STATISTICAL ANALYSIS PLAN

A Phase I Open-Label, Dose Escalation Study to Assess the Safety and Tolerability of Intravitreal ONL1204 in Patients with Macula-off, Rhegmatogenous Retinal Detachment

PROTOCOL NO.: ONL1204-RRD-001

PRODUCT CODE: ONL1204

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SAP APPROVAL

By my signature, I confirm that this SAP has been reviewed by ONL Therapeutics and has been approved for use on the ONL1204-RRD-001 study:

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List of Abbreviations

Abbreviation	Description
R^2 adj	Corrected goodness-of-fit
AE	Adverse Event
ATC	Anatomical Therapeutic Class
AUC	Area under the curve
BCVA	Best Corrected Visual Acuity
BLQ	Below limit of quantification
nBLQ	Number of participants below limit of quantification
BMI	Body Mass Index
CS	Clinically Significant
CSR	Clinical Study Report
CI	Confidence Intervals
CV	Coefficient of Variation
DA	Dark Adaptometry
DBP	Diastolic blood pressure
ECG	12-Lead Electrocardiogram
ETDRS	Early Treatment Diabetic Retinopathy Study
FSH	Follicle Stimulating Hormone
eCRF	Electronic Case Report Form
GCP	Good Clinical Practice
IOP	Intra-Ocular Pressure
ITT	Intent-to-Treat
MedDRA	Medical Dictionary for Regulatory Activities
N/A	Not Applicable
NCA	Non-compartmental analysis
NCS	Not Clinically Significant
NK	Not Known
PI	Principal Investigator
PD	Pharmacodynamic
PK	Pharmacokinetic
PO	per oral
PT	Preferred Term
RBP4	Retinol-Binding Protein 4
SAD	Single Ascending Dose
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SBP	Systolic blood pressure
SD	Standard Deviation
S.I.	International System of Units
SOC	System Organ Class

1. INTRODUCTION

The following Statistical Analysis Plan (SAP) provides the outline for the statistical analysis of data collected from the ONL1204 study (Protocol Version 1.3 dated 3 July 2019, protocol version 1.5 dated 10th May 2020, protocol version 1.6 dated 27th July 2020).

Protocol v1.3 and v1.4 had a total of 10 visits and all these visits are captured for patients 111-001, 111-002, 111-005, 222-002 only.

Protocol v1.5 had a total of 8 visits and are applicable to the rest of the patients.

Protocol v1.6 was introduced only for the use of site 444, to allow for the pre-injection B Scan Ultrasonography to be optional at Visit 1/Day1. This was brought about as the site did not have access to the relevant equipment.

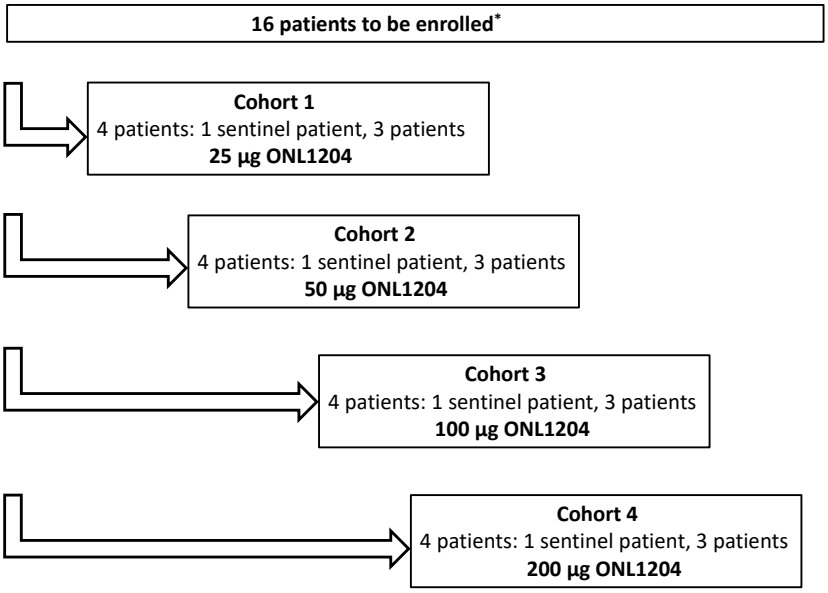
The planned analyses identified in this SAP may be included in clinical study reports (CSRs), regulatory submissions, or future manuscripts. Also, post hoc exploratory analyses not necessarily identified in this SAP may be performed to further examine study data. Any post hoc, or unplanned, exploratory analyses performed will be clearly identified as such in the final CSR.

2. PROJECT OVERVIEW

Study Design

This proposed study is a single-injection, open-label, dose escalation study to assess the use of intravitreal ONL1204 in patients with macula-off rhegmatogenous retinal detachment (RRD). Approximately 16 patients with RRD will be enrolled. The expected duration of patient participation in the study is approximately 24 weeks (treatment, then 24 weeks of follow-up).

Figure 1: Study Design



*Patients may be replaced who do not receive an injection of study medication, or from discontinuing from the study for reasons unrelated to treatment before completing Visit 4.

Sixteen patients will be enrolled in 4 ascending dose groups.

After signing informed consent and meeting all eligibility criteria, patients will undergo a vitreous tap for cytokine and other potential molecular biomarker analysis followed by a single intravitreal injection of ONL1204. This will occur on the day of diagnosis (Day 1). Safety visits will be conducted 1-day post-injection and per Investigator's discretion until Visit 3. At Visit 3 (3+ days post-injection, in accordance with standard of care) patients will undergo surgical repair of their retinal detachment with vitrectomy (with or without scleral buckling) and gas or oil tamponade. At the time of surgery, an undiluted vitreous sample will be collected for pharmacokinetic and pharmacodynamics analyses, and an aqueous sample will be collected for pharmacodynamic analysis. Additional safety visits will be conducted 1-day post-surgery, 14 (+4 days) days post-surgery, Weeks 6, 9 (±7 days each), and Week 24 (±14 days; study exit).

As this is a first-in-human safety study of ONL1204, the first patient in the first cohort will receive the lowest dose of ONL1204. After this patient completes Visit 5 (14 +4 days post-surgery) and a safety review committee reviews the data, the remainder of the patients in that cohort will be enrolled. After all patients in the cohort have completed Visit 5, the SRC will review the available safety data to determine if dose limiting toxicity exists that prohibits enrolling the next higher dose cohort. This process will be repeated until escalating to the top dose.

The following doses will be evaluated: 25 µg, 50 µg, 100 µg, and 200 µg ONL1204. ONL1204 ophthalmic solution is available in vials with concentrations of 0.5 mg/mL and 2 mg/mL. Therefore, a volume of 0.05 or 0.1 mL will be injected into the study eye.

Objectives

2.1.1 Primary objective

The primary objective will be to determine the Maximum Tolerated Dose (MTD) of ONL1204 and select doses for Phase 2. Determination of this will be through review of the parameters measured in the Schedule of Assessments, as well as adverse event (AE) reports.

2.1.2 Secondary objective

Not Applicable

2.1.3 Exploratory objective

During the study, ocular fluid samples will be collected at the time of study drug injection and at the time of vitrectomy. These samples will be used for pharmacokinetic and pharmacodynamic analyses, the results of which will help inform the dose selection for the Phase 2 trial(s).

Endpoints

2.1.4 Primary endpoints

The primary endpoints will be the safety profile demonstrated in this study, including:

- Adverse event reporting
- Clinical evaluations including BCVA
- Clinical laboratory evaluation

2.1.5 Secondary endpoint

Not Applicable.

2.1.6 Exploratory endpoints

The exploratory endpoints will be the pharmacokinetic and pharmacodynamic analyses performed in this study, including:

- Measurement of ONL1204 drug levels in plasma and vitreous fluid for PK analysis
- Cytokine and other potential molecular biomarker measurement in aqueous and vitreous samples, taken by vitreous tap, and at vitrectomy

Sample Size

The number of patients to be enrolled has been selected to provide information for this Phase 1 study. No formal sample size calculation is provided given the open-label nature of this study.

Randomization

This is an open-label study, and all patients will be receiving study drug.

3. STATISTICAL CONSIDERATIONS

Data will be handled and processed per the sponsor's representative (Novotech) Standard Operating Procedures (SOPs), which are written based on the principles of good clinical practice (GCP).

General Considerations

All data collected on the electronic case report form (eCRF) will be presented in the data listings and will be listed and sorted by cohort, participant number and visit, where applicable. All descriptive summaries will be presented by cohort and nominal visit/time point (where applicable).

All safety, PD and PK descriptive summaries will be presented by cohort. All disposition and, concomitant medication descriptive summaries will be presented by cohort and nominal visit/time point (where applicable).

Unless otherwise stated, the following methods will be applied:

- Continuous variables: Descriptive statistics will include the number of non-missing values (n), arithmetic mean, standard deviation (SD), median, minimum, and maximum values.

  The minimum and maximum values will be displayed to the same decimal precision as the source data, the arithmetic mean, SD, and median values will be displayed to one more decimal than the source data for the specific variable.

  95% Confidence Intervals (CIs), mean differences (among treatments and from baseline) and least-square (LS-Means) values will be displayed to one more decimal than the source data for a specific variable. P-values will be displayed to 3 decimal places.

  The appropriate precision for derived variables will be determined based on the precision of the data on which the derivations are based, and statistics will be presented in accordance with the above-mentioned rules.

- PK data: The actual blood sampling dates and times relative to dosing time will be listed by participant and nominal sampling time, with time deviation calculated, for all participants with available plasma and vitreous concentration data, including participants excluded from the PK population. Individual (for each participant) and mean concentrations over time will be displayed graphically in linear and semi-logarithmic plots of concentrations versus time. The actual collection time will be used for individual plasma concentration curves and the nominal time will be used for the plots of mean plasma concentration curves. Similar plot will be provided using vitreous concentration.

  For PK concentration data, the number of non-missing values, number of below limit of quantification (BLQ) values, arithmetic mean, standard deviation, median, minimum, maximum, coefficient of variation (CV%), geometric mean and geometric coefficient of variation (geo CV%) values will be presented. For the calculation of summary statistics, unrounded data will be used and reported to three significant figures except for n, n BLQ, and CV% which will be presented to the nearest integer and one decimal place, respectively.

  For PK parameters data, the number of non-missing values, arithmetic mean, standard deviation, median, minimum, and maximum values will be presented. Individual PK parameters will be presented to three significant figures except for T_{max} which will be presented to two decimal places.

- Categorical variables: Descriptive statistics will include counts and percentages per category. The denominator in all percentage calculations will be the number of participants in the relevant analysis population with non-missing data, unless specifically stated otherwise. Percentages will be displayed to one decimal place. Proportions will be displayed to 3 decimal places.

- Unscheduled assessments: Only values collected at scheduled study visits/time points will be presented in summary tables. If a repeat assessment was performed, the result from the original assessment will be presented as the result at the specific visit/time point. All collected data will be included in the data listings.

- Assessment windows: All assessments will be included in the data listings and no visit windows will be applied to exclude assessments that were performed outside of the protocol specified procedure windows.
- Result display convention: Results will be center aligned in all summary tables and listings. Participant identifiers visit and parameter labels may be left-aligned if required.
- Date and time display conventions: The following display conventions will be applied in all outputs where dates and/or times are displayed:

  Date only: YYYY-MM-DD

  Date and time: YYYY-MM-DD HH:MM

If only partial information is available, unknown components of the date or time will be presented as 'NK' (not known), i.e., '2016-NK-NK'. Times will be reported in military time.

Key Definitions

The following definitions will be used:

- Baseline: The baseline value is defined as the last available valid (quantifiable continuous or categorical value), non-missing observation for each participant prior to first study drug administration. Repeat and unscheduled assessments will be included in the derivation of the baseline values.
- Change from Baseline: The change from baseline value is defined as the difference between the result collected/derived at a post-baseline visit/time point and the baseline value.

  The change from baseline value at each post-baseline visit/time point will be calculated for all continuous parameters using the following formula:

  Change from Baseline Value = Result at Visit/Time Point – Baseline Value

  The change from baseline value will only be calculated if the specific post-baseline visit/time point result and the baseline value for the parameter are both available and will be treated as missing otherwise.
- Study day: The study day of an event is defined as the relative day of the event starting with the date of the first study drug administration (reference date) as Day 1 (there will be no Day 0).

  The study day of events occurring before the first study drug administration will be calculated as:

  Study Day = (Date of Event - Date of First Study Drug Administration)

  For events occurring on or after Day 1, study day will be calculated as:

  Study Day = (Date of Event - Date of First Study Drug Administration) + 1

  Study days will only be calculated for events with complete dates and will be undefined for events that are 'Ongoing' at the end of the study.

  Relative days compared to an alternative reference point will be calculated similarly, but the alternative starting reference start date will be used instead of the date of the first study drug dosing.
- Prior Medications: Prior medications are defined as any medication where the use was stopped prior to the first administration of the study medication.
- Concomitant Medications: Concomitant medications are defined as any medication (other than the study drug) that was used at least once after the first administration of the study drug. Medications that were stopped on the same date as the first study drug administration will be defined as concomitant medications. If a clear determination cannot be made (partial medication end dates) the medication will be classified as concomitant.

- Actual Time from First Dose (hours) = (Date/Time of First Dose PK Sample collection) – (Date/Time of First Dose)
- Actual Time Deviation (hours) = (Actual PK Sample Collection Time Post Dose) – (Scheduled PK Sample Collection Time Post Dose).
- Treatment Emergent Adverse Events (TEAEs) are defined as adverse events that occurred following the first administration of study medication.

Inferential Analyses

Descriptive statistics will be used to summarize the safety, PD and PK data. No formal hypothesis testing is planned.

Multiple Comparisons and Multiplicity Adjustments.

Not applicable for this study.

Handling of Missing Data

For the classification of Treatment emergent adverse event (TEAE) and Concomitant medication, the following will be applied in the following order:

a. If all dates/times (start and stop) missing, the event/medication will automatically be classified as a TEAE/Concomitant medication.

b. For AEs with a missing start date/time, if the event end date/time is prior to first study drug administration, the event will not be classified as a TEAE.

c. If only the AE start year/ medication end year is present and is the same or is after the first study drug administration year unit, the event/medication will be classified as a TEAE/Concomitant medication.

d. If the AE start month and year/medication end month and year are present and are the same or after the first study drug administration month and year units, the event/medication will classified as a TEAE/Concomitant medication.

For plasma concentration-time data/profiles and PK analysis the following will be applied:

a. Concentrations that are below limit of quantification (BLQ) prior to the dosing (Pre-dose) will be set equal to zero. Post dose BLQ concentrations will be set to zero.

b. Where there are no results, these will be set to missing.

c. If more than 75% of values per nominal timepoint and treatment group are BLQ, then all descriptive statistics, except for nBLQ, will be denoted as not calculable (NC).

d. For the purpose of calculating the non-compartmental plasma PK parameters, concentrations that are BLQ prior to the first quantifiable value will be set equal to zero and BLQ values after measurable concentrations will be set to zero.

e. For the calculation of summary statistics of plasma PK parameters, all NR (not reported) and NC (not calculable) values in a data series will be set to missing.

f. The specific reasons for ‘not calculable (NC)’ and ‘not reported (NR)’ for PK parameters will be provided as footnotes in the PK results listings and summaries.

For vitreous concentration-time data/profiles the following will be applied:

a. Concentrations that are below limit of quantification (BLQ) prior to the dosing (Pre-dose) will be set equal to zero. Post dose BLQ concentrations will be set to zero.

b. Where there are no results, these will be set to missing.

c. If more than 75% of values per nominal timepoint and treatment group are BLQ, then all descriptive statistics, except for nBLQ, will be denoted as not calculable (NC).

Conversion of categorical values

In some instances, continuous variables are expressed as a range (i.e., < 10). In such cases, values may be converted to the range boundary (upper or lower limit as applicable). As an example, a value of <10 may be converted to 10. Such substitutions will be clearly documented in the footnotes of relevant outputs.

Coding of Events and Medications

Medical history and AE verbatim terms will be coded using the Medical Dictionary for Regulatory Activities (MedDRA) using version 23.0. Terms will be coded to the full MedDRA hierarchy, but the system organ class (SOC) and preferred terms (PT) will be of primary interest for the analysis.

Prior and concomitant medications will be coded using the World Health Organization Drug Dictionary using the latest version available at the time of study commencement. Medications will be mapped to the full WHO-DD Anatomical Therapeutic Chemical (ATC) class hierarchy, but PTs will be of primary interest in this analysis.

Treatment Groups

- Cohort 1 - ONL1204 25 µg
- Cohort 2: - ONL1204 50 µg
- Cohort 3 - ONL1204 100 µg
- Cohort 4 - ONL1204 200 µg
- Overall

4. ANALYSIS POPULATIONS

In this study five analysis populations are defined: The Intent-to-Treat (ITT), Safety population, Per Protocol, Pharmacokinetic (PK) and Pharmacodynamic (PD).

Furthermore, any additional exploratory analysis not identified in the SAP will be identified in the final CSR as exploratory post hoc analyses, including analyses for additional study populations or subgroups of interest.

Population Descriptions

4.1.1 ITT Population

The ITT population will be defined as all patients enrolled for the study.

All disposition and demographic data analysis will be based on the ITT population. All listings will be presented by the ITT population.

4.1.2 Safety Population

The Safety population will be defined as all patients who received the study drug and will be based on actual treatment received.

All safety analyses will be based on the Safety population.

4.1.3 Per Protocol Population

The per protocol formulation is defined as all patients who received their assigned treatment and includes all those with no major protocol deviations.

4.1.4 Pharmacokinetic Population

4.1.4.1 *Ocular PK population*

All patients who are included in the safety population and for whom quantified vitreous (ocular) concentration of ONL1204 is available will be included in the ocular PK population. Patients with protocol violations will be assessed on a patient-by-patient basis for inclusion in the ocular PK Population. Ocular PK Population determination will be made before database lock by the Study Pharmacokineticist. This ocular PK population will be used for the summaries and graphical presentation of ocular ONL1204 concentration data.

4.1.4.2 *Plasma PK population*

~~For the systemic PK data, per cohort and per protocol analysis will be performed. All patients who are included in the safety population with no major protocol deviations, who provide adequate PK samples and have a sufficient PK profile that allows the calculation of relevant PK parameters will be included in the PK population.~~

Plasma PK population is defined as all patients who are included in the safety population and for whom an evaluable plasma concentration-time profile of ONL1204 is available for the determination of at least one of the PK parameters among C_{max}, T_{max} and AUC_{0-t} with no major protocol deviations that could potentially affect the PK profile.

Plasma PK population determination will be made before database lock by the Study Pharmacokineticist.

The plasma PK population will be used for the plasma PK analysis, the summaries and graphical presentation of all plasma PK data.

4.1.5 PD Population

All patients who are included in the safety population and have at least baseline and 1 post-dose PD assessment will be included in the PD population. This population set will be derived from sample collection date for Vitreous Humor sample collected at screening and undiluted vitreous/Aqueous humor sample at time of surgery.

All PD analyses will be based on the PD population.

5. PARTICIPANT DISPOSITION AND ANALYSIS POPULATIONS

Participant disposition and analysis population will be based on the ITT population. Participant disposition and analysis populations will be summarized descriptively as described in section 3.1 (categorical descriptive analysis).

5.1.1 Participant Disposition

Participant disposition will include the number and percentage of participants who are screened, treated, completed the study as planned, participants withdrawn from the study, as well as the primary reason for early termination. Participant disposition will be summarized descriptively.

5.1.2 Analysis Populations

The number of participants included in each study population will be summarized descriptively.
In addition, the inclusion of each participant into/from each of the defined analysis populations will be presented in the by-participant data listing.

6. PROTOCOL DEVIATIONS

Protocol deviations will be presented for each participant in the by-participant data listings.

Prior to database lock, all protocol deviations will be reviewed by medical monitors and assigned a status of Important if qualifying as such.

Protocol deviations and important protocol deviations will be categorized as noted in the protocol deviation management plan as per project and communication plan.

7. DEMOGRAPHIC AND BASELINE INFORMATION

Demographic and baseline information analysis will be based on the ITT population. Demographic and baseline information will be summarized descriptively as described in section 3.1 (Continuous/ Categorical)

Demographics

The following demographic parameters will be analyzed:

Continuous descriptive analysis:

- Age (years)
- Height (cm)
- Weight (kg)

Categorical descriptive analysis:

- Sex
- Childbearing Potential as defined in the protocol
- Race
- Ethnicity

Medical History

Medical history will be coded using MedDRA® and will be presented in the by-participant data listings.

Disease History

Disease history will be presented in a separate by-participant listing.

8. TREATMENT EXPOSURE

All study drug administration information (cohort, study drug administered <Yes/No>, date of injection, time of injection will be presented in the data listings.

9. PRIOR AND CONCOMITANT MEDICATIONS

Prior and concomitant medications will be coded using the most current version of the WHO-DD available at the start of the study.

Concomitant medications will be summarized for the safety population. by Anatomical Therapeutic Chemical (ATC) class Level 3 and Preferred name as noted in section 3.1 (categorical descriptive analysis). Participant who used the same medication on multiple occasions will only be counted once in the specific category (level and preferred name). ATC level and Preferred name will be presented alphabetically.

In addition to the summaries by the coded terms, the number of participants who used at least one concomitant medication during the study will be presented.

Prior medications will be presented in the by-participant data listings.

10. PHARMACOKINETICS

10.1 Plasma PK Analysis

The plasma PK samples for ONL1204 will be at specified times as follows:

-
- Post-intravitreal injection
- 1-day post- intravitreal injection
- 3+ days post- intravitreal injection or day of surgery (vitrectomy)
- 14 days post-surgery
- Week 9
- Week 24 exit or early termination

All PK summary tables will be based on the plasma PK population. All analyses will be summarized as described in section 3.1 (PK - continuous descriptive analysis). All listings will be based on the safety population.

The following pharmacokinetic parameters will, where possible, be determined from the plasma concentrations of ONL1204 by non-compartmental method using Phoenix WinNonlin® software (Version 8.3 or higher):

Pharmacokinetic Parameters of Plasma ONL1204 Following a Single Dose Administration of ONL1204 Intravitreal Injection

~~**Parameter**~~	~~**Definition**~~
~~AUC0-last~~	~~The area under the plasma concentration-time curve, from time 0 (time of dosing) to the last time point with measurable analyte concentration, calculated by the log down, linear up trapezoidal method~~
~~AUC0-inf~~	~~The area under the plasma concentration-time curve from time 0 extrapolated to infinity. AUC0-inf is calculated as the sum of AUC0-last plus the ratio of the last measurable plasma concentration to the elimination rate constant (λz).~~
~~Cmax~~	~~Maximum analyte plasma concentration~~
~~Tmax~~	~~Time to maximum observed analyte concentration. If the maximum value occurs at more than one time point, Tmax is defined as the first time point with this value.~~
~~t½~~	~~Apparent plasma terminal elimination half-life calculated as ln (2)/λz~~

Parameter	**Definition**
C_{max}	Maximum concentration which is directly determined from the plasma concentration time profiles
T_{max}	Time to maximum concentration. If the same C_{max} concentration occurs at different time points, T_{max} is assigned to the first occurrence of C_{max}.
$AUC_{0\text{-last}}$	Area under the drug concentration-time curve, from time zero to the last measurable concentration using the 'Linear Up and Log Down' method
$AUC_{0\text{-inf}}$	Area under the drug concentration-time curve, from time zero to infinity (∞) using the following formula $AUC_{0-\infty} = AUC_{0-t} + \frac{C_t^{'}}{\lambda_z}$ Where, $C_t^{'}$ is the observed concentration at the time t (last time point with a measurable plasma concentration above the quantification limit) at which quantification was still possible, the calculation of λ_z or k_{el} is given below.

Parameter	Definition
$AUC_{\%extrap}$	The percentage of the AUC that has been extrapolated beyond the last observed data point, using the following formula $AUC_{\%extrap} = \left(\frac{AUC_{0-\infty} - AUC_{0-t}}{AUC_{0-\infty}}\right) * 100$
λ_z or k_{el}	The apparent terminal elimination rate constant, will be estimated from a regression of ln(C) versus time over the terminal log-linear drug disposition portion of the concentration-time profiles.
$t_{½}$	Apparent terminal half-life, using the following formula $T_{1/2} = \frac{Ln(2)}{\lambda_z}$
CL/F	Apparent total plasma clearance, using the following formula $CL/F = \frac{D}{AUC_{0-\infty}}$ Where D = Administered dose; $AUC_{0-\infty} = AUC_{0-inf}$
V_z/F	Apparent terminal volume of distribution, using the following formula $V_z/F = \frac{CL/F}{\lambda_z}\ Or\ \frac{Dose}{\lambda_z \times AUC_{inf}}$

Value for K_{el}, $t_{½}$ AUC_{0-inf}, CL/F and V_z/F will not be reported for cases that do not exhibit a terminal log-linear phase in the concentration versus time profile. Additional pharmacokinetic parameters may be determined where appropriate.

Pharmacokinetic analysis will be carried out using actual doses, ~~infusion durations,~~ and blood sampling times, where possible. If actual times are missing, nominal times may be used with sponsor approval.

Plasma concentrations of ONL1204 will be used as supplied by the bioanalytical laboratory. The units of concentration and resulting PK parameters, with amount or concentration in the unit, will be presented as they are received from the bioanalytical laboratory.

10.1.1 Criteria for the Calculation of an Apparent Terminal Elimination Half-Life and AUC

~~The start of the terminal elimination phase for each participant will be defined by visual inspection and will generally be the first point at which there is no systematic deviation from the log-linear decline in plasma concentrations.~~

10.1.1.1 Number of Data Points

- At least 3 data points will be included in the regression analysis and not including Cmax.

10.1.1.2 Goodness of Fit

- If data permits, the default 'best fit' method by Phoenix WinNonlin will be used for the selection of slopes to determine 'λz'. If data do not permit, then the slope selection will be done by visual inspection or by a manual method to determine λz considering the best possible the adjusted R^2 value.
- When assessing terminal elimination phases, the adjusted R^2 value will be used as a measure of the goodness of fit of the data points to the determined line.
- Regression-based parameters (K_{el}, $t_{½}$ AUC_{0-inf}, CL/F and V_z/F ~~AUCinf, t1/2, CL, and Vz~~) will only be calculated if the adjusted R^2 value of the regression line is greater than or equal to 0.7. If regression-based parameters cannot be calculated, they will be set to "NC", defined as "not calculated" in the data listings and summary tables as appropriate.

10.1.1.3 ~~Period of Estimation~~

- ~~Time period used for the estimation of t1/2, where possible, will be over at least 2 half lives.~~
- ~~Where an elimination half-life is estimated over a time period of less than 2 half lives, it will be flagged in the data listings at the discretion of the pharmacokineticist, and the robustness of the half-life value and regression-based parameters (AUC0-inf, CL, and Vz) should be discussed in the study report.~~

10.1.1.4 Calculation of AUC

- The minimum requirement for the calculation of AUC will be the inclusion of at least 3 consecutive plasma concentrations above the lower limit of quantification (LLOQ), with at least one of these concentrations following C_{max}.
- All AUC values will be calculated using the 'linear up / log down' trapezoidal rule where the linear trapezoidal rule will be used for increasing concentrations and the log trapezoidal rule for decreasing concentrations.

Pharmacokinetic Noncompartmental Analysis (NCA)

10.1.2 General Plasma NCA Pharmacokinetic Settings

• NCA Model Type	Plasma (200-202)
• Weighting	Uniform
• AUC Calculation Method:	Linear Up/Log Down
• Dose Options Type	Extravascular
• Slopes Setting Fit Method:	Best Fit. If the data does not permit best fit selection, the slope selection will be performed by visual inspection or by a manual method to determine λ_z.
• Rsq_Adjusted Criteria for 'λz'	0.70
• AUC% extrapolation Criteria	20

10.3 Inferential Analyses

No formal hypothesis testing is planned

10.4 Ocular PK analysis

An undiluted vitreous sample will be collected at the time of vitrectomy to assess ONL1204 drug levels. ~~A separate summary and listing will be provided for the same~~. Considering the sampling limitations, no ocular PK analysis will be applicable for this study and hence concentration listings and summaries along with figures will be provided based on ocular PK population.

11. PHARMACODYNAMICS

Pharmacodynamic statistical analyses will be conducted based on the proteomic analysis of cytokine biomarkers in ocular fluids.

The following PD endpoints will be assessed:

- Vitreous Humor sample collected at pre-injection baseline by vitreous tap
- Vitreous Humor sample from the undiluted sample collected at the time of surgery
- Aqueous Humor sample collected at the time of surgery

Analyte concentrations will be provided for the vitreous humor and aqueous humor samples.

All PD endpoints will be analyzed separately by ONL Therapeutics.

12. EFFICACY

No formal efficacy analyses are planned for this study, however exploratory endpoints will be evaluated.

13. SAFETY

Safety endpoints (adverse events, clinical laboratory evaluations, vital signs, BCVA, ophthalmic imaging, intraocular pressure, and other safety assessments) will be analyzed using the safety population. Safety endpoints (continuous and categorical) will be summarized descriptively as described in section 3.1.

13.1.1 Adverse Events

All AEs will be coded using MedDRA. All AE summaries will be restricted to TEAEs only. Summary tables will include the number of participants (%) experiencing an event and the number of events. Participants will be counted only once for each SOC and PT level (categorical descriptive analysis).

Ocular and non-ocular AEs will be reported separately. Ocular AEs will be split into study eye/fellow eye/Both eye events.

The TEAE summaries will include:

- Overall summary of TEAEs.
- TEAE summary by SOC and PT.
- TEAE summary of serious adverse events (SAE) by SOC and PT.
- TEAE summary by SOC, PT and Severity.
- TEAE summary by SOC, PT, and relationship to study drug.
- TEAE summary by SOC, PT, and relationship to procedure.
- TEAE summary of events leading to the study drug removal by SOC and PT.

All listings will be presented by the safety population. Separate listings will be provided for patients with SAEs, AEs leading to withdrawal from the study and AEs resulting in death, if any.

13.1.2 Safety Laboratory Assessments

Blood and urine samples will be collected at the time points specified in the Schedule of Events (refer to the Protocol) to conduct CBC, renal function, and hepatic function analyses.

The following tests will be performed within each of the specified test panels:

Complete blood count (including):

- While Blood Cell (WBC) count with differential
- Hemoglobin (hgb)
- Hematocrit (hct)
- Platelet count

Renal function test (including):

- Albumin
- Blood Urea Nitrogen (BUN)
- Bicarbonate
- Calcium
- Chloride
- Creatinine
- eGFR

- Glucose
- Phosphorus
- Potassium
- Sodium

Hepatic Function tests (including):

- Alanine transaminase (ALT)
- Alkaline phosphatase (ALP)
- Albumin
- Total protein
- Aspartate transaminase (AST)
- Bilirubin
- Prothrombin time (PT)

All laboratory data collected at scheduled and unscheduled visits will be included in the listings, but only results collected at scheduled visits will be included in the summary tables.

Results for individual parameters may be reported in different units depending on the analyzing laboratory. If required, the results (and the corresponding normal range cut-off values) for individual parameters may be converted to International System of Units (S.I.) to summarize the data.

For all the parameters where a unit value has been reported, the parameter names that will be used in the outputs will comprise of the test name and the unit of measure, for example, 'Albumin (g/L)'. For the urinalysis parameters, the parameter name will be the reported test name only. Parameters will be sorted alphabetically within tables and listings.

For all parameters where a normal range limit value was reported, the normal range will be derived based on the available lower and upper limit values and any reported mathematical symbols. If both a lower and upper limit value is available, the normal range will be presented as '(Lower, Upper)'.

The reported results for each parameter with a defined normal range will be classified ('Low', 'Normal', 'High') in relation to the defined normal range limits. If a result is equal to the normal range cut-off value, the result will be considered 'Normal'.

The complete blood count, renal function test and hepatic function test result tables will present summary statistics for each laboratory parameter. For each parameter, summaries will be presented for the baseline and each scheduled post-baseline visit. In addition, summaries will be presented for the change from baseline values at each scheduled post-baseline visit (continuous descriptive analysis).

The decimal precision to which the summary for each parameter is reported will be based on the maximum number of decimals to which the reported result or the normal range limits are presented to in the raw data. The results and normal ranges will be displayed to the same decimal precision in the listings.

13.1.3 Vital Signs

The following vital signs measurements will be taken at the time points specified in the Schedule of Events (refer to the Protocol):

- Heart Rate (beats/min)
- Sitting Systolic Blood Pressure (SBP) (mmHg)
- Sitting Diastolic Blood Pressure (DBP) (mmHg)
- Respiratory Rate (breaths/min)
- Temperature (ºC)

All vital signs data collected at scheduled and unscheduled visits will be included in the listings, but only results collected at scheduled visits will be included in the summary tables.

The parameter names that will be used in the outputs will comprise of the test name and the unit of measure, for example, 'Systolic Blood Pressure (mmHg)'. Parameters will be sorted in the order that the measurements were collected in on the Vital Signs eCRF page within the tables and listings.

Vital signs measurements will present summary statistics for the results at the baseline and each scheduled post-baseline visit for each of the parameters. In addition, summaries will be presented for the change from baseline values at each scheduled post-baseline visit (continuous descriptive analysis).

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

13.1.4 Ophthalmologic Assessments

The following parameters will be assessed in study eye and contralateral eye for Safety Population.

13.1.5 Best Corrected Visual Acuity (BCVA)

BCVA will be performed in the study eye and fellow eye using the logMAR score. The logMAR score will be converted from the ETDRS visual acuity measure at time points specified in the Schedule of Assessments (refer to protocol) to assess visual acuity.

LogMAR (Logarithm of the minimal angle of resolution) score will be calculated using the formulae logMAR = -0.02 * ETDRS + 1.7

If the patient cannot read the ETDRS chart at 1 meter, the assessment result reported in counts fingers, hand motions, light perception, or no light perception will be utilized and converted to a logMAR score (CF = 2.0 logMAR, HM = 3.0 logMAR, LP = 3.9 logMAR, NLP = 4.0 logMAR). BCVA will be performed prior to procedures requiring pupil dilation.

LogMAR BCVA will be assessed and summarized as described in section 3.1 (continuous descriptive analysis) in study eye and fellow eye:

Summary of total letters read for each eye along with change from baseline will be presented by visit for both eyes (continuous descriptive analysis). All other assessments will be listed.

Low Luminance ETDRS BCVA and Low Contrast ETDRS BCVA will also be summarized as continuous variables only.

13.1.6 Color Fundus Photography (CFP)

CFP will be performed in study eye and contralateral eye at the time points specified in the Schedule of Assessments (refer to protocol).

Separate listing will be presented for the same.

13.1.7 Fundus Autofluorescence

Fundus Autofluorescence will be performed at Visit 8 in both eyes. Listing will be presented for the same.

13.1.8 Indirect Ophthalmoscopy

Ophthalmic Exam will be performed via indirect ophthalmoscopy at baseline and at other times according to the Schedule of Assessments (please refer to Protocol).

The following parameters will be assessed and summarized as described in section 3.1 (continuous/categorical descriptive analysis) in study eye and fellow eye:

- Posterior Vitreous Detachment
- C/D Ratio
- Macula
- Vessels
- Periphery

All grading assessments of different eye structures will be summarized descriptively by abnormalities, cohort and study visit for both eyes (categorical descriptive analysis as mentioned in section 3.1).

All data recorded on the eCRF page 'Indirect Ophthalmoscopy (Retinal Exam)' will be listed.

In addition, separate by-patients listing will be provided for pre- and post-surgery of the retinal examination.

13.1.9 Slit Lamp Biomicroscopy

Biomicroscopy will be performed with a slit-lamp to examine the anterior and posterior chambers of the eye. The examination will include following assessments as per Schedule of Events please refer to the Protocol).

- Anterior Segment
    - Anterior Chamber Cell
    - Anterior Chamber Flare
- Posterior Segment
    - Optic Disc
    - Vitreous Cell
    - Vitreous Haze
- Lens Status
    - Nuclear Sclerosis
    - Cortical Cataract
    - Posterior subcapsular cataract

All grading assessments of different eye structures will be summarized descriptively by abnormalities, cohort and study visit for both eyes (categorical descriptive analysis as mentioned in section 3.1).

Listing will be provided for the same.

13.1.10 Intraocular Pressure (IOP)

IOP measurements will be done using applanation tonometry (either Goldmann (standard preferred method) or Tonopen® (use only if required)). For any given participant, the same IOP procedures will be followed for the entire duration of the study. Below measurement will be taken for both eyes.

- IOP (mmHg)

Summary of IOP measured for both eyes will be presented as per visit.

13.1.11 Pin Hole Visual Acuity

Pin Hole Visual Acuity measurements (taken at 1 Day Post-Surgery only) will be summarized using descriptive statistics described in section 3.1 for continuous data by cohort by LogMAR Score.

13.1.12 Contrast Sensitivity Testing

Contrast Sensitivity Testing measurements will be measured at the time points specified in the Schedule of Assessments (refer to protocol). Scores will be summarized for both the eyes. Results will be reported in log values.

13.1.13 Farnsworth D15 Color vision test

Farnsworth D15 color vision measurements will be assessed in both study eye and fellow eye. Separate listing will be provided.

13.1.14 Spectral Domain Optical Coherence Tomography (SD-OCT)

SD-OCT will be performed in study eye and contralateral eye at the time points specified in the Schedule of Assessments (refer to protocol). Measurements of retinal thicknesses and volumes based on determinations relying on typical retinal layers as identified by SD-OCT will be calculated. Specifically, the following results will be presented from all assessments except for the pre-injection baseline measurement. The results for the parameters below will be summarized as described in section 3.1 (continuous descriptive analysis):

- Average thickness central 1 mm (central subfield thickness)
- Average thickness central 3 mm
- Average thickness central 6 mm
- Total centerpoint thickness
- Central subfield volume
- Macular volume
- Subretinal fluid thickness at the foveal center

The following parameters will be summarized as described in section 3.1 (categorical descriptive analysis) for the collected categories with each parameter:

- Vitreomacular traction deforming the fovea
- Intraretinal fluid (cystoid macular edema)
- Epiretinal membrane
- Epiretinal membrane with deformation in the center 1 mm
- Anomaly

The results from the analyses of the parameters listed in this section above will be prepared by ONL and presented as an addendum to the tables, listings, and figures report.

The following parameters with continuous numerical results will also be obtained from all assessments except for the Screening measurement. These results may be analyzed and reviewed, with or without descriptive statistics, internally. The results for the following parameters will be available for the central 1 mm, 3 mm, and 6 mm of the scan, as well as the volume of the central 1 mm region.

- Boundaries ILM to inner aspect of ONL
- Boundaries inner aspect of ONL to Bruch's Membrane
- Boundaries ELM to Bruch's Membrane

The following parameters with categorical results will also be obtained from all assessments except for the Screening measurement. These results may be analyzed and reviewed, with or without categorical descriptive statistics, internally.

- Intraretinal fluid in the central subfield
- Intraretinal fluid at the foveal center
- Subretinal fluid in the center 3 mm
- Subretinal fluid in the center 1 mm
- Subretinal fluid at the foveal center
- Anomaly details
    - Retinal pigment epithelium (RPE) elevation, RPE atrophy, outer retinal lucency
    - Potential risk management
    - Other

The results from the analyses of the parameters identified in this section above will be included in the addendum to the tables, listings, and figures report only if deemed pertinent and potentially informative by expert review.

Pregnancy Test Results

All information related to pregnancy testing (urine) and contraception status will be presented in the by-participant data listings.

14. IMMUNOGENICITY

Not Applicable.

15. CHANGES TO THE PLANNED ANALYSIS

Not Applicable

16. FINAL ANALYSIS (END OF STUDY)

Final Analysis

The final analysis will be conducted after all participants have completed the study, the clinical database has been locked and the analysis populations have been approved.

The final analysis will be based on the final version of the SAP. Any deviations from the planned analysis will be documented in the CSR.

17. SOFTWARE

- SAS® Version 9.2 or higher (SAS Institute, Cary, North Carolina, USA).
- Phoenix WinNonlin® (Certara, USA) Version 8.3 or higher.

18. TABLES

19. LISTINGS

Listing Number	Listing Title	Population
16.2.1	**Subject Disposition**	
16.2.1.1	*Analysis Populations*	*ITT*
16.2.1.2	*Subject Disposition*	*ITT*
16.2.1.3	*Screen Failures*	*SF*
16.2.2	**Protocol Deviations**	
16.2.2.1	*Protocol Deviations*	*ITT*
16.2.4	**Demographic and Other Baseline Data**	
16.2.4.1	*Demographics and Baseline Characteristics*	*ITT*
16.2.4.2	*Medical History*	*ITT*
16.2.4.3	*Duration of Macular Detachment*	*ITT*
16.2.4.4	*Inclusion/Exclusion Criteria*	*ITT*
16.2.4.5	*B Scan Ultrasonography*	*ITT*
16.2.5	**Treatment Administration**	
16.2.5.1	*Study Drug Administration*	*ITT*
16.2.6	**PK/PD**	
16.2.6.1	*Plasma ONL1204 Concentrations*	*Safety*
16.2.6.2	*Plasma ONL1204 Pharmacokinetic Parameters*	*PK*
16.2.7	**Adverse Events**	
16.2.7.1	*Adverse Events*	*ITT*
16.2.7.2	*Serious Adverse Events*	*ITT*
16.2.7.3	*Adverse Events Leading to Study Drug Removal*	*ITT*
16.2.7.4	*Adverse Events with Outcome of Death*	*ITT*
16.2.8	**Laboratory Parameters**	
16.2.8.1	*Hematology Results*	*ITT*
16.2.8.2	*Chemistry Results*	*ITT*
16.2.9	**Other Safety**	

Listing Number	Listing Title	Population
16.2.9.1	*Vital Signs*	*ITT*
16.2.10	**Prior & Concomitant Medication**	*ITT*
16.2.10.1	*Prior and Concomitant Medication*	*ITT*
16.2.11	**Ophthalmic Assessments**	*ITT*
16.2.11.1	*Best Corrected Visual Acuity (BCVA) (Letters)*	*ITT*
16.2.11.2	*LogMAR Values*	*ITT*
16.2.11.3	*Low Luminance and Low Contrast (BCVA)*	*ITT*
16.2.11.4	*Color Fundus Photography (CFP)*	*ITT*
16.2.11.5	*Fundus Autofluorescence*	*ITT*
16.2.11.6	*Indirect Ophthalmoscopy Retinal Exam*	*ITT*
16.2.11.7	*Pre surgical Retinal Examination*	*ITT*
16.2.11.8	*Post Surgery of Retinal Exam (Study Eye)*	*ITT*
16.2.11.9	*Biomicroscopy Examination*	*ITT*
16.2.11.10	*Intraocular Pressure*	*ITT*
16.2.11.11	*Pin Hole Visual Acuity*	*ITT*
16.2.11.12	*Contrast Sensitivity Testing*	*ITT*
16.2.11.13	*Farnsworth D15 color vision test*	*ITT*

21. REFERENCES

Clinical Study Protocol Version 1.6 dated 27 July 2020