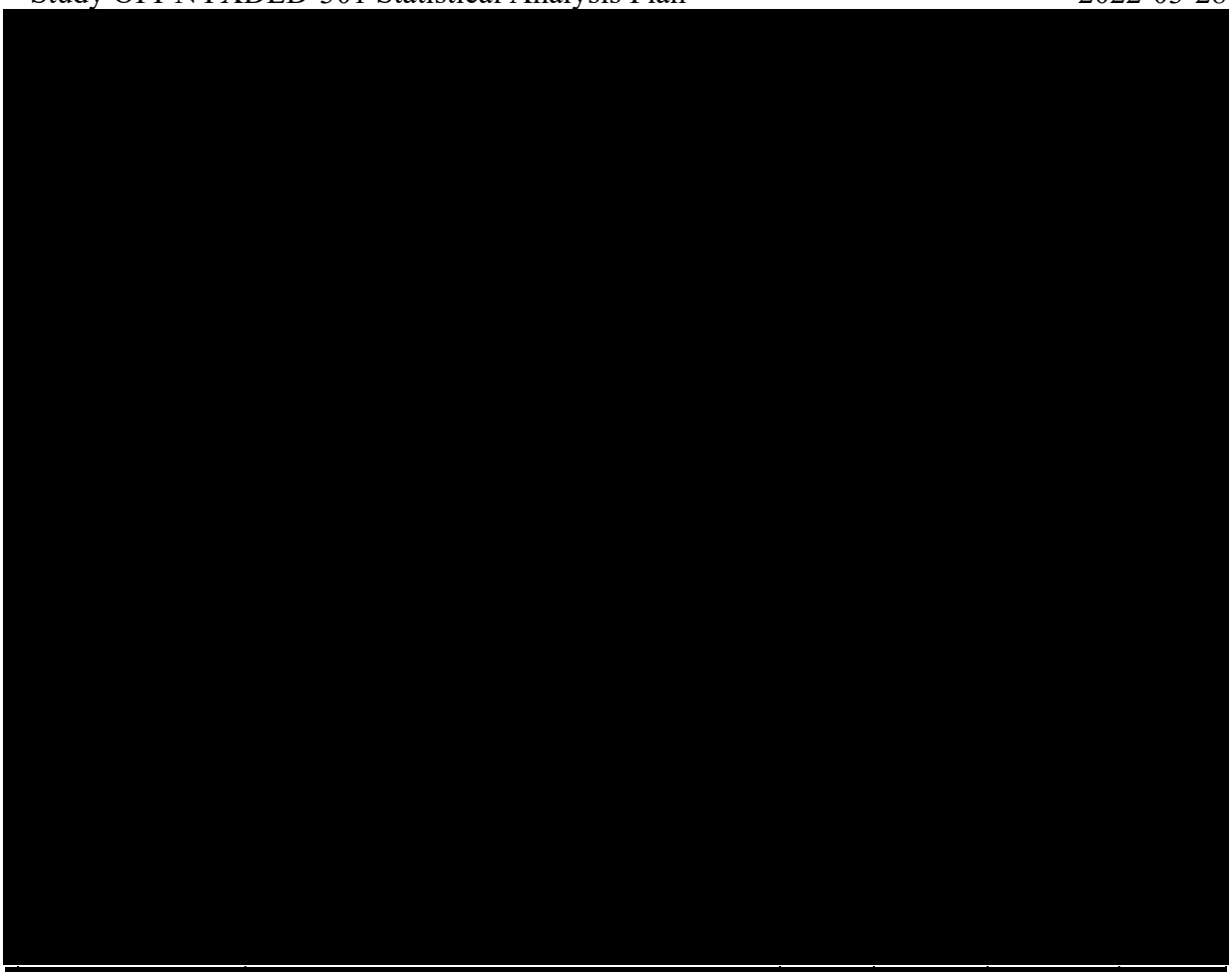
[2] Hauck WM, Anderson S, and Marcus SM, Should We Adjust for Covariates in Nonlinear Regression Analyses of Randomized Trials? *Controlled Clin Trials* 1998;19:249–256

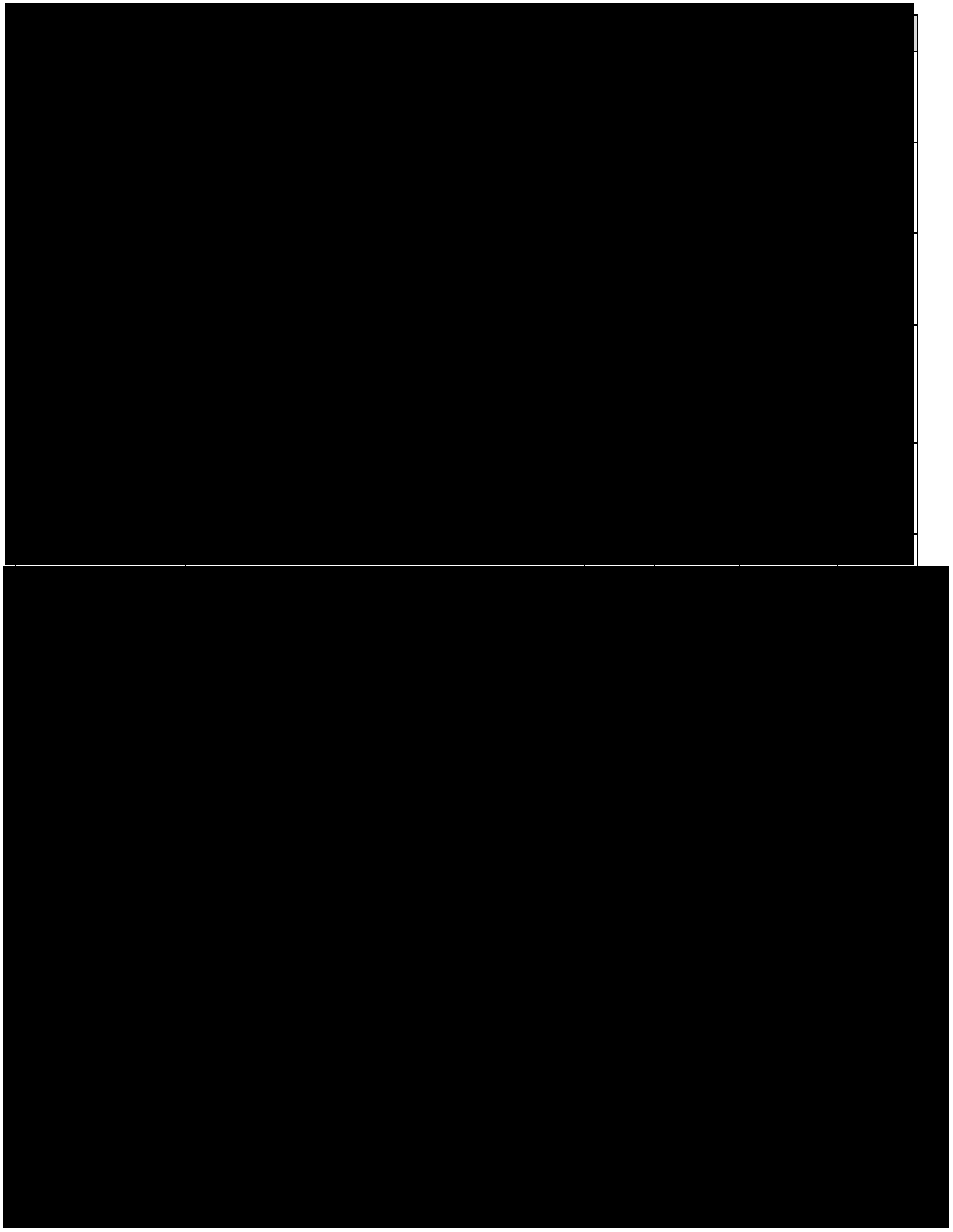
12. APPENDICES

12.1. List of Planned Tables

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







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12.2. List of Planned Listings

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12.3. Hierarchy of Sequential Testing